

# **Diversity and Management of *Phytophthora* in Southeast Asia**

**Editors: André Drenth and David I. Guest**

Australian Centre for International Agricultural Research  
Canberra 2004

The Australian Centre for International Agricultural Research (ACIAR) was established in June 1982 by an Act of the Australian Parliament. Its mandate is to help identify agricultural problems in developing countries and to commission collaborative research between Australian and developing country researchers in fields where Australia has a special research competence.

Where trade names are used this constitutes neither endorsement of nor discrimination against any product by the Centre.

ACIAR MONOGRAPH SERIES

This peer-reviewed series contains the results of original research supported by ACIAR, or material deemed relevant to ACIAR's research objectives. The series is distributed internationally, with an emphasis on developing countries.

© Australian Centre for International Agricultural Research,  
GPO Box 1571, Canberra, ACT 2601, Australia

Drenth, A. and Guest, D.I., ed. 2004. Diversity and management of *Phytophthora* in Southeast Asia.  
ACIAR Monograph No. 114, 238p.

ISBN 1 86320 405 9 (print)  
1 86320 406 7 (online)

Technical editing, design and layout: Clarus Design, Canberra, Australia

Printing: BPA Print Group Pty Ltd, Melbourne, Australia

# Foreword

The genus *Phytophthora* is one of the most important plant pathogens worldwide, and many economically important crop species in Southeast Asia, such as rubber, cocoa, durian, jackfruit, papaya, taro, coconut, pepper, potato, plantation forestry, and citrus are susceptible.

Although many plant pathologists and agronomists have been aware of the economic importance of phytophthora diseases in Southeast Asia, there is a lack of information on general aspects of *Phytophthora* species in the tropics. Numerous studies have been conducted over the past few decades but the general background information is often not outlined in detail, while specific information on the occurrence and economic impact of phytophthora disease is scattered in many different publications in a range of languages. There has never been a comprehensive compilation of which species appear where, on which hosts, or what economic impact phytophthora diseases have in the region. This publication attempts to consolidate this information.

By bringing together information on the identification of phytophthora diseases based on symptoms, their occurrence, economic impact and development of integrated disease management practices, the authors of this volume provide practical information to those who seek to limit the damage caused by phytophthora diseases.

The authors have also recognised the need for a comprehensive overview of all aspects involved in the development of integrated management strategies for phytophthora diseases. The authors provide practical information, advice and background information in such a way that a reader with a basic agronomic background is able to use this information to design and implement effective integrated disease management strategies for different phytophthora diseases in different parts of the world.

The book results from a workshop held in Chiang Mai, Thailand in November 2002 with the support of ACIAR and the ATSE Crawford Fund. The workshop was the outcome of two ACIAR projects 'A survey of the presence and importance of *Phytophthora* in Southeast Asia', led by Dr André Drenth of the Cooperative Research Centre for Tropical Plant Protection, Brisbane and 'Management of *Phytophthora* diseases of durian' led by Dr David Guest of the University of Melbourne, Dr Somsiri Sangchote of Kasetsart University, Thailand and Dr Nguyen Minh Chau of the Southern Fruit Research Institute in Vietnam.

The workshop was also part of the First International Conference on Tropical and Subtropical Plant Diseases, organised by the Thai Phytopathological Society.

This publication is the latest in ACIAR's monograph series and is also available from our website at <[www.aciar.gov.au](http://www.aciar.gov.au)>.



Peter Core  
Director  
Australian Centre for  
International Agricultural  
Research

# Contents

Foreword	3
<b>1 Introduction</b>	<b>7</b>
<i>André Drenth and David I. Guest</i>	
<b>2 Economic Impact of Phytophthora Diseases in Southeast Asia</b>	<b>10</b>
<i>André Drenth and Barbara Sendall</i>	
<b>3 Biology of <i>Phytophthora</i></b>	<b>29</b>
3.1 <i>Phytophthora</i> in the Tropics	30
<i>André Drenth and David I. Guest</i>	
3.2 Infection Biology of <i>Phytophthora palmivora</i> Butl. in <i>Durio zibethinus</i> L. (Durian) and Responses Induced by Phosphonate	42
<i>Emer O'Gara, Somsiri Sangchote, Laura Fitzgerald, Damon Wood, Ang Ching Seng and David I. Guest</i>	
3.3 Morphological and host range variability in <i>Phytophthora palmivora</i> from durian in Thailand	53
<i>R. Pongpisutta and S. Sangchote</i>	
<b>4 Occurrence of <i>Phytophthora</i> in Southeast Asia</b>	<b>59</b>
4.1 <i>Phytophthora</i> Diseases in Malaysia	60
<i>B.S. Lee and K.Y. Lum</i>	
4.2 <i>Phytophthora</i> Diseases in Indonesia	70
<i>Agus Purwantara, Dyah Manohara and J. Sony Warokka</i>	
4.3 <i>Phytophthora</i> Diseases in Thailand	77
<i>Somsiri Sangchote, Srisuk Poonpolgul, R. Sdoodee, M. Kanjanamaneesathian, T. Baothong and Pipob Lumyong</i>	
4.4 <i>Phytophthora</i> Diseases in Vietnam	83
<i>Dang Vu Thi Thanh, Ngo Vinh Vien and André Drenth</i>	
4.5 <i>Phytophthora</i> Diseases in the Philippines	90
<i>L.A. Portales</i>	
<b>5 Isolation of <i>Phytophthora</i> from Infected Plant Tissue and Soil, and Principles of Species Identification</b>	<b>94</b>
<i>André Drenth and Barbara Sendall</i>	
<b>6 Major Crops Affected by <i>Phytophthora</i></b>	<b>103</b>
6.1 <i>Phytophthora</i> on Cocoa	104
<i>Peter McMahon and Agus Purwantara</i>	
6.2 <i>Phytophthora</i> Diseases of Coconut in the Philippines	116
<i>Erlene Concibido-Manohar</i>	
6.3 Distribution and Progression of <i>Phytophthora</i> Bud Rot Disease of Coconut in Selected Areas in the Philippines	124
<i>Nemesia San Juan-Bachiller</i>	

6.4	<i>Phytophthora capsici</i> on Black Pepper in Indonesia <i>D. Manohara, K. Mulya, A. Purwantara and D. Wahyuno</i>	132
6.5	Phytophthora Diseases of Rubber <i>Ratana Sdoodee</i>	136
6.6	Phytophthora Diseases of Durian, and Durian-Decline Syndrome in Northern Queensland, Australia <i>Emer O’Gara, David I. Guest, Lynton Vawdrey, Peter Langdon and Yan Diczbalis</i>	143
<b>7</b>	<b>Managing Phytophthora Diseases</b>	<b>153</b>
7.1	Principles of Phytophthora Disease Management <i>André Drenth and David I. Guest</i>	154
7.2	Nursery Practices and Orchard Management <i>David I. Guest</i>	161
7.3	The Use of Mounds and Organic and Plastic Mulches for the Management of Phytophthora Root Rot of Papaya in Northern Queensland <i>L.L. Vawdrey, K.E. Grice and R.A. Peterson</i>	167
7.4	Root Infusion of Phosphorous Acid for the Control of Phytophthora Foot Rot in Black Pepper ( <i>Piper nigrum</i> L.) <i>Mee-Hua Wong</i>	171
7.5	Biological Control of Black Pod Disease on Cocoa in Malaysia <i>M.J. Ahmad Kamil, S. Shari Fuddin and C.L. Bong</i>	174
<b>8</b>	<b>Phytophthora in Durian</b>	<b>179</b>
8.1	Botany and Production of Durian ( <i>Durio zibethinus</i> ) in Southeast Asia <i>Emer O’Gara, David I. Guest and Nik Masdek Hassan</i>	180
8.2	Occurrence, Distribution and Utilisation of Durian Germplasm <i>Emer O’Gara, David I. Guest and Nik Masdek Hassan</i>	187
8.3	Screening for Resistance to <i>Phytophthora</i> <i>Emer O’Gara, Lynton Vawdrey, Tania Martin, Somsiri Sangchote, Huynh van Thanh, Le Ngoc Binh and David I. Guest</i>	194
8.4	Durian Propagation and Nursery Practice <i>Nguyen Minh Chau, Huynh Van Tan, Yan Diczbalis and David I. Guest</i>	200
8.5	Durian Tree Phenology and the Control of Phytophthora Diseases of Durian Using Phosphonate Trunk Injection <i>Y. Diczbalis, L. Vawdrey, G. Alvero, D. Campagnolo, Huynh Van Thanh, Mai Van Tri, L.N. Binh, N.T.T. Binh, H.V. Tan, Nguyen Minh Chau, Emer O’Gara and David I. Guest</i>	206
8.6	Control of Postharvest Diseases in Durian <i>Do Minh Hien, Huynh Van Thanh, Phan Quang Danh and Emer O’Gara</i>	217
8.7	Integrated Management of Phytophthora Diseases of Durian: Recommendations and Benefit–Cost Analysis <i>David I. Guest, Nguyen Minh Chau, Somsiri Sangchote, Lynton Vawdrey and Yan Diczbalis</i>	222
<b>9</b>	<b>Conclusions and a Vision for Future Research Priorities</b> <i>André Drenth and David I. Guest</i>	<b>227</b>
	<b>Appendix: Table of <i>Phytophthora</i> pathogens and hosts in Southeast Asia</b>	<b>233</b>

# I Introduction

André Drenth<sup>1</sup> and David I. Guest<sup>2</sup>

There are about 60 species in the genus *Phytophthora*, all of them plant pathogens. *Phytophthora*, the 'plant destroyer', is one of the most destructive genera of plant pathogens in temperate and tropical regions, causing annual damages of billions of dollars.

Phytophthora diseases have been well studied in the temperate regions of the world, ever since the potato late blight epidemic in Europe in 1845–47 provided the impetus for the development of plant pathology as a scientific discipline. Throughout the wet tropics, agricultural production of a large range of crops is seriously reduced due to the wide range of *Phytophthora* pathogens causing a large number of different diseases. This chapter will explore the reasons why phytophthora diseases are so devastating in the wet tropics.

There are a number of host and pathogen factors which, together with features of their interactions, make phytophthora diseases so troublesome in the wet tropics. One of the important factors to consider is that the genus *Phytophthora* does not belong to the fungal kingdom. It is an Oomycete, closely related to diatoms, kelps and golden brown algae in the Kingdom Stramenopila (Beakes 1998). These organisms thrive in the environments found commonly in the wet tropics. There are a number of additional reasons why phytophthora diseases cause so much damage in the tropics. We have grouped these into pathogen, host, environmental and agronomic factors in Table 1.1.

All *Phytophthora* species need high humidity for sporulation and the germination of sporangiospores and zoospores to initiate infections. Frequent or seasonal heavy rainfall, and high levels of humidity, are common throughout the tropical lowlands. Tropical highlands have the added problem of

heavy mist and dew during the morning and/or late afternoon, producing free water throughout the night and providing almost daily opportunities for sporangiospores to be formed, transported and start new infections.

Another important factor in the pathogenicity of *Phytophthora* is that sporangia release motile zoospores that are attracted by chemotaxis (Carlile 1983) and electrotaxis (Morris and Gow 1993) to the roots of their host plants. The ability to seek out susceptible host tissue, coupled with zoospore motility, makes these propagules extremely efficient, even at low numbers.

Another characteristic of *Phytophthora* species, and *P. palmivora* in particular, is their ability to cause multiple diseases on the same host. In this monograph, two examples discussed in detail are cocoa and durian, and while the symptoms expressed on each host are not independent of each other, they demonstrate how numerous interactions form complex disease cycles. On cocoa, *P. palmivora* causes seedling dieback, root rot, stem canker, chupon wilt, leaf blight, cherelle wilt and black pod rot. Thus, both inoculum and susceptible host tissue are continuously available, and the disease potential is always present. These factors make *P. palmivora* an important pathogen of cocoa, and demand an integrated disease management approach.

In addition to causing multiple diseases on the same host, *P. palmivora* can also attack a wide range of different host species that are widespread and/or cultivated throughout the tropics. An appendix to this monograph tabulates *Phytophthora* pathogens and their hosts in Southeast Asia. This array of potential hosts increases the amount of inoculum and the resulting disease pressure. Furthermore, the large number of perennial host crops grown in the tropics means that susceptible host material is available all year round. Consequently, there is rarely, if ever, a break in the disease cycle. Infected plant material continuously produces large numbers of sporangia that have the ability to spread and infect new host-plant material.

<sup>1</sup> CRC for Tropical Plant Protection, Indooroopilly Research Centre, 80 Meiers Road, Indooroopilly, Queensland 4068, Australia.

<sup>2</sup> Department of Botany, The University of Melbourne, Parkville, Victoria 3010, Australia.  
Current address: Faculty of Agriculture, Food and Natural Resources, The University of Sydney, NSW 2006, Australia

Breeding for host resistance to *Phytophthora* has given mixed results. In annual crops like soybean and potatoes, breeding started for resistance that led to the selection of specific R-genes that in some cases were quickly overcome by virulent races of the pathogen. The use of race-specific R-genes led, in some cases, to boom–bust cycles, which subsequently shifted the emphasis in resistance breeding to increasing levels of non-specific resistance, which do not completely stop infection and colonisation but slow down the rate of spread of an epidemic. Although this has led to considerable success for many annual row crops, the selection for non-specific resistance in perennial tree crops is still in its infancy, and requires a serious long-term commitment. The range of diseases caused by one *Phytophthora* pathogen further complicates breeding for non-specific resistance. Screening for resistance on leaf discs and cocoa pods may not necessarily give high levels of resistance to chupon wilt and tree cankers. Without a more complete understanding of disease cycles of the pathogen and various expressions of disease resistance in different tissue of the host plant, it is difficult to make significant steps forward by focusing on isolated aspects of phytophthora diseases.

Many agronomic practices that improve production give rise to higher levels of susceptibility, disease severity and impact in the presence of the pathogen. Flood irrigation, high levels of nitrogen fertilisers, quick-growing varieties of plants, monocultures with limited genetic diversity and high orchard density are part of modern agriculture, but these features also make these agricultural systems extremely vulnerable to phytophthora diseases. It is important to seek a broader approach in agricultural production and take account of the multitude of correlated factors in an integrated manner in order to lift production and profitability. Considering the prevalence and host range of *P. palmivora*, which can cause diseases in a large range of different host species of economic importance in the tropics, disease management efforts must move beyond controlling specific diseases on a single host and consider the whole agricultural production system. Issues like intercropping with hosts susceptible to the same *Phytophthora* pathogen need to be studied in more detail. While it may seem self-evident that interplanting susceptible hosts should increase disease severity, there are no data demonstrating that mixed farming is more vulnerable to epiphytotics caused by *Phytophthora* than are monocultures. The truth may not be so simple.

**Table 1.1** Characteristics that make *Phytophthora* species so successful as pathogens in the tropics.

Environment	Pathogen	Host	Agronomic practices
<ul style="list-style-type: none"> <li>• High rainfall</li> <li>• High humidity</li> <li>• Suitable temperature</li> <li>• Host plants available all year round</li> </ul>	<ul style="list-style-type: none"> <li>• Spread in air and/or water</li> <li>• Short generation time</li> <li>• Rapid multiplication of inoculum</li> <li>• Motile zoospores</li> <li>• Zoospores attracted to their host by electro taxis and chemotaxis</li> <li>• Chlamydospores and oospores for survival outside host</li> <li>• Wide host range, e.g. <i>P. palmivora</i></li> <li>• Disease cycle driven by free water and high humidity</li> </ul>	<ul style="list-style-type: none"> <li>• Perennial host crops – host tissue present all year round</li> <li>• Multiple diseases caused by the same <i>Phytophthora</i> species in different tissues of the same host</li> <li>• Multiple host susceptibility to same <i>Phytophthora</i> pathogen</li> <li>• Lack of resistance in many hosts</li> <li>• Abundance of insect vectors</li> <li>• Stem, root borers and nematodes provide entry points for infection</li> </ul>	<ul style="list-style-type: none"> <li>• Over-use of and/or inappropriate irrigation</li> <li>• Poor drainage creates ponding</li> <li>• Irrigation with <i>Phytophthora</i>-infected water</li> <li>• Orchard established on infested soil</li> <li>• Susceptible planting materials</li> <li>• Monoculture of susceptible species</li> <li>• Narrow spacing of trees in orchard</li> <li>• No break in crop cycle</li> <li>• Shading practices that increase humidity</li> <li>• Emphasis on selection and breeding for rapid growth and high yield with little resistance</li> <li>• Failure of chemical control in high rainfall areas</li> <li>• High-nitrogen inorganic fertilisers</li> </ul>

Due to the nature of the two ACIAR projects, the first part of this monograph focuses on the *Phytophthora* pathogens present in Southeast Asia, their hosts, general biology and economics as an output of ACIAR project PHT/1996/153 (Survey and importance of *Phytophthora* in Southeast Asia). The second part of the monograph is focused on the development of integrated disease management of *Phytophthora* on durian, 'the king of fruit', as an output of ACIAR project PHT/1995/134 (Management of *Phytophthora* diseases in durian). Some of the methods described in this monograph have been implemented and have already made a significant contribution to reducing losses due to phytophthora disease in durian. There is significant scope for further implementation of the integrated disease management practices developed — on a much larger geographic scale in durian, and similar approaches could be trialled for a number of other crops, for which a range of methods are also discussed in this monograph. Our ACIAR projects show that, for this to happen, further improvements in existing technologies, and strengthening of extension networks and training, are needed throughout Southeast Asia.

Several other crops, in addition to those described in this monograph, are under serious threat by *Phytophthora* and need a similar scientific input in an effort to reduce disease losses. We hope that this book will provide the nucleus for this effort and become a valuable resource to researchers throughout the region and beyond. We hope that by producing this monograph we will eventually help smallholders throughout the tropics reduce their losses due to phytophthora diseases.

## References

- Beakes, G.W. 1998. Evolutionary relationship among protozoa. In: Coombs, G.H., Vickerman, K., Sleight, M.A. and Warren, A., ed., The Systematics Association Special Volume Series 56. Dordrecht, Netherlands, Kluwer Academic Publishers.
- Carlile, M.J. 1983. Motility, taxis and tropisms in *Phytophthora*. In: Erwin, D.C., Bartnicki-Garcia, S. and Tsao, P.H., ed., *Phytophthora: its biology, taxonomy, ecology and pathology*. St Paul, Minnesota, USA, APS Press, 95–107.
- Morris, P.F. and Gow, N.A.R. 1993. Mechanism of electro taxis of zoospores of phytopathogenic fungi. *Phytopathology*, 83, 877–882.



A young Australian boy enjoys durian and Vegemite™—symbolic of the links forged between Asia and Australia through ACIAR collaboration.



## 2 Economic Impact of Phytophthora Diseases in Southeast Asia

André Drenth and Barbara Sendall<sup>1</sup>

### Abstract

A number of important crops grown in Southeast Asia, such as cocoa, durian, rubber, coconut, pepper, potato and citrus, are susceptible to different species of *Phytophthora*. In this chapter, we give some background on a range of crops troubled by phytophthora and discuss the economic impact of phytophthora diseases in the region. Our assessment indicates that the economic damage on the seven crops above in the five Southeast Asian countries may be as high as 2.3 billion US dollars annually.

### Introduction

Many plants grown for food and fibre suffer from a range of pest and diseases. This lowers production, increases the risk of crop failure, threatens food security and reduces the profitability of agricultural enterprises. Crop production is subject to variations in the natural environment, most notably rainfall and temperature. The complex biological and chemical interactions between the crop, mineral nutrients, and the weather give rise to considerable differences in yield and quality between seasons. The presence of diseases not only requires management inputs that reduce the profitability of crop production, but also significantly increases the risk of crop failure. The presence of diseases and pests that have the ability to significantly reduce the quality and quantity of agricultural crops is superimposed on the seasonal variability of these production factors. Thus, pests and diseases lower production, reduce product quality, increase management costs, and increase the risk of crop failure. In addition, chemical control measures may have negative collateral impacts on human health and the environment.

In order to determine the economic impact of phytophthora in Southeast Asia, the background crop production figures and an estimate of the crop

value are given for each country. Although disease losses vary enormously between different regions, seasons, different plant varieties, and under different management practices, we have tried to estimate the average crop losses experienced. The economic impact we report on is a combination of disease losses experienced on average.

In order to reduce losses due to phytophthora, disease management practices are needed. Diseases and pests can be managed in a number of ways, such as the use of resistant varieties, removal of infected plant material, pruning, tree injection, improving soil health, and application of chemicals. Each management practice imposes direct and indirect costs on the grower.

Plant pathologists need to have good tools for the assessment of disease incidence, disease severity and disease impact. These tools enable a reliable assessment of:

- the presence of the disease
- economic losses due to disease
- relative disease losses in different varieties
- field experiments comparing different disease management options
- cost-effectiveness of disease management options that improve the profitability of crop production.

Although a large range of disease assessment tools is available for our target crops, they have not been used routinely. Because of the lack of robust data, many

<sup>1</sup> CRC for Tropical Plant Protection, Indooroopilly Research Centre, 80 Meiers Road, Indooroopilly, Queensland 4068, Australia.

previous assessments of the overall disease impact in the region have been based on educated guesses.

The aim of this chapter is to (i) establish the importance and economic value of target crops in Southeast Asia, (ii) describe the importance of phytophthora diseases on these crops, and (iii) provide an overall assessment of the economic impact of phytophthora in Southeast Asia.

## Cocoa

Cocoa (cacao), *Theobroma cacao*, is native to the central and western Amazon region of South America. The Mayas, Toltecs and Aztecs cultivated cocoa more than 3000 years ago (Pereira 1992). Cocoa plants were introduced to Southeast Asia via the Philippines in the 1760s (Blaaha 1992). Cocoa is now produced by small landholders and plantations across the humid lowland tropics in Africa, Asia and the Americas (Smith et al. 1992). The major producers of cocoa are the Côte d'Ivoire (Ivory Coast), Ghana, Indonesia, Malaysia, Brazil and Papua New Guinea (Table 2.1).

### Indonesia

Cocoa cultivation in Asia started in Indonesia in 1779 when the Batavian Society of Arts and Sciences offered an award to the first person to plant at least 50 cocoa trees (Blaaha 1992). Cocoa has been produced on a larger scale in East Java and North Sumatra since the 1940s, with plantings covering around 6500 ha and producing about 2000 t of dry beans annually (ICCO 1998). Production was dominated by large estates or plantations, which produced high (fine) quality cocoa. Since most of the estates were planted on Java, the exported cocoa was referred to as 'Java cocoa'. During the 1970s, the area planted to cocoa increased rapidly, and within 15 years, the number of hectares planted had tripled. Importantly, smallholder plantings increased and, by 1986, comprised 58% of the total area planted (Effendi 1992). During this period of rapid expansion, Sulawesi, Kalimantan and Sumatra joined Java as production centres (ICCO 1998).

Sulawesi, with 400,000 smallholder cocoa growers, now produces 300,000 t of dry beans per annum.

The growth has been assisted by a free economy combined with government grants to buy land, low production costs and application of management practices used in plantations in Malaysia (ICCO 1998). The current prospects for growing cocoa in Indonesia are considered good, both in terms of exports and production for local consumption. Indonesia has one of the best performances among major producing countries in terms of average yields, achieving close to 1 t/ha/year; most other producing countries have substantially lower average yields (ICCO 1998). There has been a shift in production from fine cocoa to unfermented (bulk) cocoa, due to more favourable prices for the latter. In addition, the production costs for bulk cocoa are much lower than that for fine cocoa (Effendi 1992). Pod rot and stem canker caused by *Phytophthora palmivora* often cause severe losses in Indonesia. Infestation with *P. palmivora* has been reported to be heavy in Maluku, while it is sporadically found in all provinces where cocoa is grown, especially in humid environments (Soehardjan 1992).

### Malaysia

Although cocoa cultivation was first reported during the 1770s in Peninsular Malaysia, widespread cultivation of cocoa did not begin until after the Second World War. The first commercial plantings were established in the 1950s in Jerangau, Peninsular Malaysia and Sabah. The introduction of new hybrids led to a rapid expansion in cocoa cultivation. The State of Sabah is the major producer of cocoa, accounting for 70% of national production. Most of the cocoa is exported as cocoa beans, while some is processed into primary products such as cocoa butter and cocoa powder before export. The majority of the cocoa products produced in Sabah are sent to Peninsular Malaysia for processing into value-added products such as chocolate and chocolate-based products (Sabah Government 2001a).

There has been a trend away from estate production of cocoa in Malaysia over the last 20 years. In 1980,

**Table 2.1** Production of cocoa in selected countries (FAO 2003).

Country	Area planted (ha)	Production (t of dry beans)	Export value (USD '000)
Indonesia	490,000	426,000	788,952
Malaysia	48,000	47,661	88,268
Philippines	12,000	6,000	11,112
Thailand	800	400	741
Vietnam	na	na	na

na = data not available.

63% of the cocoa produced was grown on estates, while in 2000, only 30% of the total crop was produced on estates; 70% was produced by small landholders. The smallholder achieves lower yields than the estates, which generally have more suitable land and greater resources. In Malaysia in 1990, the average yield for a smallholder was 610 kg/ha while the estates averaged 1100 kg/ha. Production in Malaysia peaked in 1989/90 at 243,000 t (ICCO 1998). Production has declined since 1990 however, and in 2000, Malaysia produced only 98,000 t of dry cocoa beans (FAO 2001a). Malaysia has now become an importer of cocoa beans (FME 2001a). The decline is attributed to low world prices, which caused farmers to abandon cocoa and turn to more profitable crops such as oil palm. In addition, damage due to the cocoa pod borer moth (*Conopomorpha cramerella*), labour shortages, and government incentives to grow other crops caused some growers to diversify out of cocoa. Neglect of plantations has led to pests and diseases becoming a serious problem (ICCO 1998).

### Philippines

Cocoa was introduced in the Philippines in 1670, and it was the first country in Asia to plant cacao and consume chocolate drinks prepared from cocoa beans. Commercial cocoa farms were planted in the mid-1950s, and the industry expanded further in the 1960s as processing facilities were constructed. In the mid-1980s, the industry expanded further still due to investment in commercial farms and grinding and processing facilities. Southern Mindanao is the largest producing region, contributing to approximately 72% of the total production for the Philippines.

Historically, Malaysia purchased most of the cocoa beans exported, while the majority of the cocoa powder and cocoa paste were shipped to Korea. Now, the United States of America (USA) is the Philippines' major market for cocoa butter while India is the sole market for Philippine cocoa paste/cocoa cake. In 1998, the Philippines imported more than 50% of its requirement for cocoa beans, the majority coming from Indonesia (DA-AMAS 1999).

### Vietnam

The Vietnamese government plans to make cocoa an important crop. Vietnam has land that is suitable to grow cocoa in the south and centre of the country and low labour costs compared with countries such as Malaysia. In the Central Highlands of Vietnam, 1500–2000 ha of cocoa will be planted annually, with the target of having planted 10,000 hectares by 2006–07. It is believed that Vietnam could be exporting cocoa by as early as 2005. Vietnam could become a significant cocoa producer in Asia by 2010 (FME 2001a).

### Economic importance of phytophthora diseases in cocoa

*Phytophthora* spp. infect the flowers, cherelles, pods, roots, stems, and leaves of cocoa plants (Thurston 1984). Black pod caused by *Phytophthora* spp. is the most destructive disease of cocoa worldwide, causing estimated losses in production in Asia, Africa and Brazil of 450,000 t annually, worth an estimated value of USD423 million. Annual crop losses may range from 30–90% (Bowers et al. 2001). The impact of the disease varies from country to country. Black pod rot occurs in almost all cocoa-producing countries, with worldwide losses estimated at 10% (Padwick 1956). Direct crop losses of up to 90% occur in wetter areas such as Nigeria (Gregory and Maddison 1981). Pod rot and stem canker caused by *P. palmivora* often cause severe losses in Indonesia. Infestation with *P. palmivora* has been reported to be heavy in Maluku, while it is sporadically found in cocoa estates with humid environments (Soehardjan 1992). A long-term field trial over a period of 10 years at Keravat in Papua New Guinea showed a mean pod loss of 17%, with a range of 5–39% (Holderness 1992). Outbreaks of black pod disease can be so severe that cocoa plantings must be abandoned. Black pod rot is attributed to four species of *Phytophthora*: *P. palmivora*, *P. capsici*, *P. citrophthora* and *P. megakarya*. The relative impact of each of these species of *Phytophthora* varies from region to region. In Southeast Asia, *P. palmivora* seems to be the principal pathogen, while *P. megakarya* has only been found in West Africa (Brasier et al. 1981). In Africa, *P. megakarya* tends to be the principal pathogen, while in the Americas, *P. capsici* and *P. citrophthora* are the main causal agents of pod rot (Erwin and Ribeiro 1996).

*Phytophthora palmivora* also causes stem canker and chupon wilt of cocoa. The combination of different *Phytophthora* diseases of cocoa causes losses of 20–30% of the cocoa crop worldwide (Erwin and Ribeiro 1996). A conservative, long-term average estimate for crop losses and the cost of disease management practices is in the range of 15–20%.

### Durian

The 'king of fruits', durian (*Durio zibethinus* L.), is widely cultivated in the tropics of Asia. The major producers of this fruit are Thailand, Malaysia, Indonesia, and, increasingly, Vietnam (Nanthachai 1994) (Table 2.2). Durian is indigenous to the hot equatorial rainforests of Borneo, Malaysia and Indonesia. Consequently, it prefers a hot (average maximum 33°C, average minimum 22°C) humid tropical environment with high annual rainfall of 2000–3000 mm (Lim 1998a). The fruits of the durian

tree are large, weighing between 1 and 8 kg. The fruit pulp has a rich, unique flavour but it also has a strong sulphurous aroma. The pulp of the fruit is eaten raw, cooked, frozen or dried while the seeds are used to make confectionery (Smith et al. 1992).

Durian is one of the most popular and widely eaten seasonal fruits in Southeast Asia and the fruit attracts a premium price. Production in Indonesia is mainly for domestic consumption, and Malaysia still imports a significant amount of durian in its off-season. The Philippines and Vietnam also produce durian for domestic consumption (Lim 1998b). The majority of production occurs in short seasons of two or three months, although there are two fruiting seasons in Malaysia and Indonesia because the fruit is grown in areas subject to different monsoon seasons (Lim 1998b). Production in Thailand and Malaysia is highest between June and July, while harvest peaks in Indonesia from October to February (Graef and Klotzbach 1995).

#### Indonesia

Indonesia exported 331 t of durian in 1993, its main market being Singapore (Graef and Klotzbach 1995). Most of the fruit is produced in Java, Sumatra, Kalimantan and Sulawesi (Lim 1998b).

#### Malaysia

In 1991, Malaysia was a big exporter of fresh durian, its main export market being Singapore (Graef and Klotzbach 1995). Approximately 90% of the product was exported to Singapore. However, during the off-season in Malaysia, durian is imported from Thailand. Durian is grown in Peninsular Malaysia, Sarawak and Sabah. Like Thailand, there are more than 200 varieties of durian registered, but only 20 are widely used. Durian has traditionally been produced on small orchards 0.5–1.0 ha in size, but more recently 12–120 ha commercial orchards have been established (Lim 1998b).

#### Philippines

Although the durian industry is rapidly expanding in the Philippines, demand continues to outweigh supply. Durian is a high-value crop with great prospects for export, owing to its late fruiting season (August–November) compared to other Southeast Asian countries. The Philippines is actively pushing to increase durian production, especially in the typhoon-free areas of Mindanao. Local consumption of durian in the Philippines is only 0.2 kg/person/year, which is only a fraction of the per person consumption in the other Southeast Asian countries (e.g. Thailand, 14 kg/person/year). There is a need to plant an additional 30 000 ha of durian to meet domestic demand if consumption rises to 2 kg/person/year (Anon. 2000).

#### Thailand

Almost half of the durian produced worldwide is grown in Thailand. Consequently, Thailand supplies 80% of the world export trade (Guest et al. 1998). In 1993, Thailand exported 10% of its durian export as frozen product. Its main market for fresh durian is Hong Kong, but it also exports to Malaysia, Taiwan, Canada, USA, Singapore and Indonesia (Graef and Klotzbach 1995). In 1995, the area planted to durian was approximately 128,000 ha, which accounts for 11% of the total area planted for fruit production. Most of the durian production is based on four commercial cultivars, although there are more than 200 cultivars in use. Flowers are hand-pollinated to improve fruit set and yield. The harvesting process occurs between April and September, with a constant supply between the months of May and August. This is because of the diversity of cultivars and growing regions. In 1996, durian exports amounted to about 5.5% of the total production which still amounts to USD48 million to the Thai economy (Lim 1998b).

**Table 2.2** Production of durian in selected countries in Southeast Asia.

Country	Area planted (ha)	Production (t)	Value (USD '000)
Indonesia <sup>a</sup>	36,024	200,000 <sup>3</sup>	780,000
Malaysia <sup>b</sup>	106,860	200,000	1,020,000
Philippines <sup>a</sup>	8000	145,000	522,000
Thailand <sup>a</sup>	138,024	927,200	2,686,000
Vietnam <sup>c</sup>	40,000	110,000	330,000

<sup>a</sup> Figures are for 1993–94 (Nanthachai 1994).

<sup>b</sup> Figures are for 1998 (Lim 1998b).

<sup>c</sup> Figures are for 1998 (Chau 1998).

## Vietnam

The durian industry in Vietnam is small but rapidly expanding, catering mainly for the domestic market, with some export trade with Taiwan (Chau 1998). The majority of local plantings have been established from seed, rather than from selected varieties. Durian is one of the crops targeted for improvement and expansion by the Southern Fruit Research Institute (SOFRI) (Lim 1998b). Production has increased steadily over a number of years, especially in the south-east and central highlands, and the Mekong Delta region. In the past, durian orchards were established from seedlings, but grafting onto rootstocks has become more popular. Trees are rarely pruned and flowers are not hand-pollinated as they are in Thailand. On some farms, the trees are actively water-stressed to induce off-season flowering and the farmer receives a premium price for off-season fruit.

### Economic importance of *Phytophthora* diseases on durian

The high rainfall conditions under which durian are grown are conducive to the development of *Phytophthora* diseases. The most serious diseases of durian are caused by *P. palmivora*. *Phytophthora palmivora* causes seedling dieback, leaf blight, root rot, trunk cankers, and preharvest and postharvest fruit rots (Lim 1998a). Postharvest fruit rots result in 10–25% losses of durian fruits (Lim 1998b).

Patch canker caused by *P. palmivora* is considered to be a major disease of durian in Malaysia (Agrolink 2001), while fruit rot caused by the same pathogen causes losses of 30% (Chau 1998). In Sabah, *P. palmivora* and, on a few occasions, *P. nicotianae* have been reported as the causal agents of durian root rot and canker (Bong 1990).

Fruit and root rot are the most serious diseases of durian in Thailand (Pongpisutta 1998). Root rot of durian caused by *P. palmivora* was first reported in Thailand in 1966 (Phavakul and Jangsri 1969). *P. palmivora* is also responsible for many other diseases of durian in Thailand.

In Vietnam, fruit and root rot are the major diseases of durian (see Chapter 4.4). In some areas, however, damage caused by *P. palmivora* due in the form of leaf blight, patch and stem canker and fruit rot is considered to be minor (Chau 1998). Stem canker and leaf blight are more widely spread than fruit rot (van Tri 1998). The incidence and severity of *Phytophthora* diseases of durian is increasing, particularly in the Mekong Delta region, which experiences periodic waterlogging. In the Soc Trang Province of the Mekong Delta region, up to 50% of durian trees were killed by stem canker.

Since multiple diseases are caused by *P. palmivora* on durian, it is difficult to estimate the economic importance. Fruit losses due to *P. palmivora* are the easiest to assess but the influence of the tree canker on the production capacity of the durian orchard is difficult to estimate. Stem cankers can kill trees, causing loss of production over a large number of years. On average, it is estimated that disease losses and the cost of control of *P. palmivora* in durians is in the range of 20–25% of production.

## Rubber

*Hevea brasiliensis*, para rubber, has its origins in the Amazon forests of South America, and produces latex that is used to make high-quality rubber. Rubber is a major plantation crop in Southeast Asia and supplies more than 95% of the world's natural rubber, with Malaysia, Indonesia and Thailand being major producers (Smith et al. 1992).

Natural rubber is an important agricultural commodity essential for the manufacturing of a wide range of products. The largest market for natural rubber is the tyre industry. Natural rubber is sold through a complex chain of local, national and international dealers on world markets. Production of rubber from *H. brasiliensis* makes a significant contribution to the economy of many developing countries. Over 80% of production comes from small farms, each typically 2 ha or less. Thailand is the largest producer of rubber, followed by Indonesia and then Malaysia (Table 2.3). Traditionally, natural

**Table 2.3** Production of rubber in selected countries in Southeast Asia in 2000 (FAO 2001b).

Country	Area planted (ha)	Production (t)	Value (USD '000)
Indonesia	2,150,000	1,488,300	839,204
Malaysia	1,400,000	768,900	521,201
Philippines	91,474	70,000	11,756
Thailand	1,520,000	2,235,680	986,268
Vietnam	412,000	290,800	250,000

rubber was an export commodity and, until recently, processing and use was mainly in the industrialised countries. In the past few years, most of the producing countries are moving to downstream processing, converting a significant proportion of their production into manufactured products for domestic use and export.

### Malaysia

Rubber is the third most important commercial crop planted in Sabah after oil palm and cocoa and is mainly grown by smallholders. Rubber sheets and latex are imported to Peninsular Malaysia for downstream processing into high-value-added rubber-based products. The government is also encouraging the cultivation of rubber for the production of rubber wood, which is used to make furniture (Sabah Government 2001b). Rubber production in Malaysia fell by approximately 20% in 2000 because many estates and smallholders continued to switch from rubber to oil palm and other products (FAO 2001b).

### Indonesia

Natural rubber is one of the more important export commodities in Indonesia. This commodity provides both a source of foreign exchange and also of cash income for more than 12 million people. Rubber planters in Indonesia are predominantly smallholders (84%), and hence the quality and quantity of Indonesian rubber depends mainly on the conditions used by rubber smallholders. The two main constraints to rubber production are the traditional technology using unselected seedlings, poor soil conservation, low fertiliser input, low plant maintenance, high planting density, over-tapping, and poor soil fertility. Agricultural research institutes and the government cooperate to increase smallholder productivity by providing recommended planting materials through local farmer groups, and by developing regimes for intercropping during the period before rubber trees reach maturity. Intercrops have the dual role of providing additional income as well as providing cover to reduce soil erosion. Recommended food crops for intercropping include corn, upland rice, soybean and cowpea. Pineapple/banana and chilli are the recommended horticultural crops. Chilli has a good market in Indonesia where it is an important food ingredient. Studies have shown that both food and horticultural crops can be intercropped while rubber trees are immature, with no negative effect on rubber growth (Rosyid et al. 2001).

### Philippines

The area devoted to rubber plantations is approximately 92,000 ha, more than 50% of which is in western Mindanao. Of this area, 36,000 hectares are due for replanting because the trees have reached/are near their maximum productivity. If a replanting program is not implemented, it is projected that the Philippines will be a net importer of rubber within the next 10 years. Although the potential for expansion of the industry is high, production over a 10-year period increased by an average of only 3.3% per annum, and planted area increased by only 1.4%. However, over this period, the yield increased from 1810 kg/ha in 1985 to 2170 kg/ha of raw latex. The Philippines exports about 40% of its natural rubber production, its main markets being Malaysia, China and Singapore (Anon. 2001).

### Thailand

Over 90% of Thailand's natural rubber and products made from rubber are exported to overseas markets. The industry in this country is highly dependent on the world market, making it sensitive to price fluctuations in international trade, which, in turn, are influenced by the prevailing global demand for natural rubber. Strong competition from other major natural-rubber-producing countries, like Malaysia and Indonesia, and climatic conditions are also important factors that significantly affect the rubber industry in Thailand. At present, the global market situation is favourable to Thai latex producers as the global demand for natural rubber products continues to grow. Malaysia, having significantly reduced its own natural rubber production, is now importing latex concentrate from Thailand for the manufacture of rubber products (Thaitex 1998).

### Vietnam

The first rubber plantation was founded in Vietnam in 1897, during the era of French colonialism. After the Vietnam War ended, the Vietnamese government aimed to re-establish Vietnam as a major exporter of natural rubber. The industry was revitalised by a USD32 million loan from the World Bank in 1996 to improve rubber latex processing technology to international standards. Most of the rubber tree plantations in Vietnam are located in the southern region of the country. In 2000, the total rubber tree plantation area in Vietnam was 412,000 ha and the average annual output of natural rubber 290,800 t. Vietnam's output of natural rubber is growing at a rate of 15% per year due to the establishment of new plantings, and young trees reaching maturity. The area planted to rubber in the year 2005 is forecast to be 700,000 ha, with plantation

ownership split equally between state and privately owned companies. Vietnam's largest rubber company is the state-owned Vietnam Rubber Corporation, which has almost 60% of the total plantation area in the country, accounting for approximately 65% of latex production. Private and provincial companies own the balance and are expected to grow dramatically in the years ahead. Collection of latex from the rubber tree begins when it reaches six years of age, and the product is harvested continuously until the tree reaches 30 years of age. Latex production by rubber trees peaks at 12 years. In Vietnam, the highest latex yield is obtained from October to December, during the months immediately following the rainy season (CBC Vietnam 1998).

### **Economic importance of *Phytophthora* diseases in rubber**

The bark of rubber trees is regularly cut to tap the latex, and hence there are a number of important wound parasites, *Phytophthora* species being the most important (Watsie 1975). Several diseases of rubber are attributed to a number of species of *Phytophthora*, including *P. botryosa*, *P. heveae*, *P. meadii*, *P. palmivora* and *P. nicotianae*. However, *P. palmivora* and *P. meadii* are isolated most frequently as the causal agents of black stripe, patch canker, green pod rot, green twig blight, and abnormal leaf fall. Of these diseases, black stripe is the most severe disease of para rubber caused by *Phytophthora* (Erwin and Ribeiro 1996) followed by leaf fall. In wet tropical areas such as southern Thailand, leaf fall is very common and can give cause a 40% drop in yield. Black stripe is most troublesome but can be kept under control by regular management of the tapping panel. Losses due to *Phytophthora* can be high if not kept under control. The losses due to *Phytophthora* and the cost of disease control is estimated at 5–10% and has been declining recently due to the planting of more resistant rubber clones.

### **Coconut**

Coconut (*Cocos nucifera*) are one of the most valuable plant species in the tropics, providing oil, coconut milk, fibre from the husk, palm wine, and timber for furniture and construction. It is believed that coconuts originated in Asia, with some secondary centres of origin in Central and South America. Humans have distributed coconuts throughout the tropics, and since the nuts can float, the spread has also been assisted by ocean currents.

Coconut palms are tall, unbranched trees and typically grow to 20–30 m for tall varieties, while dwarf palms only reach 10 metres. The nuts are

large, 20–30 cm in diameter, weigh up to 1 kg, and have a thick, fibrous mesocarp. The hard shell (endocarp) surrounds the seed, which contains the white, meaty endosperm that envelops the coconut water. The endosperm is high in oil and when this is dried, it is called copra. Copra contains about 60–70% oil. Coconut oil is widely used in the production of margarine, food processing, and in the production of soaps and cosmetics. The market for coconut oil has suffered in recent times because of fears the highly saturated fats are linked to increases in blood cholesterol levels. Although coconut oil contains no cholesterol, it has been largely replaced by aggressively marketed soybean and maize oils from subsidised farms in Europe and the USA.

Production of coconuts starts when the trees are 6–7 years old and may be sustained for over a century. Typical production will range from 30–70 nuts/tree/year for seedling trees but hybrids may produce more. Traditionally, the coconut tree requires little attention throughout its life span of over 50 years, and therefore it is known as a 'lazy man's crop'. Smallholders produce the majority of coconuts. Large commercial farms, however, are tended and developed for improved productivity (Agustin 2001).

The substitution of coconut oil with oil palm is another factor that is affecting the global demand for coconut oil (FME 2001b). Approximately 93% of world production of coconut occurs in the Asia-Pacific region (Table 2.4). In 1996, Indonesia supplied 26% of world production of coconut, the Philippines 23%, Thailand 5%, Vietnam 2% and Malaysia 1.5% (Food Market Exchange 2001). In the 1960s, over 1 million t of copra (dried coconut meal) was traded worldwide a year. The volume declined to about 900,000 t a year in the 1970s, further declining to an annual average of 350,000 t in the 1980s. This dramatic decline was the result of establishment of domestic copra-processing plants in response to the desire of the producing countries to obtain more value-added products. The downtrend in copra exports is likely to continue (Punchihewa and Arancon 2000).

In contrast to copra, coconut oil exports increased markedly. The world annual tonnage of coconut oil exported for 1990–1994 averaged 1.6 million t, with about 55% from the Philippines (Punchihewa and Arancon 2000). World trade in coconut oil rose 75% during the 1970s and the market further improved to an average of 1.2 million t in 1980s. Coconut oil accounts for 80% of total coconut production in the Philippines. Indonesia uses the bulk of their production internally, both as food nuts and as

coconut oil. Apart from copra and coconut oil, other exports include desiccated coconut, copra meal, cocochemicals (fatty acids, fatty alcohol, methyl ether), shell charcoal and activated carbon, fibre products, coconut cream, and coconut milk powder (Punchihewa and Arancon 2000). Coconut water is used for drinking. The white meat (copra) is processed to produce coconut milk, desiccated coconut, coconut powder, and cosmetic and pharmaceutical products (MARDI 2000).

Phytophthora diseases of coconut are important in Southeast Asia, particularly in Indonesia and the Philippines where West African-bred hybrids were widely planted in the 1980s.

### Malaysia

Commercial planting of coconut started as early as 1900. In Malaysia, most of the coconuts are planted along the coastal region of Peninsular Malaysia and the states of Sabah and Sarawak. Of the 246,015 ha of coconut in Malaysia in 1993, 93% were smallholder plantings. In worldwide terms, Malaysia is a small producer of coconut and many coconut growers are opting to grow the economically more attractive oil palm (MARDI 2000).

### Indonesia

Production of coconut and copra is important to the economy of Indonesia. Copra produced in Indonesia accounts for 26% of world production from 32% of the world area planted to coconut. Ninety-eight per cent of coconuts are produced by smallholders who under-plant coconut with other cash and food crops (Mady 1992). Average yields of coconut are relatively low because of the advanced age of the palms, and poor crop maintenance and disease control. The introduction of high-yielding hybrids has not improved productivity significantly, despite government support schemes (Darwis 1992). Coconut is frequently planted as a shade tree for cocoa plants (Lolong et al. 1998).

### Philippines

One-third of the country's arable agricultural land (which amounts to 3.31 million ha) is planted to coconut. At present, there are more than 300 million coconut trees, bearing an annual average of 12 billion nuts. In the last five years, the average production has been 2.3 million t. The Philippines supplies 64% of global coconut oil requirements. Coconut is a major source of foreign exchange – the Philippine coconut exports accounting for some 65% of the world traded coconut products. Exports earn an average of USD800 million a year. It is the top export earner on a net basis given that its raw materials and labour components are domestically based, unlike other export products. One-third of the Philippine population (approximately 24 million people) directly or indirectly benefit from the coconut industry.

The productivity of Philippine coconut plantation per hectare per year is one tonne, compared with a potential of 2–4 t/ha/year. The poor productivity is due to a lack of agricultural inputs, limited access to credit, lack of irrigation facilities, inadequate transport and roads, poor postharvest and processing facilities, the indiscriminate removal of productive trees, and the conversion of coconut lands to other commercial and agricultural enterprises. In addition, the average gross annual income of a coconut farmer is below the poverty line. Intercropping with corn, legumes, root crops or fruit trees is not widely practised, and thus the income of growers remains poor (Philippine Department of Agriculture 1999).

### Thailand

Thailand is only a small producer of coconut on a worldwide basis. During 1995, most coconut produced was consumed domestically. Its main exports of coconut products are shelled coconut, coconut oil and desiccated coconut. Exports of coconut products peaked during 1995 because production of the two biggest producers, the Philippines and Indonesia, declined (FME 2001b).

**Table 2.4** Production of coconut in selected countries in Southeast Asia in 2000 (FAO 2001a).

Country	Area harvested (ha)	Production (t)	Value (USD '000)
Indonesia	2,800,000	2,342,000	140,069
Malaysia	180,000	683,000	2789
Philippines <sup>a</sup>	3,076,647	5,761,000	686,000
Thailand	333,000	1,373,162	2870
Vietnam	161,900	939,900	1100

<sup>a</sup> The Philippines also produced 57,610 t of coconut seed in 2000.



## Vietnam

Like Thailand, Vietnam is only a small exporter of coconut.

### Economic importance of *Phytophthora* disease in coconut

Rots caused by *Phytophthora* spp. lead to palm death (by bud rot) and/or yield reduction (by premature nut fall) (Waller and Holderness 1997). While most of the coconut-growing regions of the world are affected by *Phytophthora* rots, Indonesia and the Philippines are the worst affected due to the introduction of very sensitive MAWA hybrids developed in West Africa (see Chapter 6.3) (Renard 1992). In Malaysia (Sarawak), Indonesia and the Philippines, *P. palmivora* seems to be the main causal agent of disease (Blaha et al. 1994). Coconut bud rot has an irregular distribution in the field, but the highest incidence seems to correlate with the wettest areas (Waller and Holderness 1997).

*Phytophthora* diseases were not a major problem in the tall coconut varieties grown in Southeast Asia, causing disease losses of 5–10% (Brahmana et al. 1992). *Phytophthora palmivora* was first reported in the Philippines on coconut in 1919 as *P. faberi* (Reinking 1923). In the 1980s, 500,000 ha of land were replanted with a MAWA coconut hybrid in order to replace old and non-productive trees. This hybrid proved to be highly susceptible to *P. palmivora*. As a result, bud rot infections led to the death of thousands of palms (Concibido-Manohar and Abad 1992). Chapter 6.3 provides the full details of this disastrous germplasm-introduction program, which gave rise to financial hardship to all who planted these hybrids as they succumbed to *Phytophthora* bud rot.

Bud rot and nut fall were first reported in Indonesia in 1985, the causal agents being identified as *P. palmivora* and *P. nicotianae* (Bennett et al. 1986). During this time, outbreaks of the disease resulted in severe damage to plantations planted with MAWA germplasm (Renard 1992). Since that time, almost all areas planted with MAWA coconut in Indonesia have suffered serious damage from bud rot, with losses in excess of 80% (Darwis 1992). In some areas, stand losses of 43% can occur due to bud rot. Premature nut fall, which is the more common disease, affects nuts of 3–7 months old (Lolong et al. 1998), and can cause losses of 50–75% (Brahmana et al. 1992). The incidence of bud rot is higher in the lowland areas of Indonesia, which are poorly drained, compared to the highland areas. Resistance among coconut varieties to infection and damage by *Phytophthora* varies with location, and therefore it is recommended that several varieties be planted to

minimise damage caused by the pathogen (Mangindaan et al. 1992).

In Indonesia, although *P. palmivora* seems to be the main causal agent of bud rot and nut fall in coconut (Blaha et al. 1994; Waller and Holderness 1997), *P. arecae* and *P. nicotianae* have also been found in association with these diseases (Thevenin 1994). *Phytophthora nicotianae* is rarely encountered, and it is usually associated with cocoa and infested soil (Waroka and Thevenin 1992). Bud rot and premature nut fall are the major disease problems affecting coconut in Indonesia (Lolong et al. 1998). The highest incidence of bud rot generally corresponds to the wettest areas.

Due to the high level of susceptibility of these hybrids to bud rot they are no longer planted. Breeding and selection programs aim to produce high-yielding varieties with good levels of resistance.

This example can be used as a timely reminder that large-scale planting of highly susceptible plant material can have drastic economic consequences. A conservative estimate of the economic impact of *Phytophthora* on coconuts is 0–5%, while 10–15% losses occurred in Indonesia and the Philippines due to the large-scale plantings of the MAWA hybrid.

## Pepper

Black pepper (*Piper nigrum* L.) is a member of the tropical family Piperaceae, and it is believed to be indigenous to the state of Kerala in south-western India. It is a perennial woody climbing vine with three central climbing stems and lateral stems which bear inflorescences that produce pepper berries (Holliday and Mowat 1963). Propagation of *P. nigrum* is vegetative because seedlings take longer to bear fruit than cuttings and produce highly variable dioecious progeny. The three main climbing stems are pruned frequently to stimulate the growth of lateral fruiting branches. Fruit production begins within two years of planting, and the vines can produce fruit for 12–15 years. The flower spikes are harvested at regular intervals over a 2–3 month period (Purseglove et al. 1981). Cuttings are planted in a mound of soil in which a post made from termite-resistant wood is inserted. Alternatively, concrete posts, cut-off shade trees, or brick towers may be used to support the vines. As the vines grow, they are trained around the post. There are several different types of pepper, all derived from the berries produced by *P. nigrum*.

Black pepper is prepared by drying the mature, still-green berries in the sun for 3–4 days. White pepper is prepared from fully ripened berries that are yellow to

red in colour. The pericarp is removed from the berries by soaking in water for approximately two weeks (PMB 2001). Green pepper is prepared from unripe, green berries. The berries are artificially dried, or preserved in brine, vinegar or citric acid (IPC 1999). Long pepper is derived from *P. longum*, and is not consumed on a large scale in Western society (Purseglove et al. 1981). It is, however, used widely in India (Katzer 2000). Black pepper is regarded as the world's most important spice in terms of its use and trade value (Thurston 1984). Trade in black pepper has been known since 400–300 BC (Holliday and Mowat 1963), being described by the philosopher/botanist Theophrastus (Purseglove et al. 1981).

Pepper is known as the 'king of spices', dominating 34% of the world spice trade in volume. The demand for pepper increases by about 2.5% annually, and more than 60% of pepper is used by the food industry. Prices vary substantially because of fluctuations in supply (IPC 1999). Pepper requires heavy and well-distributed rainfall and high temperatures for optimum productivity. The International Pepper Community (IPC) comprises Indonesia, Malaysia, Thailand, Sri Lanka, India and Brazil. The IPC accounts for more than 80% of the world production and export of pepper (Table 2.5). If Vietnam joins the organisation, the IPC will control 95% of world production and export (IPC 1999). Many pepper-producing countries are developing value-added pepper products for export (PMB 2001).

**Table 2.5** Production of pepper<sup>a</sup> in selected countries in Southeast Asia (FAO 2000).

Country	Area planted (ha)	Production (t)	Export value (USD '000)
Indonesia	80,000	52,188	191,241
Malaysia <sup>b</sup>	12,000	21,000	106,783
Philippines	na	na	224
Thailand	2500	7000	3082
Vietnam	15,000	34,000	103,000

<sup>a</sup> Figures include white, long and black pepper.

<sup>b</sup> Data provided by Board (2001).

Note: na = data not available; no reference has been found to pepper production in the Philippines.

### Malaysia

The British organised plantings of pepper in Malaysia early in the 19th century (Purseglove et al. 1981). Malaysia is now the fourth largest producer of black pepper in the world (PMB 2001). Currently, 95% of the pepper produced in Malaysia is grown in Sarawak (PMB 2001).

### Indonesia

Hindu colonists probably took pepper to Java between 100 BC and AD 600, and thus it has a long history of cultivation in Indonesia (Purseglove et al. 1981). Black pepper is considered to be one of the oldest export commodities of Indonesia (Sitepu 1993). Until the Second World War, when supply was cut off by the Japanese invasion, Indonesia was the largest supplier of black pepper in the world (Purseglove et al. 1981). It is now the second largest producer after India (PMB 2001). Mainly small landholders produce pepper and approximately 600,000 people depend upon this commodity for their livelihood (Sitepu 1993; Wahid and Zaubin 1993). Foot rot of black pepper was first recorded in Indonesia in 1936 (Muller 1936), and since then has caused large economic losses (Tsao et al. 1985).

### Vietnam

Vietnam increased its production and export of pepper four-fold over a 10-year period, increasing from 8000 t in 1990 to 34,000 t in 2000 (PMB 2001). It is now the world's second-largest pepper exporter. The price for Vietnamese pepper is usually 10–20% lower than the price offered by other pepper-exporting countries. This is due to a combination of poor quality and poor marketing (Nhan Dan 2001).

### Economic importance of Phytophthora diseases in pepper

*Phytophthora capsici* causes foot rot of black pepper. This disease is also referred to as 'sudden wilt'. An epidemic of the disease in Sarawak in the mid-1950s caused crop losses of almost 100% (Holliday and Mowat 1963), while crop losses of 40–50% due to foot rot have been recorded in other areas (Erwin and Ribeiro 1996). Foot rot is clearly the most important and destructive fungal disease of black pepper, occurring wherever the crop is grown (Holliday 1980). The disease was originally attributed to *P. palmivora* (Muller 1936; Holliday and Mowat 1963), although a number of studies on the disease recognised that the isolates from black pepper were morphologically distinct from *P. palmivora* isolates from other hosts (Holliday 1980), being grouped with *P. palmivora* MF4 (morphological form 4) types (Tsao et al. 1985). After extensive morphological and molecular studies (Tsao and Alizadeh 1988; Tsao 1991; Mchau and Coffey 1995), the causal agent of foot rot was determined to be *P. capsici* and not *P. palmivora*. *Piper betle* L. (betle vine), the leaves of which are used as a masticatory in Asia, is also attacked by *P. capsici* (Holliday and Mowat 1963).

Foot rot caused by *P. capsici* in pepper has been reported to cause an estimated annual loss of 5–10%

in Malaysia (Kueh 1979). This would be average for a situation where the disease is managed and kept under control. Thus, in Indonesia and Malaysia, a significant amount of management is applied and experience exists to control foot rot. In other countries, the disease losses are higher (10–15%), while in Vietnam they are higher still (15–20%) due to inexperience in managing foot rot, high ground water tables in some areas, and the use of susceptible varieties.

## Citrus

The genus *Citrus* contains a large number of species that provide a diversity of fruits and uses. In addition, there are many species hybrids, such as the Citrange, Citrumelo and Tangelo. Most species of citrus are cultivated for fresh fruits and to make fruit juices, jams or confectionaries. All commercially important citrus fruits have originated from species native to Southeast Asia. Citrus trees and shrubs occur naturally throughout the region, and selections are widely cultivated (Table 2.6). However, little is known about the domestication process but it most likely started a long time ago since citrus already were taken from Southeast Asia for growing in the Mediterranean during the great Greek civilisation. Many other species were established in the Mediterranean during the middle ages.

**Table 2.6** Production of citrus in selected countries in Southeast Asia (FAO 2000).

Country	Area planted (ha)	Production (t)	Value (USD '000)
Indonesia	100,000	680,000	260,000
Malaysia	5220	28,500	11,000
Philippines	34,674	177,266	67,000
Thailand	91,400	1,079,500	413,000
Vietnam	71,300	450,200	172,000

There are up to twelve different species of *Phytophthora* reported to cause diseases of citrus (see Table 17.1 in Erwin and Ribeiro 1996). However, the most common species causing *Phytophthora* disease in citrus in the tropics are *P. nicotianae*, *P. palmivora*, *P. citricola* and *P. citrophthora*.

*Phytophthora nicotianae* may be considered as the main pathogen of citrus and causes root rot, foot rot and gummosis, although it seldom causes problems higher up in the canopy. *P. nicotianae* produces abundant chlamydospores that enhance its survival in the soil. *P. citrophthora* is another important *Phytophthora* species that causes root rot, foot rot, and gummosis but also causes brown rot in citrus. *P. citrophthora* isolates do not produce

chlamydospores. *P. palmivora* can also cause severe brown rot on the fruit under wet conditions. *P. citricola* has mainly been reported as causing brown fruit rot in citrus.

*Phytophthora* species attacking citrus are present in the soil. Infection occurs under wet conditions when the *Phytophthora* species are induced to produce zoospores. This typically happens during prolonged periods of wet weather, especially when flooding occurs. This typically leads to infection of the roots. However, the damage due to the root rot often shows up late during the dry season when the diseased root system is unable to keep up the supply of water and nutrients.

## Economic importance of *Phytophthora* diseases in citrus

*Phytophthora* diseases are economically important in most citrus-growing regions. Due to the number of *Phytophthora* species and the number of different diseases involved, the economic impact is difficult to estimate. In addition, the relationship between root rot and yield loss are not proportional. Losses due to *Phytophthora* vary a lot with seasonal and climatic conditions. This is especially true for brown rot, which can lead to serious losses under wet conditions while virtually absent in years with below-average levels of rainfall. Over all different citrus species, the yield losses in the USA were estimated to range from 3–6% a year. In the wet tropical areas of Southeast Asia, the yield loss is estimated to be 6–12% a year as weather conditions are more favourable for disease, and the trees are more stressed due to the presence of other diseases and extensive monsoonal wet periods.

## Potato

The common potato, *Solanum tuberosum*, is a member of the large and important family Solanaceae that includes eggplant and tomato. Europeans first saw the potato in 1537 when the Spanish landed in what is now called Colombia, South America. The potato was brought back to Europe around 1570, and it was cultivated throughout the continent before 1600, and in Ireland by 1663. The cultivated potato was first introduced into North America in 1621. Potatoes are the leading starchy root crop of the subtropical countries, and one of the eight leading staple food crops of the world. Annual production of potatoes is approximately twice that of all other edible root crops combined (Ozero 1984).

The potato is becoming increasingly important in Asia and, although rice is synonymous with food in

most of Asia, almost one-quarter of the world's potatoes are now grown in Asia (Table 2.7). The potato is a short-duration crop that produces a large amount of calories in a short period of time. In addition, potato fits well into the cereal-based cropping systems found throughout Asia. The introduction of improved, short-duration varieties of rice have provided a niche for the potato crop in the agricultural production calendar (van der Zaag 1983), but unlike cereals, the potato crop does not need to grow to full maturity before harvest. The introduction of better-adapted varieties, inorganic fertilisers, fungicides and pesticides has significantly improved productivity per unit area. Improvements in transportation and postharvest handling have reduced losses, increased marketable yields and reduced marketing costs (Horton et al. 1987). However, the main reason for the expansion of potato production in Asia has been the desire by farmers to satisfy expanding markets and changing consumer preferences. Population growth and urbanisation has expanded the market for food crops and rising per capita income has stimulated the demand for more exotic foods to diversify diets. Probably the most significant factor influencing potato consumption in Asia is the growth in the fast-food industry. Over the last three decades, potato production in Asia has tripled to exceed 60 million t.

**Table 2.7** Production of potato in selected countries in Southeast Asia in 1999 (FAO 2000).

Country	Area harvested (ha)	Production (t)	Value (USD '000)
Indonesia	62,776	924,058	1,892,000
Malaysia	na	na	311,000
Philippines	5216	63,520	12,704
Thailand	900	7000	38,000
Vietnam	28,022	315,950	63,190

Note: na = data not available.

However, while productivity has increased from an average of 5.9 t/ha (1961) to 13.3 t/ha over a 30-year period, the average yield of potato crops in Asia remains low in contrast to yields of approximately 39 t/ha in temperate areas. It is considered unlikely that similar yields to those achieved in the temperate zones can be realised in Asia, and a major constraint to potato production in Asia is the inadequate supply of reasonably priced, good-quality seed tubers of the desired varieties. Substantial gains in productivity can be achieved by promoting the production and use of certified seed to reduce the risk of distributing tuber-borne pathogens. Degeneration of locally sourced planting material is

rapid due to infections with several important viral diseases. Australia produces seed potatoes under a certification system and supplies the seed to Asia. Trials in Vietnam and the Philippines with Australian-produced seed potato have resulted in significant increases in productivity (Batt 1999b).

The main biotic constraints for potato production are late blight caused by *Phytophthora infestans*, bacterial wilt disease, viruses, and potato tuber moth. Researchers estimate that developing-country farmers spend \$700 million annually to control such pests. The susceptibility of potato to these pests and diseases makes the crop the number-two user of agricultural pesticides worldwide, following cotton. Results of breeding work at the International Potato Centre (CIP, Lima, Peru) with South American potatoes is aimed at developing resistance to *P. infestans* (CGIAR 2001).

## Indonesia

The Dutch introduced the potato to West Java in Indonesia around 1794. By 1811, the crop was found on other Indonesian islands such as Sumatra. However, due to the warm climate, the potato never became a food of general consumption compared to the yam, arum, and sweet potato. The area planted to potato increased steadily to over 67,000 ha in 1995–97. Yields per hectare have also increased from around 6 t/ha in the early 1970s to 11.5 t/ha in 1985. Java accounts for 65% of national production, Sumatra for 10%, while the rest occurs mainly in southern Sulawesi. The potato, along with cabbage and tomato, is an important cash crop in certain highland areas where they are produced on non-irrigated land and compete with forestry. Land temporarily cleared from trees is sometimes planted with potatoes and other vegetables. Rotations found in irrigated areas include rice–potato–cabbage, rice–potato–maize, and cabbage–potato–cabbage. Rotations found in non-irrigated areas include potato–cabbage–maize and potato–maize–fallow. Many potato pests and diseases are found in Indonesia, late blight being the most important. Small farmers cultivating less than 0.5 ha often have limited access to capital – they will tend to keep their own seed tubers which are small in size and produce low yields. More affluent farmers will plant seed potatoes imported mainly from the Netherlands and Australia. Little information is available on storage of potatoes in Indonesia, but storage periods are fairly short and losses are high. Although a small portion of the annual production is exported to Singapore and Malaysia, most Indonesian potatoes are consumed domestically. Rice remains the basic staple for the general

population, supplemented by varying amounts of maize, cassava, sago and sweet potato. In Indonesia, potato is an expensive vegetable consumed only on special occasions (CIP 1988).

### Malaysia

The British introduced the potato into Malaysia in the 1930s. As in the rest of Southeast Asia, the potato was a minor vegetable in production systems and was consumed largely by the non-Asian populations. Due to unfavourable growing areas, lack of seed sources, and severe late-blight problems, the area of production of potato has not increased significantly over the past few years. However, there has been a growing demand for potato, largely met through imports, due to continuing economic prosperity. The area planted to potato also increased because of government incentives, but was limited by continued late-blight attacks in the 1980s. Potatoes are produced in Malaysia in two main highland areas on Peninsular Malaysia and Sabah, where temperatures are cool (average monthly maximum 23°C, minimum 15°C). They are typical vegetable-producing areas that cater mainly to urban markets. Potatoes are cultivated in rotation with other vegetables, especially cabbage, cauliflower, tomatoes and onions. The terrain is steep and sloping, and consequently the crops are planted on terraces. Potatoes can be cultivated throughout the year in Malaysia. Approximately 30% of the potato crop produced on Sabah is exported to Singapore. Raw potato imports into Malaysia come primarily from China, the Netherlands, Taiwan and Indonesia (van der Zaag 1983).

### Philippines

Although the Spanish most likely brought the potato to the Philippines, the precise date or circumstances of the introduction are unknown. However, the potato is believed to have been present in the Philippines by the late 18th century. By the late 1930s, potatoes were being produced in large quantities. Today, the potato is a high-priority crop because of its high potential yield and nutritional qualities. Potatoes generate higher returns per hectare than most other food crops. The potato has been selected by the government as one of three national priority crops for commercial development (Batt 1999a). More than 90% of the production of potatoes takes place in the highland areas of northern Luzon, followed by upland production areas of Mindanao. Scattered, but very limited, production is found in the mountainous areas of the Visayas. Almost 90% of production occurs at altitudes between 1600 and 2400 m. Domestically consumed potatoes are purchased

primarily as a luxury vegetable or a snack food (Batt 1999a; SHEL 2001).

### Thailand

Potatoes were introduced during the late 19th century to the tribes of northern Thailand either from Burma (now Myanmar) or China. The potato crop gained greater attention from both growers and the government in 1955 after the successful introduction of the variety Bintje from the Netherlands. Bintje and subsequent imported varieties have replaced most local varieties. Since the late 1970s, potato-growing has been stimulated by international agencies seeking alternatives to opium poppy as a cash crop. The growth of the tourist trade and hotel industry in Thailand has led to an increase in demand for potatoes and should stimulate further interest in the crop. Potatoes are grown mainly in the mountainous regions of northern Thailand. The two production zones include the highlands where potatoes are grown all year round but primarily in the wet season; and the lower-lying valleys where potatoes are grown on flat paddy areas after rice has been harvested. In the valleys, production occurs during the cool, dry season. Hence, production takes place virtually all year long in northern Thailand. The average mean temperature during the main growing season in the northern and north-eastern highlands is around 15–20°C, with high average annual rainfall. In the lowland valley zone, potatoes are grown on irrigated, flat paddy land using imported seed, chemical fertilisers and pesticides. They are produced by specialised potato producers who are close to the major markets and have greater opportunity to get technical advice from government extension officers. Hill tribe farmers are geographically isolated from markets and grow potatoes on rain-fed slopes, using few inputs, and locally obtained seed. They are often engaged in off-farm labour activities and are relatively isolated from extension efforts due to language and cultural barriers. The vast majority of growers of any type do not cultivate more than 1 or 2 ha/year, often at different times of the season, and hence the amount of land in potatoes at any one period may be less than 0.25 ha. A few large growers have between 5 and 10 ha of land. The vast majority of farmers sell their potatoes immediately or soon after harvesting because they lack storage facilities and need rapid cash returns. Producers eat few, if any, potatoes. Thirty per cent of total potato production is consumed locally and the rest is transported to other provinces and neighbouring countries. The government does not support producer prices, and hence potato prices are governed by the market and

by seasonal variations in supply (Rhoades et al. 1988a).

### Vietnam

Although the first published reference to potatoes in Vietnam was made in 1807, it is claimed that European missionaries introduced the potato to the Red River Delta in 1890. The potato remained a minor vegetable in Vietnam until the 1970s when population growth and annual typhoon damage of the rice crop motivated the government and farmers to use the dry season from November to February for potato production. Potatoes now rank third in importance after rice and maize.

A national potato program was established in 1981. Most of the potatoes in Vietnam are produced in the lowlands of the Red River Delta, being planted after rice. The use of high-yielding, early-maturing rice varieties make it possible to harvest two rice crops within 8 months, leaving 4 months in the winter for potato production. Some production occurs in the highlands of Dalat. The temperature in the main production areas fluctuates from 17°C to 26°C. The crop is allowed only a short growing time, frequently resulting in premature harvest. The main production unit in the Red River Delta is the agricultural cooperative: 30% of the crop is sold, the cooperative members consume 35%, and 30% is stored for seed for up to 9 months. Virus infection is high in Vietnam and seed degeneration is rapid. Seed potatoes are stored by farmers in their homes in areas characterised by darkness and high temperatures, usually above 25°C, and sometimes as high as 32–35°C. The storage period can be as long as 8 months, which results in decay and losses as high as 45–60%. Hanoi and Ho Chi Minh City are the main consumption centres for potatoes (Rhoades et al. 1988b).

### Economic importance of Phytophthora diseases in potato

*Phytophthora infestans* causes late blight of potato, and leaf, stem and fruit blights of many solanaceous hosts including tomato (*Lycopersicon esculentum*) and eggplant (*Solanum melongena* L.) (Erwin and Ribeiro 1996). Late blight is listed as a major disease of potato in Malaysia, Indonesia, the Philippines, Thailand and Vietnam van der Zaag 1983; CIP 1988; Rhoades et al. 1988a,b; SHEL 2001).

Late blight is the most important disease of potatoes in Indonesia, the Philippines, in the hills of northern Thailand, and Vietnam (CIP 1988; Rhoades 1988a,b; SHEL 2001). Postharvest rot of tubers caused by *P. infestans* is also a significant problem. In Malaysia, late blight is a problem during the rainy months

from April to November. As a result, the main growing period is from December to April when less rainfall occurs (van der Zaag 1983).

All these reports indicate that late blight caused by *P. infestans* is the most important potato disease in the Southeast Asian region. Losses vary enormously between regions, varieties, and the wet and dry seasons. Potatoes are frequently sprayed with protectant fungicides to prevent infection. This intense management comes at a significant cost to the grower and we estimate that 15–20% of the crop is lost due to late blight.

### Overall Impact of Phytophthora in Southeast Asia

In this chapter, we have tried to give a realistic picture of disease losses experienced due to Phytophthora in seven major crops that are grown on a large scale in Southeast Asia. Disease impact varies between varieties, cropping methods, regions, seasons, and years, and our overall disease assessment gives no more than a sweeping overview of the situation.

If we combine all disease assessments in Table 2.8 and add up the subsequent disease losses, we come to an average figure of USD2.4 billion for disease losses, with a minimum of USD2.1 billion and a maximum of USD2.7 billion. These figures are derived from the sum of losses for each country for the seven crops under discussion (Table 2.9). Disease losses for *P. infestans* in potatoes and *P. sojae* in soybean at a global scale have been estimated at USD3 billion and USD1.2 billion per annum, respectively.

**Table 2.8** Summary of losses (USD '000) due to Phytophthora in seven main crops in five Southeast Asian countries.

Country	Minimum	Maximum	Average
Indonesia	639,272	886,444	762,859
Malaysia	295,949	399,111	347,531
Philippines	181,203	247,413	214,308
Thailand	617,412	828,041	722,727
Vietnam	351,249	386,433	368,841
Total	2,085,085	2,747,442	2,416,266

In addition to the crops outlined here, there are a large number of important tropical crops that also suffer from Phytophthora. We know that significant disease losses are experienced in tomato, tobacco, vanilla, eucalypt forestry, papaya, longan and chilli pepper.

Another important aspect of diseases is that they increase the risk of production of crops. Many smallholders have severe credit restrictions. In practical terms, this means that they do not have the funds to buy inputs to control and manage diseases. Thus, these smallholders are exposed to epidemics and risk large losses. Some *Phytophthora* diseases may also kill mature trees such as durian, citrus and cocoa, severely reducing the production capability

of the small holder. Hence, in order to reduce losses due to *Phytophthora* we need to provide cheap and effective disease control methods that can be adapted with very little inputs. In addition more resistant germplasm is needed and made available to small holders to reduce the enormous impact currently imposed on smallholders by *Phytophthora* pathogens.

**Table 2.9** Details of losses due to *Phytophthora* in seven different crops in five different countries in Southeast Asia.

		USD '000				
		Value	Disease loss (%)	Minimum loss	Maximum loss	Average loss
Cocoa	Indonesia	788,952	15–20	118,343	157,790	138,067
	Malaysia	88,268	15–20	13,240	17,654	15,447
	Philippines	11,112	15–20	1,667	2,222	1,945
	Thailand	741	15–20	111	148	130
	Vietnam	na	15–20	-	-	-
	Total				133,361	177,815
Durian	Indonesia	780,000	20–25	156,000	195,000	175,500
	Malaysia	1,020,000	20–25	204,000	255,000	229,500
	Philippines	522,000	20–25	104,400	130,500	117,450
	Thailand	2,686,000	20–25	537,200	671,500	604,350
	Vietnam	330,000	20–25	66,000	82,500	74,250
	Total			1,067,600	1,334,500	1,201,050
Rubber	Indonesia	839,204	5–10	41,960	83,920	62,940
	Malaysia	521,201	5–10	26,060	52,120	39,090
	Philippines	11,756	5–10	588	1176	882
	Thailand	986,268	5–10	49,313	98,627	73,970
	Vietnam	250,000	5–10	250,000	250,000	250,000
	Total			367,921	485,843	426,882
Coconut	Indonesia	140,069	10–15	14,007	21,010	17,509
	Malaysia	2789	0–5	0	139	70
	Philippines	686,000	10–15	68,600	102,900	85,750
	Thailand	2870	0–5	-	144	72
	Vietnam	1100	0–5	-	55	28
	Total			82,607	124,248	103,429
Pepper	Indonesia	191,241	5–10	9562	19,124	14,343
	Malaysia	106,783	5–10	5339	10,678	8009
	Philippines	224	10–15	22	34	28
	Thailand	3082	10–15	308	462	385
	Vietnam	103,000	15–20	15,450	20,600	18,025
	Total			30,681	50,898	40,790

**Table 2.9** (Cont'd) Details of losses due to Phytophthora in seven different crops in five different countries in Southeast Asia.

		USD '000				
		Value	Disease loss (%)	Minimum loss	Maximum loss	Average loss
Citrus	Indonesia	260,000	6–12	15,600	31,200	23,400
	Malaysia	11,000	6–12	660	1320	990
	Philippines	67,000	6–12	4020	8040	6030
	Thailand	413,000	6–12	24,780	49,560	37,170
	Vietnam	172,000	6–12	10,320	20,640	15,480
	Total				55,380	110,760
Potato	Indonesia	1,892,000	15–20	283,800	378,400	331,100
	Malaysia	311,000	15–20	46,650	62,200	54,425
	Philippines	12,704	15–20	1906	2541	2223
	Thailand	38,000	15–20	5700	7600	6650
	Vietnam	63,190	15–20	9479	12,638	11,058
	Total				347,535	463,379

## References

- Agrolink 2001. Fruit technology durian [accessed 28 June 2001]. On the Internet: <<http://agrolink.moa.my/doi/Bi/Croptech/durian.html>>.
- Agustin, Y.V. 2001. The coconut palm – tree of life. United Coconut Associations of the Philippines (UCAP) [accessed 28 June 2001]. On the Internet: <<http://www.ucap.org.ph>>.
- Anon. 2000. The Philippines recommends for durian. Philippines recommends series no. 87. Los Baños, Laguna, Philippine Council for Agriculture, Forestry and Natural Resources Research and Development.
- – – 2001. Rubber industry situationer report. Republic of the Philippines Department of Agriculture, Agribusiness and Marketing Assistance Service.
- Batt, P.J. 1999a. Potato production in the Philippines. World Potato Congress. On the Internet: <<http://www.potatocongress.org/sub.cfm?source=136>>.
- – – 1999b. An alternative seed system for Asia: seed potato exports from Western Australia. World Potato Congress. On the Internet: <<http://www.potatocongress.org/sub.cfm?source=135>>.
- Bennett, C.P., Roboth, O., Sitepu, G. and Lolong, A. 1986. Pathogenicity of *Phytophthora palmivora* (Butl.) causing premature nutfall disease of coconut (*Cocos nucifera* L.). Indonesian Journal of Crop Science, 2, 59–70.
- Blahe, G. 1992. Criteria for the identification of *Phytophthora* species causing pod rot on cocoa in West Africa. In: Keane, P.J. and Putter, C.A., ed., Cocoa pest and disease management in Southeast Asia and Australasia. Food and Agriculture Organization of the United Nations (FAO) Plant Production and Protection Paper 112. Rome, Italy, FAO.
- Blahe, G., Hall, G., Warokka, J.S., Concibido, E. and Ortiz-Garcia, C. 1994. *Phytophthora* isolates from coconut plantations in Indonesia and Ivory Coast: characterisation and identification by morphology and isozyme analysis. Mycological Research, 98, 1379–1389.
- Bong, C.L. 1990. Destructive diseases of selected fruit trees and spices. Paper presented at in-house Seminar and Workshop on Fruits, Nuts and Spices. Lagud Sebrang, Tenom, Malaysia.
- Bowers, J.H., Bailey, B.A., Hebbar, P.K., Sanogo, S. and Lumsden, R.D. 2001. The impact of plant diseases on world chocolate production. The American Phytopathology Society [accessed 22 June 2001]. On the Internet: <http://www.apsnet.org/online/feature/cacao/top.html>>.
- Brahamana, J., Lubis, A.U. and Chenon, R.D. 1992. Evolution of coconut bud disease and strategy of control. Paper presented at Coconut *Phytophthora* Workshop, in Manado, Indonesia.
- Brasier, C.M., Griffin, M.J. and Maddison, A.C. 1981. The cocoa black pod *Phytophthora*s. In: Gregory, M.P.H., ed., Epidemiology of *Phytophthora* on cocoa in Nigeria. Kew, England, Commonwealth Mycological Institute.
- CBC (Connell Bros. Co. Ltd) Vietnam 1998. Vietnam natural rubber [accessed 21 June 2001]. On the Internet: <<http://www.vietnamrubber.com>>.
- CGIAR (Consultative Group on International Agricultural Research) 2001. Potato (*Solanum tuberosum*). CGIAR [accessed 24 July 2001]. On the Internet: <<http://www.cgiar.org/areas/potato.htm>>.
- Chau, N.M. 1998. Current status of durian production and handling. In: Guest, D.I., ed., Management of *Phytophthora* diseases in durian. Australian Centre for International Agricultural Research (ACIAR) Project PHT95/134 Workshop No. 1. Melbourne, Australia, University of Melbourne.



- CIP (International Potato Centre) 1988. International Potato Centre, Indonesia, 1988 [accessed 25 July 2001]. On the Internet: <<http://gis.cip.cgiar.org/gis/potato/atlas/asia/Indonesia.htm>>.
- Concibido-Manohar, E.C. and Abad, R.G. 1992. Notes on the incidence of *Phytophthora* infection on coconut cultivars in the Philippines. Paper presented at the Coconut *Phytophthora* Workshop, Manado, Indonesia.
- DA-AMAS (Department of Agriculture, Agribusiness and Marketing Assistance Service) 1999. Cacao industry situationer report [accessed 4 July 2001]. Republic of the Philippines, Department of Agriculture. On the Internet: <<http://www.da.gov.ph/agribiz/amas.html>>.
- Darwis, S.N. 1992. *Phytophthora* in relation to climate and coconut cultivar. Paper presented at the Coconut *Phytophthora* Workshop, Manado, Indonesia.
- Effendi, S. 1992. Policy on development of cocoa in Indonesia. In: Keane, P.J. and Putter, C.A., ed., *Cocoa pest and disease management in Southeast Asia and Australasia*. Food and Agriculture Organization of the United Nations (FAO) Plant Production and Protection Paper 112. Rome, Italy, FAO.
- Erwin, D.C. and Ribeiro, O.K. 1996. *Phytophthora* diseases worldwide. St Paul, USA, American Phytopathological Society Press.
- FAO (Food and Agriculture Organization of the United Nations) 2000. Food and Agriculture Organisation Commodities and Trade Division [accessed 18 June 2001]. On the Internet: <<http://apps.fao.org/page/collections?subset=agriculture>>.
- — — 2001a. Tropical fruits commodity notes. Food and Agriculture Organization Commodities and Trade Division, February 2001 [accessed 22 June 2001]. On the Internet: <<http://www.fao.org/waicent/faoinfo/economics>>.
- — — 2001b. Rubber commodity notes. Food and Agriculture Organization Commodities and Trade Division, February 2001 [accessed 23 July 2001]. On the Internet: <<http://www.fao.org/waicent/faoinfo/economic/ESC/esce/cmr/cmnotes>>.
- — — 2003. Food and Agriculture Organisation Commodities and Trade Division 2003 [accessed 2003]. On the Internet: <<http://faostat.fao.org/faostat/collections?subset=agriculture>>.
- FME (Food Market Exchange) 2001a. Vietnam could be a major player in cocoa market. Food Market Exchange [accessed 28 June 2001]. On the Internet: <[http://www.foodmarketexchange.com/datacenter/news/dc\\_ns\\_index\\_detail.php3?newsid=3384](http://www.foodmarketexchange.com/datacenter/news/dc_ns_index_detail.php3?newsid=3384)>.
- FME (Food Market Exchange) 2001b. Coconut production. World production. Statistics from: Statistical Yearbook 1996, Asian and Pacific Coconut Community (APCC) 2000–2001 [accessed 26 June 2001]. On the Internet: <[http://www.foodmarketexchange.com/datacenter/product/fruit/coconut/dc\\_po\\_ft\\_coconut02.htm](http://www.foodmarketexchange.com/datacenter/product/fruit/coconut/dc_po_ft_coconut02.htm)>.
- Graef, J. and Klotzbach, T. 1995. World market for durian. RAP Market Information Bulletin, No. 3.
- Gregory, P.H. and Maddison, A.C. 1981. Epidemiology of *Phytophthora* on cocoa in Nigeria. *Phytopathological Paper* No. 25.
- Guest, D.I., Sangchote, S. and Chau, N.M. 1998. Management of *Phytophthora* diseases in durian. Australian Centre for International Agricultural Research (ACIAR) Project Proposal PHT95/134. Canberra, ACIAR.
- Holderness, M. 1992. Biology and control of *Phytophthora* diseases of cocoa in Papua New Guinea. In: Keane, P.J. and Putter, C.A., ed., *Cocoa pest and disease management in Southeast Asia and Australasia*. Food and Agriculture Organization of the United Nations (FAO) Plant Production and Protection Paper 112. Rome, Italy, FAO.
- Holliday, P. 1980. *Fungus diseases of tropical crops*. Cambridge, United Kingdom, Cambridge University Press.
- Holliday, P. and Mowat, W.P. 1963. Foot rot of *Piper nigrum* L. (*Phytophthora palmivora*). *Phytopathological Paper* No. 5, 1–62.
- Horton, D.E., Collins, W., Iwanaga, M., Mendoza, H. and Collins, M. 1987. Constraints to production and utilization of potatoes and sweet potatoes. Paper presented at The Social Sciences at CIP (International Potato Centre). Report of the third social science planning conference, September 7–10, 1987, at International Potato Center (CIP), Apartado 5969, Lima, Peru.
- ICCO (International Cocoa Organization) 1998. Information on the history and development of cocoa bean production in Indonesia, specifically Sulawesi. Cocoa Organisation International [accessed 19 June 2001]. On the Internet: <<http://www.icco.org/questions/indonesia.htm>>.
- International Pepper Community 1999. Spurt in black pepper export from Vietnam world pepper industry [accessed 4 July 4]. On the Internet: <<http://www.ipcnet.org/link.htm>>.
- Katzer, G. 2000. Pepper. Gernot Katzer's spice pages 2000 [accessed 4 July 2001]. On the Internet: <[http://www-ang.kfunigraz.ac.at/~katzer/engl/generic\\_frame.html?Pipe\\_nig.html](http://www-ang.kfunigraz.ac.at/~katzer/engl/generic_frame.html?Pipe_nig.html)>.
- Kueh, T.K. 1979. *Pests, diseases and disorders of black pepper in Sarawak*. Lee Ming Press.
- Lim, T.K. 1998a. Durian. In: Hyde, K., ed., *The new rural industries. A handbook for farmers and investors*. Canberra, Rural Industries Research and Development Corporation, 281–287. Also available on the Internet [accessed 15 May 2001]: <<http://www.rirdc.gov.au/pub/handbook/durian.html>>.
- — — 1998b. Durian production in the world and status of *Phytophthora palmivora*. In: Guest, D.I., ed., *Management of Phytophthora diseases in durian*. Australian Centre for International Agricultural Research (ACIAR) Project PHT95/134 Workshop No. 1. Melbourne, Australia, University of Melbourne.
- Lolong, A., Smith, J.J. and Holderness, M. 1998. Characterisation of *Phytophthora* diseases of coconut in Indonesia. Paper presented at the International Congress of Plant Pathology, Edinburgh, Scotland. Paper No. 3.7.87.

- Mady, H.B. 1992. Present status of coconut bud rot disease in Indonesia. Paper presented at the Coconut *Phytophthora* Workshop, Manado, Indonesia.
- Mangindaan, H.F., Thevenin, J.M., Kharie, S. and Motulo, H.F. 1992. The susceptibility of coconut varieties to *Phytophthora* in Indonesia: the effect of environmental factors. Paper presented at the Coconut *Phytophthora* Workshop, Manado, Indonesia.
- MARDI (Malaysian Agricultural Research and Development Institute) 2000. MARDI Net. The webpage of MARDI [accessed 19 June 2001]. On the Internet: <<http://www.mardi.my/>>.
- Mchau, G.R. and Coffey, M.D. 1995. Evidence for the existence of two distinct subpopulations in *Phytophthora capsici* and a redescription of the species. *Mycological Research*, 99, 89–102.
- Muller, H.R.A. 1936. The *Phytophthora* foot rot of black pepper (*Piper nigrum* L.) in the Netherlandish Indies. Cited in: *Review of Applied Mycology* 1937, 16, 559.
- Nanthachai, S. 1994. Durian – fruit development, postharvest physiology, handling marketing in ASEAN. Kuala Lumpur, Malaysia, Association of South-East Asian Nations (ASEAN) Food Handling Bureau. Cited in: Lim (1998b).
- Nhan Dan 2001. Vietnam, world's second largest pepper exporter. *Nhan Dan Economy* [accessed 6 July 2001]. On the Internet: <<http://www.nhandan.org.vn/>>.
- Ozero, N.H. 1984. {paper title?} In: Massey, H.F., Understanding the production of the major tropical/sub-tropical root crops cassava, potatoes, sweet potatoes, yams and cocoyams. Virginia, USA, Volunteers in Technical Assistance.
- Padwick, G.W. 1956. Losses caused by plant diseases in the Colonies. Volume 1, phytopathological papers. Kew, England, Commonwealth Mycological Institute.
- PMB (Pepper Marketing Board) 2001. Sarawak Black Pepper [accessed 4 July 2001]. On the Internet: <<http://www4.jaring.my/sarawakpepper/>>.
- Pereira, J.L. 1992. Cocoa and its pathogens in the region of origin: a continued risk. In: Keane, P.J. and Putter, C.A., ed., Cocoa pest and disease management in Southeast Asia and Australasia. Food and Agriculture Organization of the United Nations (FAO) Plant Production and Protection Paper 112. Rome, Italy, FAO.
- Phavakul, K. and Jangsri, V. 1969. Root rot of durian. In: *Plant disease control: Agricultural Science Society of Thailand (in Thai)*. Cited in: Pongpisutta (1998).
- Philippine Department of Agriculture 1999. Coconut program. PCA program for food security and farmers' welfare. Republic of the Philippines Department of Agriculture [accessed 19 June 2001]. On the Internet: <<http://www.da.gov.ph/programs/coconut/coco.html>>.
- Pongpisutta, R. 1998. *Phytophthora palmivora* (Butl.) Butl., causing root and fruit rot of durian in Thailand. In: Guest, D.I., ed., *Management of Phytophthora diseases in durian*. Australian Centre for International Agricultural Research (ACIAR) Project PHT95/134 Workshop No. 1. Melbourne, Australia, University of Melbourne.
- Punchihewa, P.G. and Arancon, R.N. 2000. Coconut: post-harvest operations (Chapter XV). World Trade Asian and Pacific Coconut Community. On the Internet: <<http://www.apcc.org.sg>>.
- Purseglove, J.W., Brown, E.G., Green, C.L. and Robbins, S.R. 1981. Pepper. In: *Spices*, volume 2. London, Longman Scientific and Technical.
- Reinking, O.A. 1923. Comparative study of *Phytophthora faberi* on coconut and cacao in the Philippine Islands. *Journal of Agricultural Research*, 25, 267–284.
- Renard, J.L. 1992. Introduction to coconut *Phytophthora* diseases. Paper presented at the Coconut *Phytophthora* Workshop, Manado, Indonesia.
- Rhoades, R.E., Hijmans, R.J. and Huaccho, L. 1988a. Thailand. International Potato Center [accessed 24 July 2001]. On the Internet: <<http://gis.cip.cgiar.org/gis/potato/atlas/asia/Thailand.htm>>.
- — — 1988b. Vietnam. International Potato Center [accessed 24 July 2001]. On the Internet: <<http://gis.cip.cgiar.org/gis/potato/atlas/asia/Vietnam.htm>>.
- Rosyid, M.J., Wibawa, G. and Gunawan, A. 2001. Rubber based farming systems development for increasing smallholder income in Indonesia [accessed 20 July 2001]. On the Internet: <<http://www.irrd.org/agronomy/smincome.htm>>.
- Sabah Government 2001a. Sabah cocoa sector. Sabah Government, Malaysia [accessed 22 June 2001]. On the Internet: <[http://www.ssl.sabah.gov.my/clh/english/economy/agriculture\\_cocoa.htm](http://www.ssl.sabah.gov.my/clh/english/economy/agriculture_cocoa.htm)>.
- Sabah Government 2001b. Sabah rubber sector. Sabah Government, Malaysia [accessed 21 June 2001]. On the Internet: <[http://www.ssl.sabah.gov.my/clh/english/economy/agriculture\\_rubber.htm](http://www.ssl.sabah.gov.my/clh/english/economy/agriculture_rubber.htm)>.
- Salakpeth, S. 2000. Durian production in Thailand. Hawaii Tropical Fruit Growers Tenth Annual Tropical Fruit Conference, Hilo, Hawaii, 21 October 2000.
- SHEL (Sustainable Human Ecosystems Laboratory) 2001. Information on potato production in the Philippines. SHEL, the Philippines. University of Georgia. On the Internet: <<http://lanra.dac.uga.edu/potato/asia/malaysia.htm>>.
- Sitepu, D. 1993. Disease management on pepper. *Indonesian Agricultural Research and Development Journal*, 15(2), 31–37.
- Smith, N.J., Williams, J.T., Plucknett, D.L. and Talbot, J.P. 1992. *Tropical forests and their crops*. New York, USA, Cornell University Press.
- Soehardjan, M. 1992. Government policy on crop protection for cocoa in Indonesia. In: Keane, P.J. and Putter, C.A., ed., *Cocoa pest and disease management in Southeast Asia and Australasia*. Food and Agriculture Organization of the United Nations (FAO) Plant Production and Protection Paper 112. Rome, Italy, FAO.

- Thaitex 1998. Introduction. Thai Rubber Latex Corporation [accessed 22 June 2001]. On the Internet: <<http://www.thaitex.com/profile/frame.htm>>.
- Thevenin, J.M. 1994. Coconut diseases in Indonesia – etiological aspects. Paper presented at the Coconut Phytophthora Workshop, Manado, Indonesia. Cited in: Waller and Holderness (1997).
- Thurston, H.D. 1984. Tropical plant diseases. St Paul, Minnesota, USA, APS Press.
- Tsao, P.H. 1991. The identities, nomenclature and taxonomy of *Phytophthora* isolates from black pepper. Paper presented at Diseases of Black Pepper. Proceedings of the International Pepper Communication Workshop on Pepper Diseases, Goa, India.
- Tsao, P.H., and Alizadeh, A. 1988. Recent advances in the taxonomy and nomenclature of the so-called "*Phytophthora palmivora*" MF4 occurring on cocoa and other tropical crops. Paper presented at the 10th International Cocoa Research Proceedings, Santo Domingo, 17–23 May 1987.
- Tsao, P.H., Kasim, R. and Mustika, I. 1985. Morphology and identity of black pepper *Phytophthora* isolates in Indonesia. Food and Agriculture Organization of the United Nations (FAO) Plant Protection Bulletin, 33, 61–66.
- van der Zaag, P. 1983. Malaysia. Sustainable Human Ecosystems Laboratory. On the Internet: <<http://lanra.dac.uga.edu/potato/asia/malaysia.htm>>.
- van Tri, M. 1998. Durian cultivation and Phytophthora diseases in Vietnamese uplands. In: Guest, D.L., ed., Management of Phytophthora diseases in durian. Australian Centre for International Agricultural Research (ACIAR) Project PHT95/134 Workshop No. 1. Melbourne, Australia, University of Melbourne.
- Wahid, P. and Zaubin, R. 1993. Crop improvement and cultivation of black pepper. Indonesian Agricultural Research and Development Journal, 15(2), 27–30.
- Waller, J.M. and Holderness, M. 1997. Beverage crops and palms. In: Hillocks, R.J. and Waller, J.M., ed., Soilborne diseases of tropical crops. Wallingford, CAB International.
- Waroka, J.S. and Thevenin, J.M. 1992. *Phytophthora* in Indonesian coconut plantations: populations involved. Paper presented at the Coconut *Phytophthora* Workshop, Manado, Indonesia.
- Watsie, R.L. 1975. Diseases of rubber and their control. Pest Articles and News Summaries, 21, 268–288.

# 3

## Biology of *Phytophthora*



## 3.1 *Phytophthora* in the Tropics

André Drenth<sup>1</sup> and David I. Guest<sup>2</sup>

### Abstract

Most of the 60 described *Phytophthora* species are important in temperate as well as tropical regions. The various species, and in some cases the same species, can cause a wide array of different diseases on the same and on different crops. An understanding of typical symptoms is therefore important to recognise phytophthora disease problems in the field. An understanding of the evolutionary placement, life cycle and disease cycle of *Phytophthora* is paramount to developing sustainable disease-control strategies. Phytophthora diseases impose major limitations on the productivity and viability of many tropical and subtropical crops. Effective management of these diseases need to be based on a sound understanding of the biology of the pathogen, including its modes of survival and dissemination, host range and the role of environmental factors in the disease cycle. Examples in these proceedings, drawn from research on phytophthora diseases of cocoa, coconut, durian and other hosts, illustrate these points.

### The Genus *Phytophthora*

*Phytophthora* de Bary 1887 is a cosmopolitan genus of Oomycete obligate plant pathogens containing approximately 60 described species (Erwin and Ribeiro 1996). The *Phytophthora* genus is a member of the Order Peronosporales within the Phylum Oomycota. *Phytophthora* species attack a wide range of plants, and are responsible for some of the world's most destructive plant diseases – examples include the European potato famine of the 19th century caused by *P. infestans* (Bourke 1964). Phytophthora diseases have been well studied in the temperate regions of the world. However, they are very common also throughout the wet tropical regions of the world and cause significant disease losses in many tropical fruit crops in the form of root rots, collar rots, stem cankers, leaf blights and fruit rot. *P. palmivora* alone, for example, causes a myriad of severe diseases on many different crops including: black pod of cocoa; root, stem and fruit rot of pawpaw; root rot and fruit rot of citrus; bud

rot in palms; black stripe in rubber; and root rot, trunk canker, and fruit rot in durian.

### Evolutionary Placement

There has been considerable debate in the 20th century about the evolutionary placement of Oomycetes. First they were placed in the Fungal Kingdom but then moved to the Protists followed by the Kingdom Chromista, recently renamed to the Stramenopiles Kingdom (Hawksworth et al. 1995; van de Peer et al. 1996; Beakes 1998) (Table 3.1.1). The Oomycetes share many characteristics of ecology and life history with the true fungi. However, they are clearly distinguished from the Basidiomycetes and Ascomycetes by their genetics and reproductive mechanisms (Erwin and Ribeiro 1996). Their placement in the Kingdom Chromista (Cavalier-Smith 1986) and later the Stramenopiles was supported by a large number of characteristics, including variation in metabolic pathways (Hendrix 1970; Wang and Bartnicki-Garcia 1973; Elliott 1983), the presence of  $\beta$ -glucans rather than chitin in cell walls (Bartnicki-Garcia and Wang 1983), production of motile heterokont zoospores (Desjardins et al., 1969), and predominance of the diploid stage in the lifecycle (Erwin and Ribeiro 1996). The Oomycetes includes four orders, two of which, the Saprolegniales and the Peronosporales, contain important plant pathogens. The other two orders

<sup>1</sup> CRC for Tropical Plant Protection, Indooroopilly Research Centre, 80 Meiers Road, Indooroopilly, Queensland 4068, Australia.

<sup>2</sup> Department of Botany, The University of Melbourne, Parkville, Victoria 3010, Australia.  
Current address: Faculty of Agriculture, Food and Natural Resources, The University of Sydney, NSW 2006, Australia.

contain small groups of mainly aquatic fungal-like organisms. Within the Peronosporales, the family Pythiaceae contains a number of genera, the best known of which are *Phytophthora* and its sister group, *Pythium*, a genus of approximately 120 species (van der Plaats-Niterink 1981).

### Phytophthora as Plant Pathogens

Almost all species within the genus *Phytophthora* are formidable plant pathogens. Hence, we have to ask the question: What makes these organisms such effective plant pathogens? The following factors are involved:

- The ability to produce different types of spores such as sporangia and zoospores for short-term survival and spread, and chlamydospores and oospores for longer term survival.
- Rapid sporulation on host tissue within 3–5 days of infection. This results in a rapid build-up of secondary inoculum in a multicyclic fashion, leading to epidemics under suitable favourable environmental conditions.
- Ability of zoospores of *Phytophthora* to be attracted to root tips through a chemical stimulus (positive chemotaxis) as well as root-generated electric fields (electrotaxis) (van West et al. 2002), coupled with the mobility of zoospores to actually swim to the actively growing root tips, encyst, and infect young, susceptible root tissue.
- Ability to survive in or outside the host tissue as oospores or chlamydospores for long periods. Oospores are also known to survive passage through the digestive systems of animals such as snails.
- Production of sporangia, which can be airborne and may travel reasonable distances in raindrops, run-off and irrigation water, and on wind currents, to infect neighbouring fields. These sporangia can directly infect host tissue. These same sporangiospores also have the ability to differentiate into 4–32 zoospores under humid

and cool conditions and cause multiple infections from the one sporangium. Nevertheless, zoospores can travel only short distances, as they are susceptible to desiccation.

- *Phytophthora* pathogens belong to the Kingdom Stramenopiles and as such have different biochemical pathways to the true fungi. Many fungicides are therefore not very effective against phytophthora pathogens.
- *Phytophthora* pathogens thrive under humid and wet conditions, which makes them difficult to control, as protectant fungicides are difficult to apply and least effective under such conditions.

### Symptoms of Phytophthora Diseases

*Phytophthora* pathogens can cause many different diseases and disease symptoms on a wide range of plant species. In the next section, the disease symptoms most often encountered are discussed.

#### Root rot

Seedlings of many plants are very susceptible to root rot and damping off caused by phytophthora. The early symptoms are the wilting and yellowing of young seedlings. General symptoms of root rot are that plants appear water stressed, chlorotic, and are often stunted in their growth. New leaves are often small and show a light green to yellow colour and wilting occurs even in the presence of sufficient water. Affected root tissue is soft, watersoaked and discoloured to dark brown rather than the creamy white colour of healthy roots. Advanced root rot leads to the lack of secondary and tertiary roots and a lack of healthy root tips (Figure 3.1.1).

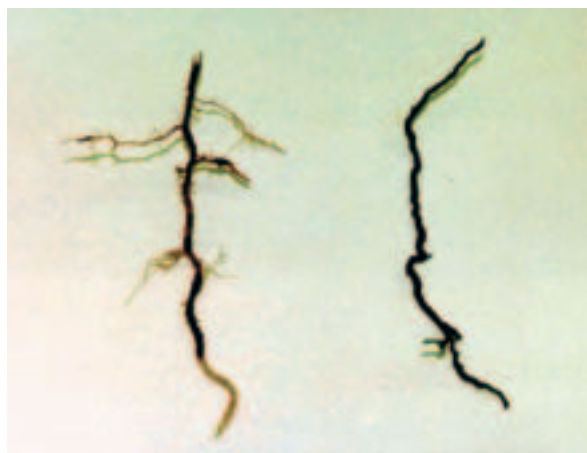
#### Collar rot

Collar rot, sometimes called foot rot, often develops at or just below ground level. The infection frequently moves upwards from the roots, rotting the lower bark tissue and discolouring the lower stem. Exudation of gum often occurs in the affected parts. The affected bark area is often irregular in shape and size and first

**Table 3.1.1** Classification of the Oomycetes (Hawksworth et al. 1995; Beakes 1998).

Kingdom	Class	Order	Family	Genus
Stramenopiles	Oomycetes	Lagenidiales	Saprolegniaceae	<i>Achlya</i>
		Leptomitales		<i>Saprolegnia</i>
		Saprolegniales		<i>Pythium</i>
		Peronosporales		<i>Phytophthora</i>
			Peronosporaceae	<i>Bremia</i>
			Albuginaceae	<i>Peronospora</i>
				<i>Albugo</i>

appears as a water-soaked lesion, before drying, becoming sunken and giving rise to cracks in the bark that usually show dark-brown discoloration. Above-ground symptoms appear as wilting, reduction of foliage, and dieback of branches such as the symptoms caused by *P. capsici* in pepper (Figure 3.1.2). Bark and cortex tissues often have a swollen and cracked appearance, separating easily from the underlying tissue. The disease may also progress around the trunk giving rise to girdling of the main roots or the trunk.



**Figure 3.1.1** Roots of pineapple affected by *Phytophthora cinnamomi*.

### Tree canker

Many species of *Phytophthora* can form cankers on the stems of host plants. These cankers have various names, including stripe canker (cinnamon), patch canker (durian) or trunk canker (cocoa). The first sign of canker is usually the appearance of wet lesions on the bark surface (Figure 3.1.3), often close to the branch points at the lower end of the trunk. Bark discoloration and exudation of reddish



**Figure 3.1.2** (Left) Wilting of pepper due to *Phytophthora capsici*. (Right) Section of the main root affected by *P. capsici*.

brown, resinous substance frequently accompany necrosis. When the bark is stripped away, the cortical tissues and wood appear dull and discoloured from cream coloured to reddish brown (Figure 3.1.4). Wood lesions are often very irregular in shape but are well defined. Expanding lesions severely restrict water and nutrient flow to the connecting branches, leading to wilting. If the lesion girdles the tree branch, dieback is more widespread in the crown and the tree may lose all its leaves.



**Figure 3.1.3** Lesion on the bark of cocoa tree due to *P. palmivora*.

### Stem lesions

Some species of *Phytophthora* attack leaves as well as stems. For example, *P. infestans* on potato and tomato, and *P. nicotianae* on tobacco. In advanced stages, dry, dark-brown or black lesions develop in the cortical tissue on the stem. Lesions frequently start near the soil line and subsequently expand upward and may cover as much as half the length of the stem in the case of black shank on tobacco. Expanded lesions often girdle the stem and give rise to wilting and death of the upper branches and leaves.





**Figure 3.1.4** Reddish brown canker on cocoa tree caused by *Phytophthora palmivora*.



**Figure 3.1.6** Bud rot symptoms in coconut palm caused by *Phytophthora palmivora*.



**Figure 3.1.5** Stem lesion in tomato caused by *Phytophthora infestans*.



**Figure 3.1.7** Lesions of *Phytophthora palmivora* on the heart of a bud rot affected palm.

Young immature stems are often most susceptible, as in stem blight of tomato caused by *P. infestans* (Figure 3.1.5).

### Bud rot

Bud rot (sometimes called heart rot), is a serious problem in many species of palms. It is caused predominantly by *P. palmivora*. The symptoms of bud rot of palm are exhibited over a period of months, often following severe storms, which facilitate infection and spread of phytophthora. Symptoms first appear as discoloration and wilting of the spear leaf and one or more of the newest

leaves, which become chlorotic (Figure 3.1.6). These new leaves may exhibit lesions from infection that has occurred in the spear. As the infection in the bud of the palm progresses, newly emerging leaves show increasing amounts of damage. Eventually, the spear leaves can be pulled out easily because they are rotted at the base, where some white mycelial growth may be observed. The fronds will turn yellow, then brown, and will fall off, finally leaving only a naked, dead trunk. In the base of the bud, small lesions can be seen (Figure 3.1.7), but secondary invaders soon move in, and fluid starts to collect giving off a foul smell. The tissue below the



bud shows discolouration from reddish brown to brown. It is hard to isolate *Phytophthora* from palms with advanced bud rot due to bacterial decay of the bud. Trees that are beginning to show symptoms with an advancing margin on the bud should be used instead, as they are often still relatively free of secondary invaders.

### Heart rot

A number of *Phytophthora* species cause heart rot, but a common one in tropical regions is heart rot of pineapple caused by *P. nicotianae* and *P. cinnamomi*. Young pineapples with heart rot show chlorotic foliage and necrotic leaf tips (Figure 3.1.8). The heart leaves towards the centre of the plant are easily pulled out and show rotting at the base with a characteristic delimited brown lesion indicating the growth of the pathogen (Figure 3.1.9). Under wet conditions, a foul odour accompanies the rotting of the base of the leaves and invasions of secondary pathogens. Heart rot is most common on young plants, while older plants may show restricted lesions slightly higher up the stem.



**Figure 3.1.8** Symptoms of heart rot in pineapple caused by *Phytophthora nicotianae*.

### Leaf blight

Several *Phytophthora* species cause leaf blight. These include *P. infestans* on potato and tomato, *P. palmivora* on rubber and a large number of tropical fruit species including durian (Figure 3.1.10), and *P. colocasiae* on taro (Figure 3.1.11). These blights on leaves are first seen as small flecks, but within 3–5 days they expand to produce large lesions. Initially, infected tissue is watersoaked but becomes necrotic (brown or black) in a few days. Often the lesions are surrounded by a halo of light green tissue. Spores appear as white velvety growth at the edge of the lesions, primarily at the underside of the leaf. It is this white growth that distinguishes phytophthora

leaf blight from several other foliar diseases. Large amounts of sporangiospores are often produced as 1–4 sporangiophores extend from the stomata at the underside of the leaf. Sporangiospores can become airborne and lead to rapid spread of the disease.



**Figure 3.1.9** Brown lesions on the bottom of pineapple leaves affected by heart rot.

### Fruit rot

Fruit rot caused by *Phytophthora* species is common in a large number of different plant species, including citrus, durian, cocoa, papaya and chilli pepper. It appears as watersoaked lesions with light-brown centres 3–5 days after infection, depending on the host. The lesions expand rapidly and can completely rot an entire fruit. Under conditions of

high humidity, white/grey mycelium may be found behind the advancing margin of the lesions (Figure 3.1.12). Often the fruit does not drop and may mummify on the tree. The infection can also be internal, as in the case of *P. palmivora* in papaya where mycelial growth can be seen on the seeds after cutting open infected fruit. Brown rot on citrus develops as an expanding circular lesion with a dull-brown colour. Typical of many fruit rots caused by phytophthora is that the diseased tissue remains firm as it darkens in colour. In the case of brown rot in citrus, a strong odour coming from the fruit is another characteristic of the disease.

### Tuber and corm rot

Tubers of potato and corms of taro are considered to be enlarged stem pieces and are susceptible to infection by phytophthora. Potato tubers can be infected by zoospores of *P. infestans* washed down by rain from the leaves. Tuber infections are characterised by patches of brown to purple discolouration on the potato skin (Figure 3.1.13). Cutting just below the skin reveals a dark, reddish-brown, dry corky rot. Heavy infection



Figure 3.1.10 Lesion of *P. palmivora* on durian leaf.



Figure 3.1.11 *P. colocasiae* lesion on taro leaf.

can give rise to severe rot and total loss of the tubers. Light infections can occur and are difficult to detect. However, if such potatoes are used as seed potatoes they can infect the emerging stems and start off a new epidemic in the next planting season. This is probably how most late blight epidemics start. Potato can also be infected by *P. erythroseptica*, causing the so-called pink rot disease. Infected tubers have a dull brown appearance and exude water under pressure. The cut surface of tubers becomes faint pink after exposure to air. After 30 minutes, the entire cut surface of the tuber turns bright pink. If corms of taro are infected with *P. colocasiae*, they stay firm and leathery, which is typical of phytophthora dry rot. Under favourable conditions, the corms may rot completely after about one week.

### Life Cycle

The life cycle of *Phytophthora* may involve up to three asexual spore forms – sporangia, zoospores, and chlamydospores – in addition to oospores, the sexual spore form (Figure 3.1.14). Diploid vegetative mycelium produces asexual sporangia, which may germinate directly, or differentiate to produce 8–32 zoospores, each of which passes through a cycle of dispersal and encystment before germinating. Some species, such as *P. cinnamomi*, also produce significant numbers of asexual chlamydospores from the mycelium. Sexual reproduction results in the production of oospores. All spore types are potentially infective, and chlamydospores and oospores also function as overwintering or resting structures. All species of *Phytophthora* have a soil-borne resting stage. In addition, some species, such as *P. palmivora*, are also aerially dispersed, primarily as caducous (deciduous) sporangia.

### Host Range

Species of *Phytophthora* vary greatly in their degree of host specificity. *P. fragariae* var. *rubi* infects a single host species (Kennedy and Duncan 1995), while *P. cinnamomi* is able to attack over 1000 different host-plant species (Erwin and Ribeiro 1996), and other species lie in the range between these two extremes.

In the tropics, the most commonly encountered *Phytophthora* species is *P. palmivora* which has a large host range. *P. nicotianae* is also common and occurs on many different host species. *P. capsici* has a slightly more restricted host range but is still able to infect over 40 different crop plant species. *P. hevea* and *P. katsurae* are considered to have a narrow host range when it comes to crop plants, but are commonly found in some samples obtained from

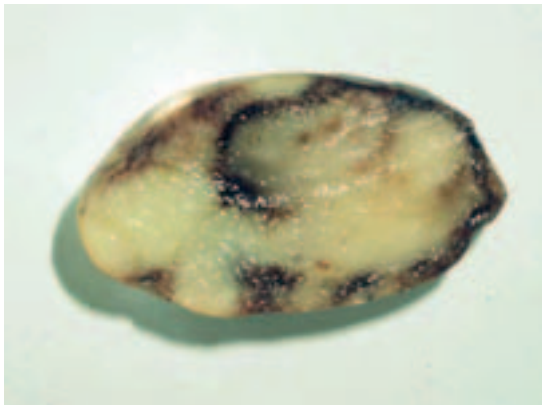
rainforest soils. Some *Phytophthora* species in the tropics are very host specific, such as *P. colocalisae* on taro. It is not difficult to understand that control of, for example, *P. palmivora* on a particular crop involves a far more complex approach that has to involve the alternative hosts, than, say, control of *P. colocalisae* on taro which has a very restricted host range (Erwin and Ribeiro 1996; Zentmyer 1980).

### Mating System

All isolates of *Phytophthora* are potentially bisexual; that is, they are able to produce both male and female sexual structures, or gametangia (Galindo



**Figure 3.1.12** Advancing lesion of *P. palmivora* on durian fruit. Note the white sporulation in the centre of the lesion



**Figure 3.1.13** Tuber infection of potato caused by *P. infestans*.

and Gallegly 1960). However, only about half of the species of *Phytophthora* are homothallic, and able to produce oospores rapidly and abundantly in single culture. The remaining species are heterothallic, and produce gametangia only in response to chemical stimulation from an isolate of the opposite mating type (Brasier 1992; Ko 1978).

The system of heterothallism involving A1 and A2 mating types is universal throughout the genus. Isolates of opposite mating types from different species are often able to reciprocally stimulate gametangial formation (Ko 1978). The mating system of a *Phytophthora* species determines its ability to outbreed: homothallism allows frequent selfing, whereas heterothallism encourages outbreeding. However, both homothallic and heterothallic species do have a range of reproductive options. Homothallic species have recently been shown to undergo low levels of outbreeding in vitro (Whisson et al. 1994), while heterothallic species have been shown to inbreed at low levels (Goodwin et al. 1994).

Sexual reproduction has a number of roles in the life cycle of phytophthora. It allows for recombination of the existing alleles in the case of heterothallic *Phytophthora* species, while for both homothallic and heterothallic species, oospores may act as a structure permitting survival for long periods in the absence of a host plant. Oospores may also remain in infected host tissue to overcome adverse conditions for further colonisation such as hot and dry weather.

At present we do not know the relative importance of sexual reproduction in most *Phytophthora* species. Although the role of oospores in the epidemiology has to a large degree been evaluated for *P. infestans* in temperate regions, the role of sexual reproduction and the formation of oospores in the tropics are not well understood for any *Phytophthora* species.

### Morphological Variation

Details of the morphological properties and pathology of many of the 60 described species of *Phytophthora* are collated in Erwin and Ribeiro (1996). In traditional taxonomy, species were discriminated mainly on the structure of the sporangium (non-papillate, semi-papillate, or papillate), the form of the antheridium (amphigynous or paragynous) and on whether the taxon is inbreeding (homothallic), or outbreeding with A1 and A2 sexual incompatibility, or mating types (heterothallic) (Tucker 1931; Waterhouse 1963). Heterothallic taxa are exclusively amphigynous while homothallic taxa may be

amphigynous, paragynous or, in some cases, have antheridia of both types. Waterhouse (1963) assigned *Phytophthora* taxa to six morphological groups which have provided the framework for a number of traditional identification keys (e.g. Stamps et al. 1990).

Many researchers have observed that there are considerable levels of morphological variation within and between *Phytophthora* species, which makes identification of some isolates to species level difficult. A number of *Phytophthora* species have been reclassified over the years. Numerous changes have taken place especially in the *P. megasperma* species complex. A number of species were morphologically indistinguishable from the complex although this was recognised and the use of *formae speciales* promoted (Hansen and Maxwell 1991).

*P. megasperma* was first described by Drechsler (1931). This first description was later broadened (Tompkins et al. 1936). In the 1950s a disease found on soybean in Illinois was designated as being caused by a new species. Since the morphology of this species was highly similar to the previously described *P. megasperma*, Hildebrand (1959) renamed it *P. megasperma* var. *sojae* as it showed high levels of host specificity towards soybean. In a reevaluation of the species, Kuan and Erwin (1980) showed a continuous distribution of oogonial size between the various

varieties within the *P. megasperma* complex and proposed the use of *formae speciales*. Since no simple morphological character distinguished the different *formae speciales*, this system was used quite extensively. However, with the advent of molecular taxonomy, the genetic relationships between the various species within this complex were tested and it was shown that various distinct biological species were lumped together in the *P. megasperma* species complex (Forster et al. 1989). The taxonomic status was subsequently reviewed by (Hansen and Maxwell 1991); one species was (*P. sojae*) reinstated and two others (*P. medicaginis* and *P. trifolii*) were created. The genetic relationship between these species is confirmed and illustrated in a more recent phylogeny of the genus *Phytophthora* (Cooke et al. 2000).

*Phytophthora palmivora*, which is probably the most important *Phytophthora* species in the tropics, also has undergone several changes in classification since Butler (1919) first described it. *Phytophthora palmivora* shows considerable morphological and pathological variation and, since the original description, a number of additions and delineations have been proposed. First *P. palmivora* strains were grouped together based on the host from which they were collected (Gadd 1924). This sometimes correlated with mating type, giving rise to further confusion, as reviewed by Zentmyer et al. (1977).

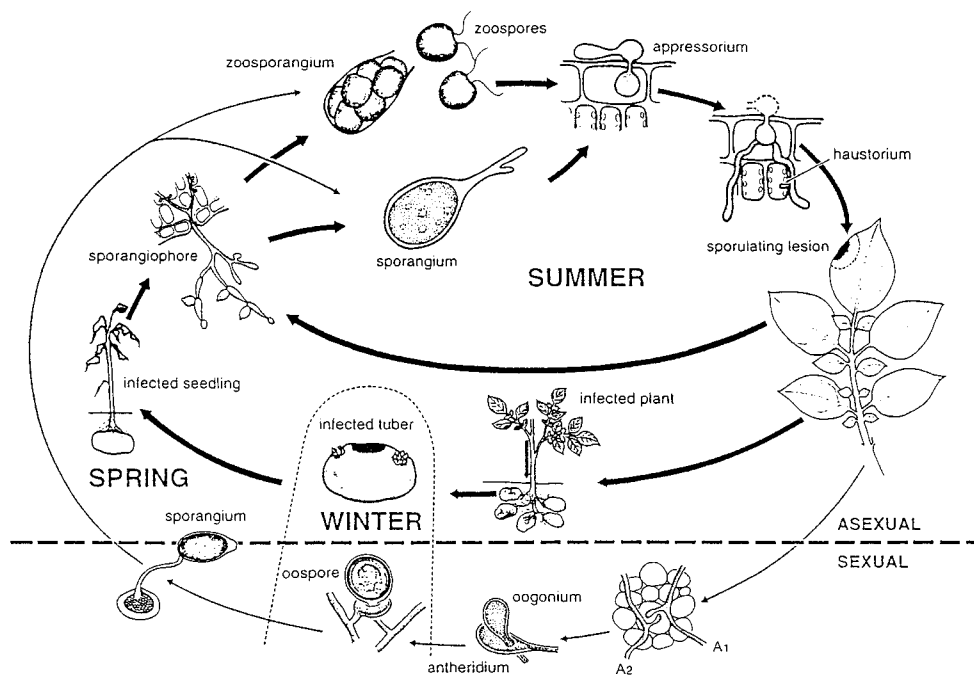


Figure 3.1.14 Life cycle of *Phytophthora infestans*. Reproduced from Dreth (1994).

Once it became clear that species other than *P. palmivora* could infect cocoa, and that each of these had a different and sometimes extensive host range (Brasier et al. 1981), a better delineation of species began to emerge. The species was first split into four different morphological groups, MF1–MF4. MF1 was the typical form of *P. palmivora*, while MF2 was, for some time, deemed to be morphological different. It was later found to be insufficiently different and lumped with MF1 again. MF3 was renamed *P. megakarya* (Brasier and Griffin 1979) based on pedicel length and chromosomal differences. MF4 was found to be closely related to *P. capsici* and thus this group was placed in the redescribed species *P. capsici* (Tsao 1991). These species reclassifications were later confirmed in the evolutionary analysis of the *Phytophthora* genus by (Cooke et al. 2000).

The above two examples illustrate the significant amount of variability in morphological and physiological characters that have to be taken into account when trying to classify organisms. From the outset one does not know the extent of variation present, while the boundaries that define biological species are not always straightforward. If we study only a few isolates of a few species at any point in time we are trying to complete an evolutionary jigsaw puzzle while holding only a few of the pieces. Determining where these few pieces go in this evolutionary puzzle often turns out a difficult task. It is clear that without large collections of the material from different hosts and regions under investigation, and an understanding of variability within and between species, it is difficult to resolve these matters. In the past decade, molecular taxonomy has provided an enormous insight into phylogenetic relationships between the various species. This has allowed testing of hypotheses concerning the delineation of difficult species complexes.

## Disease Cycle

### Primary inoculum

*Phytophthora* is basically a soil-borne organism, although species including *P. palmivora* are well adapted to attack aerial parts of plants causing diseases such as cankers, leaf blights and fruit rots. Primary inoculum initiates epidemics when environmental conditions are conducive. In the monsoonal tropics, this usually means the wet season, but in the wet tropics conditions conducive to the development of phytophthora diseases may persist throughout the year, enabling an unbroken disease cycle.

Primary inoculum of *Phytophthora* spp. survives as mycelium and chlamydospores in infected roots, soil, bark cankers and mummified fruits or pods. For example, unharvested, infected cocoa pods become mummified, develop sporangia during the rainy season and drop inoculum onto pods below every time it rains, for up to three years. Untreated bark and flower cushion cankers also develop and release sporangia that are carried in run-off water down the stem. Although both mating types are often present, oospores are relatively rarely formed in tropical species of *Phytophthora*. The role of oospores as a source of inoculum of heterothallic species, such as *P. palmivora*, in the tropics is poorly understood.

### Secondary inoculum

Once conditions conducive to the disease are present, primary inoculum germinates and establishes an infection. If this infection succeeds, a generation of secondary inoculum is produced which fuels propagation of the epidemic. The rate of propagation and the success of these propagules in causing new infections determines the slope of the disease progress curve — explosive epidemics are caused by the rapid increase in secondary inoculum. For example, although it only takes a single zoospore to initiate the infection of a cocoa pod, the lesion spreads rapidly and will release 4 million sporangia from a single pod within a week (Medeiros 1976). Sporangia also form on infected debris and roots on the surface of soil, and are released into water pooling on the surface, or into creeks, rivers and dams.

Sporangia are dislodged by water, wind, rapid changes in humidity or by contact with vertebrate or invertebrate vectors. Sporangia of many species germinate in the presence of free water, either in the soil, in ponds, or on films of water on aerial plant surfaces, to release around 30 zoospores. Zoospores swim and are attracted chemotactically and electrotactically to suitable penetration sites, such as stomata or anticlinal wall junctions. Zoospores may remain motile for several hours, but usually encyst within 30 minutes if host tissues are present. Encystment involves shedding of the flagella and the rapid deposition of a cell wall around the zoospore. Cysts germinate to form a germ tube that is also tactically attracted to suitable penetration sites.

While indirect germination of sporangia, through the release of zoospores, is common, some species may also germinate directly to form a germ tube. If no suitable hosts are located, a secondary sporangium may form. If host tissue is located, the germ tube forms an appressorium that attaches to

the host surface, then penetrates and infects. Successful colonisation results in the development of further infective or resting propagules.

### Movement of inoculum

A close examination of disease symptoms can give valuable insights into the biology and disease cycle of the pathogen. For example, phytophthora lesions may be initiated on various parts of a cocoa pod (Figure 3.1.15). Lesions beginning at the peduncle reflect either direct contact with a stem canker lesion, or with ant tents constructed with contaminated soil. Lesions beginning at the distal end of the pod indicate that the inoculum was borne in drops of water contaminated with pathogen propagules, most likely originating from pod mummies or stem cankers higher in the canopy, or from soil-splash on pods close to the ground. Lesions beginning on the side of pods are mostly associated with damage caused by flying insects, mammals or knife wounds.



**Figure 3.1.15** Naturally occurring *P. palmivora* pod rot on cocoa showing lesions initiated at (from left to right) the distal end, the peduncle end and at the pod equator.

This simple analysis reveals several sources of inoculum and several modes of dissemination of *P. palmivora* within cocoa canopies in Papua New Guinea. Inoculum moves from the soil into the canopy as a result of human activity, rain and soil-splash, tent-building ants, termites, slugs and flying beetles. The beetles breed in discarded pod cases, visit flowers and are attracted to pod lesions (Konam and Guest 2004). When they bore into pod lesions they release large amounts of easily dispersed, contaminated frass. Once in the canopy, secondary inoculum spreads to infect pods, cankers, flower cushions, leaves and chupons. Secondary inoculum moves to pods by direct contact, contaminated

implements, raindrops, ants, flying beetles and mammals.

Black stripe and patch canker of rubber caused by *P. palmivora* presents an unusual situation. The tapping operation creates a wound that facilitates pathogen entry, especially if the panel is close to soil splash from the ground. The tapping knife itself provides another means for spreading secondary inoculum from tree to tree.

Footrot, cankers, gummosis, seedling and leaf blights are commonly initiated by soil splash inoculum, where raindrops dislodge sporangia and zoospores on the soil surface or in pools and puddles of water onto the base of the stem and low-lying leaves. Root rots and root cankers are almost always initiated by the migration of zoospores in the soil water.

### Environment

The activation of primary inoculum, production and release of secondary inoculum and infection all depend on humidity and free moisture. Although symptoms appear year-round, the most severe epidemics coincide with the proliferation of sporangia and insect vectors during the wet season. Zoospores generally need 20–30 minutes in free water on the plant surfaces for the start of encystment and germination; then, given sufficient atmospheric moisture, those that have germinated will continue to grow. If susceptible plant surfaces remain wet for several hours, there is a high probability of infection if zoospores are present. Temperature rarely limits the development of phytophthora diseases in the tropics, other than in highland environments.

### Implications for disease management

Phytophthora disease cycles are complex, involve numerous sources of primary and secondary inoculum and several modes of dissemination. As an organism it is flexible and very well adapted to monsoonal and wet tropical environments.

Integrated disease management strategies should address numerous components of the disease cycle by selecting disease-resistant planting material, and preventing or disrupting the dissemination of primary inoculum from the soil into the canopy and the movement of secondary inoculum from one part of the canopy to another. Mixed plantings of genetically diverse plants, that include medicinal plants, herbs, fruit, vegetables and timber trees, may prevent rapid inoculum build-up and sustain farm productivity over a longer period. Treatments that increase soil microbial activity reduce the survival of

chlamydospores and mycelium in infected debris. Postharvest disease can be suppressed through fungicide treatments and low temperature storage.

## References

- Bartnicki-Garcia, S., and M.C. Wang. 1983. Biochemical aspects of morphogenesis in *Phytophthora*. In: Erwin, D.C., Bartnicki-Garcia, S. and P. H. Tsao, P.H., ed., *Phytophthora: its biology, taxonomy, ecology and pathology*. St Paul, Minnesota, USA, American Phytopathological Society.
- Beakes, G.W. 1998. Evolutionary relationship among protozoa. In: Coombs, G.H., Vickerman, K., Sleight, M.A. and Warren, A., ed., *The Systematics Association Special Volume Series 56*. Dordrecht, Netherlands, Kluwer Academic Publishers.
- Bourke, P.M. 1964. Emergence of potato blight. *Nature*, 203, 805–808.
- Brasier, P.M. 1992. Evolutionary biology of *Phytophthora*: I. Genetic systems, sexuality and the generation of variation. *Annual Review of Phytopathology*, 30, 134–135.
- Brasier, C.M. and Griffin, M.J. 1979. Taxonomy of *Phytophthora palmivora* on cocoa. *Transactions of the British Mycological Society*, 72, 111–143.
- Brasier, C.M., Griffin, M.J. and Maddison, A.C. 1981. The cocoa black pod *Phytophthoras*. In: Gregory, M.P.H. and Kew, A.C. *Epidemiology of Phytophthora on cocoa in Nigeria*. England, Commonwealth Mycological Institute.
- Butler, E.J. 1919. Report of the imperial mycologist 1910–1919. In: Scientific report, Research Institute of Pusa, India 1910–1919, 82.
- Cavalier-Smith, T. 1986. The kingdom Chromista: origin and systematics. In: Round, I. and Chapman, D.J., ed., *Progress in phycological research*. Bristol, England, Biopress.
- Cooke, D.E.L., Drenth, A., Duncan, J.M., Wagels, G. and Brasier, C.M. 2000. A molecular phylogeny of *Phytophthora* and related Oomycetes. *Fungal Genetics and Biology*, 30, 17–32.
- Desjardins, P.R., Zentmyer, G.A. and Reynolds, D.A. 1969. Electron microscopic observations of the flagellar hairs of *Phytophthora palmivora* zoospores. *Canadian Journal of Botany*, 47, 1077–1079.
- Drechsler, C. 1931. A crown rot of hollyhocks caused by *Phytophthora megasperma* n.sp. *Journal of the Washington Academy of Science*, 21, 513–526.
- Drenth, A. 1994. Molecular genetic evidence for a new sexually reproducing population of *Phytophthora infestans* in Europe. PhD thesis, Wageningen University, The Netherlands.
- Elliott, C.G. 1983. Physiology of sexual reproduction in *Phytophthora*. In: Erwin, D.C., Bartnicki-Garcia, S. and P. H. Tsao, P.H., ed., *Phytophthora: its biology, taxonomy, ecology and pathology*. St Paul, Minnesota, USA, American Phytopathological Society.
- Erwin, D.C. and Ribeiro, O.K. 1996. *Phytophthora* diseases worldwide. St Paul, Minnesota, USA, American Phytopathological Society Press.
- Forster, H., Kinscherf, T.G., Leong, S.A. and Maxwell, D.P. 1989. Restriction fragment length polymorphisms of the mitochondrial DNA of *Phytophthora megasperma* isolated from soybean, alfalfa, and fruit trees. *Canadian Journal of Botany*, 67, 529–537.
- Gadd, C.H. 1924. *Phytophthora faberi* Maubl. *Annals of the Royal Botanic Garden Peradeniya (Ceylon)*, 9, 47–89.
- Galindo, A.J. and Gallegly, M.E. 1960. The nature of sexuality in *Phytophthora infestans*. *Phytopathology*, 50, 123–128.
- Goodwin, S.B., Cohen, B.A., Deahl, K.L. and Fry, W.E. 1994. Panglobal distribution of a single clonal lineage of the Irish potato famine fungus. *Proceedings of the National Academy of Science*, 91, 11591–11595.
- Hansen, E.M. and Maxwell, D.P. 1991. Species of the *Phytophthora megasperma* complex. *Mycologia*, 83, 376–381.
- Hawksworth, D.L., Kirk, P.M., Sutton, B.C. and Pegler, D.N. 1995. *Ainsworth and Bisby's dictionary of the fungi*, 8th ed. Wallingford, UK, CAB International.
- Hendrix, J.W. 1970. Sterols in growth and reproduction of fungi. *Annual Review of Phytopathology*, 8, 111–130.
- Hildebrand, A.A. 1959. A root and stalk rot of soybeans caused by *Phytophthora megasperma* Drechsler var. *sojae* var. nov. *Canadian Journal of Botany*, 37, 927–957.
- Kennedy, D.M. and Duncan, J.M. 1995. A papillate *Phytophthora* species with specificity to *Rubus*. *Mycological Research*, 99, 57–68.
- Ko, W.H. 1978. Heterothallic *Phytophthora*: evidence for hormonal regulation of sexual reproduction. *Journal of General Microbiology*, 107, 15–18.
- Konam, J.K. and Guest, D.I. 2004. Role of flying beetles (Coleoptera: Scolytidae and Nitidulidae) in the spread of *Phytophthora* pod rot of cocoa in Papua New Guinea. *Australasian Plant Pathology*, in press.
- Kuan, T.L. and Erwin, D.C. 1980. *Formae speciales* differentiation of *Phytophthora megasperma* isolates from soybean and alfalfa. *Phytopathology*, 70, 333–338.
- Medeiros, A.G. 1976. Sporulation of *Phytophthora palmivora* (Butl.) Butl. in relation to epidemiology and chemical control of black pod disease. PhD thesis, University of California, Riverside, California, USA. Cited in Pereira (1992).
- Pereira, J.L. 1992. Cocoa and its pathogens in the region of origin: a continued risk. In: Keane, P.J. and Putter, C.A., ed., *Cocoa pest and disease management in Southeast Asia and Australasia*. Rome, Italy, FAO Plant Production and Protection Paper, No. 112.
- Stamps, D.J., Waterhouse, G.M., Newhook, F.J. and Hall, G.S. 1990. Revised tabular key to the species of *Phytophthora*. *Agricultural Bureau of International Mycology Institute, Institute of Mycology Paper*, No. 162.

- Tompkins, C.M., Richards, B.L., Tucker, C.M. and Gardner, M.W. 1936. Phytophthora rot of sugar beet. *Journal of Agricultural Research*, 52, 205–216.
- Tsao, P.H. 1991. The identities, nomenclature and taxonomy of *Phytophthora* isolates from black pepper. Paper read at Diseases of black pepper. In: Proceedings of the International Pepper Communication Workshop on Pepper Diseases, Goa, India.
- Tucker, C.M. 1931. Taxonomy of the genus *Phytophthora* de Bary. University of Minnesota, Agriculture Experimental Station, Research Bulletin, 153, 207p.
- van de Peer, Y., van der Auwera, G. and De Wachter, R. 1996. The evolution of stramenopiles and alveolates as derived by substitution rate calibration of small ribosomal subunit RNA. *Journal of Molecular Evolution*, 42, 201–210.
- van der Plaats-Niterink, A.J. 1981. Monograph of the genus *Pythium*. Baarn, Netherlands, Centraalbureau voor Schimmelcultures, Studies in Mycology No. 21.
- van West, P., Morris, B.M., Reid, B., Appiah, A.A., Osborne, M.C., Campbell, T.A., Shepherd, S.J. and Gow, N.A.R. 2002. Oomycetes plant pathogens use electric fields to target roots. *Molecular Plant–Microbe Interactions*, 15, 790–798.
- Wang, M.C. and Bartnicki-Garcia, S. 1973. Novel phosphoglucans from the cytoplasm of *Phytophthora palmivora* and their selective occurrence in certain life cycle stages. *Journal of Biological Chemistry*, 248, 4112–4118.
- Waterhouse, G. M. 1963. Key to the species of *Phytophthora* de Bary. Kew, Surrey, England, Commonwealth Mycological Institute, Mycological Papers.
- Whisson, S.C., Drenth, A., Maclean, D.J. and Irwin, J.A.G. 1994. Evidence for outcrossing in *Phytophthora sojae* and linkage of a DNA marker to two avirulence genes. *Current Genetics*, 27, 77–82.
- Zentmyer, G.A. 1980. *Phytophthora cinnamomi* and the diseases it causes. St Paul, Minnesota, American Phytopathological Society, Monograph No. 10.
- Zentmyer, G.A., Kaosiri, T. and Idosu, G. 1977. Taxonomic variants in the *Phytophthora palmivora* complex. *Transactions of the British Mycological Society*, 69, 329–332.



## 3.2 Infection Biology of *Phytophthora palmivora* Butl. in *Durio zibethinus* L. (Durian) and Responses Induced by Phosphonate

Emer O’Gara,<sup>1,2</sup> Somsiri Sangchote,<sup>3</sup> Laura Fitzgerald,<sup>1</sup>  
Damon Wood,<sup>1</sup> Ang Ching Seng<sup>1</sup> and David I. Guest<sup>1,4</sup>

### Abstract

We investigated the infection biology of *Phytophthora palmivora* on durian leaf and fruit. Zoospores of *P. palmivora* are preferentially attracted to fresh wounds in durian and such wounds are shown to be key infection courts. Overlapping layers of peltate trichomes cover the stipules, lower surface of the leaf, petiole, young stem and fruit, and are the first point of contact between the pathogen and the host on these tissues. The pathogen binds randomly to the surface of the trichomes but is unable to penetrate the heavily lignified walls, however the hypha can grow over the edge of the trichome until it reaches the epidermal surface beneath. The stomata that occur beneath the trichomes on all tissues are readily infected by the advancing hyphal strands, and are also major infection courts. When infection occurs through fresh wounds in leaves, lesions appear within 2 days and leaves are entirely diseased within 6 days. Treatment of durian seedlings with phosphonate before inoculation with *P. palmivora* led to a significant restriction of the pathogen, but only if leaves were inoculated while still attached to the tree, and not if they were excised before inoculation. Phenylalanine ammonia lyase activity was not stimulated in excised inoculated leaves from phosphonate-treated durian seedlings, compared to untreated seedlings.

### Introduction

An understanding of the infection biology of a host/pathogen interaction is essential in understanding the disease cycle, and ultimately in formulating effective disease management strategies.

*Phytophthora palmivora* Butl. is the most important pathogen of durian (*Durio zibethinus* L.), but there is no readily available information on the processes of

infection. However, there is a wealth of published information on a number of other phytophthora ‘pathosystems’ that cause significant economic and ecological damage. It has been repeatedly demonstrated in these other systems that morphological, anatomical and biochemical characteristics of the host largely determine the outcome of an encounter with the pathogen. The physical characteristics of the host become even more important when there is susceptibility to *Phytophthora* spp. with caducous sporangia, as is the case with durian and *P. palmivora*, as the pathogen has access not only to the root zone but to all the aerial tissues.

Chemical control of plant diseases is moving away from a total dependence on fungicides to the use of systemic compounds that alter the biochemistry of the interaction between host and pathogen, inducing the plant’s natural defence responses. One

<sup>1</sup> School of Botany, University of Melbourne, Victoria 3010, Australia.

<sup>2</sup> Current address: Centre for Phytophthora Science and Management, School of Biological Sciences, Murdoch University, Western Australia 6150, Australia.

<sup>3</sup> Department of Plant Pathology, Kasetsart University, Bangkok 10900, Thailand.

<sup>4</sup> Current address: Faculty of Agriculture, Food and Natural Resources, The University of Sydney, New South Wales 2006, Australia.

such compound, 'phosphonate' (salts or esters of phosphonic acid), has proven highly successful in controlling phytophthora diseases in a number of crops including avocado (Darvas et al. 1984), cocoa (Holderness 1990; Guest et al. 1994) and, more recently, black pepper (see Chapter 7.4) and durian (see Chapter 8.5). The mode of action of phosphonate is thought to be due to the disruption of phosphate metabolism in the pathogen, which causes the release of pathogen stress metabolites that activate host defence responses (Guest and Grant 1991). A key step in plant defence responses is activation of the enzyme phenylalanine ammonia-lyase (PAL) that is involved in the biosynthesis of phenylpropanoids, including phytoalexins, salicylic acid, lignin and suberin.

The major objective of the current study was to examine how the morphological and anatomical characteristics of durian influence infection by *P. palmivora*, and to investigate the effect of phosphonate on the infection process.

## Methods

The results presented in this chapter were derived from a number of separate studies conducted at The University of Melbourne in Australia and Kasetsart University in Thailand. As Melbourne is well outside the climatic range for durian, seedlings were grown in a temperature-controlled glasshouse and the histopathological studies concentrated on shoot tissues. The biology of fruit infection was studied in Thailand where durian fruit was readily available. Axenic cultures of *P. palmivora* were used as inoculum in all studies, as it is the species predominantly associated with durian diseases in Australia and Southeast Asia (see Chapter 6.7).

Durian fruit was inoculated with a sporangial suspension (400 sporangia per cm<sup>2</sup> leaf) and incubated at 25°C and approximately 98% relative humidity. Shoot tissues were inoculated with a motile zoospore suspension, covered with a plastic bag to maintain a high humidity and incubated in a temperature-controlled glasshouse. For some studies, leaves were wounded by deliberate removal of trichomes with adhesive tape. Standard histological techniques were used for the preparation of samples for examination by either light or scanning electron microscopy.

Durian seedlings were treated with phosphonate by pouring 500 mL of a 1 g/L a.i. solution of Foli-R-Fos 200 (UIM Agrochemicals (Aust.) Pty Ltd) onto the surface of the potting mix. Trays were placed beneath the pots to capture any drainage, plastic bags were placed around the tray and pot and tied

around the main stem for 24 hours to minimise the loss of the liquid through drainage and/or evaporation. To determine the effect of phosphonate on symptom development, leaves were wounded (see Chapter 8.3) and the wound inoculated with sporangia, either while still attached to the tree, or after the leaves were excised. Excision of leaves (or other organs including fruit or stems) before inoculation is a standard bioassay technique for ranking resistance to infection in germplasm collections (see Chapter 8.3). Attached leaves were covered with a plastic bag and aerated each day when symptoms were monitored, while the seedlings were maintained in the glasshouse. Excised leaves were incubated in a humid chamber in a constant temperature cabinet at 28°C and symptoms monitored daily.

Leaves from phosphonate-treated durian seedlings were excised and inoculated with *P. palmivora* sporangia. Leaves were not wounded before inoculation. Activity of PAL was determined by measuring the amount of L-phenylalanine converted to cinnamic acid in extract from tissue immediately surrounding the region of inoculation, according to the methods of El Modafar et al. (2001).

As lesion development and changes in cinnamic acid concentrations over time were linear, the data were analysed by calculating the slope of the lines for each treatment by regression and comparing slopes by analysis of variance (ANOVA) (Minitab Inc., Version 14).

## Surface Features of Durian and Their Influence on Pre-penetration Events

Motile zoospores of *P. palmivora* bind randomly and individually in low numbers to the smooth upper surface of the durian leaf, which has a continuous cuticle with no stomata or trichomes. The encysted zoospores readily germinate on the upper leaf surface but growth of the germ-tubes appears random, unlike growth at sites of preferential attraction as described below.

In *D. zibethinus*, trichomes occur on the lower leaf surface, petiole, young stem, the external surface of the stipule, and on fruit (except in the trough between the spines). Three distinct trichome types were identified on durian leaves: (i) glandular trichomes which are not lignified; (ii) stellate trichomes which vary in the level of lignification; and (iii) peltate trichomes which are heavily lignified and form the external layer (Figure 3.2.1) giving the lower leaf surface a silver to golden hue. Stomata occur in a random arrangement beneath the

trichomes on the petiole, young stem, lower leaf (although absent from the major veins) and fruit (although absent from the trough between the spines). There are no trichomes on durian roots.

A higher proportion of *P. palmivora* spores bind to the lower surface of the leaf, which is a function of the rough topography caused by the indumentum and trapping of spores at the ragged edges of the overlapping peltate trichomes. Under optimal environmental conditions, *P. palmivora* can bind, germinate, produce extensive hyphae and re-sporulate, thus completing its life cycle on the surface of the durian tissue within eight hours of inoculation (Figure 3.2.2).

Although successful penetration of heavily lignified peltate trichomes by *P. palmivora* was never observed, attempted penetration was marked by appressoria-like swellings and some dissolution of the trichome surface in the region of attachment (Figure 3.2.3). An unsuccessful attempt to penetrate was often followed by the formation of a hyphal branch from the swelling, growth of this hypha and attempted penetration at another site. This process could be repeated numerous times by a single zoospore/cyst (Figure 3.2.4). Invariably, some hyphae grow over the edge of the trichome and down to the surface of the tissue (Figure 3.2.5), where infection occurred through open stomata.

When trichomes were deliberately removed from the lower leaf surface *P. palmivora* did not show preferential attraction to the exposed stomata, and occasionally hypha grew across the stomatal pore with no attempt at penetration (Figure 3.2.6).

*Phytophthora palmivora* also showed no attraction to the axillary shoots of durian, probably due to the impressive trichome armour (Figure 3.2.7), which is already well developed on the leaf buds and external sides of the stipules before emergence.

### Infection Courts of Durian and Penetration by *Phytophthora palmivora*

Although there is no evidence that *P. palmivora* zoospores are preferentially attracted to stomata, they are clearly important infection courts as, more often than not, hyphae will infect through open stomata as they grow with apparent randomness across the surface of the tissue (Figure 3.2.8).

*Phytophthora palmivora* is preferentially attracted to fresh wounds in durian tissue. When trichomes were deliberately removed from leaves, taxis of zoospores

to the resulting fresh wound was evident through heavy and localised spore binding, and docking of the cysts with the side of germ-tube emergence directed toward the wound (Figure 3.2.9). The demonstrated importance of fresh wounds as infection courts led to the investigation of wound healing in durian leaves. Using histological stains, suberin was detected in the remnants of the trichome stalk within 24 hours of trichome removal (Figure 3.2.10), while lignin and callose were detected within 48 hours. The intensity of lignin (Figure 3.2.11) and callose (Figure 3.2.12) staining increased with time, which coincided with a decrease in number of spores binding to the wound.

*Phytophthora palmivora* can directly penetrate the cuticle and epidermis on the upper surface of the durian leaf and in the trichome-free region between the spines of the durian fruit (Figure 3.2.13), usually at the anticlinal wall between epidermal cells.

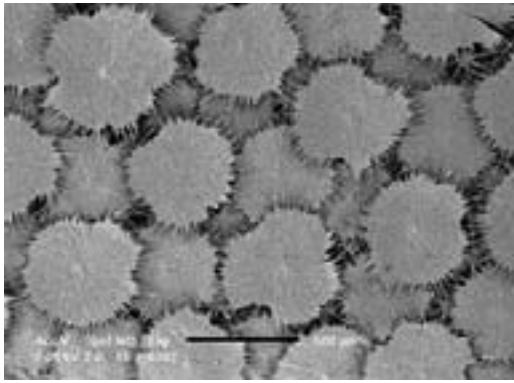
### Colonisation of Durian Tissues by *Phytophthora palmivora* and Symptom Development

*Phytophthora palmivora* rapidly colonised the entire leaf lamina when infection occurred through fresh wounds, and lesions were visible within 2 days of inoculation. The appearance of lesions resulting from a single *P. palmivora* isolate can be highly variable within and between trees, ranging from dark brown/black with a distinct margin to water-soaked light grey with a diffuse border.

When the pathogen infects through stomata of a durian leaf, colonisation is initially intercellular, particularly in the relatively open structure of, and surrounding, the sub-stomatal cavity. However, as the pathogen progresses through the leaf lamina into the more compacted mesophyll tissues, colonisation becomes increasingly intracellular (Figure 3.2.14).

Infection and symptom development in excised durian fruit did not occur unless high relative humidity (98%) was maintained for at least 72 hours after inoculation with *P. palmivora*.

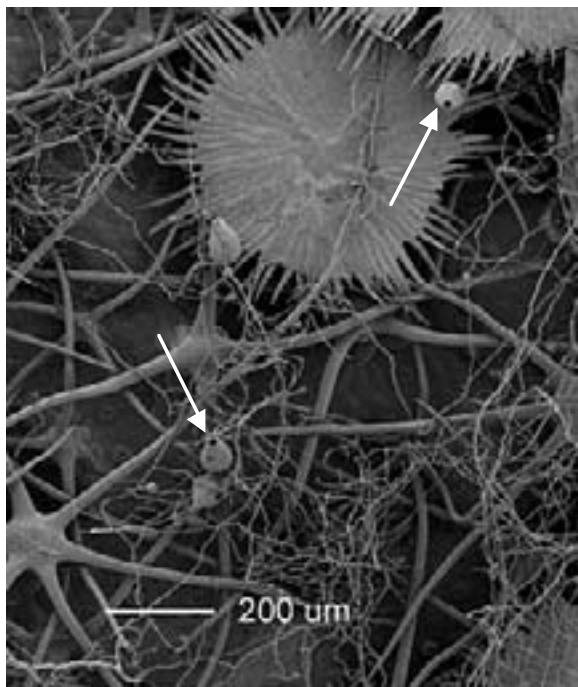
When penetration and infection is successful, the pathogen proliferates within the host and sporangiophores exit either through stomata or by erupting through the epidermis (Figure 3.2.15), releasing a new generation of sporangia into the environment (Figure 3.2.16). In disease-affected durian orchards, this is often seen as a whitish bloom on severely infected organs, particularly fruit (Figure 3.2.17).



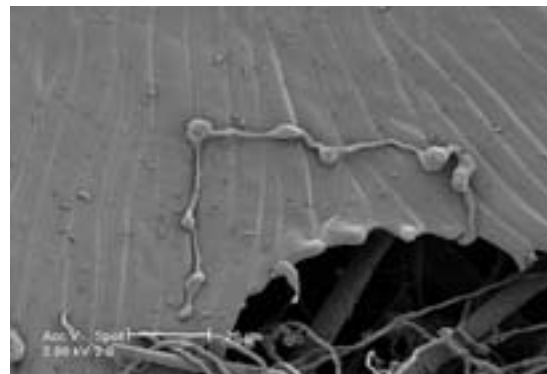
**Figure 3.2.1** A scanning electron micrograph of the overlapping peltate trichomes that form the external layer of the underside of the durian leaf. Scale bar = 500 μm.



**Figure 3.2.3** A scanning electron micrograph of a zoospore cyst of *Phytophthora palmivora* attempting to penetrate the surface of a lignified peltate trichome. The inset enlargement shows partial dissolution of the trichome surface (arrow). Scale bar = 10 μm.

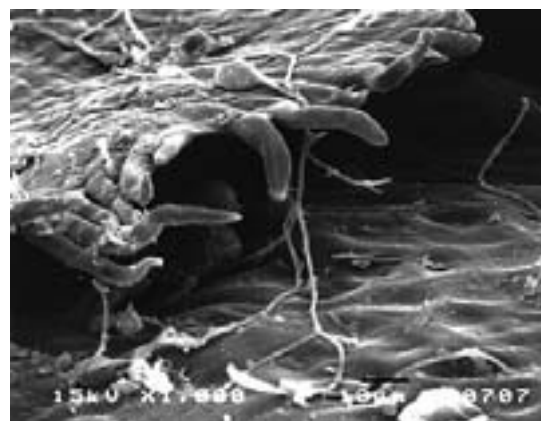


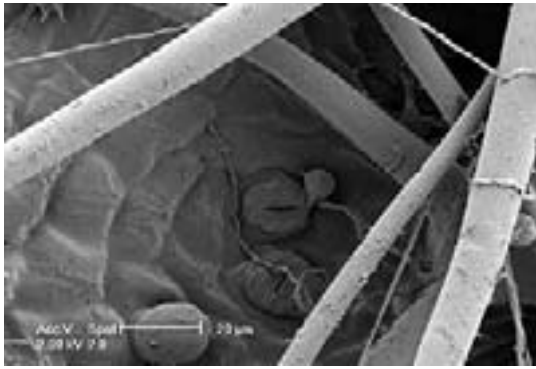
**Figure 3.2.2** Proliferation of *Phytophthora palmivora* among the trichomes on the lower surface of the durian leaf. Sporangia have formed and the open exit pores (arrows) indicate that the zoospores have been released. Scale bar = 200 μm.



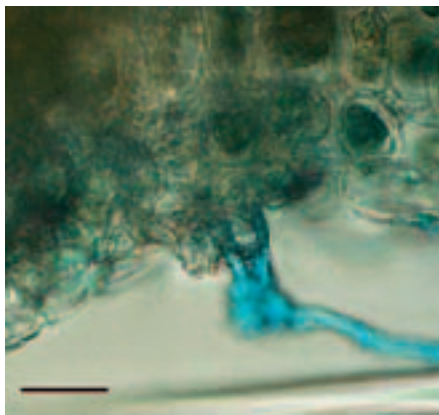
**Figure 3.2.4** A hypha of *Phytophthora palmivora* makes repeated attempts to penetrate a lignified peltate trichome of durian. Each attempt is marked by an appressorium-like swelling. Scale bar = 20 μm.

**Figure 3.2.5** A scanning electron micrograph of a *Phytophthora palmivora* sporangium which has germinated on the surface of a peltate trichome and grown over the edge to the epidermal surface of the durian fruit below. Scale bar = 10 μm.

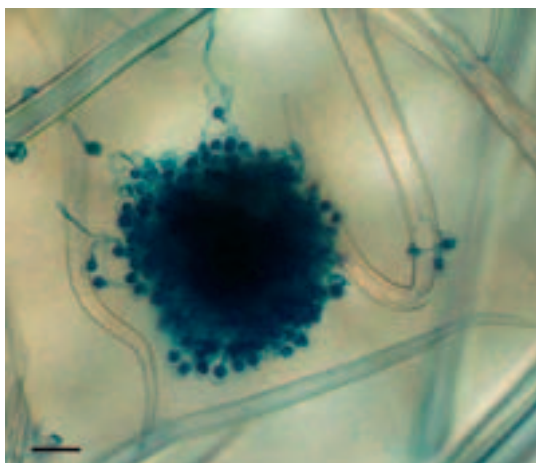




**Figure 3.2.6** A germinated cyst of *Phytophthora palmivora* which has grown over the top of a stoma on the lower surface of a durian leaf, with no attempt at penetration. Note: glandular trichome (arrow). Scale bar = 20  $\mu\text{m}$ .



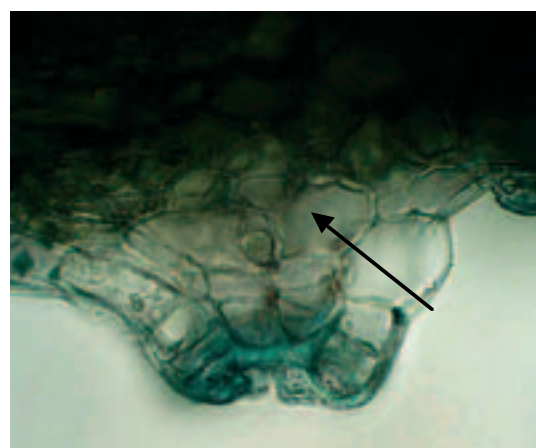
**Figure 3.2.8** A light micrograph of a whole leaf mount showing penetration of a stoma on the lower surface of a durian leaf by *Phytophthora palmivora* (stained with lactophenol cotton blue). Scale bar = 25  $\mu\text{m}$ .



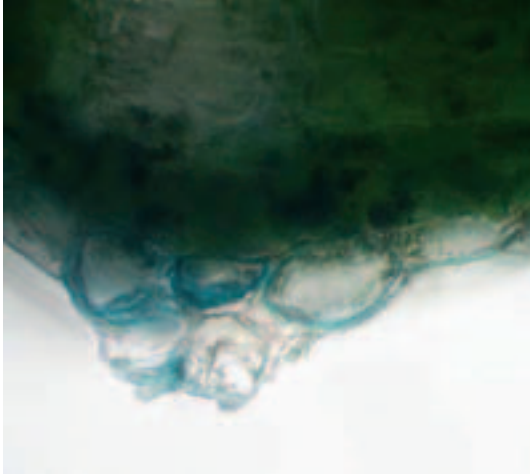
**Figure 3.2.9** A light micrograph of a cleared whole leaf mount showing large numbers of *Phytophthora palmivora* zoospore cysts (stained with lactophenol cotton blue) preferentially attracted to, and germinated on, a fresh wound on the lower surface of a durian leaf. Scale bar = 25  $\mu\text{m}$ .



**Figure 3.2.7** An axillary shoot of durian. One stipule has been excised and placed to the right to show the newly formed and as yet still folded leaf (arrow) and a new generation of shoots within the fused stipules on the left. The external surface of the stipules, folded leaf and young bud are covered with peltate trichomes, but the inner side of the stipule (circle) is free of peltate trichomes. Bars on left = 1 mm spacings.



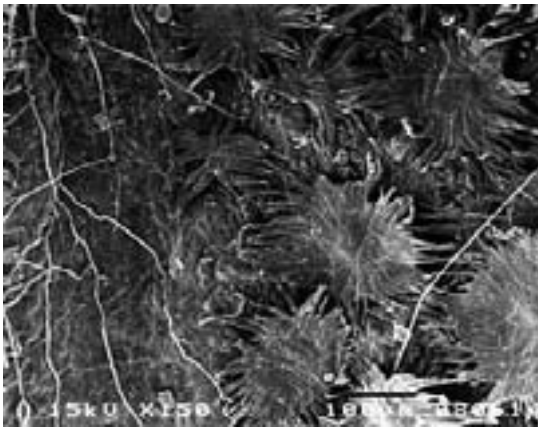
**Figure 3.2.10** Deposition of suberin (blue/grey stain) 48 hours after the deliberate removal of a peltate trichome from a durian leaf. Suberin was detected with Sudan Black B and is deposited mainly in the cells at the point where the trichome was attached ( $\times 400$ ).



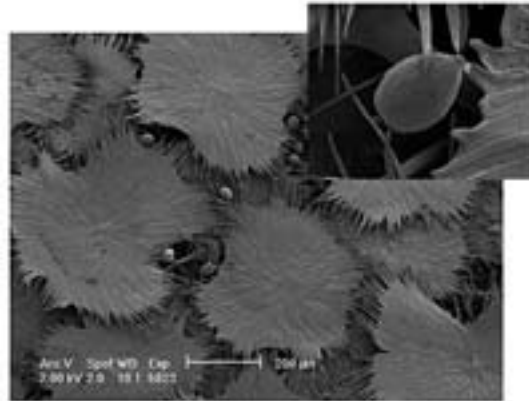
**Figure 3.2.12** Deposition of callose (blue staining) 2 weeks after the deliberate removal of a peltate trichome from a durian leaf. Callose was detected with resorcinol blue and was deposited around the wall of cells throughout the trichome mound ( $\times 400$ ).



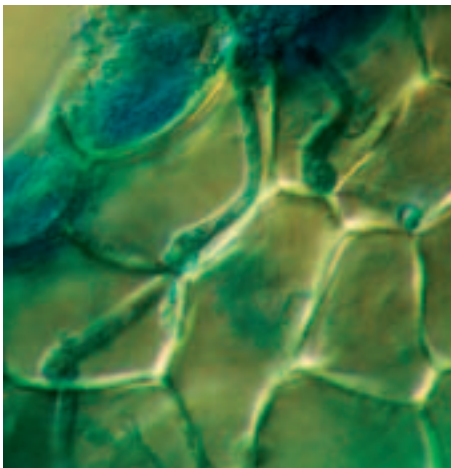
**Figure 3.2.15** A scanning electron micrograph of sporangia of *Phytophthora palmivora* produced as secondary inoculum and emerging above the peltate trichomes on a heavily infected durian fruit. Scale bar = 10  $\mu\text{m}$ .



**Figure 3.2.13** A scanning electron micrograph of zoospore cysts of *Phytophthora palmivora* that have bound and germinated in the trichome-free region between the spines on the durian fruit. Scale bar = 100  $\mu\text{m}$ .



**Figure 3.2.16** Sporangia of *Phytophthora palmivora* at the surface of a durian leaf where they can be readily distributed to the wider environment by water, insects and possibly wind. Sporangia have become detached from the hyphal body that has erupted through the surface of infected tissues beneath the trichomes. The inset picture is an enlargement of the sporangium within the square.



**Figure 3.2.14** Differential interference contrast micrograph showing intracellular growth of *Phytophthora palmivora* in a durian leaf, stained with lactophenol cotton blue. Note the swelling of the hypha at the point of wall penetration indicating the direction of growth ( $\times 400$ ).



**Figure 3.2.17** A durian fruit infected with *Phytophthora palmivora*. The white bloom in the middle of the lesion is hyphae and sporangia that have formed on the surface of the lesion.

### The Effect of Phosphonate on Symptom Development and the Activity of PAL in Durian

The treatment of durian seedlings with 1 g a.i. phosphonate led to significantly smaller lesions ( $F_{1,8} = 8.14; p = 0.02$ ) when attached leaves were inoculated with *P. palmivora* (Figure 3.2.18). Within 9 days of inoculation, all leaves from the untreated trees were totally diseased or had abscised. In contrast, leaves from the phosphonate-treated seedlings were still attached 21 days after inoculation and lesion development was restricted (Figure 3.2.19).

No significant difference in lesion size between phosphonate-treated and untreated seedlings was observed when leaves were detached before inoculation (Figure 3.2.18), although lesions from the untreated seedlings were surrounded by a chlorotic halo not present in leaves from phosphonate-treated seedlings (Figure 3.2.20).

Activity of PAL significantly ( $P < 0.05$ ) increased within 48 hours of inoculating detached leaves of durian with *P. palmivora* whether the leaves were from treated or untreated seedlings (Figure 3.2.21). The apparently higher levels of PAL activity in inoculated leaves from phosphonate-treated seedlings compared to leaves from untreated seedlings were not significant (Figure 3.2.21).

### Discussion

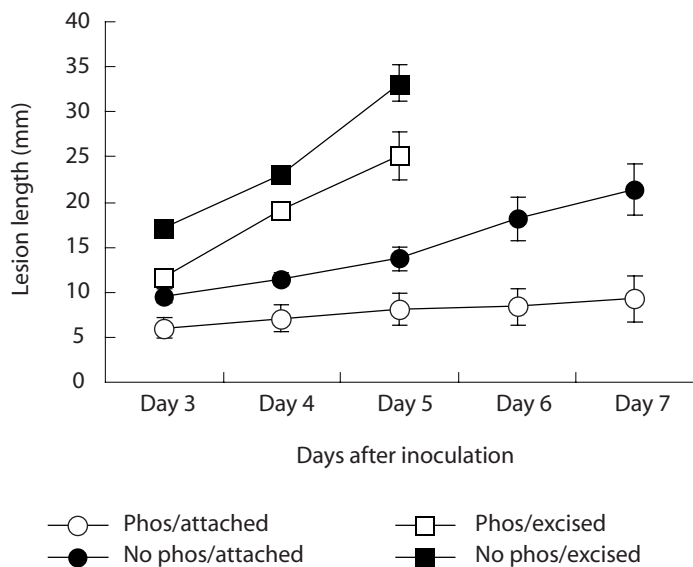
*Phytophthora palmivora* is attracted to fresh wounds in durian which make them key infection courts. Taxis of zoospore/cysts was evident from the manner in which they amassed on the wound with germ-tubes aligned toward it, as *Phytophthora cinnamomi* propagules have been shown to do in zones of chemotaxis on roots (Hardham and Gubler 1990).

Natural wounds or those caused by pruning are considered key infection courts of *Phytophthora syringae* in apple (Sewell and Wilson 1964) and *P. citricola* in avocado (El-Hamalawi et al. 1995). Leaf scars have been identified in apple and peach as infection courts for *Nectria galligena* (Crowdy 1952) and *Leucostoma* spp. (Biggs 1997), respectively, and should be examined in durian as potential sites of ingress for *P. palmivora*, given the pathogen's attraction to fresh wounds and the potential for tree injury during typhoons.

We have shown that it takes 24–48 hours from the time of wounding for suberin, callose and lignin to become visually detectable in durian leaves, and this is likely to take longer in woody organs. We have also shown that under optimal conditions, the pathogen infects, ramifies in tissue and reproduces very rapidly and would thus be able to produce many generations of propagules in the time taken to wound healing. Consequently, care should be taken to prune durian when weather conditions are not conducive to disease, and treatment of cut surfaces should be considered.

Stomata have been identified in a previous study as infection courts of *P. palmivora* in cocoa pods (Iwara et al. 1997). While this is also the case in durian, there appeared to be no preferential attraction and stomata were penetrated by a single hypha.

Trichomes are a common feature of species in the Bombacaceae family (Metcalf and Chalk 1950), but are more complex in *Durio* than in other genera (Baas 1972). The absence of the overlapping peltate trichomes from the trough between the spines on the durian fruit make it particularly vulnerable to infection. However, trichomes on other organs such as the young stem, petiole and the underside of the leaf do not always protect from infection, as the pathogen can grow extensively before penetration (presumably utilising an endogenous nutrient supply) with the potential to grow over the side of the trichome to the underlying epidermis, including the stomata.



**Figure 3.2.18** Mean lesion length resulting from inoculation with *Phytophthora palmivora* of leaves from phosphonate-treated or untreated durian seedlings when leaves were either excised before inoculation or inoculated while still attached to the seedling.

Although fresh wounds and stomata are considered key infection courts, *P. palmivora* is capable of direct penetration of leaf and fruit tissues. However, direct penetration by hyphae was observed rarely, compared to extremely common stomatal infections. It is unlikely that direct penetration would cause significant disease in healthy tissues but this mode of infection probably becomes increasingly important in ripening fruit.

Opportunistic infections such as stomatal and direct infections are made possible by the ability of *P. palmivora* zoospores to randomly bind and germinate on the rough surface of durian produced by trichomes. Randomly bound spores of *P. palmivora* attempted to penetrate the heavily lignified trichomes at 'appressoria-like' swellings, but were apparently unsuccessful as hypha emerged from the swelling and resumed growth across the tissue. According to Emmett and Parbery (1975), the definition of a 'true' appressorium is any structure that adheres to the host surface, with the ability to germinate and penetrate through the production of an infection peg.

'Appressoria-like' swellings were also produced by *P. cinnamomi* with apparently unsuccessful attempts to penetrate phellem cells of *Eucalyptus marginata* (O'Gara 1998). These types of structures have been observed in other phytophthora pathosystems (Beagle-Ristaino and Rissler 1983; Swiecki and McDonald 1988). Hardham (2001) suggests they are often associated with attempted penetration of

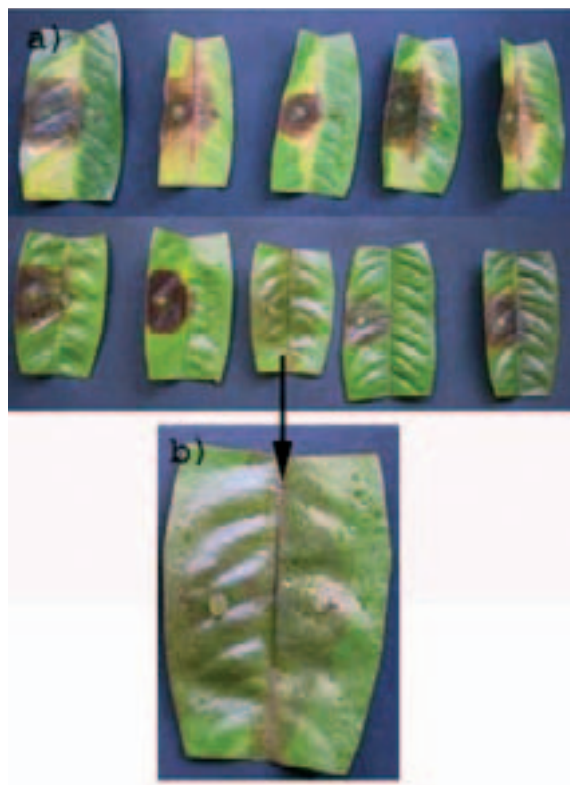
relatively resistant tissues — for example, they occur when *P. cinnamomi* penetrates the periclinal root wall of onion but not the anticlinal wall.

*Phytophthora cinnamomi* is preferentially attracted to the axillary shoots of jarrah and they are considered key infection courts (O'Gara 1998). In contrast, *P. palmivora* showed no attraction to the axillary shoots of durian. However, the emerging shoots of durian are well protected by trichome-covered stipules and by the time of bud opening, the lower surface of the leaf has a trichome covering, while emerging jarrah shoots are devoid of stipules, trichomes and indeed the leaf cuticle is either extremely thin or absent (O'Gara 1998).

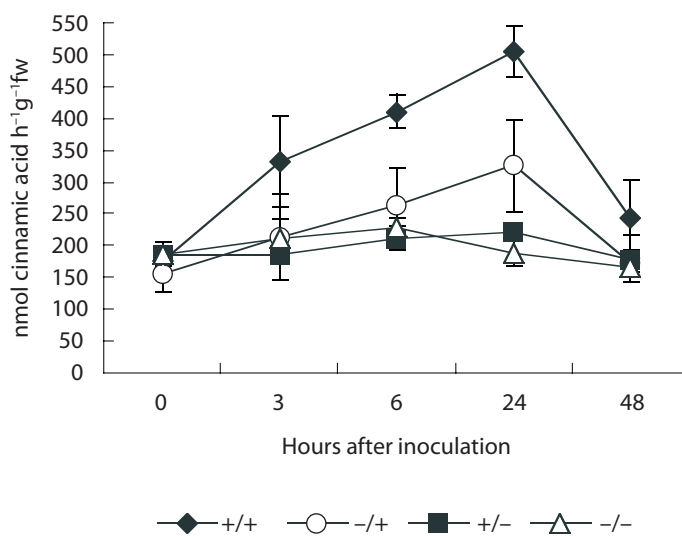


**Figure 3.2.19** A highly restricted lesion on a leaf from a phosphonate-treated durian seedling, 21 days after inoculation with *Phytophthora palmivora*. Scale bar = 1 cm.





**Figure 3.2.20** Lesion development in excised durian leaves 4 days after inoculation with *Phytophthora palmivora* on the left side of the mid-vein (a). Although there was no difference in size, lesions from untreated seedlings (top row) were surrounded by a chlorotic halo while leaves from phosphonate-treated seedlings (bottom row) were not. Note the variability in lesion appearance between leaves from the phosphonate-treated (bottom row) seedlings. There appears to be no lesion on the middle leaf, but the enlarged photo (b) shows that it has a water-soaked, pale lesion with diffuse boundary which has almost entirely covered the leaf.



**Figure 3.2.21** Phenylalanine ammonia-lyase (PAL) activity in leaves from phosphonate-treated and untreated seedlings of durian that were inoculated with *Phytophthora palmivora* after excision. Treatment combinations include: (+/+) = phosphonate and *P. palmivora*; (-/+) no phosphonate and *P. palmivora*; (+/-) = phosphonate and no *P. palmivora*; and (-/-) = no phosphonate and no *P. palmivora*.

It was originally hoped that an excised leaf bioassay, such as that developed to assess natural resistance to *P. palmivora* (see Chapter 8.3), could also be used to estimate phosphonate concentrations in durian tissues. However, the effect of phosphonate could not be demonstrated in leaves that were excised before inoculation, but was readily demonstrated

when attached leaves were inoculated, through highly restricted symptom development. It appears that the excision of leaves may interrupt the phenylpropanoid pathway signalling, as there was no significant difference in lesion development or PAL activity in excised inoculated leaves from untreated or treated seedlings.

The information gathered in the current study, coupled with field observations, improves our understanding of infection biology in durian orchards. Under mild weather conditions, the pathogen is at relatively low levels and individual spores infect individual stomata (if they can first negotiate the trichome armour), or isolated propagules directly penetrate through the epidermis. However, the host can resist these limited attacks and disease levels remain low. When conditions are extreme though, such as during typhoons, cyclones, or when 5–6 days of continuous rainfall occur, as can happen during the monsoon, the inoculum levels increase rapidly and tree injury provides numerous infection courts to which large numbers of zoospore/cysts are attracted. The pathogen reproduces faster than the infection-court-wounds heal. The synergism of the amassed spores enables the pathogen to overcome the host's capacity to impede the growth of a single hypha (Hinch et al. 1995). The pathogen ramifies in the infected tissues, and erupts through the surface of the organ, releasing more propagules to fuel the epidemic. The 'multi-cyclic' nature of the infection biology just described in durian is similar to that of *P. palmivora* in cocoa, which has been called a 'compound continuous interest' disease (MacKenzie et al. 1983; see also Chapter 6.2).

In conclusion, the current study has provided new information on the infection biology of *P. palmivora* in durian. While the information presented in this paper is extremely valuable from a purely academic perspective, it has also assisted in the understanding of the disease aetiology and epidemiology and was a key component in the formulation of integrated disease management options for phytophthora diseases in durian. However, there is much more that could be learnt about the host/pathogen interaction at a cellular and molecular level, which would enable fine-tuning of current recommendations.

## References

- Baas, P. 1972. The vegetative anatomy of *Kostermansia Malayana* Soegeng. *Reinwardtia*, 8(2), 335–344.
- Beagle-Ristaino, J.E. and Rissler, J.F. 1983. Histopathology of susceptible and resistant soybean roots inoculated with zoospores of *Phytophthora megasperma* f. sp. *glycinea*. *Phytopathology*, 73, 590–595.
- Biggs, A.R. 1997. Genetic and temporal variation in abscission zone formation in peach leaves in relation to peach canker disease. *Canadian Journal of Botany*, 74, 717–722.
- Crowdy, S.H. 1952. Observations on apple canker 4. The infection of leaf scars. *Annals of Applied Biology*, 39, 569–580.
- Darvas, J.M., Toerien, J.C. and Milne, D.L. 1984. Control of avocado root rot by trunk injection with fosetyl-Al. *Plant Disease*, 68, 691–693.
- El-Hamalawi, Z.A., Menge, J.A. and Guillemet, F.B. 1995. Infection court and factors affecting the expansion of stem canker of avocado caused by *Phytophthora citricola*. *Plant Disease*, 79(4), 384–388.
- El Modafar, C., Tantaoui, A. and El Boustani, E. 2001. Differential induction of phenylalanine ammonia-lyase activity in date palm roots in response to inoculation with *Fusarium oxysporum* f. sp. *albedinis* and to elicitation with fungal wall elicitor. *Journal of Plant Physiology*, 158, 715–722.
- Emmett, R.W. and Parbery, D.G. 1975. Appressoria. *Annual Review of Phytopathology*, 13, 147–167.
- Guest, D.I., Anderson, R.D., Phillips, D.A., Foard, H.J., Worboys, S. and Middleton, R.M. 1994. Long-term control of *Phytophthora* diseases of cocoa using trunk-injected phosphonate. *Plant Pathology*, 43, 479–492.
- Guest, D.I. and Grant, B.R. 1991. The complex action of phosphonates in plants. *Biological Reviews*, 66, 159–187.
- Hardham, A.R. 2001. The cell biology behind *Phytophthora* pathogenicity. *Australasian Plant Pathology*, 30, 91–98.
- Hardham, A.R. and Gubler, F. 1990. Polarity of attachment of zoospores of a root pathogen and pre-alignment of the emerging germ tube. *Cell Biology International Reports*, 14, 947–956.
- Hinch, J.M., Wetherbee, R., Mallett, J.E. and Clarke, A.E. 1985. Response of *Zea mays* roots to infection with *Phytophthora cinnamomi*. 1. The epidermal layer. *Protoplasma*, 126, 178–187.
- Holderness, M. 1990. Efficacy of neutralised phosphonic acid (phosphorous acid) against *Phytophthora palmivora* pod rot and canker of cocoa. *Australasian Plant Pathology*, 19(4), 130–131.
- Iwano, A.D., Sreenivasan, T.N. and Umaharan, P. 1997. *Phytophthora* resistance in cacao (*Theobroma cacao*): influence of pod morphological characteristics. *Plant Pathology*, 46, 557–565.
- MacKenzie, D.R., Elliott, V.J., Kidney, B.A., King, E.D., Royer, M.H. and Theberge, R.L. 1983. Application of modern approaches to the study of the epidemiology of diseases caused by *Phytophthora*. In: Erwin, D.C., Bartnicki-Garcia, S. and Tsao, P.H., ed., *Phytophthora: its biology, taxonomy, ecology and pathology*. St Paul, Minnesota, USA, APS Press, 303–313.
- Metcalf, C.R. and Chalk, L. 1950. *Anatomy of dicotyledons*, volume 1. Oxford, Clarendon Press. 724p.
- O'Gara, E. 1998. Infection and disease of *Eucalyptus marginata* (jarrah) caused by *Phytophthora cinnamomi* in rehabilitated bauxite mines in the south-west of Western Australia. PhD thesis, Murdoch University, Western Australia.

Sewell, G.W.F. and Wilson, J.F. 1964. Death of maiden apple trees caused by *Phytophthora syringae* Kleb. and a comparison of the pathogen with *P. cactorum* (L. & C.) Schroet. *Annals of Applied Biology*, 53, 275–280.

Swiecki, T.J. and MacDonald, J.D. 1988. Histology of chrysanthemum roots exposed to salinity stress and *Phytophthora cryptogea*. *Canadian Journal of Botany*, 66, 280–288.

### 3.3 Morphological and Host Range Variability in *Phytophthora palmivora* from Durian in Thailand

R. Pongpisutta<sup>1,2</sup> and S. Sangchote<sup>2</sup>

#### Abstract

Comparative morphological, physiological, and pathological tests showed that all isolates of *Phytophthora* isolated from durian orchards in Thailand are *Phytophthora palmivora*. Sporangia of 26 isolates were caducous with short pedicels (2.8–4.2 µm), but were variable in shape and size. The cultures produce ovoid, ellipsoid, obpyriform, ovoid-obpyriform, and spherical sporangia, average 35 to 90 µm in length and 22 to 62 µm in breadth, and have a length/breadth ratio of 1.6 to 2.0. The *P. palmivora* isolates also caused brown lesions on black pepper and rubber.

#### Introduction

The oomycete *Phytophthora palmivora* Butl. is a serious pathogen of durian, causing trunk canker, fruit and root rot in Thailand. *P. palmivora* infects durian fruit at the ripening stage and causes a soft brown lesion on the skin (Lim 1990; Pongpisutta and Sangchote 1994). Many durian plantation areas in the south and east of Thailand are close to rubber and black pepper plantation areas, and some growers plant these trees as intercrops in durian orchards. Orellana (1959) studied the pathogenicity of *P. palmivora* isolated from cocoa in which it was causing black pod rot, and from rubber a *Phytophthora* species causing fruit rot and defoliation. When unwounded leaves, petioles and terminal buds of young rubber seedlings were inoculated with *P. palmivora* from cocoa, disease occurred within 7–8 days, but isolates of the *Phytophthora* species from rubber did not cause symptom development after inoculation on comparable parts of cocoa plants under the same conditions in the greenhouse and laboratory. Since at least seven different *Phytophthora* species (Erwin and Ribeiro 1996) have the ability to cause disease on rubber, the identification of this particular isolate remains unknown. Tsao and Tummakate (1977)

collected a number of *Phytophthora* isolates causing foot and root rot disease from a black pepper plantation Amphur Palien in Trang province in southern Thailand. The Thai black pepper isolates produced narrow, ellipsoid, obovoid, pyriform sporangia with a tapered base, instead of the rounded or hemispherical base common in *P. palmivora*. However, using Tsao (1991), these isolates would probably at present be identified as *P. capsici* (= *P. palmivora* MF4).

The aims of the research described in this paper were: (i) to determine the *Phytophthora* species occurring on durian in Thailand; (ii) to determine the range of morphological characteristics from *Phytophthora* isolates obtained; and (iii) to investigate the host range of *Phytophthora* from durian. This information may be useful for identifying *Phytophthora* species, for choosing planting sites and for determining intercropping practices. Since *P. palmivora* has a wide host range including a number of important food crops in the tropics, recommendations for intercropping need to be backed up by long-term field experiments involving intercropping of hosts susceptible to *P. palmivora* in this area.

#### Materials and Methods

##### Isolation of the pathogen

Isolates of *P. palmivora* were obtained from soils and diseased leaves, branches, and stems of durian from

<sup>1</sup> Faculty of Agriculture, Food and Natural Resources, University of Sydney, New South Wales 2006, Australia.

<sup>2</sup> Department of Plant Pathology, Kasetsart University 10900, Thailand.

different locations in eastern Thailand (Table 3.3.1). Isolations from durian were assessed using a tissue transplanting method. Tissue was cleaned under running tap water and plated on selective agar containing benomyl (10 ppm), nystatin (50 ppm), pentachloronitrobenzene (PCNB) (25 ppm), ampicillin (500 ppm), rifampicin (10 ppm) and hymexazol (45 ppm). For soil samples, a baiting technique was used. Small pieces of fresh durian leaves were exposed to soil for 2 days before placing on fresh selective agar.

### Morphological characteristics

Colony characteristics were assessed on potato dextrose agar (PDA) after incubation at 25°C under near ultraviolet (UV) light for 5 days.

*Phytophthora* cultures were grown on carrot agar (CA) and incubated at 25°C under near UV for 7 days. Morphological characteristics of the asexual structure assessed included sporangia morphology (shape, size and length–breadth ratio), presence of papilla, caducity and chlamydospore production. These characters were determined by light microscopy of lactophenol-mounted slides.

### Growth temperatures

Small discs of agar were cut from all isolates using a 5 mm cork borer, then placed on CA and incubated

at 10, 15, 20, 25, 30, 35, and 37°C for 7 days, after which the colony diameter was measured.

### Pathogenicity test

Isolates of *Phytophthora* from durian were tested for pathogenicity against durian, rubber and black pepper by artificial inoculation of wounded leaves. A needle was used to wound leaves before placing 5 mm mycelial discs from each isolate upon separate wounds. The inoculated leaves were incubated in plastic bags for 24 hours. Pathogenicity was measured as the length–breadth ratio of brown lesions 5 days after inoculation.

## Results

### *Phytophthora* cultures obtained

Twenty- six isolates were recovered from infected parts of durian such as fruit, stem, leaf and branch (Table 3.3.1). These morphological characteristics of the isolates were compared to the *P. palmivora* description in Erwin and Ribeiro (1996).

### Morphological characteristics

Most cultures grew on PDA with a stellate pattern, except for P09, P27, P31, and P33, which were radiate, irregular and slightly fluffy, slightly petallate and stoloniferous colonies, respectively (Table 3.3.2 and Figure 3.3.1).

**Table 3.3.1** Sources of *Phytophthora* isolated from durian in Thailand.

Isolate no.	Host tissue	Location	District	Province	Year of collection
P01	fruit	Toong Benja,	Tamai	Chantraburi	1997
P03	fruit	Toong Benja	Tamai	Chantraburi	1997
P04	fruit	Sagthai	Tamai	Chantraburi	1997
P05	fruit	Khao Baisri	Tamai	Chantraburi	1997
P07	fruit	Khao Baisri	Tamai	Chantraburi	1997
P09	fruit	Khao Baisri	Tamai	Chantraburi	1997
P10	fruit	Khao Baisri	Tamai	Chantraburi	1997
P12	fruit	Toong Benja	Tamai	Chantraburi	1998
P14	fruit	Toong Benja	Tamai	Chantraburi	1998
P17	soil	Sagthai	Tamai	Chantraburi	1998
P19	stem	Ta Chang	Mueng	Chantraburi	1998
P21	fruit	Mueng	Mueng	Prachin Buri	1998
P22	stem	Mueng	Mueng	Prachin Buri	1998
P23	stem	Mueng	Mueng	Chantraburi	1999
P25	leaf	Praneet	Khao Saming	Trat	1999
P26	branch	Praneet	Khao Saming	Trat	1999
P27	leaf	Khao Saming	Khao Saming	Trat	1999
P29	stem	Sagthai, Tamai	Tamai	Chantraburi	1999
P31	branch	Toong Kwai Hin	Klang	Rayong	1999
P32	leaf	Toong Kwai Hin	Klang	Rayong	1999
P33	branch	Toong Kwai Hin	Klang	Rayong	2000
P35	fruit	Ta Chang	Mueng	Chantraburi	2000
P36	fruit	Ta Chang	Mueng	Chantraburi	2000
P37	fruit	Khao Saming	Khao Saming	Trat	2000
P38	stem	Lang Suan	Lang Suan	Chumphon	2000
P39	branch	Lang Suan	Lang Suan	Chumphon	2000

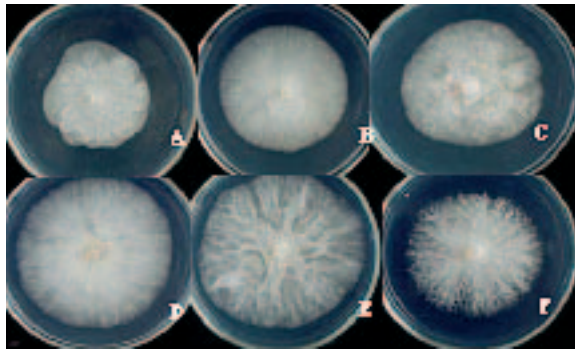
Table 3.3.2 Asexual characteristics of *Phytophthora* from durian in Thailand

Isolate no.	Chlamydospores			Sporangia				
	Colony pattern	Type	Average diameter (µm)	Pedicel length (µm)	Length (µm)	Breadth (µm)	L:B ratio	Shape
P32	stellate	terminal	36	3.9	48-72	28-38	1.8	Ovoid-obpyriform, Ellipsoid
P25	stellate	terminal & intercalary	34	3.4	48-72	28-40	2.0	Ovoid-obpyriform, Ellipsoid, Obpyriform
P31	slightly petallate	terminal	34	3.0	48-72	25-38	1.8	Ovoid-obpyriform, Ellipsoid, Obpyriform
P35	stellate	terminal	37	4.1	50-75	28-42	1.8	Ovoid-obpyriform, Ellipsoid, Obpyriform
P39	stellate	terminal	33	3.0	48-80	28-42	1.8	Ovoid-obpyriform, Ellipsoid, Obpyriform
P07	stellate	terminal	34	2.9	43-80	28-40	1.8	Ovoid-obpyriform, Ellipsoid, Obpyriform, Ovoid
P01	stellate	terminal & intercalary	36	4.0	50-73	28-45	1.9	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P04	stellate	terminal	36	4.2	38-75	25-35	2.0	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P09	radiate	terminal	34	2.8	40-65	28-45	1.6	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P10	stellate	terminal	32	2.9	41-65	29-44	1.7	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P14	stellate	terminal	37	2.8	48-78	28-42	1.9	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P19	stellate	terminal & intercalary	36	3.5	48-70	28-48	1.7	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P22	slightly stellate	terminal	38	4.4	50-70	28-42	1.8	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P23	stellate	terminal	34	3.5	45-72	25-42	1.8	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P27	irregular	terminal	34	3.3	45-78	25-40	1.9	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P29	stellate	terminal	34	3.4	40-78	28-40	1.8	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P36	stellate	terminal	34	3.6	42-85	22-40	1.9	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P38	stellate	terminal	39	3.3	45-90	28-50	1.8	Ovoid-obpyriform, Ellipsoid, Obpyriform, Spherical
P12	stellate	terminal	38	3.2	45-72	28-45	1.6	Ovoid-obpyriform, Obpyriform, Spherical
P17	stellate	terminal & intercalary	32	3.4	42-75	25-38	1.8	Ovoid-obpyriform, Obpyriform, Spherical
P21	stellate	terminal	36	3.0	48-82	28-42	1.8	Ovoid-obpyriform, Obpyriform, Spherical
P33	stoloniferous	terminal	30	3.9	35-65	22-38	1.7	Ovoid-obpyriform, Obpyriform, Spherical
P37	stellate	terminal	34	3.4	42-72	28-42	1.7	Ovoid-obpyriform, Obpyriform, Ovoid, Spherical,
P26	stellate	terminal	36	3.2	52-78	25-38	2.0	Ovoid, Ellipsoid, Obpyriform
P05	stellate	terminal	35	3.5	48-78	28-45	1.8	Ovoid, Ellipsoid, Obpyriform, Spherical
P03	stellate	terminal & intercalary	35	3.9	39-74	26-35	1.9	Ovoid, Obpyriform, Spherical

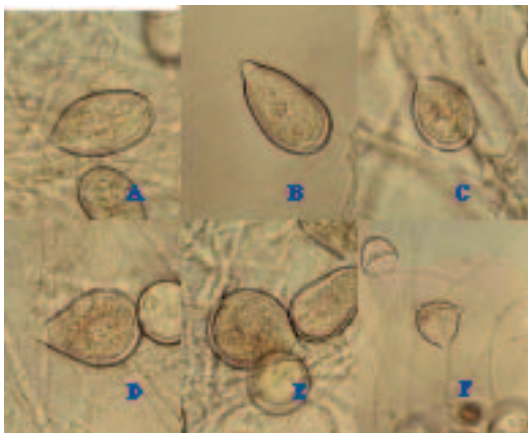
Twenty-six isolates produced ovoid, ellipsoid, obpyriform, ovoid-obpyriform, and spherical sporangia (Figure 3.3.2). The sporangia were caducous, with a short pedicel (2.8–4.2  $\mu\text{m}$ ) and conspicuously papillate. A few isolates showed bipapillate sporangia. The sporangia were variable in size (Table 3.3.2), averaging 35–90  $\mu\text{m}$  in length and 22–62  $\mu\text{m}$  in breadth, with a length–breadth ratio of 1.6–2.0. All isolates produced globose chlamydospores, which were terminal and intercalary in the mycelium. The average diameter of chlamydospores was 30–39  $\mu\text{m}$ . Based on these morphological characteristics, all isolates belonged to *P. palmivora*.

### Growth temperatures

All isolates grew at 10°C, with colony diameters less than 6 mm after 7 days. The optimum temperature was 25°C, with diameters of most isolates about 80–90 mm, and the maximum temperature 35°C, with



**Figure 3.3.1** Colony types of *Phytophthora* isolated from durian on PDA after 5 days incubation: A, stellate pattern; B, slightly stellate pattern; C, slightly petallate pattern; D, radiate pattern; E, irregular pattern and slightly fluffy; F, stoloniferous pattern.



**Figure 3.3.2** Morphology of sporangia: A, ellipsoid; B, obpyriform; C, ovoid; D, ovoid-obpyriform; E, spherical; F, bipapillate sporangium.

growth diameters around 55–72 mm. The exception was isolate P14, which could not grow at that temperature. No isolates grew at 37°C. Cardinal temperatures were thus minimum < 10°C, optimum 25°C and maximum 35°C.

### Pathogenicity test

All *Phytophthora* isolates from durian were successful in infecting wounded leaves of durian, black pepper and rubber. The isolates produced lesions of variable size on different host plants. They caused large lesions on durian leaves, and brown lesions on black pepper and rubber. Most of the isolates from durian were more aggressive on rubber than on black pepper (Table 3.3.3).

**Table 3.3.3** Results of pathogenicity tests of *Phytophthora* isolates from durian on wounded leaves of durian, black pepper and rubber.

Isolate no.	Diameter of disease lesion (mm)		
	Durian	Black pepper	Rubber
P01	14.9	9.7	7.5
P03	10.9	5.3	5.7
P04	17.2	5.3	7.8
P05	14.1	4.7	14.2
P07	14.7	9.1	8.5
P09	16.0	9.7	10.4
P10	14.5	6.8	9.6
P12	14.2	0	9.0
P14	16.1	11.4	10.4
P17	11.4	8.3	10.5
P19	12.8	6.1	8.7
P21	17.0	6.3	8.9
P22	21.1	9.1	9.2
P23	13.7	9.4	12.9
P25	9.0	8.9	16.0
P26	10.5	8.0	9.4
P27	10.2	9.8	12.2
P29	13.2	8.1	7.8
P31	9.5	7.9	9.0
P32	11.3	0	10.0
P33	13.0	0	7.5
P35	13.3	7.5	7.4
P36	12.9	8.2	9.9
P37	18.4	7.3	8.7
P38	15.7	9.1	8.5
P39	10.6	9.8	14.3

### Discussion

Several researchers have described the features of *P. palmivora* that distinguish it from other heterothallic species with conspicuous papillate sporangia. The sporangia are variable in shape, depending on isolate, mostly elliptical to ovoid, and prominently papillate. They are caducous with a

short pedicel (< 5 µm), and are variable in size but average 40 to 60 µm in length and 25 to 35 µm in breadth, with length–breadth ratio of 1.4 to 2.0 µm (Ho 1990; Erwin and Ribeiro 1996). On a few occasions we also observed spherical bipapillate sporangia

Many reports have shown that *P. palmivora* produces globose chlamydospores. Chlamydospore diameters have been reported to measure 32 to 42 µm (Holliday 1980), averaging 33 µm (Waterhouse 1974), 36 µm (Ashby 1929) and 36.2±9.6 µm (Mchau and Coffey 1994).

Most isolates produced stellate colony types. Waterhouse et al. (1983) reported that *P. palmivora* colonies were stellate. In our study, only one isolate (P33) showed a stoloniferous growth pattern, but other morphological characters confirmed the identity of this isolate as *P. palmivora*. The data produced on the isolates in our study fall within this range, confirming the species identity.

Waterhouse (1974) studied the effect of temperature on the growth of *P. palmivora* and reported the minimum temperature as 11°C, the optimum as 27.5–30°C, and the maximum as near 35°C.

Pathogenicity tests by many researchers have shown that *P. palmivora* isolates that cause black stripe and patch canker of rubber, and leaf and collar rot of black pepper in Southeast Asia, can also cause patch canker of durian (Belgrave and Norris 1917; Navaratnam 1966; Tsao and Tummakate 1977; Suzui et al. 1979). *Phytophthora nicotianae* has also been reported as infecting durian, causing patch canker, and fruit, crown, foot, and root rot of black pepper, especially in Malaysia and Thailand (Liu 1977; Suzui et al. 1979). However, all isolates collected in this study were identified as *P. palmivora*.

The results of earlier research on the causes of leaf and collar rot of black pepper have often given *P. palmivora* as the likely causative agent. However, a more recent reclassification of pepper isolates of *P. palmivora* MF4 as *P. capsici* indicate that further taxonomic and genetic studies are needed to more clearly define the boundaries between these *Phytophthora* species (Tsao and Alizadeh 1988; Tsao 1991).

This study revealed that there is variation in the *P. palmivora* population obtained from durian in Thailand. This variation within as well as between species makes identification of these species more difficult. Some of the phenotypic variation observed may also be due to environmental factors such as the media and temperature used to culture *Phytophthora*

species. More accurate species identification may be achieved through the use of molecular-based identification methods. This is especially important considering the wide range of *Phytophthora* species occurring in the tropics.

With intercropping gaining in popularity it is important to know the species of *Phytophthora* involved in disease. Many host plants susceptible to *P. palmivora* are grown throughout Southeast Asia. Intercropping of hosts susceptible to the same pathogens may give rise to an increased build-up of inoculum and thus be responsible for disease problems more severe than those encountered in monoculture situations.

## Acknowledgments

We thank Dr Brett Summerell from the Royal Botanic Gardens, Sydney and Sophie Peterson from the University of Sydney for critically reading this manuscript. This research was supported by the Australian Centre for International Agricultural Research (ACIAR).

## References

- Ashby, S.F. 1929. Strains and taxonomy of *Phytophthora palmivora* Butler (*P. faberi* Maubl.). Transactions of the British Mycological Society, 14, 18–38.
- Belgrave, W.N.C. and Norris, F. de la M. 1917. Notes on bark cankers and their treatment. Federated Malay States Agricultural Bulletin, 6, 2–10.
- Erwin, D.C. and Ribiero, O.K. 1996. *Phytophthora* diseases worldwide. St Paul, Minnesota, American Phytopathological Society Press.
- Ho, H.H. 1990. Taiwan *Phytophthora*. Botanical Bulletin Academia Sinica, 31, 89–106.
- Holliday, P. 1980. Fungus diseases of tropical crops. Cambridge, UK, Cambridge University Press, 607 p.
- Lim, T.K. 1990. Durian diseases and disorders. Kuala Lumpur, Malaysia, Tropical Press Sdn. Bhd.
- Liu, P.S.W. 1977. Diseases caused by *Phytophthora* and *Pythium* in Sabah, Malaysia Technical Bulletin, 3, 48 p.
- Mchau, G.R.A. and Coffey, M.D. 1994. Isozyme diversity in *Phytophthora palmivora*: evidence for a Southeast Asia centre of origin. Mycological Research, 98, 1035–1043.
- Navaratnam, S.J. 1966. Patch canker of the durian tree. Malayan Agricultural Journal 45, 291–294.
- Orellana, R.G. 1959. Variation in *Phytophthora palmivora* isolated from cacao and rubber. Phytopathology, 49, 210–213.
- Pongpisutta, R. and Sangchote, S. 1994. *Phytophthora* fruit rot of durian (*Durio zibethinus* L.). In: Champ, B.R., Highley, E., and Johnson, G.I., ed., Postharvest handling of tropical fruits: proceedings of an international conference held at



- Chiang Mai, Thailand, 19–23 July 1993. Canberra, ACIAR Proceedings No. 50, 460–461.
- Suzui, T.J., Kueprakone, U., and Kamphangridthrong, T. 1979. *Phytophthora* spp. Isolated from some economic plants in Thailand. Technical Bulletin Tropical Agricultural Research Center, 12, 32–41.
- Tsao, P.H. 1991. The identities, nomenclature and taxonomy of *Phytophthora* isolates from black pepper. Paper read at Diseases of black pepper. Proceedings of the International Pepper Communication Workshop on Pepper Diseases, at Goa, India.
- Tsao, P.H., and Alizadeh, A. 1988. Recent advances in the taxonomy and nomenclature of the so-called "*Phytophthora palmivora*" MF4 occurring on cocoa and other tropical crops. Paper read at 10th International Cocoa Research Proceedings, Santo Domingo, 17–23 May 1987.
- Tsao, P.H. and Tummakate, R. 1977. The identity of a *Phytophthora* species from black pepper in Thailand. *Mycologia*, 69, 631–637.
- Waterhouse, G.M. 1974. *Phytophthora palmivora* and some related species. In: Gregory, P.H. ed., *Phytophthora* disease of cacao. London, Longman, 51–70.
- Waterhouse, G.M., Newhook, F.J. and Stamps, D.J. 1983. Present criteria for classification of *Phytophthora*. In: Erwin, D.C., Bartnicki-Garcia, S. and Tsao, P.H., ed., *Phytophthora: its biology, taxonomy, ecology, and pathology*. St Paul, Minnesota, American Phytopathological Society, 139–147.

# 4

## Occurrence of *Phytophthora* in Southeast Asia



# 4.1 Phytophthora Diseases in Malaysia

B.S. Lee<sup>1</sup> and K.Y. Lum<sup>2</sup>

## Abstract

This chapter provides a historical overview of the *Phytophthora* species found in Malaysia and details on the occurrence, impact and control of the main phytophthora diseases affecting Malaysia's major agricultural crops: rubber, cocoa, durian and pepper.

## Introduction

Malaysia is made up of two geographical regions, namely Peninsular Malaysia on the southeastern tip of mainland Asia, and the states of Sabah and Sarawak on the island of Borneo. The South China Sea separates the two regions. Situated just north of the equator, the climate is typically hot and humid tropical. It has an annual rainfall of 2000–4000 mm, falling in 150 to over 200 days per annum. For example, the foothills of the Cameron Highlands in Peninsular Malaysia and Kuching in Sarawak experience about 250 rainy days per annum. Average daily temperature under shade ranges from 23 to 29°C with relative humidity in the range 70–90%. These climatic conditions are ideal for year-round cultivation of tropical crops. They are also excellent for the development and spread of tropical plant diseases.

Agriculture in Malaysia is dominated by mega plantations, with extensive planting of monocultures of rubber, oil palm, cocoa and coconut. This export-oriented agricultural system was first introduced into the country in the late 1800s. Phytophthora diseases are common on rubber and cocoa. There are no reports of phytophthora on oil palm. The incidence of phytophthora on coconut is sporadic, although *Phytophthora nicotianae* (syn. *P. parasitica*) has occasionally been isolated from infected palms.

Among non-plantation crops, durian (*Durio zibethinus* L.) is, next to rice, the most important crop, in terms of area planted. It is also the most popular fruit in Southeast Asia, the centre of biodiversity for *Durio* species. One of the most important factors limiting the planting and production of durian is trunk and root rot caused by *Phytophthora palmivora*.

About 60,000 farm families, mostly from Sarawak, are involved in pepper cultivation. This makes pepper the most important cash crop in Sarawak. Foot rot caused by *P. capsici* is the most serious disease limiting the successful cultivation of the crop. Johor in Peninsular Malaysia was at one time an important centre for pepper production, but most of the farms have now been converted to non-agricultural uses.

Research into the genus *Phytophthora* in the country started about 80 years ago and is centred mostly on rubber, cocoa, durian and pepper, which together occupy an area of about 1.6 million ha. Some phytophthora research, mostly limited to identification and control, had also been carried out on crops such as citrus, papaya, guava, passionfruit, jackfruit, roselle, tomato, potato, yam and orchids.

## Phytophthora Species and Their Recorded Hosts

Plant pathological work in the early 1900s was focused mainly on rubber and spices. The person who contributed most to the understanding of phytophthora in the early 1900s was A. Thompson. In 1925, he recorded a *Phytophthora* species that caused patch canker of rubber (*Hevea brasiliensis* (H.B.K.) Mull. Arg.) (Thompson 1925). A year later,

<sup>1</sup> AGR Smart/MARDI, No. 65, Jalan SS2/43, 47300 Petaling Jaya, Selangor, Malaysia.

<sup>2</sup> Malaysian Agricultural Research and Development Institute, GPO Box 12301, 50774 Kuala Lumpur, Malaysia.

he recorded another *Phytophthora* species on betel vine (*Piper betle* L.), which he described as either *P. parasitica* or *P. colocasiae* (Thompson 1926). In his preliminary report on *Phytophthora* species in Malaysia, Thompson (1928) noted that there were only two recorded species. In 1929, he recorded three more species, namely *P. palmivora*, *P. heveae*, and *P. meadii* on rubber (Thompson 1929).

Sudden death of pepper (*Piper nigrum* L.), possibly caused by phytophthora, was first reported by Holl (1929) in Sarawak. Thompson (1941) and Holliday and Mowat (1957) isolated a species of *Phytophthora* from infected pepper vines. Several years later, Holliday and Mowat (1963) identified the fungus as an atypical strain of *P. palmivora*. The first report of the disease in Johor was by Loh (1970).

Sharples (1930) recorded *P. nicotianae* (described as *P. parasitica*) on *Hibiscus sabdariffa* L., a plant grown for its fibre at that time. In the early 1990s, this crop was reintroduced for juice extraction on a commercial scale in the east coast of Peninsular Malaysia. On mineral soil, *P. nicotianae*, causing sudden wilt symptoms, was frequently isolated from infected roots and collars of the plant, especially during the wet monsoon months from October to January (B.S. Lee, unpublished data). Interestingly, the crop was free of phytophthora symptoms when planted on irrigated sandy soil.

Thompson (1934a) described for the first time the occurrence of *P. palmivora* as the causal agent of patch canker on durian (*Durio zibethinus*) in Penang. He observed that the disease had been present in the locality for at least the previous 10 years, and that it had killed many mature trees. Subsequently, the disease was extensively studied by Chan and Lim (1987), Lee (1999), Lee and Varghese (1974), Lim and Chan (1986), Lim and Yassin (1985), Navaratnam (1966), and Tai (1971). Although durian trees in Penang remain badly affected by phytophthora (Hashim et al. 1991), the state has remained famous for its unique varieties of durian.

Thompson (1934b) reported the occurrence of *Phytophthora infestans* on potato (*Solanum tuberosum* L.) and tomato (*Lycopersicon esculentum* Mill.) in the Cameron Highlands, while *P. parasitica* was recorded on tomato several years later (McIntosh 1951). Leaf blight and fruit rot on tomato, caused by *P. infestans*, are still limiting factors to about 700 ha of tomato in the Cameron Highlands. Tolerant varieties, fungicidal sprays, and planting of tomato under plastic rain shelter have reduced the problem.

Thompson (1940) identified *P. palmivora* as the causal agent of root and collar rot on papaya (*Carica*

*papaya* L.). Fruit rot caused by *P. palmivora* is common under wet weather conditions. *P. parasitica* is another species commonly isolated from infected fruit, collar and roots of papaya (Singh 1973; B.S. Lee, unpublished data). In general, phytophthora is not a serious problem on papaya, except when replanting is practised (Lee and Chan 1980). The disease is often localised and occurs in situations where soil drainage is poor (Lim and Yaakob 1989).

Cocoa (*Theobroma cacao* L.) was first introduced in the 1940s, but it was not until the 1950s that it was successfully planted on a commercial scale. Barcroft (1961) reported the first incidence of black pod caused by *P. palmivora* in the country. Ten years later, Chee and Phillips (1971) reported the occurrence of stem canker.

Wong and Varghese (1966) spent several years researching on the biology, ecology and control of foot and root rot of citrus. They attributed the diseases to *P. nicotianae*. Chee (1969b) studied the sudden outbreak of abnormal leaf fall of *Hevea* rubber in the northern states of Perlis, Kedah and Perak and described the pathogen as a new species, which he named as *P. botryosa* Chee. An extensive search for *Phytophthora* in the early 1970s resulted in the isolation of *P. capsici* from bell pepper (*Capsicum annuum* var. *grossum* L.), *P. heveae* from cocoa, and *P. nicotianae* from brinjal (*Solanum melongena* L.) and strawberry (*Fragaria* sp.) (Lee 1972).

Singh (1973), in his compilation of plant diseases, made several additions to the increasing list of *Phytophthora* species in the country: *P. colocasiae* on yam (*Colocasia esculenta* Schott.) and *Piper betle*; *P. palmivora* on Vanda orchids; *Phytophthora* sp. on avocado (*Persea gratissima*), *P. nicotianae* on *Salvia splendens* and *P. nicotianae* on *Vinca rosea*.

*Phytophthora cinnamomi* was reported to cause root rot and dieback of quinine tree (*Cinchona ledgeriana* Moens and *C. succirubra* Pav. Ex. Klotzsch) in the Cameron Highlands (Thompson 1940). Lee (1974) isolated *P. cinnamomi* from infected roots of cloves (*Eugenia aromatica* Baill.). Kueh and Khew (1982) isolated *P. meadii* from roots of *Piper betle*. Chan and Lim (1987) reported *P. nicotianae* as the causal agent of leaf blight of guava (*Psidium guajava* L.). This was the last published record of a new incidence of phytophthora on any crop, although species of *Phytophthora* were isolated from various hosts from time to time (*P. nicotianae* from passionfruit and orchid, and *P. citrophthora* from jackfruit) (B.S. Lee, unpublished data).

Table 4.1.1 summarises this section.

## Phytophthora Diseases of Economic Importance

### Rubber

The earliest plantation crop in Malaysia was *Hevea* rubber. A number of seedlings from Brazil were sent to Kew Garden, then to the Botanical Garden of Singapore in 1877. That same year, some plants were sent to Malaysia. These few plants became the progenitors of all the large rubber plantings in Southeast Asia.

Total area planted with rubber has steadily declined from 1.69 million ha in 1995 to 1.43 million ha in 2000 (Table 4.1.2). This is expected to shrink further before stabilising at about 1.1 million hectares. The declining trend is due to the decline in the prices of primary commodities and the acute shortage of plantation labour. Despite the reduction, rubber cultivation will remain an important element in the Malaysian economy.

Most of the studies on the biology and control of phytophthora on rubber were done in the 1960s and 1970s (e.g. Chee 1968a,b, c; 1969a,b; 1970; 1971a,b; Lim and Abdul Aziz 1978; Tan et al. 1977; Tan 1979). In general, the stem, shoot, leaf and pod of the tree are attacked by two *Phytophthora* species, *P. palmivora* and *P. botryosa*. *P. palmivora* causes black stripe of the tapping panel and patch canker on the untapped bark, pod rot and leaf fall. On the other hand, *P. botryosa* is the main cause of leaf fall and pod rot diseases, although it may also cause black

stripe under conditions favourable to it. Leaf fall and black stripe are important diseases during the rainy seasons from July to October in the northern states of Perlis, Kedah and Perak, and from October to January in Kelantan. Most of the research has been centred on black stripe and leaf fall as they occurred more frequently than other phytophthora diseases (Tan 1979). Pathogenicity studies indicated that *P. palmivora* and *P. botryosa* from rubber were capable of infecting other crops including cocoa, durian, pepper, mango, citrus and orchid (Chee and Hashim 1971). No *Phytophthora* species has been recorded on rubber roots, although rubber root diseases caused by other groups of fungi are major constraints to the rubber industry.

Traditional methods of controlling rubber diseases rely heavily on the use of chemicals. Against leaf diseases, the application of fungicides on mature rubber trees is difficult due to the height of the trees, unsuitable terrain, poor accessibility and uneconomic plot size. Adequate control of black stripe is achieved by early detection and application of fungicides such as oxadixyl, metalaxyl and folpet. Against leaf fall, a pre-monsoon thermal fogging of copper-in-oil at 1.2 kg/ha has proven effective in controlling the disease (Lim 1982). Excellent control of leaf fall was also achieved by trunk injection with neutralised phosphorous acid (Lim and Lee 1990). Direct injection into the basal portion of the stem is easy and it dispenses with repeated rounds of expensive ground or aerial spraying. It also overcomes the problem with height of the rubber

**Table 4.1.1** Host list of *Phytophthora* species isolated in Malaysia since 1925.

Species	Host	Collector
<i>Phytophthora</i> sp.	Rubber	Thompson (1925)
<i>P. colocasiae?</i>	Betel vine	Thompson (1926)
<i>P. palmivora</i>	Rubber	Thompson (1929)
<i>P. heveae</i>	Rubber	Thompson (1929)
<i>P. meadii</i>	Rubber	Thompson (1929)
<i>Phytophthora</i> sp.	Pepper	Holl (1929)
<i>P. nicotianae</i> ( <i>P. parasitica</i> )	Roselle	Sharples (1930)
<i>P. palmivora</i>	Durian	Thompson (1934a)
<i>P. infestans</i>	Potato, tomato	Thompson (1934b)
<i>P. cinnamomi</i>	Quinine	Thompson (1940)
<i>P. palmivora</i>	Papaya	Thompson (1940)
<i>P. capsici</i> ( <i>P. palmivora</i> atypical)	Pepper	Holliday and Mowat (1963)
<i>P. palmivora</i>	Cocoa	Barcroft (1961)
<i>P. nicotianae</i>	Citrus	Wong and Varghese (1966)
<i>P. botryosa</i>	Rubber	Chee (1969b)
<i>P. capsici</i>	Bell pepper	Lee (1972)
<i>P. heveae</i>	Cocoa	Lee (1972)
<i>P. nicotianae</i>	Brinjal, strawberry	Lee (1972)
<i>P. nicotianae</i>	Papaya	Singh (1973)
<i>P. colocasiae</i>	Yam	Singh (1973)

trees, the difficult terrain or high equipment, labour and chemical costs.

The Environmax planting strategy implemented since the early 1970s has been quite successful in controlling phytophthora diseases (Lim 1980; Ho et al. 1984; Ismail and Mohd 1984). This involves the avoidance of planting susceptible clones in areas conducive to disease development. Under this program, tolerant clones are recommended for planting in pre-demarcated areas. To sustain growth and productivity of susceptible clones already planted, short-term remedial measures using chemicals are recommended. This includes disease forecasting, which has been used successfully to control leaf fall (Lim 1980).

**Table 4.1.2.** Area planted to rubber in Malaysia, 1995–2000.

Year	Area ('000 ha)
1995	1.688
1996	1.644
1997	1.616
1998	1.556
1999	1.465
2000	1.431

Source: Anon. (2002)

## Cocoa

A native of South America who attempted to grow cocoa commercially in Malaysia in the early 1900s did not succeed. Following a report on cocoa by Cheesman in 1948 (unpublished), the Malaysian Department of Agriculture in the late 1940s devoted a great deal of attention to cocoa as a possible crop for diversification (McIntosh 1948). Experimental planting of cocoa using local Trinitario and imported Amelonado failed because of dieback

problems (Haddon 1960). In the 1960s, trial plantings with Upper Amazon as an inter-crop in Peninsular Malaysia and as a mono crop in Sabah proved successful, paving the way for a rapid expansion of cocoa in the country.

Production of cocoa in Malaysia has steadily declined from 9% of world cocoa bean output in 1990–1991 to 2% in 1999–2000. The decline is attributed to the falling price of cocoa, which resulted in growers moving to other crops, especially oil palm.

Table 4.1.3 shows the rapid decline in area under cocoa over the last 10 years. In 1992, the area under cocoa was estimated at 380,000 ha, but by 2001 this had dropped to 70,000 ha, a decline of about 80%.

The dominant *Phytophthora* species on cocoa is *P. palmivora* (A2 mating type) with occasional A1 mating type reported in Sabah (C.L. Bong, pers. comm.). The fungus is present in all cocoa-growing areas. The sporangia are typically caducous, with a rounded base, short pedicel and having a prominent papilla. Length–breadth (L/B) ratio varies from 1.0 to 2.1, but most sporangia lie in the range of 1.4 to 1.6. Some cultures in our laboratory resembled *P. nicotianae*, with ovoid sporangia and L/B ratios of 1.1–1.3 and with no pedicel. *P. heveae* had been isolated from cocoa rhizosphere and was pathogenic to cocoa pods (Lee 1972). In addition, *P. meadii*, *P. heveae* and an unidentified *Phytophthora* species were occasionally isolated from infected pods in Sabah (Liu 1977).

In laboratory tests, several *Phytophthora* species from other host plants were pathogenic to cocoa: *P. cinnamomi* from clove (Lee 1974), *P. capsici* and *P. nicotianae* from capsicum and brinjal (Lee 1972), and *P. botryosa* from rubber (Chee and Hashim 1971). The potential threat of these species to cocoa is significant.

**Table 4.1.3** Area planted to cocoa in Malaysia, 1992–2001.

Year	Estate plantings (ha)	Smallholder plantings (ha)	Total (ha)
1992	168,058 (44%)	210,482 (56%)	378,540
1993	145,646 (49%)	154,349 (51%)	299,995
1994	130,232 (48%)	141,107 (52%)	271,339
1995	96,053 (51%)	49,074 (49%)	190,127
1996	73,503 (44%)	94,716 (56%)	168,219
1997	50,270 (36%)	90,629 (64%)	140,899
1998	37,045 (31%)	80,634 (69%)	117,679
1999	27,937 (28%)	72,866 (72%)	100,803
2000	22,439 (30%)	53,327 (70%)	75,766
2001	20,526 (30%)	48,922 (70%)	69,448

Source: Malaysian Cocoa Board

In Malaysia, black pod is the most common phytophthora disease on cocoa. Chan and Lee (1973) reported low incidence of black pod in the early 1970s. The situation was similar in Sabah, where low incidence was attributed to environmental conditions unfavourable for disease development at the time (Liu 1977; Liu and Liew 1975). Incidence and severity of black pod has since increased, due to the planting of highly susceptible clonal materials. In areas of high rainfall and poor agronomic practices, incidence as high as 30% was common (Tey 1983). *P. palmivora* infects pods of all ages, including young cherelles. Lee and Chan (1980) reported that, in localities of high rainfall and poor management, incidence of cherule wilt caused by *P. palmivora* could be as high as 30%. Epidemiological studies of black pod were undertaken by Tey et al. (1986). They showed that incidence of black pod was related to weather conditions and fruiting patterns. Heavy infection occurred during the months of high rainfall, which coincided with the main fruiting season. The abundance of susceptible host tissue under conditions favourable for disease development resulted in high incidence of the disease.

First reported by Chee and Phillips (1971), stem canker is the next most important phytophthora disease on cocoa. Infection starts from anywhere along the trunk, branches or jorquettes. Lesions can also form just above the soil line, and often extend into the soil as well. Incidence and severity of stem canker are closely related to rainfall and management practices. In general, areas with high incidence of black pod also have high incidence of stem canker. An outbreak of stem canker in the mid 1980s in Perlis in northern Peninsular Malaysia was attributed to improper use of drip irrigation. The damp and waterlogged conditions created by the drip around the base of the trees induced the disease to develop (Tey and Musa 1987).

Seedling blight caused by *P. palmivora* was first reported by Chee (1969a). Seedlings of up to 4 months old in polybags could be affected (Lim 1980). Although localised, losses of up to 20% are common (Chan and Lim 1987).

In areas where the incidence of black pod is low, control is achieved by regular removal of infected pods, which are then either buried or burnt. Maintenance pruning is practised to improve ventilation, quicken the drying of pods and stem surfaces, and to prevent disease build-up.

Fungicides are used in most plantations. They include copper-based products such as copper

hydroxide, copper oxychloride, cuprous oxide, copper-mancozeb mixtures, triphenyltin acetate, etridiazole, metalaxyl, and fosetyl aluminium. Depending on the size of trees, most plantations used either pressurised knapsack sprayers or motorised mist blowers. Excellent control of black pod was achieved by injecting the trunk of affected trees with neutralised phosphorous acid (Tey and Lee 1994). Continued exposure of the pathogen to sublethal doses of systemic fungicides can lead to the development of resistant strains. This was demonstrated by Tey (1984) when he exposed mycelium to sublethal doses of metalaxyl and milfuram.

Considerable progress has been made to develop high-yielding varieties with favourable secondary characters such as disease tolerance (Chong and Shepherd 1986; Tey 1987; Tiong and Kueh 1986). Current research includes clonal selection for disease tolerance and biological control studies.

## Durian

Southeast Asia is the centre of origin of *Durio* species, with the majority originating from the island of Borneo. There are some 28 *Durio* species in Malaysia, of which about 11 are edible. *Durio zibethinus* is the only species cultivated commercially. All the registered 'D' clones are from this species. There are still many wild and semi-wild varieties of *D. zibethinus* waiting to be assessed in proper trials on performance, susceptibility and yield. Area under durian has steadily increased since the early 1990s (Table 4.1.4). The drop in farm prices of first grade durian in the last three years has put a damper on durian production. Many farms are being neglected, resulting in the increased incidence of pests and diseases including phytophthora diseases.

**Table 4.1.4** Area under durian cultivation in Malaysia, 1990–1997.

Year	Area (ha)
1990	57,000
1991	62,000
1992	62,000
1993	83,000
1994	107,000
1995	108,000
1996	110,000
1997	112,000

Source: Department of Agriculture, Malaysia

The dominant species attacking durian is *Phytophthora palmivora*, although more than one

species may be involved. L/B ratio of the sporangia varies from 1.3 to 2.2, with majority falling between 1.8 and 2.1.

All parts of the durian tree are attacked by *P. palmivora*: the trunk, twigs, branches, fruit, leaves, flowers and the underground portion of the stem and roots. Entry of pathogen is through wounds caused by mechanical injury or through natural openings (Lee 1999). With regular inspection, above-ground lesions can be easily treated. Treatment is difficult when the lesions have penetrated deep into the wood or have completely girdled the tree. Stem and root lesions formed below ground are difficult to detect or treat. Fruit rot is an important disease and, depending on weather conditions, 20–30% of the fruits in an orchard may be affected.

The incidence of durian canker is high in most orchards. In a survey of six locations in Penang, 30% of nearly 2000 trees examined were severely affected by stem canker (Hashim et al. 1991). This figure is representative of most of the orchards in Malaysia. If one assumes that 10–20% of mature durian trees in the country are affected with canker and 50,000 hectares are of fruit-bearing age, there will be a total of half to one million infected trees in the country. Untreated trees will eventually die.

Control of the disease is limited to foliar application of fungicides in the nursery to protect the young seedlings (Chan and Lim 1987) and bud-wood nurseries, and curative treatment to control stem canker in the field. This is achieved through extensive and laborious tree surgery to remove the infected bark and the underlying wood tissue, followed by painting with protective and curative fungicides such as fixed copper fungicides, dimethomorph, triphenyltinacetate, oxadixyl, metalaxyl and fosetyl aluminium. Lim and Yassin (1985) found metalaxyl and fosetyl aluminium to be readily translocated to nearby tissues when these chemicals were painted onto the surface of the scraped branches. Lee et al. (1988) reported excellent control of phytophthora in durian seedlings when the seedlings were trunk injected with phosphorous acid. Trunk injection of mature trees with metalaxyl and fosetyl aluminium (Lee 1994) and phosphorous acid (Lim and Lee 1990) also provided good control. Foliar application with 0.4% phosphorous acid also gave excellent protection of one-year-old seedlings (Table 4.1.5).

While all *D. zibethinus* clones are susceptible, there is variability in susceptibility. Screening of clonal materials through wound inoculation showed that D24 and D66 were the most susceptible while D2 and D10 were the least susceptible (Tai 1973). Nik

(2000) also reported mixed reactions of durian clones to phytophthora. In an attempt to overcome the disease, several hybrid clones have been developed by the Malaysian Agricultural Research and Development Institute (MARDI) in recent years, some with very promising anti-phytophthora properties. In a study of 10-year-old clonal hybrids subjected to heavy inoculum pressure and high annual rainfall, Lee (1999) found MDUR 79, MDUR 88 and MDUR 78 to be the least susceptible. These were hybrids derived from D10 and D24 crosses. In the same study, the most susceptible clone was D24.

**Table 4.1.5** Effect of phosphorous acid on control of durian stem canker.

Treatment	Lesion length (mm) <sup>a</sup>
Foliar spray	16.6
Soil drench	44.5
Control	71.9

<sup>a</sup> Mean of eight one-year-old seedlings

D24 is a tree that grows vigorously and has excellent fruit quality. The extensive planting of this clone, with its vigorous growth, thick foliage and high branching system, has contributed significantly to the incidence and severity of the disease throughout the country in recent years. However, when bark of D24 seedlings was artificially inoculated without wounding, the stem remained healthy. This indicated that mechanical injury was an important factor in disease initiation. This observation has led Nik and Lee (2000) to develop a rain-fast wound dressing specifically for durians. Wounds treated with this dressing were protected against infection in the field. The protection could last for at least six months, long enough for the wounds to be naturally healed.

In an extensive study on the potential of tolerant rootstock to overcome patch canker in durian, Lee (1999) studied the possibility of using *Durio lowianus* as rootstock. Excellent survival of D24 trees grafted onto *D. lowianus* rootstock in a naturally infested field after 13 years of planting indicated that *D. lowianus* has good potential for commercial use to prevent premature death due to *P. palmivora*. Nearly 50% of D24 trees grafted onto normal rootstock died of canker within 13 years while close to 100% survived when they were grafted onto *D. lowianus* rootstock.

The use of suppressive soil for controlling phytophthora diseases has been well documented (e.g. Broadbent and Baker 1974; Ko and Nishijima 1985; Ko and Shiroma 1989). Lee (1999) reported possible suppression when durian trees were planted in limestone soil high in soil pH, cation



exchange capacity, exchangeable calcium and micronutrients such as Mn, Zn and Cu. The presence of relatively high copper content in these soils is interesting because copper ions are strongly fungitoxic to *P. palmivora*. While copper deficiency causes dieback of durian trees.

### Pepper

*Piper nigrum* L is native to the state of Kerala in India. Hindu migrants to Indonesia first introduced the crop into Southeast Asia as early as 100 BC. India and Indonesia are the main producers of pepper, accounting for more than 50% of world production. In recent years, Vietnam has become an important producer as well.

In the early 1800s, the crop spread to Sarawak, which is now the main pepper-producing state in Malaysia (Table 4.1.6). Pepper orchards are generally small, averaging about 0.25 ha or 400 vines, and situated on hill slopes, often without ground cover. The high-yielding but susceptible Kuching variety is the most widely cultivated variety in Malaysia. Average yield is between 2 and 3 kg of dried pepper per vine, with some progressive farmers reporting a yield of 4 kg or more (Anon. 2002).

Pepper requires a tropical climate with well-distributed annual rainfall of 2000–4000 mm, a mean air temperature of 25–30°C and relative humidity of 65–95%. It grows best at altitudes below 500 metres, but may grow up to 1500 metres above sea level, and on soils ranging from heavy clay to light sandy clay. Soils should be deep, well drained and with good water-holding capacity to deal with water stress during the dry period.

*Phytophthora capsici* affects the leaves, spikes, berries, branches, climbing stems, underground stems and roots, i.e. all parts of the pepper vine. Initiation of infection takes place during wet weather when black necrotic spots with typical fimbriate margins develop on the lower leaves as a result of rain splash. These infected leaves subsequently drop off, resulting in the built up of soil inoculum. Roots and underground stem infection is indicated when the leaves turn pale and flaccid. Leaf and spike fall indicate a late stage of infection. Eventually, the vine is completely defoliated and is left standing with only the climbing stems and lateral branches. Infection may start at soil level or at any point along the underground stem to a depth of 20 cm. Lesions on stem and roots are dark brown in colour with a sharp margin of demarcation.

*Phytophthora capsici* grows best in a humid environment of 25–30°C and a pH of 5.5 to 6.0. The

identification and taxonomy of this species has been well described by Alizadeh (1983) and Alizadeh and Tsao (1985). In the 1970s, when pepper was widely grown in Johor, the species frequently isolated was *P. nicotianae*. Tsao (1986) also reported the presence of *P. nicotianae* in Thailand. From his study of pepper phytophthoras from around the world, Tsao (1986) concluded that there was no typical *P. palmivora* on pepper.

The biology, spread and control of pepper foot rot in Sarawak had been studied by Kueh (1977) and Kueh and Khew (1982). Inoculum is spread by rain splash, root contact, snails (*Achatina fulica* and *Hemiplecta crossei*), and wooden posts from infected fields, farm tools and man. The fungus could survive in soil in the absence of a host for at least 18 months. Fungal propagules were found mainly in the first 15 cm of the soil profile, with very low counts at a depth of 30–45 cm. The optimum soil moisture for survival was 25–45% water-holding capacity and soil pH 6.5–7.0.

**Table 4.1.6** Area (ha) under pepper cultivation in Malaysia, 1999 and 2000.

State	1999	2000
Sarawak	12,196	12,996
Johor	43	43
Sabah	48	45
Total	12,287	13,084

Source: Department of Statistics, Malaysia

Lee (1973) studied the mating types of pepper isolates from Johor and Sarawak, and concluded that the Johor isolate (probably *P. nicotianae*) was of the A1 mating type while the Sarawak isolate was of the A2 mating type. In addition, an atypical strain from Sarawak that formed oospores in single culture was reported by Turner (1962).

Lee (1973) reported the use of culture filtrate as a possible method to screen for resistance. From his study, two distinct groups of *Piper* spp. could be differentiated: the resistant group consisting of *Piper colubrinum* and *Piper sarmentosum*, and the susceptible group consisting of *Piper nigrum* varieties Kuching, Bangka, Djambi, Belantung and Uthirancotta, with the Kuching variety being the most susceptible and Uthirancotta the least susceptible. Similar results were obtained by Kueh and Khew (1980) when they used different fungal propagules as inoculum. Attempts to use *P. colubrinum* as resistant rootstock had met with little success due to late incompatibility and high susceptibility of *P. colubrinum* to other root diseases. Development of resistant planting material is

urgently needed. Certain varieties showed some tolerance, but infection and spread of disease in the field was only retarded rather than controlled. The ideal strategy for foot rot control is to adopt an integrated approach that involves cultural practices, chemical and biological control, and the exploitation of host resistance (Kueh and Khew 1980)

## Future Directions

Public research institutions funded by the federal government have been established to carry out research on specific crops in Malaysia. For example, research on rubber is carried out by the Malaysian Rubber Board, oil palm by the Malaysian Palm Oil Board and cocoa by the Malaysian Cocoa Board. Research on all other crops is carried out by MARDI and universities involved in biological sciences. Research on forestry is carried out by the Forest Research Institute of Malaysia. The departments of agriculture in Sabah and Sarawak have their own research centres to cater for their own regional needs. In these institutions, priority has always been given to phytophthora research. Directions that need to be developed or further strengthened are:

1. accurate detection and identification of species and strains within species through DNA based diagnostics and DNA fingerprinting
2. studies on the nature and diversity of *P. palmivora* and to develop, if feasible, national breeding and selection programs for crops such as rubber, cocoa, durian and pepper
3. integrated control of phytophthora diseases, including the use of resistant genes from wild plant species
4. training of plant pathologists in specific area of phytophthora research (isolation, identification, ecology, biological control, epidemiology, and disease management)
5. regional collaboration through the formation of a phytophthora working group for Southeast Asian countries sharing common phytophthora problems.

## Acknowledgments

We thank ACIAR and the Crawford Fund for their financial support and for organising the workshop at which this paper was presented.

## References

Alizadeh, A. 1983. Comparative morphology and reproductive physiology of *Phytophthora capsici* and *Phytophthora palmivora* MF 4 from black pepper and other hosts. Riverside, CA, USA, University of California.

Alizadeh, A. and Tsao, P.H. 1985. Effect of light on sporangium formation, morphology, ontogeny and caducity of *Phytophthora capsici* and *P. palmivora* MF4 from black pepper and other hosts. Transactions of the British Mycological Society, 85, 47–69.

Anon. 2002. Report on Malaysia's primary commodities 2001. Kuala Lumpur, Ministry of Primary Industries.

Barcroft, A.L. 1961. Annual report 1959, Department of Agriculture, Federation of Malaya.

Broadbent, P. and Baker, K.F. 1974. Behaviour of *Phytophthora cinnamomi* in soils suppressive and conducive to root rot. Australian Journal of Agricultural Research, 25, 121–137.

Chan, C.L. and Lee, B.S. 1973. A preliminary survey of cocoa diseases in West Malaysia. MARDI Research Bulletin, 1, 22–31.

Chan, L.G., and Lim, T.K. 1987. Control of *Phytophthora palmivora* on cocoa and durian seedlings. Journal of Plant Protection in the Tropics, 4, 9–13.

Chee, K.H. 1968a. Phytophthora leaf disease in Malaysia. Journal of the Rubber Research Institute Malaya, 21, 79–86.

– 1968b. Variability of *Phytophthora* species from *Hevea brasiliensis*. Paper presented at First International Congress of Plant Pathology, London, 1968.

– 1968c. Patch canker of *Hevea brasiliensis* caused by *Phytophthora palmivora*. Plant Disease Reporter, 52, 132–133.

– 1969a. Hosts of *Phytophthora palmivora*. Review of Applied Mycology, 48, 337–344.

– 1969b. Variability of *Phytophthora* species from *Hevea brasiliensis*. Transactions of the British Mycological Society, 52, 425–436.

– 1970. *Phytophthora heveae* and *Pythium vexans* of *Hevea*. Journal of the Rubber Research Institute Malaya, 23, 13–14.

– 1971a. Host adaptability to strains of *Phytophthora palmivora*. Transactions of the British Mycological Society, 57, 175–178.

– 1971b. Some new disorder of the stem and panel of *Hevea*. Paper read at Rubber Research Institute of Malaya.

Chee, K. H., and M. Hashim. 1971. Pathogenicity to some cultivated plants of *Phytophthora palmivora* and *Phytophthora botryosa* from *Hevea brasiliensis*. Malay Agricultural Journal, 48, 54–56.

Chee, K.H. and Phillips, T.A. 1971. Phytophthora stem canker of cacao. The Planter, Kuala Lumpur, 47, 43–46.

Cheesman, E.E. 1948. Report on potentialities for the cultivation of cocoa in Malaysia, Sarawak and North Borneo. London: H. M. Stationery Office.

Chong, C.F. and Shepherd, R. 1986. Promising Prang Besar cocoa clones. In: Pusparajah, E. and Chew, P.S., ed., Cocoa and coconut: progress and outlook. Kuala Lumpur, Incorporated Society of Planters.

Haddon, A.V. 1960. Variety trials of seedling cocoa in Malaysia. Malay Agricultural Journal, 43, 169–205.

- Hashim, L., M. Suhaimi, and H. Othman. 1991. Pest and disease survey for durian in the state of Penang. Paper read at Technical paper presented at the MAPPS Durian seminar, at Penang, 7–8 August, 1991, 13 pp.
- Ho, H.H., Liang, Z.R., Zhuang, W.Y and Yu, Y.N. 1984. *Phytophthora* spp. from rubber tree plantations in Yunnan province of China. *Mycopathologica*, 86, 121–124.
- Holl, E.S. 1929. Pepper: Annual Report for 1929. Department of Agriculture, Sarawak.
- Holliday, P. and Mowat, W.P. 1957. A root disease of *Piper nigrum* L. in Sarawak caused by a species of *Phytophthora*. *Nature*, 179, 543–544.
- 1963. Foot rot of *Piper nigrum* L. (*Phytophthora palmivora*). Kew, Surrey, UK, Commonwealth Mycological Institute, 62 p.
- Ismail, H. and Mohd, A.S. 1984. Integrated management of diseases and insect pests of *Hevea* rubber. In: Lee, B.S., ed., Integrated pest management in Malaysia. Kuala Lumpur, Malaysian Plant Protection Society.
- Ko, W.H. and Nishijima, K.A. 1985. Nature of suppression of *Phytophthora capsici* in a Hawaiian soil. *Phytopathology*, 75, 683–685.
- Ko, W.H. and Shiroma, S.S. 1989. Distribution of *Phytophthora cinnamomi* suppressive soil in nature. *Journal of Phytopathology*, 127, 75–80.
- Kueh, T.K. 1977. Recent advances in pepper foot rot research. Paper presented at the Technical Sessions of the 14th Research Officers Annual Conference, Department of Agriculture, Malaysia.
- Kueh, T.K. and Khew, K.L. 1980. A screening technique useful in selecting for resistance in black pepper to *Phytophthora palmivora*. *Malaysian Agricultural Journal*, 52, 37–45.
- 1982. Survival of *Phytophthora palmivora* in soil and after passing through alimentary canals of snails. *Plant Disease*, 66, 897–899.
- Lee, B. S. 1972. Research into the genus *Phytophthora* in Malaysia. MAgSci thesis, Kuala Lumpur, University of Malaya, 160p.
- 1973. The use of toxin for the screening of black pepper for foot rot resistance. *MARDI Research Bulletin*, 1, 10–14.
- 1974. *Phytophthora cinnamomi*: A new pathogen on cloves in Peninsular Malaysia. *MARDI Research Bulletin*, 2, 26–30.
- 1994. Control of *Phytophthora* patch canker of durian with metalaxyl and fosetyl aluminium. Paper presented at 4th International Conference on Plant Protection in the Tropics, Kuala Lumpur.
- 1999. Biological approaches for controlling *Phytophthora* root and trunk rot of durian. Paper presented at National Horticulture Conference, 16–17 November 1999, Kuala Lumpur.
- Lee, B.S. and Chan, Y.K. 1980. Replanting problem of papaya (*Carica papaya* L.) in Malaysia. Paper presented at National Fruit Seminar, Universiti Putra Malaysia, 5–7 November 1980, 15 p.
- Lee, B.S., Tey, C.C. and Musa, M.J. 1988. Trunk injection with phosphorous acid to control *Phytophthora palmivora* on cocoa and durian. Paper presented at *Phytophthora* Workshop, 5th International Congress of Plant Pathology, Kyoto, Japan.
- Lee, B.S., and Varghese, G. 1974. Studies on the genus *Phytophthora* in Malaysia. Isolation techniques, comparative morphology and physiology and reaction to antibiotics. *Malaysian Agricultural Research*, 3, 13–21.
- Lim, T.K., and Chan, L.G. 1986. Fruit rot of durian caused by *Phytophthora palmivora*. *Pertanika*, 9, 269–276.
- Lim, T.K. and Lee, B.S. 1990. Control of *Phytophthora* on four temperate and tropical tree crops by trunk injection with phosphorous acid. Paper presented at the 3rd. International Conference on Plant Protection in the Tropics, 20–23 March, Kuala Lumpur.
- Lim, T.M. 1980. A forecasting system for use in the chemical control of *Phytophthora* leaf fall on plantation rubber in Malaysia. Paper presented at workshop on *Phytophthora* diseases on tropical cultivated plants, Kerala, India, 1980 (Abstract).
- 1982. Fogging as a technique for controlling rubber leaf diseases in Malaysia and Brazil. *Planter*, 58, 197–212.
- Lim, T.M. and Abdul Aziz, S.A.K. 1978. Thermal fogging – a promising new method for controlling rubber leaf diseases. Paper presented at Plant Protection Conference, Kuala Lumpur.
- Lim, W.H. and Yassin, I. 1985. Chemical control of *Phytophthora* patch canker of durian. *Durio zibethinus* Murr. *Teknologi Buah-buahan*, 1, 31–36.
- Lim, W.H. and Yaakob, D. 1989. Major diseases of papaya and their treatment. Paper presented at MARDI-MAPPS Seminar on Eksotika papaya, Johor, 35–42.
- Liu, P.S.W. 1977. Diseases caused by *Phytophthora* and *Pythium* in Sabah, Malaysia. *Technical Bulletin*, 3, 48.
- Liu, P.S.W. and Liew, P.S.C. 1975. Cocoa diseases in Sabah. Sabah, Department of Agriculture, *Technical Bulletin* No. 1, 82.
- Loh, C.F. 1970. *Phytophthora* foot rot of pepper (*Piper nigrum* L.) in West Malaysia: Kuala Lumpur, Research Branch, Division of Agriculture.
- McIntosh, A.E.S. 1948. Implementation of the recommendation of the Cheesman Report. *Malaysian Agricultural Journal*, 31, 211.
- 1951. Annual Report, Department of Agriculture, Malaya.
- Navaratnam, S.J. 1966. Patch canker of the durian tree. *Malaysian Agricultural Journal*, 45, 291–294.
- Nik, M.H. 2000. Relative resistance/susceptibility to *Phytophthora palmivora* of durian clones and hybrids in MARDI. Paper presented at durian seminar, Ipoh, Perak.
- Nik, M.H. and Lee, B.S. 2000. Wound dressing for the prevention of canker in durian. Paper presented at durian seminar, Ipoh, Perak.

- Sharples, A. 1930. Report of the Division of Mycology, Department of Agriculture, S.S. and F.M.S., 1929, Bulletin 3, General Series.
- Singh, K.G. 1973. A check-list of host and diseases in Peninsular Malaysia, volume 132, Ministry of Agriculture and Fisheries, Malaysia.
- Tai, L.H. 1971. Studies on *Phytophthora palmivora*, the causal organism of patch canker disease of durian. Malay Agricultural Journal, 48, 1-9.
- 1973. Susceptibility of durian clones to patch canker disease. MARDI Research Bulletin, 1, 5-9.
- Tan, A.M. 1979. Phytophthora diseases of rubber in Peninsular Malaysia. Planters' Bulletin Rubber Research Institute of Malaysia, 158, 11-19.
- Tan, A.M., Leong, M.W., John, C.K. and Tan, K.J. 1977. Current status of Phytophthora diseases of rubber in Peninsular Malaysia. Paper presented at Rubber Research Institute Malaysia Planters' Conference, Kuala Lumpur.
- Tey, C.C. 1983. Black pod and brown pod of cocoa in Jerangau. MAPPs Newsletter, 7, 9-10.
- 1984. Strains of *Phytophthora palmivora* (Butl) resistant to metalaxyl and milfuram. MAPPs Newsletter 8, 6-7.
- 1987. Annual Report for 1987, Cocoa and Coconut Research Division, MARDI, Malaysia.
- Tey, C.C. and Lee, B.S. 1994. Controlling Phytophthora black pod of cocoa by trunk injection with fosetyl-al and metalaxyl. Paper presented at 4th International Conference on Plant Protection in the Tropics, Kuala Lumpur.
- Tey, C.C. and Musa, M.J. 1987. Outbreak of stem canker in irrigated cocoa. MAPPs Newsletter, 11, 3.
- Tey, C.C., Musa, M.J. and Lee, B.S. 1986. Preliminary observations on the incidence of black pod disease in Amelonado cocoa in Trengganu, Peninsular Malaysia. In: Pusparajah, E. and Chew, P.S., ed., Cocoa and coconut: progress and outlook. Kuala Lumpur, Incorporated Society of Planters.
- Thompson, A. 1925. A preliminary note on a Phytophthora associated with patch canker of *Hevea brasiliensis* in Malaya. Malayan Agricultural Journal, 13, 139-141.
- 1926. A disease of the betel vine caused by a species of *Phytophthora*. Malayan Agricultural Journal, 14, 1-7.
- 1928. A preliminary note on *Phytophthora* species found in Malaya. Malayan Agricultural Journal, 16, 40-47.
- 1929. *Phytophthora* species in Malaya. Malayan Agricultural Journal, 17, 53-100.
- 1934a. A disease of the durian tree. Malayan Agricultural Journal, 22, 369-371.
- 1934b. Annual Report, Department of Agriculture.
- 1940. Notes on plant diseases in 1939. Malayan Agricultural Journal, 28, 400-407.
- 1941. Notes on plant diseases in 1940. Malayan Agricultural Journal, 29, 241-245.
- Tiong, R.H.C. and Kueh, T.K. 1986. Studies on some cocoa diseases in Sarawak. In: Pusparajah, E. and Chew, P.S., ed., Cocoa and coconut: progress and outlook. Kuala Lumpur, Incorporated Society of Planters.
- Tsao, P.H. 1986. Black pepper *Phytophthora* species: importance of proper identification to the study of their ecology and control. Paper presented at 2nd International Conference on Plant Protection in the Tropics, Kuala Lumpur.
- Turner, G.J. 1962. Production of fusion organs by the species of *Phytophthora* which causes foot rot of *Piper nigrum* L. Nature, 195, 201.
- Wong, T.K., and Varghese, G. 1966. Assessing the susceptibility of local citrus species to Phytophthora root rot in Malaya. Experimental Agriculture, 2, 305-308.

## 4.2 Phytophthora Diseases in Indonesia

Agus Purwantara,<sup>1</sup> Dyah Manohara<sup>2</sup> and J. Sony Warokka<sup>3</sup>

### Abstract

This review summarises the species of *Phytophthora* recorded in Indonesia, their hosts, distribution, and current control measures. Some advances in research and control of phytophthora diseases have been made, but there is still a long way to go before sustainable disease-management practices are available for the wide range of diseases caused by different species of *Phytophthora*.

### Introduction

Indonesia is often referred to as the world's largest archipelago, consisting of 17,000 islands (6000 inhabited) scattered around the equator. It has a tropical, hot, humid climate with more moderate conditions prevailing in the highlands. Terrain is mostly coastal lowland, whereas the larger islands have interior mountains. Having a tropical climate with high levels of rainfall and humidity in most areas, several phytophthora diseases cause significant damage and are difficult to control.

*Phytophthora* spp. cause important diseases in agricultural, horticultural and industrial crops in Indonesia. At least 11 species of *Phytophthora* are reported to cause economic losses in Indonesia. *Phytophthora palmivora* has been identified as the most economically important *Phytophthora* species in Indonesia. It causes diseases on the largest number of economically important plant species (Table 4.2.1). In fact, it has been recorded as attacking more than 138 plant species. *Phytophthora palmivora* causes approximately 25–50% yield loss on cocoa, whereas *P. capsici* causes 52% yield reduction in pepper. However, the disease losses on most plants have not been accurately quantified.

*Phytophthora* spp. infect various parts of plants including roots, stems, leaves, and fruits. Disease symptoms vary depending on the host, species involved and the prevailing conditions.

*Phytophthora cinnamomi* is known to infect stems causing bark canker in cinchona and cinnamon, whereas *P. palvimora* infects all parts of cocoa, causing root rot, stem canker, pod rot, leaf blight and chupon blight.

Also known as a water mould, the life cycle of *Phytophthora* reflects adaptation to an aquatic environment. A tropical climate with prolonged wet conditions and relatively stable temperatures is very conducive for the pathogen. Disease epidemics normally occur during the wet season. Sporangia can either germinate directly by forming a germ tube, or differentiate into up to 50 biflagellate zoospores. Using their flagella, the zoospores can move actively in water for short distances before they encyst and germinate to initiate infections. In the soil, zoospores are attracted to the roots of plants. This mobility of zoospores to their host is a very important characteristic for the local spread and development of epidemics by *Phytophthora* species.

This review summarises the species of *Phytophthora* recorded, their hosts, distribution, and current control measures in Indonesia. The biology, epidemiology and control of the two most important species, namely *P. palmivora* and *P. capsici*, will also be presented. The review concludes with a discussion on the future research and implementation of integrated management to control phytophthora diseases.

<sup>1</sup> Research Institute for Industrial Crops, Jalan Cimanggu No. 3, Bogor 161111, Indonesia.

<sup>2</sup> Biotechnology Research Unit for Estate Crops, Jalan Taman Kencana No. 1, PO Box 179, Bogor 16151, Indonesia.

<sup>3</sup> Research Institute for Coconut and Palms, PO Box 1004, Manado 95001, Indonesia.

**Table 4.2.1** *Phytophthora* species, economically important hosts and diseases caused in Indonesia

<i>Phytophthora</i> species	Host	Scientific name	Diseases caused	Regions where pathogen has been recorded	Reference
<i>P. cactorum</i>	Apple Avocado	<i>Malus pumila</i> <i>Persia americana</i>	Collar rot Root rot, stem canker	Java	Sukirman and Semangun (1972) Triharso et al. (1975)
<i>P. capsici</i>	Chili Pepper	<i>Capsicum</i> spp. <i>Piper nigrum</i>	Fruit rot Foot rot	Java, Sumatra, Bangka, Kalimantan	Muller (1936) Anon. (1987)
<i>P. cinnamomi</i>	Cinchona Cinnamomum	<i>Chincona ledgeriana</i> <i>Cinnamomum burmannii</i>	Stem canker Stem canker	Java, Sumatra	Djafaruddin (1975) Winarto et al. (1980)
<i>P. citricola</i>	Cinchona	<i>Chincona ledgeriana</i>	Seedling dieback	Java	Semangun et al. (1977)
<i>P. citrophthora</i>	Citrus	<i>Citrus</i> spp.	Foot rot, gummosis	Java, Sumatra, Kalimantan	Toxopeus (1932) Muller (1939) Anon. (1987)
<i>P. colocasiae</i>	Taro	<i>Colocasia esculenta</i>	Leaf blight	Java	Semangun (1991a)
<i>P. infestans</i>	Potato Tomato	<i>Solanum tuberosum</i> <i>Lycopersicon esculentum</i>	Late blight Leaf blight, fruit rot	Java, Sumatra, Bali, Lombok, Sulawesi	Thung (1947), Suhardi et al. (1976) Anon. (1987)
<i>P. nicotianae</i>	Tobacco	<i>Nicotiana tabacum</i>	Black shank	Java, Sumatra	Thung (1938) Hartana and Soepeno (1978)
	Citrus Pineapple Roselle Vanilla	<i>Citrus</i> spp. <i>Ananas comosus</i> <i>Hibiscus sabdariffa</i> <i>Vanilla planifolia</i>	Foot rot Heart rot, root rot Black foot rot Pod rot	Java, Sumatra, Bali	Semangun (1991b) Toxopeus (1934) Oka and Prajati (1972) Anon. (1987)
<i>P. palmivora</i>	Cacao Citrus Coconut	<i>Theobroma cacao</i> <i>Citrus</i> spp. <i>Cocos nucifera</i>	Pod rot, stem canker Gummosis Bud rot, premature nutfall	All regions	Purwantara (1987) Triharso et al. (1975) Wahyuni and Hadisutrino (1976) Purwantara and Darmono (2003)
	Durian Orchid Papaya Rubber	<i>Durio zibethinus</i> <i>Vanda</i> spp. <i>Carica papaya</i> <i>Hevea brasiliensis</i>	Patch canker Black rot Root rot, fruit rot Black stripe, leaf fall		Prayudi (1988) Dwiastuti (1985)
<i>P. porri</i>	Shallot	<i>Allium ascalonicum</i>	Blight	Java	Anon. (1987)

## Diseases of Major Economic Importance

### Phytophthora diseases in cocoa

Cocoa is an important commodity for Indonesia. The total area planted to cocoa was 532,000 ha in late 1999. Just over 70% of cocoa farmers are smallholders. Indonesia is the world's third largest cocoa exporter. It produces 335,000 tonnes/year, which is valued at USD294 million. *Phytophthora palmivora* is a serious pathogen of cocoa, causing pod rot, stem and cushion cankers, leaf, chupon and seedling blights and sudden death (Sri-Sukanto 1985; Purwantara 1987). At the beginning of the last century, canker was very serious in Java, leading to the eradication of the very susceptible Criollo types of cocoa (Van Hall 1912, 1914). However, canker is no longer a menace in this area since Criollo has been replaced by Forastero types (Tollenaar 1958). In most areas, direct losses by pod infection leading to black pod rot are the most common cause of the problem. Newly set fruit up to fully mature pods are susceptible to infection.

There is a positive correlation between disease intensity and the number of pods per tree. Higher-yielding trees had a higher percentage of black pods than lower-yielding ones, as most of the latter escaped infection (Tollenaar 1958). When the incidence is high, an abundance of sporangia is produced. This, in turn, makes it more difficult to control the disease than when the incidence of pod infection is low. For the same reason, disease control becomes gradually easier in an area where the control measures have been executed systematically year after year.

Production of spores and the risk of infection are increased by high humidity. The incidence of phytophthora diseases can be very high in wet years and in humid areas. A combination of high rainfall and high humidity during the crop season will lead to severe losses. In West Java, cocoa plantations in areas with an annual rainfall of approximately 4000 mm at 400–600 m above sea level suffered very high incidence of pod rots and cankers (Purwantara 1990). Even in the absence of rain, infections still occur in these areas, as the humidities of nearly 100% that occur for a few hours during the night provide enough free water to initiate infection (Purwantara and Pawirosoemardjo 1990; Purwantara 2003). Poor drainage of plantations, high humidity due to heavy canopies, and low branching of trees increased disease incidence in mountainous areas of Java. Pruning of cocoa and removal of low branches provide some reduction in disease incidence. For

this reason, pod rot usually becomes increasingly serious as soon as the canopy has closed, this occurring after 5 to 7 years (Van Hall 1912).

Originally, disease control was attempted by removing the newly infected pods and burying them. Removal should be done every other day, as new spores are produced on pods within 2 days after the first symptoms are visible (Tollenaar 1958). However, even daily removal of the infected pods did not reduce disease incidence below economic threshold levels (A. Purwantara, unpublished data). It seems that infected pods are not the only source of infection in plantations. Other sources, including infected cushions and cankers, soils and insects are part of the disease cycle (Konam 1999).

Chemical spraying using copper-based fungicides has been practised in several cocoa plantations. However, because of wash-off in the wet season, these sprays provide only limited protection. Trunk injection with phosphonates provides good control of pod rot and stem canker in East Java (Y.D. Junianto, unpublished data), but this control technique is not widely adopted by growers. They are reluctant to drill holes in the trees because of they have limited information about the healing process of wounds from multiple and regular injections. The current recommendation for controlling the diseases is integrated management, including the reduction of inoculum from the soil by ground-cover management and removal of tent-building ants, adoption of wide plant spacing and regular pruning to reduce humidity in the canopy, removal of infected pods, frequent harvest to remove sources of secondary inoculum from the canopy, and trunk injection with phosphonate.

### Phytophthora diseases in coconut

The production of coconut and copra are extremely important activities in Indonesia. Annual copra production is 2.342 million tonnes (26% of world production) from 1.384 million ha (32% of world coconut area). The bulk of production is by smallholders, with other cash and food crops generally planted under coconut, which notably serves as a shade tree for cocoa. Breeding for improved varieties represents a national priority. The improved hybrid variety PB121 (MAWA) was successfully adopted by many smallholders, but in the early 1980s a disease of coconut causing budrot and premature nut fall was identified on this variety and now rates as the most significant disease affecting coconut production in the country.

Rots caused by *Phytophthora* species lead to palm death (by bud rot) and/or yield reduction (by

premature nut fall) (Waller and Holderness 1997), and are the major disease problems affecting coconut in Indonesia (Lolong et al. 1998). While most of the coconut-growing regions of the world are affected by phytophthora rots, Indonesia and the Philippines are the worst affected (Renard 1992). In Indonesia and the Philippines, *P. palmivora* seems to be the main causal agent of disease (Blaha et al. 1994). Coconut bud rot has an irregular distribution in the field, but the highest incidence seems to correlate with the wettest areas (Waller and Holderness 1997) and with plantings of the susceptible hybrid, PB121 (MAWA).

Bud rot and nut fall were first reported in Indonesia in 1985, the causal agents being identified as *P. palmivora* and *P. nicotianae* (Bennett et al. 1986). During this time, outbreaks of the disease resulted in severe damage to plantations (Renard 1992). Since that time, almost all areas planted to coconut in Indonesia have suffered serious damage from bud rot, with losses above 80% (Darwis 1992). The severity of disease is linked to the introduction of high-yielding hybrid breeding lines from West Africa (MAWA). These are highly susceptible to phytophthora (see also Chapter 6.2). In Indonesia, although *P. palmivora* seems to be the main causal agent of bud rot and nut fall in coconut (Blaha et al. 1994; Waller and Holderness 1997), *P. arecae* and *P. nicotianae* have also been found in association with these diseases (Thevenin 1994) in a small number of cases. Nut damage is usually most severe in immature bunches during the rainy season. *Phytophthora* spores proliferate and then spread horizontally (by contact between bunches) or vertically (between nuts within a bunch) (Renard and Darwis 1992). The diseases cause extensive losses of both stands and nut production. In some areas, stand losses of 43% can occur due to bud rot. Premature nut fall, which is the more common disease, affects nuts 3–7 months old (Lolong et al. 1998), and can cause losses of 50–75% (Brahmana et al. 1992). The incidence of bud rot is higher in the lowland areas of Indonesia than in the highlands. Resistance among coconut varieties to infection and damage by phytophthora varies with location, and therefore it is recommended that several varieties be planted to minimise the risk of damage caused by the pathogen (Mangindaan et al. 1992).

From field observations and inoculation studies, some varieties have been found to be resistant in Sulawesi, but knowledge of the variation in the pathogen populations is required for successful resistance breeding programs. The susceptibility of PB121 to *P. palmivora* has been linked to its parental lines, yet these parental lines continue to be used in

breeding programs due to their favourable early and high-yielding characteristics. National plant breeding programs are on the way to ensure that the next generation of recommended coconut varieties planted in Indonesia is not susceptible to *P. palmivora*. An increased understanding of this disease will enable the development of improved disease-management procedures

### Foot rot in pepper

*Phytophthora capsici* Leonian causes the most destructive and economically significant disease of black pepper (*Piper nigrum* L.). The fungus attacks all parts and growth stages of the black pepper plant. If it attacks the root or collar, it causes sudden death. This disease was first reported in Lampung in 1885, and has been known as foot rot disease since 1928 (Muller 1936). The causal agent was first identified as *P. palmivora* var. *piperis* (Muller 1936), then in 1985 it was recognised as *P. palmivora* MF4 (Tsao et al. 1985) and later renamed *P. capsici sensu lato* (Tsao and Alizadeh 1988). Nowadays, the disease is found in almost all areas where pepper is cultivated in Indonesia.

Pepper (black and white final products) is the seventh most important export income earner for Indonesia. The total area planted is about 136,450 ha. The crop is produced by over 132,000 farmers, most of whom are smallholders. They care for and control their cultivations when the pepper price is high, but neglect them if the price falls. All cultivated pepper varieties grown in Indonesia are susceptible to the disease. Vines more than 3 years old seem to be the most susceptible to foot rot (Holliday and Mowat 1963).

### Population Biology

Some understanding of the biology and epidemiology of phytophthora diseases has been achieved. Populations of heterothallic *P. palmivora* attacking coconut and cocoa in Indonesia consist of only one mating type (A1). In contrast, populations from papaya consist of mating type A2. No oospores have been reported in the field so far. Molecular analysis of this *P. palmivora* population showed limited genetic diversity amongst isolates originating from coconut. *P. palmivora* affecting cocoa was shown to be genetically distinct from that isolated from coconut and this distinction was confirmed by pathogenicity assessments. Two mating types have been reported in *P. capsici* attacking black pepper (Manohara et al. 2002). However, the importance of sexual reproduction in enhancing genotypic diversity in *P. capsici* populations and the importance of the formation of



oospores as long-term survival structures have not been determined

## Control of *Phytophthora* Diseases in Indonesia

Most phytophthora diseases can be cost-effectively controlled only through a well thought out integrated disease management program that incorporates, in an appropriate way, the several control measures that are available. There is still considerable scope for research into several aspects of the resistance of plants and the genetics of the pathogen, especially in understanding mechanism of pathogenesis, host specificity of the phytophthora pathogens and in the development of sustainable and cost-effective integrated disease-management practices.

The impact of phytophthora disease can be reduced through manipulation of the environment, such as by reducing humidity in orchards through pruning, weeding and good drainage. Sanitation and crop rotation also provide good control, such as in black shank of tobacco, where crop rotation and monitoring of the pathogen population in the soil through baiting with tobacco leaves has been implemented since before the 1940s (Semangun 1991a,b). Planting resistant varieties will be the best option for controlling the disease. However, such material is not available for all disease systems and against all species of *Phytophthora*. Selection and breeding for resistance to phytophthora have provided an effective means of controlling some phytophthora diseases of economic importance in Indonesia. In some plant species, such as in tobacco, resistance has been identified. Nevertheless, the genetics of resistance in many tropical crops, especially tree crops, is not fully understood and needs significantly more investigation. In disease systems, such as black pepper-*P. capsici*, sources of resistance appear to be limited. Introductions of plant materials with higher levels of resistance to phytophthora diseases are urgently needed. Late blight of potato and black shank of tobacco have been sufficiently controlled by moderately resistant cultivars. Chemical control relies on copper-based fungicide as protective measures, and systemic fungicides such as metalaxyl and phosphonates.

Biological control may become an alternative control for phytophthora diseases, as it is considered as an environmentally safe form of disease control. In an attempt to reduce the use of fungicides in response to increases in price and environmental concerns, research on biological control has been conducted

for several *Phytophthora* species. Various fungi, actinomycetes and bacteria have been isolated and proven to control the pathogen in glasshouse trials. However, the effectiveness of these biological control agents needs to be demonstrated and validated in the field.

## References

- Anon. 1987. Daftar organisme pengganggu tumbuhan penting yang dilaporkan telah terdapat di dalam wilayah Republik Indonesia. Jakarta, Pusat Karantina Pertanian, 138 p. Cited in Semangun (1991a).
- Bennett, C.P., Roboth, O., Sitepu, G. and Lolong, A. 1986. Pathogenicity of *Phytophthora palmivora* (Butl.) causing premature nutfall disease of coconut (*Cocos nucifera* L.). Indonesian Journal of Crop Science, 2, 59–70.
- Blaaha, G., Hall, G., Warokka, J.S., Concibido, E. and Ortiz-Garcia, C. 1994. *Phytophthora* isolates from coconut plantations in Indonesia and Ivory Coast: characterisation and identification by morphology and isozyme analysis. Mycological Research, 98, 1379–1389.
- Brahamana, J., Lubis, A.U. and Chenon, R.D. 1992. Evolution of coconut bud disease and strategy of control. Paper presented at coconut *Phytophthora* workshop, Manado, Indonesia.
- Darwis, S.N. 1992. *Phytophthora* in relation to climate and coconut cultivar. Paper presented at coconut *Phytophthora* workshop, Manado, Indonesia.
- Djafaruddin. 1975. *Phytophthora cinnamomi*, penyebab penyakit kanker pada kayu manis Padang di Sumatera Barat. Proceedings of the Indonesian Plant Pathology Society Conference 1976, Bogor. Cited in Semangun (1991b).
- Dwiastuti, M.E. 1985. Penelitian penularan dan pengendalian penyakit busuk buah pepaya. Proceedings of the Indonesian Plant Pathology Society Conference 1985, Jakarta, 112–113. Cited in Semangun (1991a).
- Hartana, I. and Soepeno 1978. Pewarisan ketahanan terhadap penyakit kolot basah (*Phytophthora parasitica* var. *nicotianae*) pada tembakau cerutu Indonesia. Menara Perkebunan, 46, 55–60. Cited in Semangun (1991b).
- Holliday, P. and Mowat, W.P. 1963. Foot rot of *Piper nigrum* L. (*Phytophthora palmivora*). Phytopathological Paper, No. 5, 1–62.
- Konam, J.K. 1999. Integrated management of *Phytophthora palmivora* diseases of cocoa in Papua New Guinea. PhD thesis, University of Melbourne, 255 p.
- Lolong, A., Smith, J.J. and M. Holderness, M. 1998. Characterisation of *Phytophthora* diseases of coconut in Indonesia. Paper presented at International Congress of Plant Pathology, Edinburgh, Scotland. Paper number 3.7.87.
- Mangindaan, H.F., J.M. Thevenin, S. Kharie, and H.F. Motulo. 1992. The susceptibility of coconut varieties to *Phytophthora* in Indonesia: the effect of environmental factors. Paper presented at coconut *Phytophthora* workshop, Manado, Indonesia.

- Manohara, D., Mulya, K. and Purwantara, A. 2002. *Phytophthora capsici* on black pepper in Indonesia. Paper presented at International Conference on Tropical and Subtropical Plant Pathology, Chiang Mai, Thailand.
- Muller, H.R.A. 1936. The Phytophthora foot rot of black pepper (*Piper nigrum* L.) in the Netherlands Indies. Cited in Review of Applied Mycology, 16, 559.
- 1939. Overzicht van de belangrijkste citrus ziekten in Ned. Indie. Mededelingen Inst. Plziekten 94. Cited in Semangun (1991a).
- Oka, I.N. and Prajati, S. 1972. Percobaan resistensi lapangan dari beberapa nomor seleksi *Hibiscus sabdariffa* L. dan *H. cannabinus* L. terhadap *Phytophthora sabdariffae*. Bulletin Lembaga Penelitian Tanaman Industri, 13, 1-5. Cited in Semangun (1991b)
- Prayudi, B. 1988. Kajian ketahanan *Phytophthora palmivora* dan *P. infestans* terhadap beberapa fungisida. Yogyakarta, Universitas Gadjah Mada, Disertasi, 150 p. Cited in Semangun (1991a).
- Purwantara, A. 1987. Penyebab penyakit Phytophthora pada tanaman kakao di Jawa. In: Proceedings of the Indonesian Plant Pathology Society Conference 1987, Surabaya, 283-290.
- Purwantara, A. 1990. Pengaruh beberapa faktor cuaca terhadap infeksi *Phytophthora palmivora* pada buah kakao. Menara Perkebunan, 58, 78-83.
- Purwantara, A. 2003. Epidemiology and control of Phytophthora diseases of cocoa in Java, Indonesia. Paper presented at International Congress of Plant Pathology, at Christchurch, New Zealand. Paper number 28.1.
- Purwantara, A. and Darmono, T.W. 2003. Incidence of Phytophthora leaf fall of Hevea rubber in Java. Paper presented at International Congress of Plant Pathology, Christchurch, New Zealand. Paper number 28.2.
- Purwantara, A. and Pawirosoemardjo, S. 1990. Fluktuasi intensitas penyakit *Phytophthora* pada buah kakao di daerah basah. Menara Perkebunan, 58, 44-50.
- Renard, J.L. 1992. Introduction to coconut *Phytophthora* diseases. Paper presented at coconut *Phytophthora* workshop, Manado, Indonesia.
- Renard, J.L. and Darwis, S.N. 1992. Report on the coconut *Phytophthora* disease seminar. Paper presented at coconut *Phytophthora* workshop, Manado, Indonesia.
- Semangun, H. 1991a. Penyakit-penyakit tanaman hortikultura di Indonesia. Yogyakarta, Gadjah Mada University Press, 850 p.
- 1991b. Penyakit-penyakit tanaman perkebunan di Indonesia. Yogyakarta, Gadjah Mada University Press, 808 p.
- Semangun, H., Martosupono, M., Saleh, N. and Rohmat 1977. Penyakit mati-ujung pada semai dan sambungan kina. Warta Balai Penelitian Teh dan Kina, 3, 159-166. Cited in Semangun (1991b).
- Sitepu, D. 1993. Disease management on pepper. Indonesian Agricultural Research and Development Journal, 15(2), 31-37.
- Sri-Sukamto 1985. *Phytophthora palmivora*, salah satu jamur penyebab penyakit pada tanaman cokelat. Menara Perkebunan, 53, 7-11.
- Suhardi, Bustamam, M., Bismo, Vermeulen, H. and Widjorini, S. 1976. The chemical control of *Phytophthora infestans* on tomatoes in Indonesia. Bulletin Penelitian Horticultura, 4, 45-54. Cited in Semangun (1991a).
- Sukirman and Semangun, H. 1972. Notes on apple diseases in Indonesia. Paper presented at South East Asia regional symposium on plant diseases in tropics, Yogyakarta, September 1972. 5 p. Cited in Semangun (1991b).
- Thevenin, J.M. 1994. Coconut diseases in Indonesia — etiological aspects. Paper presented at coconut *Phytophthora* workshop, Manado, Indonesia, cited in Waller and Holderness (1997).
- Thung, T.H. 1938. De epidemiologie van *Phytophthora parasitica* var. *nicotianae* op de Vorstenlandsche Tabak Ondernemingen. Mededelingen Proefstation Vorst. Tabak 86. Cited in Semangun (1991b).
- Thung, T.H. 1947. Over de verspreiding van plantenziekten. Landbouw 19. Cited in Semangun (1991a).
- Tollenaar, D. 1958. *Phytophthora palmivora* of cocoa and its control. Netherlands Journal of Agricultural Science, 6, 24-38.
- Toxopeus, H.J. 1932. Nadere gegevens over de gomziekte in jeruk manis (*Citrus sinensis*) en haar bestrijding. Mededelingen Inst. Plziekten 80. Cited in Semangun (1991a).
- 1934. Onderzoekingen over het invloed van temperatuur en vochtigheid op de levensproces van *Phytophthora parasitica*. Landbouw 9, 385. Cited in Semangun (1991a).
- Triharso, Kaselan, J. and Sumardiyono, C. 1975. List of diseases of important economic crop plants already reported in Indonesia. Bulletin Fakultas Pertanian Universitas Gadjah Mada, 14, 1-60. Cited in Semangun (1991b).
- Tsao, P.H. and Alizadeh, A. 1988. Recent advances in the taxonomy and nomenclature of the so-called "*Phytophthora palmivora*" MF4 occurring on cocoa and other tropical crops. Paper presented at 10th International Cocoa Research Proceedings, Santo Domingo, 17-23 May 1987.
- Tsao, P.H., Kasim, R. and Mustika, I. 1985. Morphology and identity of black pepper *Phytophthora* isolates in Indonesia. FAO Plant Protection Bulletin, 33, 61-66.
- Van Hall, C.J.J. 1912. De cacao-kanker op Java en zijn bestrijding. Mededelingen Proefstation Midden-Java, 6, 1-17.
- 1914. De bestrijding van de cacao-kanker op de onderneming Kemiri (Pekalongan). Mededelingen Proefstation Midden-Java, 14, 1-10.
- Waller, J.M. and Holderness, M. 1997. Beverage crops and palms. In: Hillocks, R.J. and Waller, J.M., ed., Soilborne diseases of tropical crops. Wallingford, Oxon, UK, CAB International.

Wahyuni, W.S. and Hadisutrisno, B. 1976. Identifikasi penyakit jamur dan bakteri pada beberapa tanaman anggrek di Daerah Istimewa Yogyakarta. Proceedings of the Indonesian Plant Pathology Society Conference 1976, Bandung, 8 p. Cited in Semangun (1991a).

Winarto, A., H. Semangun, and M. Martosupono. 1980. Penyakit kanker batang pada kina. Warta Balai Penelitian Teh dan Kina, 6, 187-198. Cited in Semangun (1991b).

## 4.3 Phytophthora Diseases in Thailand

Somsiri Sangchote,<sup>1</sup> Srisuk Poonpolgul,<sup>2</sup> R. Sdoodee,<sup>3</sup>  
M. Kanjanamaneesathian,<sup>4</sup> T. Baothong,<sup>5</sup> and Pipob Lumyong<sup>6</sup>

### Abstract

Phytophthora diseases have been recorded on durian, rubber, black pepper, cocoa, citrus, potato and pineapple in Thailand. *Phytophthora palmivora* is the predominant species and is found on many different crops. It has a wide host range and shows considerable morphological variability. Epidemiological studies indicate that rainfall has a significant influence on disease development. Control of phytophthora diseases is difficult, and research efforts are directed towards using biological and chemical control as part of integrated disease control practices.

### Introduction

Many *Phytophthora* species have been reported in Thailand: *Phytophthora nicotianae*, causing root and fruit rot of citrus (Wichiencharoen 1990); *P. palmivora*, causing pod rot of cocoa (Kasaempong 1991) and patch canker and fruit rot of durian (Bhavakul and Changsri 1969); and other crops as shown in Table 4.3.1.

### Root and Stem Rot of Durian

Durian has been grown commercially in Thailand since 1800, and there are at least 68 cultivars grown in the area around Bangkok and Nonthaburi. In 1942, there was a serious flood in the area that damaged most of the durian plantations. Since then, farmers recultivated durian by propagating the E-Luang cultivar as a monocrop. In 1966, root and stem rot of durian was reported on 20-year-old trees and, in 1967, durians in Chantaburi Province

showed symptoms of root rot, especially when grown near irrigation lines and canals.

The causal agent was identified as *P. palmivora* (Chee 1969). Monthong, E-loung and Chanee cultivars were reported as being susceptible to *P. palmivora*. The symptoms that appeared on roots and stems can be described as dark brown to black discoloration with rotting root and bark on the base of the trunk. In years when there is a long wet season and turbulent strong winds, the symptoms can be found on twigs as high as 10 m above the ground. Mycelium can infect leaves and young shoots and produce white, fluffy mycelium on the lesion under humid conditions. Some investigations have reported that beetles, termites and ants may be involved in carrying the fungus up into the canopy of the tree.

### Root and Stem Rot of Black Pepper

The causal agents have been reported as *P. palmivora*, *P. nicotianae* and *P. capsici*. The pathogens infect the roots of black pepper vine below the soil line. The first symptoms appear as dark brown to black lesions at the tip of the young root. Nodes on the upper part can be removed easily. Small lesions enlarge and merge into larger lesions and turn black with age. Infected leaves, pedicels and flowers show rot symptoms, while the fruit turns brown, dries and wilts. If young vines are infected, the plants die in 1–2 months. If the pathogen infects older vines, the plants show a significant decline in yield before dying.

<sup>1</sup> Department of Plant Pathology, Kasetsart University, Bangkok-10900, Thailand. Email: <agrsrs@ku.ac.th>.

<sup>2</sup> Plant Pathology and Microbiology Division, Department of Agriculture, Bangkok 10900, Thailand.

<sup>3</sup> Faculty of Natural Resource, Prince of Songkhla University, Hat Yai, Thailand 90112.

<sup>4</sup> Faculty of Industrial Technology, Prince of Songkhla University, Surat Thani Campus, Thailand 84100.

<sup>5</sup> Surat Thani Biological Pest Management Centre, Department of Agricultural Extension, Thailand 80220.

<sup>6</sup> Department of Plant Pathology, Chiang Mai University, Chiang Mai 50200, Thailand.

**Table 4.3.1** *Phytophthora* species reported from different plants in Thailand.

<i>Phytophthora</i> species	Host	Common name	References
<i>P. botryosa</i>	<i>Hevea brasiliensis</i>	rubber	Suzui et al. (1979)
<i>P. capsici</i>	<i>Piper nigrum</i>	black pepper	Tsao and Tummakate (1977)
<i>P. infestans</i>	<i>Solanum tuberosum</i>	potato	Tucker (1933)
<i>P. meadii</i>	<i>Hevea brasiliensis</i>	rubber	Chee and Greenwood (1968)
<i>P. nicotianae</i>	<i>Citrus</i> spp. <i>Durio zibethinus</i> <i>Ananas comosus</i> <i>Piper nigrum</i>	citrus durian pineapple black pepper	Suzui et al. (1979) Suzui et al. (1979) Suzui et al. (1979) Silayoi et al. (1983)
<i>P. palmivora</i>	<i>Piper nigrum</i>  <i>Durio lowianus</i> <i>Durio zibethinus</i> <i>Hevea brasiliensis</i> <i>Theobroma cacao</i> <i>Euphorbia longana</i> <i>Mangifera indica</i> <i>Ananas comosus</i>	black pepper  wild durian durian rubber cocoa longan mango pineapple	Krengpiem et al. (1989); Kunloun (1967); Tsao and Tummakate (1977) Kumjaipai (1974) Suzui et al. (1979) Tsao et al. (1976) Chomenansilpe et al. (1983) Bhavakul et al. 1997 Kueprakone et al. (1986) Suzui et al. 1979)

### Coconut Nut Drop

Malayan yellow dwarf, an imported coconut cultivar, showed heavy nut fall in a breeding plot at Chumporn Horticulture Research Centre in 1968. Investigations revealed that *P. palmivora* had infected the coconut. Symptoms on the nut were found at the base of the pedicel attached to the outer carp. The pathogen can infect the fruit at 2–8 months stage, and often the disease lesion starts from the pedicel base down inside the fruit to the young shell. In moist conditions, fluffy white mycelium can be seen at the early stages of infection but not in the later stages. Infected nuts die prematurely. Symptoms can also be found on shoots of the seedling while it germinates. Studies on the host range revealed that mangosteen, tangerine, lime, coffee, rambutan, black pepper, cocoa and pineapple can be infected with *P. palmivora*. So far, nut drop has not been reported from any other coconut cultivars in Thailand.

### Black Rot Disease of Vanilla

Vanilla is a crop of economic value due to its aromatic flavour that is used in the manufacture of chocolate, ice-cream, soft drinks, cakes and snacks. Black rot disease of vanilla is the most severe disease limiting vanilla production. The disease was first found at Maehae Highland Agriculture Station in Chiangmai Province. Symptoms first appeared as yellowing on leaves and stems. The pathogen, *P. palmivora*, infects the roots and foot, developing into black rot in the roots, foot, stem and leaves. Furthermore, the causal agent can directly infect the

shoot and leaves, again leading to black rot and death of the plant. The disease can become epidemic during prolonged periods of high humidity in the rainy season. Rain splash helps the dispersion of zoospores from infected soil up to the plant.

### Longkong (longan) root rot

The symptoms of longkong root rot appeared on 2-year-old longkong seedlings on langsat root stock at Chantaburi Province in 1999. Characterisation of the sporangium, chlamydospores and mating types revealed that the pathogen was *P. nicotianae*.

### Leaf Fall and Black Stripe in Rubber

Rubber is an important crop to Thailand, especially in the south, where average annual rainfall is 2000–3000 mm and average temperature is 28±2°C. There are several diseases caused by different *Phytophthora* species that limit rubber production in Thailand. The major diseases are phytophthora leaf fall and black stripe.

Leaf fall, caused in Thailand by *P. palmivora*, is characterised by individual leaves turning yellow. The lesions turn dark brown to black and often show white spots of coagulated latex in the centre of the lesion. Leaf fall is most common soon after the monsoon season has started and may give rise to serious defoliation. In addition to the leaves, the fruit may also be infected. The fruit may be covered with sporulating mycelium during periods of high humidity. In contrast to the leaves, the infected fruit turn dark but remain attached to the tree.

Black stripe is a disease of the tapping panel caused by *P. palmivora* and *P. botryosa*. Sunken discoloured areas appear on the tapping panel, and when the bark is cut away, characteristic vertical black lines become apparent. The disease develops rapidly in wet weather.

Disease surveys in rubber-growing areas in 1976, including 3 provinces in the east and 14 in the South, indicated that 10% of the total area was infested (Kajornchaiyakol 1977). Phytophthora diseases were prominent in the western coast of the South due to long periods of wet, humid weather with few periods of sunshine (Kajornchaiyakol 1977). However, phytophthora diseases have, in recent years, been less troublesome than previously, due to the introduction of more-resistant clones in the affected areas (Chantarapratin et al. 2001).

### Root Rot, Gummosis and Brown Rot in Citrus

Thailand is among the largest producers of a wide range of different citrus fruits in Southeast Asia. However, due to the prevailing wet climatic conditions, phytophthora is a major impediment to production. In Thailand, citrus is grown by smallholders as well as on large plantations. Root rot and foot rot are common in many citrus species, especially after prolonged periods of wet weather. Gummosis, a rotting of the bark due to phytophthora growing into the cambium and producing a necrosis, is often accompanied by the exudation of water soluble gum. Brown rot of the fruit is common under wet conditions.

**Table 4.3.2** Major phytophthora diseases in southern Thailand.

Crop	Disease	Disease occurrence <sup>a</sup> (%)
Rubber	Leaf fall and black stripe	10 <sup>b</sup>
Durian	Root and stem rot	3.5
Citrus	Root and stem rot	0.5
Robusta coffee	Root and stem rot	0.4

<sup>a</sup> Average percentage of infested area per year (Source: Department of Agricultural Extension, southern unit, Songkhla).

<sup>b</sup> Infested area surveyed in 1976 (Kajornchaiyakol 1977)

### Occurrence of *Phytophthora*

Although root and stem rot in fruit crops caused by *Phytophthora* was endemic in the south (Table 4.3.2), the infested area was less than 10% of the total

growing area in each year. This was an average from reports on plant pests in southern Thailand during 1998–2001 in the Thailand Department of Agricultural Extension annual report. However, in certain years the damage to durian (Table 4.3.3) was high in particular areas: 51%, 41% and 38% in Chumporn, Ranong and Surat Thani provinces, respectively, in southern Thailand in 2001. Although the percentage infestation by phytophthora disease in citrus was low in southern provinces, in other areas the disease had devastated particular orchards (Figure 4.3.1). In addition, the damage by phytophthora disease in coffee was insignificant because the disease incidences were reported only in 2001 with 0.4% infestation.



**Figure 4.3.1** Phytophthora stem rot in a Shogun mandarin orchard in southern Thailand.

**Table 4.3.3** Incidence of root and stem rot in durian in southern Thailand, 2001.

Province	Growing area (ha)	Infested area (%)
Chumporn	21,490	51
Ranong	3,732	45
Surat Thani	6,174	34
Nakan Si Thammarat	20,532	27
Phangnga	2,150	38
Krabi	1,400	20
Phuket	592	7

Source: Surat Thani Biological Pest Management Centre, Department of Agricultural Extension.

Considerable amounts of fungicides have been used to manage phytophthora diseases in fruit crops, particularly in durian. Biological control measures using antagonistic fungi (*Trichoderma harzianum*) to suppress phytophthora infestation has increased in the past 3 years. The biological control agents were

distributed to farmers through Surat Thani and Songkla Biological Pest Management Centre, Department of Agricultural Extension. Selection and utilisation of resistant varieties to control phytophthora root rot in durian is continuing. Screening for resistance to *P. palmivora* in selected durian seedlings (Figure 4.3.2) for the selection of more-resistant rootstock has been developed by the Tropical Fruit and Plantation Crop Research Centre, Prince of Songkhla University since 1997. A few promising clones have been detected from Nakorn Sri Tammarat, Songkhla and Narativat provinces (Kanjamaneeesathian et al. 2000).

In Thailand, the mating type of *P. palmivora* is reported as A1, *P. nicotianae* is both A1 and A2, and *P. botryosa* is A1 and A2. *P. palmivora* isolates obtained from cocoa and durian showed variability in their colony characteristics (Kasaempong 1991).

### Research on *Phytophthora* in Thailand

Phytophthora diseases are a major constraint to the production of many crops in Thailand. The most common *Phytophthora* species are *P. palmivora*, *P. nicotianae* and *P. botryosa*. *Phytophthora* isolates show a high level of variation and wide host range. Epidemiological studies of *Phytophthora* pathogens conducted in durian, citrus, and cocoa indicated that high levels of rainfall in many parts of Thailand are a major contributing factor to disease incidence.

Thailand has, in the more recent past, built up a considerable level of experience in plant pathology and mycology, including of *Phytophthora* species, at different universities and research organisations. However, in order to more effectively control plant diseases such as phytophthora in a range of different areas, a higher level of collaboration between the various research and extension providers is needed.



In an effort to indicate what research has been conducted on phytophthora in Thailand and to foster further collaborations we have listed the most recent research reports (Table 4.3.4) and research theses (Table 4.3.5).

### References

- Chantarapratin, U., Pattanakul, P., Changreung, N., Rojanasujit, A., Romreunsukarom, P. and Ramlee, A. 2001. Rubber diseases survey on large scale clone trail. Research Report: Rubber Research Institute Thailand.
- Chee, K.H. 1969. Hosts of *Phytophthora palmivora*. Review of Applied Mycology, 48:337-344.
- Chee, K.H. and Greenwood, J.M. 1968. Phytophthora leaf fall and pod rot of *Hevea brasiliensis* in Thailand. FAO Plant Protection Bulletin, 16(1), 1-5.
- Chomenansilpe, S., Doungpunya, A. and Nilprapai, C. 1983. Black pod disease of cocoa. Journal of Thai Phytopathological Society, 3, 222-224.
- Kajornchaiyakol, P. 1977. Survey of Phytophthora diseases in 1976. Thai Journal of Agricultural Science 10, 427-436.
- Kanjamaneeesathian, M., Te-chato, S., Chantararat, S., Luang-Aram, T. and Bunjerdpradit, B. 2000. Searching for local durians *Durio zibethinus* (Murr.) resistant to *Phytophthora palmivora* (Butl.) Butl. in Southern Thailand. Thai Journal of Agricultural Science, 32, 111-125.
- Kasaempong, Y. 1991. Black pod rot of cocoa (*Theobroma cacao* L.) caused by *Phytophthora palmivora* (Butl.) Butl. in Thailand. Bangkok, Kasetsart University.
- Krengpiem, P., Silayoy, E., Khingduang, S., Raktham, S. and Leelasattakul, K. 1989. Study on the causal organism of foot and root rot disease of black pepper. Bangkok, Annual Report, Division of Plant Pathology and Microbiology, Department of Agriculture.
- Kueprakone, U., Saengkong, S., Pienpuck, K. and Choobumroong, W. 1986. *Phytophthora palmivora* (Butl.) Butl., the casual organism of black rot of mango seedlings. Journal of Thai Agricultural Research, 4, 67-73.



Figure 4.3.2 Screening of durian leaves for resistance to *Phytophthora palmivora*.

**Table 4.3.4** Research papers listed in annual reports of the Division of Plant Pathology and Microbiology Department of Agriculture, Bangkok

Year	Title	Author
1994	The study on resistant rootstock of avocado root rot caused by <i>Phytophthora cinnamomi</i> .	Phavakul, K., Kraturisha, C., Koariyakul, S. and Tossapol, M.
1993	Studies on diseases of aloe	Chingduang, S. and Silayoy, E.
1993	Black rot disease of vanilla.	Chingduang, S., Silayoy, E., Likhitearaj, S. and Sasipalin, S.
1993	Selection of some durian rootstocks for their resistance to phytophthora root and stem rot.	Kraturisha, C., Vichitrananda, S., Pingkusol, S. and Leelasettakul, K.
1989	Study on disease of betel vine.	Krengpiem, P., Silayoy, E., Chingduang, S. and Raktham, S.
1989	Study on the causal organism of foot and root rot disease of black pepper.	Krengpiem, P., E. Silayoy, S. Khingduang, S. Raktham and K. Leelasettakul.
1988	Study on varietal reaction of black pepper to foot rot disease under field condition.	Silayoy, E., Leelasettakul, K., Krengpiem, P., Tummakate, A., Kraturisha, C. and Suksawat, S.

**Table 4.3.5** Theses, Department of Plant Pathology, Kasetsart University, Bangkok.

Thesis title	Author	Year
Genetics of the resistance to <i>Phytophthora sojae</i> in soybean ( <i>Glycine max</i> (L.) Merrill).	Sriphacet, S.	2002
Influence of organic fertiliser from the glutamic acid fermentation on <i>Phytophthora parasitica</i> (Dastur.) and other moulds in tangerine orchard soil.	Phonyangsong, P.	2000
Screening for local durian in southern Thailand resistance to <i>Phytophthora palmivora</i> (Butl.) Butl. by pathogenicity test and isozyme.	Bunjuerdpradit, B.	1999
Efficacy of antagonistic microorganisms for the protection of tangerine root rot caused by <i>Phytophthora parasitica</i> (Dastur.).	Kitjaideaw, A.	1998
Application of <i>Trichoderma harzianum</i> to control root rot of durian caused by <i>Phytophthora palmivora</i> (Butl.) Butl.	Roungwiset, K.	1997
Application of an antagonistic microorganism for the control of root rot of tangerine caused by <i>Phytophthora parasitica</i> (Dastur.).	Seemadua, S	1997
<i>Phytophthora</i> disease or rubber ( <i>Hevea brasiliensis</i> Muell.-Arg.): identification, clonal reaction and some chemical control	Srisa-arn, P.	1995
Selection and application of antagonistic microorganisms to control root and stem rot of durian caused by <i>Phytophthora palmivora</i> (Butl.) Butl	Awarun, S.	1994
Effects of antagonistic microorganism used in combination with organic fertilizer and fungicides on root rot of tangerine caused by <i>Phytophthora parasitica</i> (Dastur.).	Intasorn, S.	1994
<i>Phytophthora</i> : identification and detection of fungicide resistance by electrophoresis.	Jamjanya, S.	1994
Fungitoxicity of systemic fungicides and their control efficacy against phytophthora rot and foot rot of tangerine.	Plongbunchong, T.	1992
Studies on tissue culture derived potato plant and callus for resistance to culture filtrate of <i>Phytophthora infestans</i> (Mont.) de Bary and biological control.	Sanyong, S.	1992
Black pod rot of cocoa ( <i>Theobroma cacao</i> L.) caused by <i>Phytophthora palmivora</i> (Butl.) Butl.	Kasaempong, Y.	1991
Nutritional status in leaves of durian cv. Mon Thong infected with different levels of <i>Phytophthora palmivora</i> (Butl.) Butl.	Udomsriyothin, T.	1991
Efficacy of mono-dipotassium phosphite against <i>Phytophthora palmivora</i> (Butl.) Butl. on durian.	Bunyanupapong, K	1990
Influence of soil microorganisms on tangerine root rot caused by <i>Phytophthora parasitica</i> (Dastur.).	Termkietpisarn, A.	1990
Epidemiology and chemical preventive control of <i>Phytophthora</i> root and foot rot of tangerine at Rangsit irrigated area.	Wichiencharoen, A.	1990



- Kumjaipai, W. 1974. Citrus diseases in Thailand. Paper presented at 13th National Conference on Agriculture (Biological Sciences), Kasetsart University, Bangkok, Thailand.
- Kunloun, S. 1967. Studies on root rot of *Piper nigrum* Linn. Bangkok, Thailand, Kasetsart University, M.Sc. thesis.
- Silayoi, I., P. Krengpiem, S. Boonthai, and A. Foongkiatpaiboon. 1983. Root rot disease of betel vine. *Journal of the Thai Phytopathological Society*, 3, 10-13.
- Phavakul, K. and Changsri, V. 1969. Root rot of durian tree. *Proceedings of a seminar on plant protection, Agricultural Science Society Thailand*, 60-61.
- Phavakul, K., Teamsakul, P. and Tospol, M. 1997. Root rot disease of longan. *Thai Phytopathology*, 12, 123-128.
- Suzui, T.J., Kueprakone, U. and Kamphangridthrong, T. 1979. *Phytophthora* spp. Isolated from some economic plants in Thailand. *Technical Bulletin Tropical Agricultural Research Center*, 12, 32-41. Cited in *Review of Plant Pathology*, 59, 2090.
- Tsao, P.H. and Tummakate, R. 1977. The identity of a *Phytophthora* species from black pepper in Thailand. *Mycologia*, 69, 631-637.
- Tsao, P.H., Tummakate, A. and Bhavakul, K. 1976. Recovery of *Phytophthora* species from old, badly decayed, infected tissues of *Hevea brasiliensis*. *Transactions of the British Mycological Society* 66, 557-558.
- Tucker, C.M. 1933. Distribution of the genus *Phytophthora*. *University of Missouri Agricultural Experiment Station Research Bulletin* 184, 80.
- Wichiencharoen, A. 1990. Epidemiology and chemical preventive control of *Phytophthora* root and foot rot of tangerine at Rangsit irrigated area. Bangkok, Kasetsart University.

## 4.4 Phytophthora Diseases in Vietnam

Dang Vu Thi Thanh,<sup>1</sup> Ngo Vinh Vien<sup>1</sup> and André Drenth<sup>2</sup>

### Abstract

Phytophthora diseases have been reported from a range of crops in Vietnam. This chapter provides an overview of the *Phytophthora* species identified and the relative importance of phytophthora diseases on a range of crops including tomato, potato, pineapple, taro, durian, citrus, plum and rubber.

### Introduction

Vietnam is a country with two distinct climatic regions: the subtropical region north of the Haivan Mountains, which has four distinct seasons, and the tropical region to the south, which has only two seasons, wet and dry. The presence of mountain ranges in central and northern Vietnam further increases the variety of climatic regions, allowing for a wide range of different plant species to be grown. The subtropical climate in the north, bordering on mountain ranges, allows the growth of tropical and temperate plants in areas close to each other. Various regions in Vietnam also provide an ideal climate for *Phytophthora* species to flourish, and the genus *Phytophthora* is responsible for extensive economic damage in a wide range of different crops throughout the country, including fruit, vegetables, tree plantations and other agricultural crops.

*Phytophthora* pathogens have been reported to cause leaf blights, stem cankers, heart rots, fruit rots and root rots in a wide range of plant species. However, information on the occurrence and distribution of the various *Phytophthora* species present in Vietnam, disease transmission and progression, and suitable control methods is lacking. A strategic approach to the future study and control of phytophthora diseases is needed.

### Distribution of *Phytophthora* in Vietnam

The main information concerning the presence and distribution of phytophthora disease comes from surveys conducted by the National Institute of Plant Protection in Hanoi (NIPP) as part of a national survey of plant diseases. The information from surveys conducted in 1977–1980 has been published in the list of plant diseases in southern Vietnam and the results of a 1997–1998 survey of diseases on fruit crops was published in 1999 (Dang and Ha 1999). The information collected about phytophthora is reproduced in Table 4.4.1.

From these surveys and other field studies, 13 species of *Phytophthora* have been identified in Vietnam. Considering the array of *Phytophthora* species identified in other countries in the region, it is to be expected that many more will be identified in Vietnam. This is especially so, given the current rapid increase in the number of different food, fruit and industrial crops being grown throughout Vietnam. An increase in expertise in plant pathology and diagnostic capability is likely to further increase the number of species identified.

### Tomato and Potato

Late blight of tomato and potato is the major disease of these crops. It has been studied in the Red River Delta area since the 1960s. The causative agent is *Phytophthora infestans* and the disease occurs annually from December to March when climatic conditions are cool and humid. All tomato varieties are susceptible to the disease, and infection generally results in a 30–70% yield loss. In severe

<sup>1</sup> National Institute of Plant Protection, Plant Disease Identification Service Laboratory, Chem, Tu liem, Hanoi, Vietnam.

<sup>2</sup> CRC for Tropical Plant Protection, Indooroopilly Research Centre, Plant Pathology Building, 80 Meiers Road, Indooroopilly, Queensland 4068, Australia.

Table 4.4.1 *Phytophthora* species found in Vietnam.

Species	Host	Common name	Diseases	Year	Reference
<i>P. botryosa</i>	<i>Hevea brasiliensis</i>	rubber	leaf fall black stripe	1961	Dang Vu Thi Thanh and Ha Minh Trung (1999)
<i>P. cactorum</i>	<i>Prunus salicilis</i>	plum	fruit rot	1996	Dang Vu Thi Thanh and Ha Minh Trung (1999)
<i>P. capsici</i>	<i>Piper nigrum</i>	black pepper	foot rot	1998	Nguyen Viet Trong (2002)
<i>P. cinnamomi</i>	<i>Ananas comosus</i>	pineapple	heart rot	2001	Dang Vu Thi Thanh et al. (this paper)
<i>P. citrophthora</i>	Citrus species	citrus	fruit rot stem canker		Roger (1951)
<i>P. colocasiae</i>	<i>Colocasia esculenta</i>	taro	leaf blight	1951	Roger (1951)
<i>P. infestans</i>	<i>Solanum tuberosum</i> <i>Lycopersicon esculentum</i> <i>Solanum melongena</i>	potato tomato eggplant	late blight late blight late blight late blight	1951	Roger (1951)
<i>P. nicotianae</i>	<i>Ananas comosus</i> <i>Nicotiana tabacum</i> Citrus species	pineapple tobacco citrus	heart rot black shank stem canker	2001 1967 2002	Dang Vu Thi Thanh et al. (this paper) NIPP (1975) Dang Vu Thi Thanh et al. (this paper)
<i>P. palmivora</i>	<i>Durio zibethinus</i> <i>Cocos nucifera</i> <i>Theobroma cacao</i> <i>Hevea brasiliensis</i>	durian coconut cocoa rubber	leaf blight stem canker fruit rot leaf blight black tripe leaf fall	2000 2002 2002 1965	Dang Vu Thi Thanh et al. (this paper) Dang Vu Thi Thanh et al. (this paper) Dang Vu Thi Thanh et al. (this paper) Dang Vu Thi Thanh and Ha Minh Trung (1999)
<i>P. durian</i>	<i>Durio zibethinus</i>	durian	canker	2002	Dang Vu Thi Thanh et al. (this paper)
<i>Phytophthora</i> sp.	<i>Hevea brasiliensis</i>	rubber	stem canker	1965	Dang Vu Thi Thanh and Ha Minh Trung (1999)
<i>Phytophthora</i> sp.	<i>Zizyphus mauritania</i>	zizyphus	fruit rot	1997	Dang Vu Thi Thanh and Ha Minh Trung (1999)
<i>Phytophthora</i> sp.	<i>Dimocarpus longan</i>	longan	fruit rot leaf blight	1995	Dang Vu Thi Thanh and Ha Minh Trung (1999)

cases, the crop is totally destroyed (Vu 1973). It was noted that the incidence of disease was higher than average in areas with clay soils. Late blight in potatoes and tomatoes in the Red River Delta is controlled by application of a 1% Bordeaux spray every 7–10 days to prevent infection of the crop. The protectant fungicides, Maneb and Zineb at 0.2–0.3% a.i. have also shown a high efficacy against *P. infestans*.

In recent years crop losses in tomato due to late blight have been reduced in the north of Vietnam through the combined use of fungicide applications hybrid varieties with partial resistance to *P. infestans*. In the provinces of Hanoi, Hatay and Vinhphuc, local farmers use an extreme regime of fungicide application in an attempt to control *P. infestans* in tomato. Fungicides such as Zineb and Ridomil are applied at concentrations 2–3 times above the recommended level, and successive spraying is carried out at short time intervals, in some cases every 3–5 days (Ha Minh et al. 2002). Air and water samples taken from the immediate area were found to be contaminated with fungicides. The residue levels in many of these sprayed crops were above the legal limit, making the tomatoes unsuitable for human consumption.

The tomato industry of the provinces of the Red River Delta, such as Hanoi and Haiphong, has potential for expansion in the near future if demand from an increasingly affluent population and the demands from the food-processing industry are to be met. For this expansion to occur, and to safeguard human health, the environment and the future of the tomato industry, the Vietnamese Government needs to develop and implement a cohesive plan for the control of phytophthora diseases in the Delta. Part of this plan should include the education of local farmers in the correct dosage and application of fungicides, and in other disease management tools, in an effort to control late blight.

## Taro

Leaf blight caused by *Phytophthora colocasiae* is the major disease of taro in northern Vietnam. The disease was first recorded by Roger (1951). Warm temperatures (24–30°C) and high humidities are required for disease spread, conditions that are found throughout that part of the country. The disease occurs annually, starting between April and May and reaching a peak in July and August when temperatures are a steady 27–29°C and the average monthly rainfall is in the range 201–308 mm. Disease surveys have found leaf blight of taro in all ecological zones of northern Vietnam, with an

average disease incidence of between 21 and 66% (Table 4.4.2).

*Phytophthora colocasiae* attacks both species of taro grown in Vietnam, *Colocasia esculenta* var. *antiquorum* and *C. esculenta* var. *esculenta*. The level of genetic diversity in *P. colocasiae* was studied using isozymes. Two genotypes were identified from five isolates. Additional RAPD analysis revealed differences between the genotypes found in Vietnam and strains found in other ASEAN countries (Nguyen Van Viet et al. 2002).

**Table 4.4.2** The effect of *Phytophthora colocasiae* on taro in northern Vietnam, 2000–2001.

Location	Region	Disease incidence (%)	
		July 2000	July 2001
Kyson	Hoabinh	30	30
Vinhtuong	Vinhphuc	52	53
Hoaiduc	Hatay	30	31
Tuliem	Hanoi	35	31
Dongtrieu	Quangninh	47	66
Dongson	Thanhhoa	20	24

Source: Nguyen et al. (2002)

## Pineapple

Pineapple has become an increasingly important crop in Vietnam. In 2001, pineapples covered 32,000 ha of agricultural land, with government targets for the year 2010 set at 50,000 ha. Heart-rot disease is one of the major causes of losses in Vietnam's pineapple crop. The disease has been found in all pineapple-growing areas in the northern and central regions of the country, including Thuathien-Hue, Nghean, Hatay, Bacgiang, Thanhhoa and Ninhbinh. The pineapple variety Cayenne appears to be more susceptible to heart rot than other varieties. The disease incidence 2 months after cultivation in plantations in the Quang Nam region was 35%. After 3 months, it had risen to 60% in some plantations. Interestingly, in regions with very low soil pH levels (3.5–4.2) such as Tiengiang province and Ho Chi Minh City, heart rot disease has not been found (Table 4.4.3). However, it is unclear whether this is due to low soil pH levels or other factors.

A survey in the Donggiao Ninhbinh region revealed that 40% of the samples taken yielded *Phytophthora* after incubating leaf material on potato sucrose agar (PSA). Further identification of the strains obtained revealed the presence of both *P. cinnamomi* and *P. nicotianae* (Table 4.4.4). Samples taken from Hatrung–Thanhhoa in June 2002 were also infected with *P. cinnamomi*. To confirm that *P. nicotianae* and *P. cinnamomi* are responsible for heart-rot disease in

pineapple, glass house trials were conducted at NIPP in 2001. Fifteen days after inoculation with both *Phytophthora* species, 100% of plants displayed symptoms of heart-rot disease (Ngo et al. 2001). Application of 4% Phosacide 200 (a phosphonate source) and 0.25% Aliette 80WP (Rhône Poulenc) reduced the incidence of disease by around 96%.

In field trials at Donggiao Ninhbinh, seedlings dipped in fungicide solution before planting showed a reduced level of infection 45 days after treatment (Table 4.4.5).

Phytophthora heart rot causes significant problems in pineapple cultivation in Vietnam. Continued research into disease development in the field, and integrated disease management through cultivar selection, drainage, cultivation, fungicide application and alternative methods, is needed to establish effective integrated disease management strategies for pineapple cultivation that will allow for the continued and successful expansion of the industry.

## Citrus

*Phytophthora citrophthora* was first recorded on oranges in the Mekong Delta in the 1950s, and was not observed again until the 1970s, when it was found on orange in northern and central Vietnam.

The pathogen has since spread significantly and it now affects fruit in all citrus-growing areas, such as the Thanh tra area of Thua Thien Hue, and Ninhbinh in Tien Giang.

*P. citrophthora* attacks the stem and fruit, resulting in gummosis and fruit rot symptoms. The disease develops quickly in the rainy season, and is most severe in July and August. In March 2002, disease incidence in orange in Caophong–Hoabinh was 10% but had risen to 20–30% by August. Mandarin was more severely affected, with some orchards suffering total crop loss and the death of many plants. Samples taken from plants suffering citrus stem canker in the Tien Giang province were identified as *P. nicotianae* (A. Drenth, unpublished data).

Phytophthora diseases on citrus have been studied only sporadically in Vietnam, and have often been limited to surveys of disease incidence and severity. There has been no research into the development of control strategies for the disease, nursery management, the breeding of resistant varieties and the use of resistant rootstock. New citrus plantations have been established in Hagiang, Tuyenquang and Vinh Long, and the area devoted to citrus continues to increase. Research into phytophthora diseases and their control is required now to safeguard the future of the industry.

**Table 4.4.3** Influence of soil pH on incidence of heart-rot disease, 2000–2001.

Location	Region	Soil pH	Total number of plants surveyed	Disease incidence
Donggiao	Ninhbinh	5.7–7.9	960	211 (21.9%)
Le minh Xuan State farm	Ho Chi Minh City	3.5–4.1	880	0
Tanlap State farm	Tien Giang	3.5–4.2	750	0

**Table 4.4.4** Identification<sup>a</sup> of *Phytophthora* species causing heart-rot disease in pineapple.

Location	Year	Number of samples	<i>P. nicotianae</i> infected samples	<i>P. cinnamomi</i> infected samples
Donggiao	2001	86	28 (32.5%)	6 (7%)
Hatrung	2002	2	–	2 (100%)

<sup>a</sup> Identifications by Dr André Drenth, University of Queensland, Australia, August 2001 and June 2002.)

**Table 4.4.5** Effect of fungicide on phytophthora heart rot in pineapple in Donggiao, Vietnam, August 2001.

Treatment	Total number of plants tested	Disease incidence
0.25% Aliette 80WP	240	11 (4.6%)
4% Phosacide 200	240	12 (5.6%)
Control	240	47 (19.6%)

## Durian

Durian (*Durio zibethinus* Murr) is one of most favoured fruit crops in southern Vietnam. In recent years, the durian-growing area has rapidly expanded north to the southern and central highlands, displacing rice and other crops due to higher profitability that can be obtained from cultivating durian.

*P. palmivora* causes a wide range of diseases in durian, including root rot, stem canker, fruit rot and leaf blight. It has been found in all durian growing areas of the southern and central highlands. In 2001, the disease also affected durian growing in the lowlands, and was particularly severe in Quang Nam province. Of the 3075 plants growing in Que Trung commune, 2138 were killed by *P. palmivora* at an economic loss of 15 billion VND (USD1.5 million). Elsewhere in the country, the disease was found to be most prevalent in Cai Be, Tien Giang, with 24.6% of plants infected. Disease incidence was related to plant age, with plants more than 10 years old being most susceptible (Table 4.4.6).

An ACIAR-funded study, 'Management of *Phytophthora* disease in durians', was conducted by the University of Melbourne, Australia, Kasetsart University, Thailand, and the Southern Fruit Research Institute of Vietnam. The study identified several orchard-management practices, such as phosphonate trunk injections and improved nursery hygiene, which can be combined into an integrated disease management package specifically tailored to meet the needs of each region. Full details of this study and its outcomes can be found in chapter 8 of this monograph and will therefore not be discussed here.

## Plum

In recent years, black spot disease of plum (*Prunus salicilas*) has seriously reduced crop yields in Bac Ha and Moc Chau provinces. *Phytophthora cactorum* was identified as the causal agent. In Bac Ha in March 1996, the disease affected 300 ha of young plum fruit causing serious damage and a 20% yield loss. During 1997 and 1998, the disease was less widespread but the damage caused was more severe, with some gardens showing black spot disease in up to 50% of their crop.

Disease symptoms on plum are typically white--grey water-soaked spots on young fruit, developing into sunken black spots with brown edges as the disease progresses. In cases of severe infection, the whole fruit will shrivel and fall from the tree. The sunken spots may become covered with white mycelium in damp conditions, and *P. cactorum* conidia have been isolated from the spots in these conditions. Sporangia can be isolated and grown into culture on carrot, kidney bean or potato dextrose agar, but sporulation has not been observed in vitro.

In March 1998, the infectivity of *P. cactorum* on plum gardens was studied at Bac Ha. Initial disease symptoms were observed on all treated fruit 3–5 days after inoculation, but in many cases did not develop further and the final incidence of disease was low, probably due to the relatively high temperature (18–25°C) in the field during that time. The results of the 1998 study are summarised in Table 4.4.7.

Late February in northern Vietnam is typically cool and damp, with daytime temperatures of 12–14°C, nights that are around 10°C cooler than the days,

**Table 4.4.6** Phytophthora disease incidence in durian growing areas

Location	Region	Number of plants surveyed	Disease incidence (%)			
			Total	< 5 years	6–10 years	>10 years
Que Son	Quang Nam	370	22.0	5.1	7.5	9.4
Long khanh	Dong nai	280	21.1	5.0	6.1	10.0
Cai Be	Tien Giang	182	24.6	3.8	10.4	10.4

**Table 4.4.7** Results of inoculation of plum fruit with pure culture of *Phytophthora cactorum* at Bac Ha, March 1998.

Treatment	Number of fruit	Infected fruit	Disease incidence (%)	Time to appearance of symptoms (days)
Control - distilled water	56	0	0	-
<i>P. cactorum</i>	191	7	13.4	3–5

and frequent fog. These conditions are ideal for *P. cactorum* infection. Development of black spot disease on young plums is therefore swift in March and early April, slowing as the temperature rises towards the end of the month. It has been observed that young trees are more susceptible to black spot disease than more mature trees. In March 1998, the disease incidence on 2-year-old plum trees was 10%, while 4-year-old trees suffered only a 2.1% disease incidence.

In 1999, plums on the hills of Bac Ha suffered a widespread outbreak of black spot disease. This allowed for a study of disease distribution in relation to geographical factors. The incidence of disease was found to vary widely according to location on the hill. On 5 March 1999 the disease incidence at the summit of the hill was 0.7%, at the middle of the hill it was 3.1% and in the foothills it was 3.9%. By 20 March the pattern of disease incidence had changed dramatically; at the top of the hill it was 43%, in the middle 81% and in the foothills 26.3% (Table 4.4.8). Very similar results were obtained from a similar survey conducted in the previous year.

The reason for the high disease incidence and severity halfway up the hill is most likely microclimatic factors that lead to differences in humidity and temperature between the different sites.

## Rubber

Rubber (*Hevea brasiliensis*) is a highly valuable industrial commodity that is grown all over Vietnam. Rubber plants were imported to Vietnam in 1897 to establish plantations, with 288,000 ha devoted to its growth in 1996 and a government target of 700,000 ha by the year 2005. Most of the large rubber plantations are located in the southern and central highlands of Vietnam.

The 1960s saw the start of studies concerning diseases in rubber in Vietnam. Of the 19 diseases that affect rubber in Vietnam, leaf fall, black stripe and stem canker are caused by *Phytophthora* species. *P. palmivora* has been isolated from around 72% of

rubber plants affected with black-stripe disease, while *P. botryosa* can be found in 75–80% of leaves and fruit suffering from leaf-drop disease. Both *P. palmivora* and *P. botryosa* are known to infect trees in all parts of the country.

In the southern highlands during the wet season, disease incidence of leaf-drop and black-stripe diseases has been as high as 45% and 34%, respectively. Leaf-fall disease is generally more severe in Dong Nai and Binh Long than in Dau Tieng and Tay Ninh. The two diseases combined can reduce rubber production by 23 to 36.8% annually. Of the rubber varieties grown (PR107, PB86, RRIM600, PB310, PR255 and PB244) most are susceptible to black-stripe disease, with only one, PR107, being resistant. The application of Ridomil 72 WP and Difolatan has proven to be effective in controlling the disease. Bark-rot disease, also caused by *Phytophthora* species, is found only in the northern regions of the country.

## Conclusions

*Phytophthora* diseases are responsible for some of Vietnam's major crop losses in tomato, potato, citrus, pineapple, plum, black pepper, rubber, and durian. Identification of *Phytophthora* is currently based on disease symptoms and morphological characteristics. An increased capability is needed to accurately distinguish *phytophthora* diseases from other soil-borne diseases and to be able to identify isolates down to species level.

Very little research on *Phytophthora* and *phytophthora* diseases has been carried out in Vietnam. Progress in the following areas is urgently needed:

- obtaining expertise in and information on disease symptoms and methods to isolate potential *Phytophthora* pathogens
- gaining expertise in the identification of *Phytophthora* pathogens down to species level, including the use of molecular methods for identifying *Phytophthora* species

**Table 4.4.8** Development of black spot disease on plum fruit at Bac Ha in March 1999.

Location	5 March		10 March		15 March		20 March	
	DI <sup>a</sup> (%)	DS <sup>b</sup> (%)	DI (%)	DS (%)	DI (%)	DS (%)	DI (%)	DS (%)
Foothills	3.9	1.3	19.5	7.1	11.8	9.3	26.3	16.3
Middle of hill	3.1	1.5	43.6	17.7	49.7	18.8	81.2	46.0
Top of hill	0.7	0.2	25.0	8.0	32.0	8.4	42.6	14.2

<sup>a</sup> Disease incidence. <sup>b</sup> Disease severity.

- selection and resistance-screening methods that allow the development of crops resistant to infection by *Phytophthora* species
- information on host spectrum, disease development and control methods of the various *Phytophthora* species
- the development of an integrated pest management approach for the control of phytophthora diseases. This should include the establishment of pilot schemes, and the education and training of farmers and growers, especially about environmental protection.

Collaborative work and exchange of information and knowledge between the scientists of Vietnam, other ASEAN countries and Australia is required if the successful control of *Phytophthora* is to be achieved.

## Acknowledgments

We thank ACIAR, the Crawford Fund, and Dr Fiona Benyon for assistance in this research.

## References

Dang Vu Thi Thanh and Ha Minh Trung 1999. List of diseases on fruit crops. In: Survey results of insect pests and diseases on fruit trees, 1997–1998. Hanoi, Agricultural Publishing House, 158p. (in Vietnamese)

Ha Minh, Trung, Duy Trang Nguyen and huu Vinh va CTG. Nguyen 2002. Studying influence of agricultural chemicals on health of persons and overcoming methods (Nghien cuu anh huong cua cac hoa chat doc hai dung trong Nong nghiep toi suc khoe con nguoi – Cac giai phap khac phuc). Tuyen tap cong trinh nghien cuu BVTV 1996–2000, tr. 161–170.

NIPP (National Institute for Plant Protection) 1975. Survey results of plant diseases 1967–1968. Hanoi, Agricultural Publishing House, 207p.

Ngo, Vinh Vien, Bui Van Tuan, Le Thu Hien, Dang Luu Hoa, Benyon, F. and Drenth, A. 2001. Phytophthora fungi causing heart rot on pineapple (Nam Phytophthora gay benh thoi non dua). In: Bao cao hang nam Vien BVTV 2001.

Nguyen, Van Viet et al. 2002. Importance of taro leaf blight and study on genetic diversity of *Phytophthora colocasiae* Raciborski of taro in northern Vietnam. Paper presented at 1st National Conference on Plant Pathology and Molecular Biology, Agricultural–Forestry University of Ho Chi Minh City.

Nguyen, Viet Truong 2002. Initial diagnosis and identification on the foot rot disease of black pepper in Vietnam. Paper presented at 1st National Conference on Plant Pathology and Molecular Biology, Agricultural–Forestry University of Ho Chi Minh City.

Roger, L. 1951. Genre *Phytophthora*. Phytopathologie des pays chauds. Paris, Paul Lechevalier, 1, 627–698.

Vu, Hoan 1973. Studying *Phytophthora infestans* fungi causing late blight on tomato (Nghien cuu hinh thai va sinh thai nam *Phytophthora infestans* gay benh moc suong ca chua). KHKT Nong Nghiep, so 3.



## 4.5 Phytophthora Diseases in the Philippines

L.A. Portales<sup>1</sup>

### Abstract

An overview is provided of the phytophthora diseases reported and the species of *Phytophthora* identified in the Philippines. *Phytophthora palmivora* and *P. nicotianae* are the most common species and have caused considerable disease losses in a range of crops of significant economic importance to the Philippines.

### Introduction

Stretching 1839 kilometres north-to-south, the Republic of the Philippines has a total land area of 300,000 km<sup>2</sup> spread over 7107 islands. The Philippines is a tropical country with an average temperature of 32°C (80°F). The months of March to June are hot and dry (36°C), rains and typhoons abound from July to October, while November to February are pleasantly cool (around 23°C) and dry. In mountainous regions, temperatures can dip to about 15°C. The long wet and cool seasons are conducive to the infection of the country's agricultural crops by phytophthora diseases.

### Overview of *Phytophthora* Problems in the Philippines

The first crop reported to be infected with *Phytophthora* was coconut in 1908, but comprehensive studies on the disease did not begin until 1919 (Table 4.5.1). Between 1919 and 1933, seven important papers dealing with phytophthora on coconut, citrus, cocoa, eggplant, santol and cinchona were published (Table 4.5.1) The papers reported detailed information on the morphology and pathogenicity of *Phytophthora* species, and the mode of disease propagation, disease symptoms and control measures. From 1934–1971, only three papers reporting phytophthora diseases were published (potato late blight, pineapple heart rot

and eggplant fruit rot), but since 1972 many government research agencies and academic institutions have become involved in phytophthora research.

### Economic Importance

Although many species of *Phytophthora* have been detected and are known to cause serious crop losses in the Philippines, published data on the impact of these pathogens is not available for most crops. Disease losses for only four crops have been published, and these losses occurred in isolated areas.

Although the disease is usually of minor importance to eggplant, under favourable conditions and in dense planting, *Phytophthora* may cause serious infection and yield loss. Eggplant in the garden of the College of Agriculture in Los Baños and environs was found to be infected with *Phytophthora*. Disease losses on pineapple were reported in three municipalities of Laguna, namely Los Baños, Calauan and Alaminos. Fifty per cent of the pineapples in a 2 ha field at Alaminos were infected. The outbreak of a serious seedling blight of cinchona was first reported in 1932, from a nursery of the College of Agriculture in Los Baños, Laguna, where 45% of the plants were infected. The 1924 infection of santol (*Sandoriam koetjape*) resulted in the death of 90% of infected seedlings, the disease being manifest as blight on the different parts of the young seedlings, causing eventual collapse and decay. Table 4.5.2 presents these crops with the reported level of infection.

<sup>1</sup> Department of Agriculture, Division of Plant Protection, Bureau of Plant Industry, 629 San Andreas Street, Malate, Manila 2801, Philippines.

Table 4.5.1 *Phytophthora* diseases of various crops in the Philippines

<i>Phytophthora</i> species <sup>a</sup>	Host	Common name	Disease	Year first reported	Reference	
<i>P. cactorum</i>	<i>Theobroma cacao</i>	Cocoa	Oriental cocoa pod disease	1916	Mendiola and Espino (1916)	
<i>P. capsici</i>	<i>Piper nigrum</i>	Black pepper	Crown/root rot	1994	Tsao et al. (1994)	
<i>P. citrophthora</i>	<i>Citrus</i> spp.	Citrus	Foot rot	1968	Rosario (1968)	
			Gummosis	1968	Rosario (1968)	
			Brown rot	1974	Quimio and Quimio (1974)	
			Crown/root rot	1994	Tsao et al. (1994)	
			Crown/root rot	1994	Tsao et al. (1994)	
<i>P. colocasiae</i>	<i>Colocasia esculenta</i>	Gabi	Leaf blight	1916	Mendiola and Espino (1916)	
<i>P. heveae</i>	<i>Sandorium koetjape</i>	Santol	Crown/root rot	1994	Tsao et al. (1994)	
<i>P. infestans</i>	<i>Solanum tuberosum</i>	Potato	Late blight	1921	Lee (1921)	
<i>P. meadii</i>	<i>Hevea brasiliensis</i>	Rubber	Black thread	1926	Teodoro (1926)	
<i>P. nicotianae</i>	<i>Piper nigrum</i>	Black pepper	Crown/root rot	1994	Tsao et al. (1994)	
		Citrus	Foot rot	1921	Lee (1921)	
	<i>Solanum melongena</i> <sup>c</sup>		Gummosis	1968	Rosario (1968)	
			Fruit rot	1925	Ocfemia (1925)	
			Phytophthora blight, fruit rot	1974	Quimio and Quimio (1974)	
			Heart rot / pineapple wilt	1962	Quebral et al. (1962)	
			Watermelon	Phytophthora rot	1974	Quimio and Quimio (1974)
	<i>P. palmivora</i>	<i>Persea americana</i>	Avocado	Crown/root rot	1994	Tsao et al. (1994)
		<i>Theobroma cacao</i>	Cocoa	Black rot/stem canker	1979	PCARR (1979)
<i>Artocarpus camansi</i>		Camansi	Crown/ root rot	1994	Tsao et al. (1994)	
<i>Cinchona calisaya</i>		Cinchona	Seedling blight	1933	Celino (1933)	
<i>Citrus</i> spp.		Citrus	Phytophthora blight	1968	Rosario (1968)	
<i>Cocos nucifera</i> <sup>a</sup>		Coconut	Bud rot	1919	Reinking (1919)	
<i>Durio zibethinus</i>		Durian	Crown/root rot	1994	Tsao et al. (1994)	
<i>Artocarpus heterophyllus</i>		Jackfruit	Crown/root rot	1994	Tsao et al. (1994)	
<i>Lansium domesticum</i>		Lanzones	Crown/root rot	1994	Tsao et al. (1994)	
<i>Euphoria longana</i>		Longan	Crown/root rot	1994	Tsao et al. (1994)	
<i>Garcinia mangostana</i>		Mangosteen	Crown/root rot	1994	Tsao et al. (1994)	
<i>Azadirachta indica</i>		Neem	Crown/root rot	1994	Tsao et al. (1994)	
<i>Phalaenopsis</i> spp.		Orchid	Black rot	1968	Ela (1968)	
<i>Carica papaya</i> <sup>a</sup>		Papaya	Fruit rot	1918	Reinking (1919)	
<i>Hevea brasiliensis</i> <sup>a</sup>		Rubber	Black rot of fruit	1918	Reinking (1919)	
<i>Hevea brasiliensis</i> <sup>a</sup>		Rubber	Canker	1926	Teodoro (1926)	
<i>Annona muricata</i>		Soursop	Crown/root rot	1994	Tsao et al. (1994)	
<i>Tamarindus indica</i>		Tamarind	Crown/root rot	1994	Tsao et al. (1994)	
<i>P. phaseoli</i>		<i>Sandorium koetjape</i>	Santol	Leaf blight	1928	Clara (1928)

<sup>a</sup> The use of these names has been discontinued in the literature: *P. faberi* is the synonym of *P. palmivora*, *P. ommitoora* is the synonym of *P. cactorum*; both *P. melongenae* and *P. terrestris* are synonyms of *P. nicotianae* (= *P. parvisitica*).

## Management of Phytophthora Disease in the Philippines

Early studies on phytophthora management and control recommended a mix of cultural management, chemical control and quarantine policy. Control measures included the draining of excess water, cultivation of soil for increased aeration, increasing the space between neighbouring trees, and the pruning of excess branches to improve ventilation and to allow sunlight to enter. Maintaining clean culture by the removal and burning of infected plants and plant parts was also recommended, together with the use of resistant varieties and spraying with Bordeaux mixture.

**Table 4.5.2** Reported crops with known *Phytophthora* infection.

Crop	<i>Phytophthora</i> species	Disease	Level of infection (%)
Eggplant	<i>P. nicotianae</i>	Fruit rot	25–75
Pineapple	<i>P. nicotianae</i>	Heart rot	20–50
Cinchona	<i>P. palmivora</i>	Seedling blight	45
Santol	<i>P. phaseoli</i>	Leaf blight	90

### Nursery Management<sup>2</sup>

A survey of the prevalence of phytophthora diseases in the Philippines by the Bureau of Plant Industry resulted in the following recommendations for the management and control of phytophthora in nursery operations.

1. Sterilised soil, sand or other planting media should be used for the germination of seeds and cuttings. Leftover soil, pots and plastic containers should also be sterilised if they are to be re-used.
2. Porous materials with good aeration and drainage properties, such as sand, sawdust, or composted tree bark, should be used instead of pure soil, or in addition to pure soil, whenever possible. Adequate nutrients should be provided by the use of organic and inorganic fertilisers.
3. Clean, sterilised soil should be stored in closed containers such as soil bins, to prevent contamination. Diseased materials, foot traffic, animals, and run-off water may contaminate soil left on bare ground or in uncovered containers.
4. Only clean seeds extracted from healthy fruits should be used. Never use seeds taken from fruit

already on the ground. Avoid using infected fruit or fruits showing lesions or other signs of disease.

5. Use only clean tools and with clean hands. Tools such as shovels, trowels, shears and knives should be washed, dried and sterilised after each use, with either 70% ethanol or 10% bleach.
6. Hoses should not be left on the ground after use.
7. Water sources should be protected from contamination by soil or diseased material. Hands or tools should never be washed in water stored for watering.
8. Seed boxes and potted plants should be kept on raised benches above the ground. Where this is not possible, cover the ground with 5–8 cm of gravel to avoid contamination from standing or splashing water. Low areas in the nursery, where the risk of contamination from standing water is high, should not be used.
9. Infected or diseased plants and plant material should be removed from the propagation area and disposed of appropriately.

### Training and Extension

During 1989 and again in 1990–91 the Philippine–German Biological Plant Project and the Deutsche Gesellschaft für Technische Zusammenarbeit (PGBPPP/GTZ) funded visits by Dr Peter H. Tsao, from the University of California in Riverside (UCR) to the Bureau of Plant Industry (BPI) for 1 and 6 months, respectively, to train BPI plant pathologists on aspects of *Phytophthora* and phytophthora diseases.

There were 16 participants in a training course on ‘Detection, isolation and identification of *Phytophthora* diseases in the Philippines’ run by Dr Tsao during January 1991 at the Crop Protection Division of BPI–Manila. They included representatives from eight of the twelve Regional Crop Protection Centers, and six people from the BPI Research Centers, including one from the National Crop Protection Center at Los Baños and one from the Department of Plant Pathology of the University of the Philippines at Los Baños. During 1992, and again in 1993–95, a BPI staff member joined the research team at UCR for further training on the biology and control of phytophthora, and in isolation and identification techniques. Numerous other training courses have been conducted by overseas experts over the past decade. These were typically tied to specific research projects including the outbreak of budrot following the introduction of hybrid coconut (see chapters 6.2 and 6.3).

<sup>2</sup> See also Chapter 7.1.

## Technology Information and Dissemination

As a result of the BPI survey into the prevalence of phytophthora diseases in the Philippines, the booklet 'How to produce healthy plants' was funded and published by BPI-PGBPPP/GTZ (Tsao 1993). The booklet highlights the importance of clean soil, seed and stock, and hygienic nursery practices, in order to run a successful nursery operation.

An easy-to-understand leaflet entitled '*Phytophthora* disease diagnosis' was produced by the Crop Protection Division of BPI. It aimed at increasing growers' awareness of the disease, and contains basic information on disease diagnosis, from isolating and purifying *Phytophthora* from crop samples, through to identification and pathogenicity testing procedures. Similar material has been produced as part of an FAO-funded project on coconut bud rot.

## Conclusion

Phytophthora diseases have been detected in the Philippines since the early 1900s. The country's climate and environment mean these diseases have been detected over a large range of crops and geographic locations. All BPI field surveys have recovered *Phytophthora* samples using isolation techniques such as selective agar media and baiting procedures. However, comprehensive information on the impact of phytophthora disease on plant production in the Philippines is lacking. Thus, a concerted effort by research agencies and academic institutions into suitable management and control strategies for the disease is needed, so as to minimise and manage crop losses.

## References

Celino, M.S. 1933. Blight of cinchona seedlings. *Philippine Agriculture*, 23, 111–123.

Clara, F.M. 1928. A *Phytophthora* disease of santol seedlings. *Philippine Journal of Science*, 35, 411–425.

Ela, V.M. 1968. Notes on diseases of orchids in the Philippines. *Philippine Agriculture*, 4, 531–537.

Lee, A. H. 1921. Observations on previously unreported or noteworthy plant diseases in the Philippines. *Philippine Agricultural Review*, 14, 422–434.

Mendiola, N., and Espino, R.B. 1916. Some phycomycetous diseases of cultivated plants in the Philippines. *Philippine Agriculturalist and Forester*, 5, 65–72.

Ocfemia, G. O. 1925. The *Phytophthora* disease of eggplant in the Philippine Islands. *Philippine Agriculture*, 14, 317–328.

PCARR (Philippine Council for Agriculture and Resources Research) 1979. The Philippines recommends for cacao. Los Baños, Laguna, PCARR.

Quebral, F.C., Pordesimo, A.N., Reyes, T.T. and Tamayo, B.P. 1962. Heart rot of pineapple in the Philippines. *Philippine Agriculture*, 46, 432–450.

Quimio, T.H. and Quimio, A.J. 1974. Compendium of postharvest and common diseases of fruits in the Philippines. UPCA Technical Bulletin 34.

Reinking, O. A. 1919. *Phytophthora faberi* Maubl: the cause of coconut bud rot in the Philippines. *Philippine Journal of Science*, 14, 131–150.

Rosario, M.S. del. 1968. A handbook of citrus diseases in the Philippines. UPCA Technical Bulletin 31.

Teodoro, N.G. 1926. Rubber tree diseases and their control. *Philippine Agricultural Review*, 19, 63–73.

Tsao, P.H. 1993. How to produce healthy plants. Philippine–German Biological Plant Protection Project, Bureau of Plant Industry.

Tsao, P.H., Gruber, L.C., Portales, L.A., Gochangco, A.M., Luzaran, P.B., De los Santos, A.B. and Pag, H. 1994. Some new records of *Phytophthora* crown and root rots in the Philippines and in world literature. *Phytopathology*, 84, 871. (Abstract)

# 5 Isolation of *Phytophthora* from Infected Plant Tissue and Soil, and Principles of Species Identification

André Drenth and Barbara Sendall<sup>1</sup>

## Abstract

In order to assign the cause of a disease or disorder to a particular pathogenic organism it is important that the causative agent be identified, and that additional pathogenicity tests are conducted to show beyond reasonable doubt that the organism in question can indeed cause the disease. Although the isolation of *Phytophthora* pathogens is not difficult it is different to the isolation and identification of many true fungi. We give an overview of media, antibiotics and methods available that may be used for isolation and identification of *Phytophthora* species in the tropics.

## Introduction

It is estimated that *Phytophthora* species cause 90% of the crown rots of woody plants. However, lack of knowledge on how to isolate *Phytophthora* often leads to negative results and hence other pathogens such as *Fusarium*, *Pythium*, *Rhizoctonia* and nematodes are frequently blamed for root and crown rots (Tsao 1990). Unlike species of *Pythium* and *Fusarium*, which are generally associated as saprophytes or opportunists with plants and soil, *Phytophthora* species associated with diseased plants are likely to be the causal agent of the disease. This is because most *Phytophthora* species attack only living or freshly wounded tissue. They are primary invaders and hence do not colonise plant tissue already invaded by other microorganisms. Detection and/or isolation of *Phytophthora* from plant tissue is relatively simple and successful if the tissue is fresh and recently infected. Isolation of *Phytophthora* from necrotic plant tissue is more difficult, because most species of *Phytophthora* have poor saprophytic capabilities, and there may be very little mycelia remaining once the host tissue dies and secondary invaders move in. In addition,

dormant propagules such as chlamydospores and oospores are slow to germinate and emerge from senescent plant tissue. Isolation of *Phytophthora* directly from soil is difficult, but the use of baiting techniques markedly increases the frequency of successful isolation of *Phytophthora* from infested soils.

## Isolation Media

The Oomycetes are not true fungi (see Chapter 3.1), and therefore special techniques are required for their isolation. Most species of *Phytophthora* grow rather slowly in vitro compared with saprophytic fungi and bacteria. In addition, bacterial populations need to be kept low because they may suppress the growth of *Phytophthora* by direct competition, by antagonism caused by antibiotic production, or by direct parasitism. The use of selective media usually overcomes these problems. Antibiotics are added to isolation media in order to suppress the growth of bacteria. Also, because *Phytophthora* spp. are out-competed by many fungi, it is desirable to choose media that are nutritionally 'weak'. This reduces the growth rate of fungal contaminants, allowing colonies of *Phytophthora* to become established.

Cornmeal agar (CMA) is the most frequently used basic medium for isolation of *Phytophthora* from

<sup>1</sup> CRC for Tropical Plant Protection, Indooroopilly Research Centre, 80 Meiers Road, Indooroopilly, Queensland 4068, Australia.

infected plant tissue. However, other desirable basal media include water agar, and 2% and 4% (v/v) V8 juice agar. Alternatives to these media made with locally available ingredients are cocoa pulp, taro, coconut milk, and carrot mixed with agar-agar.

### Selective Media for Isolation from Diseased Tissue

Various media containing different antibiotics and antifungal components can be used to isolate *Phytophthora*. Corn meal agar (CMA) at 1.7% is the most common medium used as a basis. 3-P (Eckert and Tsao 1960; Eckert 1962) (Table 5.1) is suitable for the isolation of *Phytophthora* from freshly diseased tissue but not from old, decayed tissue or freshly infested soil in which the propagules are likely to be spores. This is because high levels of pimaricin can inhibit spore germination. A suitable medium for isolating *Phytophthora* from old plant tissue or soil is 3-P medium + 10 mg/mL pimaricin (Table 5.1). Plates of selective media used for isolations should not contain any free water or condensation on the lids, as water encourages the growth and spread of bacterial contaminants. Ideally, selective media containing antibiotics should be made fresh before use. Otherwise, they should be used within 2–4 weeks of preparation.

Hymexazol-25 and Hymexazol-50 (Masago et al. 1977) contain the fungicide Hymexazol (Tachigaren). This fungicide has been found to suppress most *Pythium* spp. except for *P. irregulare* and *P. vexans*. It can also inhibit some *Phytophthora* spp., including *P. cinnamomi*, *P. citrophthora* and *P. palmivora*. P10VP (Tsao and Ocana 1969) is suitable for isolating *Phytophthora* from soil and infected plant tissue. Hymexazol can also be added to a final concentration of 25–50 mg/mL. P10ARP

(Kannwischer and Mitchell 1978) and P5ARP (Papavizas et al. 1981; Jeffers and Martin 1986) are the media of choice for isolating most species of *Phytophthora* (Table 5.1).

Since the availability of media and antibiotics varies between locations a series of common antibiotics and antifungal and alternative compounds which may be used to produce media suitable for the isolation of *Phytophthora* are given in Table 5.2. In some cases, when samples are relatively clean and secondary invaders are still absent, one can also isolate directly onto media without the use of antibiotics. Infected fruit can be processed in this manner as the *Phytophthora* typically grow quite deeply in the tissue which allows one to cut away the outer part and directly place fruit tissue containing *Phytophthora* mycelium onto agar with a very high success rate.

### Isolation of *Phytophthora* from Infected Plant Material

*Phytophthora* species attack only healthy plant material, including roots. Thus, the pathogen can be present when no symptoms are obvious.

*Phytophthora* species are difficult to isolate from necrotic tissue because the tissue often harbours many secondary pathogens. Successful isolation of *Phytophthora* species from diseased tissue involves careful selection of freshly infected tissue. Therefore, it is best to obtain material from the edge of an actively growing lesion. Leaf and stem tissue selected for isolation should ideally contain part diseased and part healthy tissue. Once the tissue has been surface-sterilised, it should be transferred to the appropriate selective medium, and the plates examined regularly for the slow emergence of non-septate hyphae.

**Table 5.1** Amount of antibiotic/fungicide required for various *Phytophthora* selective media

Antibiotic/ fungicide	Stock (mg/mL)	Final antibiotic/fungicide concentration (µg/mL)							
		3-P	3-P + 10 µg/mL pimaricin	P <sub>10</sub> VP	P <sub>10</sub> ARP	P <sub>5</sub> ARP	Hymexazol 25	Hymexazol 50	
Ampicillin	100					250	250	500	500
Benomyl	Powder							10	5
Hymexazol	50							1	1
Nystatin	100							25	25
PCNB	Powder			100	100	100		25	25
Penicillin	50	50	50						
Pimaricin	25	100	10	10	10	5			
Polymixin B	50	50	50						
Rifampicin	10				10	10		10	10
Vancomycin	100			200					

*Pythium* spp. are almost invariably present on both healthy and diseased roots, crowns and lower stems of plants. There are three ways in which contamination of isolation media by *Pythium* can be minimised:

1. *Pythium* is confined to roots or badly rotted lower stems — choose other parts if possible.
2. *Pythium* is confined to the outer cortex of the root — surface sterilisation will usually kill it; alternatively choose the centre of the root.
3. Hymexazol will inhibit most species, except for *P. irregulare* and *P. vexans*. Care must be taken, however, as it can also inhibit some *Phytophthora* spp., including *P. cinnamomi*, *P. citrophthora* and *P. palmivora*. When these species are suspected, it is wise to use selective media with and without hymexazol.

### Preparation and Surface Sterilisation of Tissue

It is important to use aseptic techniques, including flame sterilisation and wiping areas with 70% ethanol, when attempting to isolate *Phytophthora* from infected plant tissue. Place well-washed roots, stems or leaves suspected to be infected with *Phytophthora* into a shallow layer of distilled water. Leave for 24–48 hours in the light, at 18–25°C and examine for sporangial development. If sporangia are found, a small infected plant piece can be cut off, surface sterilised and transferred to selective media.

Infected fruit is easily treated by cutting off the outer parts and placing small pieces of the freshly cut fruit onto selective media. Leaf tissue which is reasonably clean may be placed immediately onto selective media but it is almost always better to surface sterilise it first. Surface sterilise leaf and stem tissue by dipping in 70% ethanol for 30–60 seconds. Blot tissue dry between sterile filter paper before placing on selective media. If wet plant material is placed onto media, bacteria can grow rapidly and suppress the growth of *Phytophthora*. If the stems are particularly thick (0.5–1 cm wide), they can be dipped in 70% ethanol for 10–30 seconds, and then quickly flamed to burn off the excess ethanol. Small sections can then be taken either side of the lesion, and embedded directly into selective media.

Diseased roots often need more preparation. Place the roots in a beaker and wash them in gently running water for several hours. This process removes the bacteria and stimulates production of sporangia. After washing, cut out small sections of advancing root lesions, surface-sterilise and blot the roots dry between sterile filter paper. Transfer to

selective media. Infected root material can be surface sterilised by using either one of the two methods below: (i) dip pieces of root tissue in 70% v/v ethanol for about 1 minute, wash for 10–20 seconds in sterile distilled water and blot dry on sterile filter paper or tissue paper. Cut root pieces into 0.5 cm lengths before placing onto selective media; (ii) dip root tissue in a 1:10 dilution of commercial bleach (sodium hypochlorite; approx. 0.5% v/v final concentration) for about 30 seconds. Rinse the roots in sterile water and blot dry on sterile filter paper. Cut root pieces into 0.5 cm lengths before placing onto selective media. Sterilisation with ethanol results in fewer problems with bacterial contamination and gives good recovery of most species of *Phytophthora*. It is also important that root pieces are very well dried by blotting and pushed just under the surface of agar instead of just being placed on top. This will ensure good contact between bacteria in the tissue and the antibiotics in the media. *Phytophthora* species will grow through the media quickly leaving bacterial contaminants behind.

### Biology of *Phytophthora* from Plant Tissue and Soil

*Phytophthora* can also be isolated from infected plant tissue or soil by baiting. This method is useful for two reasons: (i) the initial steps can be performed in the field, and (ii) surface sterilisation of the baited tissue is usually not required.

The best way to go about sampling soil for *Phytophthora* is as follows: where possible, samples should be taken from moist soil, near healthy roots at least 5 cm below the soil surface. The soil surface is often dry and heated by the sun, making it an inhospitable place for *Phytophthora*. Soil samples are often best taken during or immediately after wet weather, which typically increases *Phytophthora* activity. Sampling is often best under the edge of the plant/tree canopy, as root growth is more vigorous there than immediately adjacent to the stem.

Samples should be handled carefully after collection. If soil samples are exposed to drying or high temperatures (+45°C) they will lose their viability. Therefore, samples should not be left in an enclosed vehicle in warm weather. Place your soil samples in plastic bags to prevent drying out and put them in an insulated icebox to prevent overheating. Avoid low temperatures too, as *Phytophthora* does not withstand freezing. In case the samples need to be stored, do not use a refrigerator but hold them at 10–15°C and ensure that the samples are moist (add water if the samples are dry).

It is best to process samples within a few days but soil samples can be kept like this for a few months. If soil samples dry out during storage, they can be re-moistened for 1–7 days before isolation is attempted. This can stimulate production of sporangia or germination of chlamydospores or oospores.

Many plant parts can be used to selectively bait a target species of *Phytophthora*. These include fruits, seeds, seedpods, seedlings, cotyledons, leaves, leaf discs/strips, and petals.

There are three main baiting techniques:

- insertion of soil or infected tissue into a hole made on a fleshy fruit (e.g. apple, cocoa pod, pear, watermelon) — a large fruit is desirable
- planting seeds, seedlings or rooted cuttings into field soil followed by heavy watering to induce infection
- floating or partial immersing baits of various types in a water and soil mixture, which is the most widely used method for isolating *Phytophthora* spp.

The choice of bait is dependent on the species of *Phytophthora* that is suspected to be the causal agent of disease, and the host plant. A list of baiting techniques is provided in Table 5.3.

The following method is described by Chee and Foong (1968). Core out 8 mm diameter plugs of tissue from a green (unripe) cocoa pod. Insert a wedge of diseased tissue (1 cm wide × 2 cm long) or soil into the hole and push it in so that the end is flush with the outside of the fruit. Alternatively, the pod can be cut at an angle and very fine pieces of tissue such as bark inserted into the cuts. Seal the pod in a plastic bag and incubate at room temperature. Up to six wedges can be inserted into a single pod. After 4–5 days, brown discolouration should be obvious around the plugs. A firm rot indicates the presence of *Phytophthora*, a soft rot the presence of saprophytic organisms. Take a small amount of healthy tissue from around the discoloured patch. If the tissue is taken from inside the pod, it does not require surface sterilisation. Plate tissue pieces onto selective media. Other baits such as papaya and apple may also be used if cocoa pods are not available.

For those techniques requiring partial immersion of baits in soil, or floating of baits in soil, high water:soil ratios (4:1 or greater) are desirable. It is best to use distilled or deionised water or some other source of water such as bottled drinking water free from chloride or copper ions. Dilution of the soil may also dilute inhibitors present in the soil, enhancing the

formation of sporangia and zoospores. Isolations from infected bait material should be made from healthy tissue surrounding lesions. In the case of leaf discs/strips or petals, the entire tissue may be placed on the media. Include a control of water only to ensure water or baits are not infested.

## Culturing and Storage of *Phytophthora*

### Culturing

Most *Phytophthora* species grow well on a range of media. Cultures of *Phytophthora* should be grown at 15–25°C in a dark incubator. Cultures should be transferred every 2–4 weeks to maintain vigour. For long-term storage, water storage as described below is recommended. The pathogenicity of *Phytophthora* cultures is known to decrease after prolonged storage on media. In case pathogenicity studies need to be performed, serial passage through the host plant is required. Another alternative is storage of cultures in liquid nitrogen, which seems to overcome the problem of loss of pathogenicity.

### Long-term storage in sterile water

*Phytophthora* strains should be maintained as living cultures for two reasons: (i) to provide reference strains for various studies involving pathogenicity, virulence, mating type etc. and (ii) as a source of DNA for genetic diversity and evolutionary studies.

To store cultures of *Phytophthora*, cut 8–10 small blocks from the edge of an actively growing colony culture, and place in small, screw-capped glass bottles containing autoclaved distilled water. The caps should be tightened during storage and the vials placed at room temperature in the dark. Most species of *Phytophthora* can be stored this way but the isolates will lose pathogenicity and aggressiveness during storage and cannot be used for studies in that area after prolonged storage. Ideally, cultures should be revitalised once a year or every second year. For some species, a soybean or maize seed can be added before autoclaving the water as it seems to induce oospore formation in homothallic species. Record details such accession number, identification, date, host, locality, identifier's name etc.

## Identification of *Phytophthora*

The genus *Phytophthora* has been widely acknowledged as taxonomically 'difficult' (Brasier 1983) as many of the characters used for species identification are plastic, highly influenced by environment, show overlap between species, and



have an unknown genetic basis. Nonetheless, since a major review of the genus was performed by Waterhouse (1963), morphological characters have remained the basis for species identification and taxonomy (Newhook et al. 1978; Stamps et al. 1990). Waterhouse classified species based primarily on papillation and caducity (easy detachment) of sporangia, type of antheridial attachment, and mating system. Based on this analysis, the genus was divided into 6 major groups (Table 5.4), which were intended solely as an aid to species identification, and were not meant to imply a natural classification (Waterhouse 1963).

Many species of *Phytophthora* can be easily identified. However, the morphological differences among some species are few and variable, making it difficult to classify them accurately. Identification of *Phytophthora* is based on the taxonomic keys of Waterhouse (1963) and Stamps et al. (1990). Characteristics that are used to classify species of *Phytophthora* include sporangium morphology, morphology of sexual structures such as antheridia, oogonia and oospores, presence or absence of chlamydospores, and morphology of hyphae.

### Cultures

It is important to remember that, on selective media, most *Phytophthora* species will not sporulate and form characteristic propagules for identification. Therefore, cultures should be incubated at the optimum temperature for the species suspected, on a natural medium such as V8 juice, carrot agar or Lima bean agar. In order to identify an isolate of *Phytophthora* to species level, it is necessary to induce the production of asexual and sexual structures that will aid in species identification. Characteristics of the mycelium, and whether the culture produces chlamydospores, will also assist in identification.

### Morphological characters

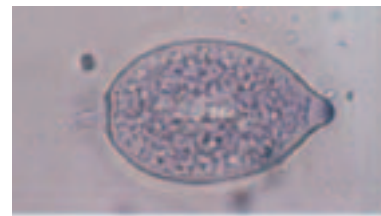
There are a number of morphological characters upon which identification of *Phytophthora* species is based. These include sporangium shape, papillation, and caducity, sporangiophore morphology,

presence of chlamydospores and hyphal swellings, antheridial attachment, and whether sexual reproduction is heterothallic or homothallic.

### Sporangia

Sporulation in *Phytophthora* cultures provides important clues for species identification. Important characters to observe are:

- sporangium morphology (shape, size, length:width ratio) papillation of the sporangium
- caducity (shedding of the sporangium at maturity) (Figure 5.1)
- length of the pedicel on the sporangium
- proliferation of sporangium (production of new sporangium within a sporangium that has germinated directly)
- branching of the sporangiophores on which the sporangia are borne.



**Figure 5.1** *Phytophthora palmivora* sporangia, papillate, caducous and with a short pedicel.

Some species of *Phytophthora* produce sporangia readily on the surface of agar media. However, many species need to be cultured in water, mineral salt solutions or dilute soil extracts before they will produce sporangia. It is important to remember that sporangia production in *Phytophthora* is dependent on light (Schmitthenner and Bhat 1994). Table 5.5 provides a general guide to which species of *Phytophthora* produce sporangia on agar media.

Sporangia can be induced by cutting blocks of 0.5 cm<sup>2</sup> agar discs from the edge of a colony that has been grown on V8 juice agar or carrot agar. Cultures 2–4 days old are most suitable. Incubate the discs in

**Table 5.4** Classification of *Phytophthora* into six groups by Waterhouse (1963).

Group	Sporangia	Antheridial attachment	Examples
I	papillate	paragynous	<i>P. cactorum</i> , <i>P. clandestina</i>
II	papillate	amphigynous	<i>P. capsici</i> , <i>P. palmivora</i>
III	semi-papillate	paragynous	<i>P. inflata</i> , <i>P. multivesiculata</i>
IV	semi-papillate	amphigynous	<i>P. infestans</i> , <i>P. ilicis</i>
V	non-papillate	paragynous	<i>P. megasperma</i> , <i>P. sojae</i>
VI	non-papillate	amphigynous	<i>P. cinnamomi</i> , <i>P. drechsleri</i>

a shallow layer of distilled water (or pond water or salt solution or soil extract) in a Petri dish, at room temperature (22–24°C). Incubation under continuous fluorescent light is recommended. Sporangia are produced within 12 hours in some species, and typically within 1–2 days.

**Table 5.5** *Phytophthora* species that produce sporangia on solid or liquid media.

Sporangia produced on agar	Sporangia produced in liquid media
<i>P. capsici</i>	<i>P. cambivora</i>
<i>P. heveae</i>	<i>P. cinnamomi</i>
<i>P. megakarya</i>	<i>P. citricola</i>
<i>P. nicotianae</i>	<i>P. cryptogea</i>
<i>P. palmivora</i>	<i>P. drechsleri</i>

### Chlamydospores and hyphal swellings

Chlamydospores are thick-walled spores that function as a resting spore. They can be intercalary (formed between hyphae) or terminal (on the ends of hyphae). They differ from hyphal swellings by having thick walls and are delimited from the mycelium by septa. The morphology of chlamydospores does not differ greatly between species and therefore these spores are of limited use in species identification. However, the presence (for example, *P. palmivora*) or absence (for example, *P. heveae*) of chlamydospores can aid species identification. Chlamydospores are generally produced readily in agar or water culture.

### Sexual structures

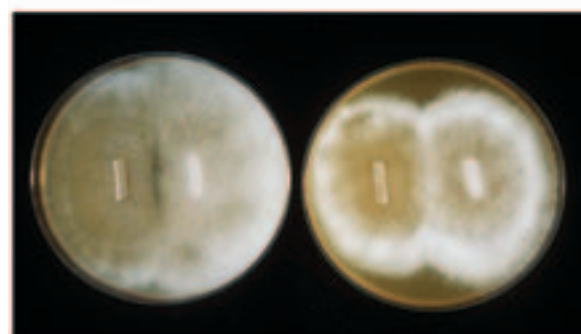
Approximately half of the species of *Phytophthora* are homothallic. They will therefore produce oogonia, antheridia, and oospores in single culture. The remainder are heterothallic, with two mating types, A1 and A2. Heterothallic species produce gametangia (oogonia and antheridia) only in the presence of an isolate of the opposite mating type on the same plate. For species identification, it is important to determine if a culture is homothallic or heterothallic, and whether the antheridium is amphigynous (Figure 5.2) (around the oogonial stalk) or paragynous (next to the oogonial stalk).

A number of media are suitable for mating type tests, including cornmeal agar, carrot agar, Lima bean agar and kidney bean agar. Kidney bean extract contains anthocyanins that are incorporated into the oogonial wall, so they stain red, making them easy to see. Although the majority of species of *Phytophthora* produce oospores in culture, some species require specialised media containing additives such as sterols to induce oospore formation. In general it is best to start with carrot

agar, which works for most species. Place a 0.5 cm<sup>2</sup> plug of culture of the unknown isolate on one side of the Petri dish. Place an agar plug from the known A1 or A2 tester isolates on the other side of the dish. Incubate plates in the dark at the optimal temperature for the species being examined. Oospores should form at the junction of the two colonies (Figure 5.3) after 7–14 days if the isolates are of different mating types.



**Figure 5.2** Amphigynous antheridia of *Phytophthora palmivora* oospore.



**Figure 5.3** Oospore tester plate.

### Differences between *Pythium* and *Phytophthora*

When isolating from soils one of the most common organisms one encounters is species of the genus *Pythium*. *Phytophthora* and *Pythium* belong to the Family Pythiaceae and hence are very closely related genera. Differences between the two include the following:

- Production of zoospores: in *Phytophthora*, the zoospores are produced within the sporangium, in *Pythium*, the zoospores develop within a vesicle produced by the sporangium. This is the most important distinguishing feature between *Pythium* and *Phytophthora*. Therefore, the second and third points below are provided for information only.

Table 5.2 Some common antibiotics: their activity, properties, preparation and alternatives.

Chemical	Activity	Target organisms	Preparation of stock solution	Stock (mg/ mL)	Range used (µg/ mL)	Comments	Alternative
Ampicillin	Antibacterial	Gram +ve bacteria	Dissolve 1000 mg (1g) powder in 10 mL distilled water. Filter-sterilise	100	250-500		Penicillin G
Benomyl	Antifungal	Most fungi except Zygomycetes and Oomycetes	Benomyl is relatively insoluble in water. Add powder to media before autoclaving. Agitate media during pouring to ensure uniform distribution of suspension.	-	10-25	Does not suppress many undesirable fungi	
Hymexazol (HMI or Tachigaren)	Antifungal	Most <i>Pythium</i> spp.	Dissolve 500 mg powder in 10 mL distilled water. Filter sterilise	50	25-50	May inhibit some <i>Phytophthora</i> spp.	
Nystatin (Mycostatin)	Antifungal	Most fungi except the Peronosporales	Dissolve 500 mg powder in 5 mL distilled water. Filter-sterilise	100	10-100	Not active against some <i>Mortierella</i> spp. Not as active as Pimaricin; used at a higher concentration	Pimaricin
Penicillin G	Antibacterial	Gram +ve and Gram -ve cocci; Gram +ve bacilli	Dissolve 500 mg powder in 10 mL distilled water. Filter-sterilise	50	50-100	Not active against Gram -ve bacilli	Supplement with polymixin B at 50-100 mg/mL
Pentachloronitrobenzene (PCNB)	Antifungal	Narrow antifungal spectrum	PCNB is not soluble in water. However, it is heat stable so that the powder can be added to the media before autoclaving.	-	10-100	Does not suppress many undesirable fungi	
Pimaricin	Antifungal	Most fungi except the Pythiaceae	Does not dissolve in water. Mix 250 mg powder in 10 mL sterile distilled water. Do not filter-sterilise	25	2-100	Not active against some <i>Mortierella</i> spp. Dosage must be restricted to $\leq 10$ µg/mL	Nystatin
Polymixin B	Antibacterial	Gram -ve bacteria	Dissolve 500 mg powder in 10 mL distilled water. Filter-sterilise	50	20-50		
Rifampicin	Antibacterial	Gram +ve bacteria; Gram -ve bacteria to a lesser extent	Dissolve 100 mg powder in 10 mL ethanol (95%). Filter-sterilise	10	10		Penicillin G and polymixin B
Vancomycin	Antibacterial	Gram +ve bacteria; Gram -ve bacteria to a lesser extent	Dissolve 1g powder in 10 mL distilled water. Filter-sterilise	100	100-200	Very expensive	Penicillin G and polymixin B

**Table 5.3** Baiting techniques for isolating *Phytophthora* from soil.

Species	Bait material	Procedure	Reference
<i>P. cinnamomi</i>	Apple or pear	Make holes in fruit. Fill with soil. Incubate covered with plastic bag at 15–27°C for 5–10 days.	Campbell (1949)
	Apple slices	Isolate from the edge of the rotted area around the hole. Suitable technique for many <i>Phytophthora</i> spp.	Gerretson-Cornell (1974)
	Avocado fruit	Immerse slices in 200 mL water to which 25 g soil has been added, for 4–10 days.	Zentmyer et al. (1960)
	Avocado seedlings	Embed fruit partially in flooded soil. Incubate at 20–27°C for 2–4 days.	Zentmyer (1980)
<i>P. citrophthora</i>	Avocado leaf pieces	Plant seedlings in wet soil. Incubate at 21–27°C for 2–3 days.	Pegg (1977)
	Apple, lemon or orange fruit	Float leaf pieces on water added to soil for 4 days.	Klotz and DeWolfe (1958)
	Lemon fruit	Insert soil or citrus tissue into fruit as per Campbell (1949) for <i>P. cinnamomi</i> . Alternatively, place lemon or orange on the surface of soil for 4 or more days	Tsao (1960)
	Lupin radicles	Immerse partially in 150 mL water to which 25 cc soil has been added. Incubate at 25°C for 6 days	Lee and Varghese (1974)
<i>P. heveae</i>	Apple fruit, eggplant fruit, cocoa pod	See Dance et al. (1975) under <i>P. cryptogea</i>	
<i>P. nicotianae</i>	Apple, lemon or orange fruit	No method is given but the baits could have soil inserted into them as per the apple method of Campbell (1949), or they could be embedded in partially flooded soil as per Zentmyer et al. (1960) for <i>P. cinnamomi</i> .	
	Citrus leaf pieces	See Klotz and DeWolfe (1958) under <i>P. citrophthora</i> .	
	Cocoa pods	Float small leaf pieces on water 1–2 cm above 100 mL soil. Incubate at 22–28°C for 3–4 days.	Grimm and Alexander (1963)
	Lemon fruit	Insert soil or diseased rubber tissues into unripe green pods as per the apple method of Campbell (1949).	Chee and Foong (1968)
<i>P. palmivora</i>	Tobacco leaves	Incubate at 26–30°C for 4–5 days.	Jenkins (1962)
	Apple fruit	See Tsao (1960) under <i>P. citrophthora</i> .	
	Apple fruit, eggplant fruit, cocoa pod	Immerse the petiole end of leaf in water–soil mixture as per Tsao (1960) under <i>P. citrophthora</i> .	
	Black pepper leaves	As per the apple method of Campbell (1949).	Lee and Varghese (1974)
	Cocoa pods	No method is given but the baits could have soil inserted into them as per the apple method of Campbell (1949), or they could be embedded in partially flooded soil as per Zentmyer et al. (1960) for <i>P. cinnamomi</i> .	
	Cocoa pods	Immerse black pepper leaves or leaf discs partially in a water–soil mixture. Used for isolation from black pepper soils.	Holliday and Mowat (1963)
	Cocoa pods	Place on or in soil. Or, inoculate pod surface with a small amount of soil suspension. Incubate in a plastic bag for 1–4 days. Used for isolation from cocoa soils.	Orellana (1959)
Cocoa pods and tissues	Insert soil beneath flaps of endocarp tissue of unripe cocoa pods. Used for isolation from cocoa soils	Turner (1965)	
Taro roots	See Chee and Foong (1968) under <i>P. nicotianae</i> .		
		Place pod or pod tissue on soil, or in flooded soil. Soil can also be placed inside the pod.	Newhook and Jackson (1977)
		Cut roots into 2.5 cm lengths, autoclave. Incubate in moistened soil (50 g/Petri dish) at 15°C for 7 days.	Satyprasad and Ramaro (1980)

- Differences in the sporangia: the sporangia of *Phytophthora* are always terminal and usually ovoid or obpyriform in shape, whereas sporangia of *Pythium* may be globulose, lobate (many lobed), or filamentous and are frequently intercalary.
- Differences in the antheridia: in *Pythium*, the antheridia are paragynous and may be attached at any point on the oogonium, whereas in *Phytophthora*, the antheridium attaches only at the lower hemisphere of the oogonium. In addition, in some species of *Pythium*, many antheridia may be attached to a single oogonium.

## References

- Brasier, P.M. 1983. Problems and prospects in *Phytophthora* research. In: Erwin, D.C., Bartnicki-Garcia, S. and Tsao, P.H. *Phytophthora: its biology, taxonomy, ecology and pathology*. St Paul, Minnesota, USA, American Phytopathological Society.
- Campbell, W.A. 1949. A method of isolating *Phytophthora cinnamomi* directly from the soil. *Plant Disease Reporter*, 33, 134–135.
- Chee, K.H. and Foong, K.M. 1968. Use of cacao pod for recovering *Phytophthora* species pathogenic to *Hevea brasiliensis*. *Plant Disease Reporter*, 52, 5.
- Dance, M. H., Newhook, F.J. and Cole, J.S. 1975. Bioassay of *Phytophthora* spp. in soil. *Plant Disease Reporter*, 59, 523–527.
- Eckert, J.W. 1962. A selective antibiotic medium for the isolation of *Phytophthora* and *Pythium* from plant roots. *Phytopathology*, 52, 771–777.
- Eckert, J.W. and Tsao, P.H. 1960. A preliminary report on the use of pimarin in the isolation of *Phytophthora* spp. from root tissues. *Plant Disease Reporter*, 44, 660–661.
- Gerrettson-Cornell, L. 1974. A comparative test of isolation of *Phytophthora cinnamomi* Rands between the lupin baiting and a newly devised apple trap. *Phyton*, 32, 35–36.
- Grimm, G.R. and Alexander, A.F. 1963. Citrus leaf pieces as traps for *Phytophthora parasitica* from soil slurries. *Phytopathology*, 63, 540–541.
- Holliday, P. and Mowat, W.P. 1963. Foot rot of *Piper nigrum* L. (*Phytophthora palmivora*). *Phytopathological Paper*, No. 5, 1–62.
- Jeffers, S.N. and Martin, S.B. 1986. Comparison of two media selective for *Phytophthora* and *Pythium* species. *Plant Disease Reporter*, 70, 1038–1043.
- Jenkins, S.F. 1962. Preliminary studies estimating the disease potential of *Phytophthora parasitica* var. *nicotianae* in infested tobacco soils. *Plant Disease Reporter*, 46, 825–826.
- Kannwischer, M.E. and Mitchell, D.J. 1978. The influence of a fungicide on the epidemiology of black shank of tobacco. *Phytopathology*, 68, 1760–1765.
- Klotz, L.J. and DeWolfe, T.A. 1958. Techniques for isolating *Phytophthora* spp. which attack citrus. *Plant Disease Reporter*, 42, 675–676.
- Lee, B.S. and Varghese, G. 1974. Studies on the genus *Phytophthora* in Malaysia. Isolation techniques, comparative morphology and physiology and reaction to antibiotics. *Malaysian Agricultural Research*, 3, 13–21.
- Masago, H., Yoshikawa, M., Fukada, M. and Nakanishi, N. 1977. Selective inhibition of *Pythium* spp. from soils and plants. *Phytopathology*, 67, 425–428.
- Newhook, F.J. and Jackson, G.V. 1977. *Phytophthora palmivora* in cocoa plantation soils in the Solomon Islands. *Transactions of the British Mycological Society*, 69, 31–68.
- Newhook, F.J., Waterhouse, G.M. and Stamps, D.J. 1978. Tabular key to the species of *Phytophthora* de Bary. Kew, Surrey, UK, Commonwealth Mycological Institute, Mycology Paper, N. 143, 20p.
- Orellana, R.G. 1959. Variation in *Phytophthora palmivora* isolated from cacao and rubber. *Phytopathology*, 49, 210–213.
- Papavizas, G.C., Bowers, J.H. and Johnston, S.A. 1981. Selective isolation of *Phytophthora capsici* from soils. *Phytopathology*, 71, 129–133.
- Pegg, K.G. 1977. Soil application of elemental sulphur as a control of *Phytophthora cinnamomi* root and heart rot of pineapple. *Australian Journal of Experimental Agriculture and Animal Husbandry*, 17, 859.
- Satyprasad, K. and Ramaro, P. 1980. A simple technique for isolating *Phytophthora palmivora* from the soil. *Current Science*, 49, 360–361.
- Schmittenner, A.F. and Bhat, R.G. 1994. Useful methods for studying *Phytophthora* in the laboratory. Wooster, Ohio, USA, Department of Plant Pathology, Ohio Agricultural Research and Development Centre.
- Stamps, D.J., Waterhouse, G.M., Newhook, F.J. and Hall, G.S. 1990. Revised tabular key to the species of *Phytophthora*. In: Agricultural Bureau of International Mycology Institute, Institute of Mycology Paper 162.
- Tsao, P.H. 1960. A serial end-point dilution method for estimating disease potentials of citrus *Phytophthora* spp. in soil. *Phytopathology*, 50, 717–724.
- 1990. Why many *Phytophthora* root rots and crown rots of tree and horticultural crops remain undetected. *OEPP/EPPO Bulletin*, 20, 11–17.
- Tsao, P.H. and Ocana, G. 1969. Selective isolation of species of *Phytophthora* from natural soils on an improved antibiotic medium. *Nature*, 223, 636–638.
- Turner, P.D. 1965. Behaviour of *Phytophthora palmivora* in soil. *Plant Disease Reporter*, 49, 135–137.
- Waterhouse, G.M. 1963. Key to the species of *Phytophthora* de Bary. In: Kew, Surrey, England: Commonwealth Mycological Institute, Mycological Papers.
- Zentmyer, G.A. 1980. *Phytophthora cinnamomi* and the diseases it causes. In: St Paul, Minnesota, American Phytopathological Society, Monograph No. 10.
- Zentmyer, G.A., Gilpatrick, J.D. and Thorn, W.A. 1960. Methods of isolating *Phytophthora cinnamomi* from soil and from host tissue. *Phytopathology*, 50, 87.