



# Germplasm Health Management

## for COGENT's Multi-site International Coconut Genebank

Robert Ikin and Pons Batugal, *editors*



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*Top (L-R)* - Coconut trees affected by lethal yellowing disease in Mexico; seednuts damaged by *Pseudotheraptus wayi* attack; palms infested by *Brontispa* in Vietnam

*Middle* - Coconut palms conserved in the International Coconut Genebank for Africa and Indian Ocean in Côte d'Ivoire

*Bottom (L-R)* - Mature seednuts ready for transport; polybagged coconut seedlings in a PEQ facility; *in vitro* cultured coconut embryo and seedling; male coconut inflorescence; pollen stored in a freezer

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***Foreword***

[to be written]

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## **Introduction**

In the past 10 years, coconut farmers have been suffering from declining farm productivity and unstable markets for their traditional coconut products which are copra (dried kernel) and oil. On the average, coconut farmers cultivate one hectare which produces one tonne of copra per year, valued at US\$150-180 at farm gate, generating a gross income below the poverty line. Coconut productivity is declining due to ageing palms, natural calamities, inadequate replanting programme, lack of suitable planting materials, poor crop management, population pressures causing crop shifts, and lack of capital for farmers to invest in coconut production. The development and use of improved coconut cultivars can markedly help solve these problems and promote increased coconut production. However, the landraces of coconut (ecotypes), which contain important genetic characters for yield, disease and pest resistance and adaptation, are under threat to genetic erosion and need to be collected, conserved, evaluated and shared more widely to develop improved varieties.

While there is a general interest to conserve and share germplasm, and collaborate in coconut breeding to produce improved coconut varieties among the 38 member countries of the International Coconut Genetic Resources Network (COGENT), there are constraints that limit this collaboration. First, while national coconut field genebanks are important sources of germplasm for exchange among COGENT member countries, many countries still lack the necessary economic and technical capacities to maintain their conserved germplasm. Second, many countries do not have the capacity to evaluate the performance of their germplasm and whatever research data they may have generated seldom allow scientific comparison of varieties/test materials. Third, multi-country negotiations for obtaining germplasm are often difficult for national breeding programmes needing to import germplasm that belong to several countries. Fourth, many researchers, who may want to share their germplasm, do not have the needed policy cover and their countries generally lack the needed facilities for ensuring the safe movement of coconut accessions. Fifth, COGENT does not have an established mechanism that could facilitate access and safe movement of germplasm to its member countries.

To address these constraints, the COGENT Steering Committee decided to establish a multi-site International Coconut Genebank (ICG) in 1995, which is now hosted by India for South Asia, Indonesia for Southeast and East Asia, Papua New Guinea for the South Pacific and Côte d'Ivoire for Africa and the Indian Ocean. Negotiations with Brazil to host the ICG for Latin America and the Caribbean are underway.

The mandate of the ICGs are: 1) to conserve nationally- and regionally-identified diversity; 2) to conserve internationally identified diversity; 3) to further assess the diversity, evaluate the performance of the conserved germplasm and disseminate related information to coconut-producing countries; 4) to make germplasm materials available to interested coconut-producing countries in accordance with existing protocols; and 5) to conduct research and training in relation to the above.

The ICGs are now in various stages of development and some of them are ready to share germplasm. However, many quarantine authorities have recognized that the importation of planting materials, in particular, plants for planting, presented the highest level of pest risk. In some countries, this level of risk is unacceptable and the resulting importation prohibitions have excluded them from the development of improved germplasm with negative economic benefits. In some cases, these prohibitions have resulted in illegal importation of germplasm of unknown

phytosanitary status. In order to devise import requirements that had technical validity, Dr Robert Ikin, formerly of the Australian Quarantine and Inspection Service, undertook to develop guidelines based upon the internationally agreed process for conducting pest risk analysis (PRA). This procedure identifies the pest risk for the various recommended forms of germplasm (seednuts, embryo or pollen) that are used, as well as the effective testing/treatment regime to address the identified risk. For many developing countries, this process requires guidelines and technical advice because of lack of local resources.

While COGENT desires to implement a progressive coconut germplasm movement programme it would, at the same time, like to protect the coconut industry of receiving countries. Following the recommended approach of Dr Ikin and based upon the international PRA standards of FAO, the International Plant Genetic Resources Institute (IPGRI), the executing agency of COGENT, with funding assistance from the Australian Centre for International Agricultural Research (ACIAR), developed and published this manual on "Germplasm Health Management for COGENT's multi-site International Coconut Genebank". ACIAR has agreed to support this very important and strategic initiative.

Chapter 1 of the manual provides the background of the ICG, the rationale for its establishment, the major stakeholders, its mandates and the status of the regional collections and activities in the host countries. Chapter 2 provides the principles and operational procedures of germplasm health management starting with the requirements for germplasm exchange under the 1993 IBPGR-FAO Guidelines for Safe Movement of Coconut Germplasm and the revision in 1997, followed by a description of PRA, its rationale and operational approach. The chapter also includes the data of identified coconut pests in COGENT member countries and prescribes the needed quarantine interventions required for safe germplasm movement among and between them. Chapter 3 recommends the operational management of germplasm movement involving seednuts, embryos and pollen as these are the recommended materials for germplasm exchange of coconuts.

The materials for Chapter 1 were provided by the COGENT Coordinator and the managers of the ICG in each of the current host countries. The materials for Chapter 2 were obtained primarily from the IPGRI consultancy report on pest risk analysis conducted by Dr Ikin for the ICG-South Asia, ICG-Southeast and East Asia, ICG-South Pacific as well as from the IPGRI consultancy report of Dr Hubert de Franqueville of the Centre de Cooperation Internationale en Recherche Agronomique pour le Developpement (CIRAD) who did a similar work for the ICG-Africa and the Indian Ocean and the proposed ICG-Latin America and the Caribbean. For Chapter 3, the section on embryo culture was written by Mrs Erlinda Rillo, embryo culture expert of the Philippine Coconut Authority (PCA); and on pollen, by Mr Gerardo Santos and Mr Ernesto Emmanuel, Centre Manager/Senior Coconut Breeder and Coconut Breeder, respectively, of PCA's Zamboanga Research Centre; and on seednuts, by Dr Ikin.

It is hoped that the manual will be useful in providing coconut genebank managers with the effective procedures for safe coconut germplasm exchange, and the quarantine officers with alternative options for making informed quarantine decisions for germplasm movement.

**Pons Batugal**  
*COGENT Coordinator*





## CHAPTER 1

# COGENT'S Multi-Site International Coconut Genebank

- COGENT's Multi-Site International Coconut Genebank (ICG)
- ICG for the South Pacific (Papua New Guinea)
- ICG for South Asia (India)
- ICG for Southeast and East Asia (Indonesia)
- ICG for Africa and the Indian Ocean (Cote d'Ivoire)
- Proposed ICG for Latin America and the Caribbean (Brazil)



## COGENT's multi-site International Coconut Genebank

### *Pons Batugal*

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### **Background of COGENT**

In 1992, the Consultative Group on International Agricultural Research (CGIAR) decided to include coconut in its research portfolio after studies indicated that international support and global coordination of research in coconut is essential to make coconut more productive and beneficial to small-scale coconut farmers. The CGIAR and its Technical Advisory Committee (TAC) recognized that international support to coconut research was needed as many coconut-producing countries lacked both the human and material resources to conduct expensive and time-consuming research. Thus, it tasked the International Plant Genetic Resources Institute (IPGRI) to undertake research on coconut genetic resources, which the CGIAR identified as one of the five priority research areas that deserved international support. Accordingly, IPGRI included coconut genetic resources in its plant genetic resources research programme and organized the International Coconut Genetic Resources Network (COGENT) to implement this mandate. Starting with 15 countries, COGENT has rapidly developed into an active global network currently involving 38 coconut-producing countries (Table 1).

**Table 1.** COGENT member countries

<b>Southeast and East Asia</b>	<b>South Asia</b>	<b>South Pacific</b>	<b>Africa/Indian Ocean</b>	<b>Latin America/ Caribbean</b>
1. China 2. Indonesia 3. Malaysia 4. Myanmar 5. Philippines 6. Thailand 7. Vietnam	1. Bangladesh 2. India 3. Pakistan 4. Sri Lanka	1. Cooke Islands 2. Fiji 3. Kiribati 4. Papua New Guinea 5. Solomon Islands 6. Tonga 7. Vanuatu 8. Samoa	1. Benin 2. Cote d'Ivoire 3. Ghana 4. Kenya 5. Madagascar 6. Mozambique 7. Nigeria 8. Seychelles 9. Tanzania	1. Brazil 2. Colombia 3. Costa Rica 4. Cuba 5. Guyana 6. Haiti 7. Honduras 8. Jamaica 9. Mexico 10. Trinidad-Tobago

COGENT's goal is to improve coconut production on a sustainable basis and increase income of coconut farmers and growers in developing countries through improved cultivation of the crop and efficient utilization of its products. The objectives of COGENT are:

- 1) To establish and maintain an international database on existing and future collections;
- 2) To encourage the protection and use of existing germplasm collections;
- 3) To identify and secure additional threatened diversity by developing and adopting suitable technologies and conservations strategies;
- 4) To promote greater collaboration among research groups in producer

countries and advance technology sources in the exchange of germplasm and the development of new techniques;

- 5) To conduct appropriate training and information dissemination; and
- 6) To secure necessary funding for network activities.

In the last 12 years, COGENT has generated modest but significant achievements. The network has been successfully established with a Steering Committee serving as its supervisory and policy making body and is fully operational with 38 coconut producing countries as members. The International Coconut Genetic Resources Database (CGRD) has been established that currently contains passport and characterization data of 1416 accessions conserved in 25 sites in 23 countries. In addition, 278 ecotypes from the Asia Pacific region have been collected and conserved. To further secure conserved germplasm, a multi-site International Coconut Genebank (ICG) is being established to conserve 200 important accessions in the regions in which COGENT operates, which is hosted by India, Indonesia, Papua New Guinea, and Côte d'Ivoire and Brazil (under negotiation). An additional 212 farmers' varieties have been identified and are currently being characterized. Multipurpose uses of coconut varieties are also being documented and promoted. The performance of 34 high-yielding hybrids are being evaluated in multilocation hybrid trials in four African and three Latin America/Caribbean countries to select varieties and hybrids that are suited to particular agroecological conditions and to determine germplasm x environment interaction. Farmers' varietal preferences in 15 countries are being evaluated. Diversity-linked income-generating activities have been initiated in 15 countries as part of a strategy to promote *in situ* and on-farm conservation, and germplasm utilization. Protocols for embryo culture, cryopreservation, morphometric and molecular marker-based methods for locating and characterizing diversity, assessing pest risks and managing germplasm health are being developed, tested and upgraded. Strategies and techniques for farmer participatory research, collecting, characterization, and *ex situ* and *in situ* conservation are being refined.

To strengthen coconut research capability of COGENT member countries, IPGRI and COGENT have, as of 2003: organized 39 country missions involving 28 experts to help COGENT member countries conduct research needs assessment and to identify priority research and training activities; conducted 41 workshops and meetings involving 994 coconut researchers to share information and technologies, discuss issues and common problems and opportunities and how to address them; conducted 40 training courses involving 765 participants from 41 countries; supported 180 research projects in 30 member countries; and led in establishing the Global Coconut Research for Development Programme (PROCORD), a global coconut research alliance with the Bureau for the Development of Research on Perennial Tropical Oil Crops (BUROTROP) and the Asian and Pacific Coconut Community (APCC). COGENT's current priority involves the further promotion of more effective conservation and use of coconut genetic resources, both regionally and globally. This includes the establishment and operation of COGENT's multi-site International Coconut Genebank (ICG).

### ***Integrated approach to coconut conservation***

COGENT's conservation strategy is anchored on promoting the sustainable protection of diversity as well as maximizing germplasm use. In developing its conservation strategy, COGENT recognized that no one method or approach of conservation can meet all conservation needs and that there is a need to employ a combination of methods to ensure the sustainable conservation of as much genetic diversity as possible.

It actively encourages the participation of its member country governments, partner organizations in both developing and developed countries, non-government organizations (NGOs) and coconut farmers themselves in conserving germplasm. The components of COGENT's conservation strategy consist of:

1. Conservation in national collections;
2. Conservation in the multi-site ICG;
3. *In vitro* embryo culture and cryopreservation;
4. *In situ* and on-farm conservation; and
5. Promoting conservation through use by developing and implementing a globally-coordinated coconut breeding programme, establishing farmer community-managed coconut seedling nurseries in at least 25 countries, linking germplasm conservation and use with the broader areas of research and development assigned to BUROTROP (agro-physiology and crop protection) and APCC (processing and marketing), developing and disseminating catalogues of conserved germplasm and farmers' varieties, and upgrading and widely disseminating the CGRD.

### ***Conservation in the multi-site International Coconut Genebank***

#### **Rationale**

World coconut production is declining due to ageing palms, natural calamities, inadequate replanting programme, lack of suitable planting materials, poor crop management, population pressures causing crop shifts, and lack of capital for farmers to invest in coconut production. The development and use of improved coconut cultivars can markedly help solve these problems and promote increased coconut production. However, the landraces of coconut (ecotypes), which contain important genetic characters for yield, disease and pest resistance and adaptation, are under threat to genetic erosion and need to be collected, conserved, evaluated and shared more widely to develop improved varieties.

Conservation and use of a wide range of coconut diversity is faced with several constraints. First, while national coconut field genebanks are important sources of germplasm for exchange among COGENT member countries, many countries still lack the necessary economic and technical capacities to maintain their conserved germplasm. Second, many countries do not have the capacity to evaluate the performance of their germplasm while the data obtained are often not comparable. Third, multi-country negotiations for obtaining germplasm are often difficult for national breeding programmes needing to import germplasm that belong to several countries. Fourth, many researchers, who may want to share their germplasm, do not have the needed policy cover and their countries generally lack the needed facilities for ensuring the safe movement of coconut accessions. Fifth, COGENT does not have a concrete mechanism that would facilitate access and safe movement of germplasm to its member countries.

To address these constraints, the COGENT Steering Committee decided to establish a multi-site ICG in 1995. Subsequently, site assessment surveys were conducted to evaluate the suitability of proposed regional genebank sites in the five host countries of Indonesia, India, Papua New Guinea, Côte d'Ivoire and Brazil. During the International Coconut Genebank workshop held from 26 to 28 February 1996 at Pekanbaru, Riau, Indonesia, representatives of IPGRI, the Centre de Cooperation Internationale en Recherche Agronomique pour le Développement (CIRAD) and the World Bank worked with representatives of COGENT member countries in developing a series of legal agreements, initial work plans and proposed

budgets, using national funds for each of the initial four genebanks to be hosted by Indonesia for Southeast and East Asia, Papua New Guinea for the South Pacific, India for South Asia and Côte d'Ivoire for Africa and the Indian Ocean.

### **ICG objectives and initial activities**

The objectives of the ICG are: 1) to conserve nationally- and regionally-identified diversity; 2) to conserve internationally identified diversity; 3) to further assess the diversity, evaluate the performance of the conserved germplasm and disseminate related information to coconut-producing countries; 4) to make germplasm materials available to interested coconut-producing countries in accordance with existing protocols; and 5) to conduct research and training in relation to the above.

Memoranda of Agreements (MOAs) for hosting of the ICGs for Southeast and East Asia (Indonesia), South Asia (India), South Pacific (PNG) and Africa and the Indian Ocean (Côte d'Ivoire) were developed and signed by the host countries and IPGRI on behalf of COGENT, with the Food and Agriculture Organization (FAO) of the United Nations serving as trustee. All MOAs were worded similarly (see MOA for ICG-SP in Annex 1.1). Negotiations are underway for Brazil to host the ICG for Latin America and the Caribbean. The host countries agreed to commit resources for their establishment maintenance and data gathering. The existing national field collections of Côte d'Ivoire and Papua New Guinea were donated to the ICG. However, COGENT is also exerting efforts to source additional funds for the maintenance of collections. COGENT has developed a sustainability strategy for the ICGs consisting of the following:

- 1) MOA committing host countries to maintain the field genebanks;
- 2) Negotiations for income from the ICG to be used for maintenance;
- 3) Superimpose research and training onto the ICG to share the cost of administration and maintenance;
- 4) Charge requesting countries for the cost of preparation, shipment and maintenance of germplasm, the latter on a pro-rata basis;
- 5) Undertake income generating activities in ICG plantations such as the production and marketing of high-value products from all parts of the coconut and integrate with intercropping and livestock raising as appropriate;
- 6) Generate external donor support; and
- 7) Generate national and provincial/state funding and institutional support.

The sites for ICG were chosen based on surveys conducted by coconut experts who considered and evaluated several important selection criteria. Thus, the basic needs of field genebanks such as safety, security, accessibility, environment, etc. have been established. Among several items that were considered, two principles were highlighted. First, the choice of material in the ICG was determined by the needs of the users and by the need for as much representation of genetic diversity and ecotypes as possible. The importance of having a balance between elite lines and accessions that represent a broad range of genetic diversity from within the country as well as from the region was recognized from the beginning. Care has been taken to ensure that each ICG accession is unique and is not a duplicate. Thus current are being further validated using molecular marker studies to eliminate duplicates. Second, decisions were made as to which accessions would be maintained regionally and nationally. It was agreed that the nationally important accessions that cannot be accommodated in the regional genebanks would be maintained in the collections of strong national programmes. Thus, from the beginning it was apparent that national collections and the ICG would complement each other to accommodate as much coconut genetic diversity as possible.

It is envisioned that the ICG at each regional site will conserve in field genebanks about 200 accessions which are important to the region. The ICG field genebanks are part of the *ex situ* collection under the International Undertaking on Plant Genetic Resources. The designated germplasm are conserved in the field genebanks and shared with coconut growing countries based on material transfer agreements. The field genebanks are established and managed by national programmes under the oversight of COGENT and IPGRI. Laboratories and facilities will also be developed to further locate diversity, identify and eliminate duplicates, conduct disease indexing, process pollen and embryos for export, conduct cryopreservation and train coconut researchers from member countries in evaluating, conserving and using germplasm. Thus, each site of the ICG will be developed as Centres of Excellence in concurrence to IPGRI's initiatives of building and upgrading the capacity of partner institutions.

### **Germplasm conservation and sharing**

In the next seven years (2004-2010), the ICG host countries aim to conserve in respective regional field genebanks a maximum of 200 accessions each, which will be contributed by coconut-producing countries in each region. Accessions will be imported in the form of excised embryos, grown *in vitro* in the embryo culture laboratory, transferred into pots in the greenhouse and eventually transplanted in the field. These accessions, which will be planted in the field genebank of about 200 hectares, will be characterized and evaluated using agronomic and molecular data methods by the ICG to determine their diversity, performance and potential for improvement work. Four types of coconut accessions will be conserved in the ICG: 1) major varieties (parents of existing hybrids and advanced generations of selected cultivars); 2) varieties/cultivars threatened with genetic erosion or total loss; 3) varieties/cultivars with special traits/genetic markers; and 4) genotypes being used for current genetic diversity studies using molecular markers.

Member countries of each region can access germplasm belonging to different countries by negotiating with each ICG host country. The requested accessions will be sent in the form of embryos or pollen to interested countries after disease indexing to ensure safe movement. Requesting countries will be charged the cost of producing the seednuts and for preparing the embryos as well as the pro-rata cost of maintenance, disease indexing and shipping. These germplasm transfers will be covered by Material Transfer Agreements (MTA).

### **Initial achievements**

Under COGENT, ICG sites in four host countries have been strengthened to some extent (i.e., ICG-South Asia (India), ICG-Southeast and East Asia (Indonesia), ICG-South Pacific (Papua New Guinea) and ICG-Africa and the Indian Ocean (Côte d'Ivoire)). IPGRI has supported the ICGs in capacity building for embryo culture technology, in terms of materials, skills and laboratory upgrading to prepare them for importing and maintaining germplasm from network member countries in their respective regions. They have also been trained on germplasm collecting, morphometric and molecular marker (microsatellite kits) methods of germplasm characterization, genebank management and on cryopreservation. Since COGENT is currently an open network, it was proposed to further strengthen germplasm conservation by executing a formal Memoranda of Agreement with COGENT member countries, at the highest government level, to formalize their membership in COGENT and to formally commit access to their coconut germplasm

Despite meagre resources, the ICGs have made some significant achievements. Table 3 shows the date of signing of the hosting agreements and the status of conserved germplasm in each of the host countries.



**Table 3.** Germplasm conserved in the multi-site ICG

Name of Genebank	Date of MOA signed	Initial number in list of designated germplasm	Designated germplasm currently conserved
1. International Coconut Genebank for the South Pacific (Papua New Guinea)	30 September 1998	55	50
2. International Coconut Genebank for South Asia (India)	30 October 1998	49	46
3. International Coconut Genebank for Southeast and East Asia (Indonesia)	26 May 1999	52	29
4. International Coconut Genebank for Africa and The Indian Ocean (Côte d'Ivoire)	14 October 1999	49	99*

\* Includes additional accessions entered into the ICG after the signing of the MOA

### Plan of action for the International Coconut Genebank

As part of the above-described conservation strategy for coconut, IPGRI and COGENT would like to undertake the following plan of action for the upgrading of the ICG in the next seven years:

1. Regeneration of old palms of 50 accessions in the ICG for Africa and the Indian Ocean;
2. Additional morphometric and molecular marker characterization of the 1,416 accessions conserved in the national collections of 23 countries to select other entries for the ICG and to upgrade the CGRD; and of the 224 accessions in the ICG to identify duplicates;
3. Integration of the CGRD with SINGER, the CGIAR-supported systems-wide genetic resources programme;
4. Importation and establishment of additional accessions into the ICG sites to complete the 200 accessions per site;
5. Upgrading of pollen processing and embryo culture laboratories, net houses and coconut seedling nurseries in each ICG site;
6. Establishment of the needed facilities in the ICG host countries (i.e., molecular marker laboratories, except for ICG-Africa and Indian Ocean; disease indexing laboratories; training and dormitory facilities);
7. Research and training support for the following strategic activities: somatic embryogenesis, embryo culture, molecular marker/microsatellite research, pest risk assessment and germplasm health management, germplasm x environment interaction and genetic distance analysis of conserved germplasm, and globally- coordinated coconut breeding; and
8. International Coconut Genebank evaluation and meeting of stakeholders.

### Conclusion

Two of the major priorities of IPGRI and COGENT are (1) saving threatened diversity and (2) promoting the use of conserved materials for developing improved varieties for national programmes and small-scale farmers. Thus accelerated effort is being placed on the movement of germplasm from COGENT's member countries to their regional ICG and the provision of breeding materials from the older ICG (i.e., Côte d'Ivoire and Papua New Guinea) to member countries and soon, from the other ICG host countries where some of the new conserved materials are now starting to bear fruits.

While IPGRI/COGENT desires to implement a progressive germplasm movement initiative, at the same time, it would like to ensure that this is done in a safe manner to protect the coconut industry of receiving countries. Thus IPGRI approached the Australian Centre for International Agricultural Research (ACIAR) to fund the development and publication of a manual on Germplasm Health Management for COGENT's multi-site International Coconut Genebank. ACIAR has agreed to support this very important and strategic initiative. This manual will be useful as a guide to genebank managers and plant quarantine officers worldwide in making informed decisions on the safe movement of coconut genetic resources.

## The International Coconut Genebank for the South Pacific (Papua New Guinea)

**Mathias Faure**

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### **Background**

In August 1995, the COGENT Genebank Task Force visited PNG to evaluate the suitability of the proposed site and the commitment of the PNG government. The proposed genebank site is the Murunas plantation currently named Stewart Research Station of the PNG Cocoa and Coconut Institute (CCRI) in Madang, which has a total area of 450 ha, 200 ha of which was made available for the ICG. The CCRI staff and laboratories in Rabaul will provide support to the field genebank in Madang. The larger vegetation was cleared in 1993 and the secondary growth will be cleared as needed. Drainage canals will also be constructed as needed.

The annual rainfall is 3500 mm, evenly distributed, and the soil is mostly silty clay loam. Following the successful site suitability evaluation and COGENT's acceptance, the PNG Stewart Station's coconut genebank has been transformed into the International Coconut Genebank for the South Pacific (ICG-SP), which to date conserves a total of 50 designated germplasm.

The Memorandum of Agreement to establish the ICG-SP was signed in November 1998 between the Government of PNG, IPGRI on behalf of COGENT and the Food and Agriculture Organization (FAO) of the United Nations serving as trustee. The list of initial designated germplasm as stipulated in the signed MOA is shown in Annex 1.2.

Since 1994 to 2004, 10 specialists visited PNG on eight technical assistance missions including assessing the country's coconut R&D capability and assist the national programme in identifying common problems and opportunities for network collaboration, identifying a suitable site for ICG-SP, evaluating embryo culture laboratories and training their staff, evaluating COGENT's germplasm collecting and conservation strategies, assessing the pest risk for the ICG-SP, assisting in the installation of machineries and training in the production of coconut virgin oil, fiber-based products and coconut candies.

To date, IPGRI/COGENT has helped the ICG-SP establish an embryo culture laboratory which is fully operational with additional stocks of glassware and chemicals purchased in June 2001 and a seedling nursery for *ex vitro* seedling production. It has also supported the training of ICG staff on embryo culture, genebank management, germplasm characterization using morphometric methods and molecular marker (microsatellite kits) methods. In addition, six local coconut researchers have undergone staff development training sponsored by COGENT on topics such as the STANTECH; coconut collecting and conservation; coconut data analysis; computer use, documentation, data analysis, dedicated statistical software; and coconut cryopreservation.

The ICG-SP contains important germplasm from the fragile ecosystem of the South Pacific. These include typhoon-resistant accessions with big trunks and fruits, which are suitable for the Pacific islands. In the last two years, through COGENT-CIRAD collaboration, precious coconut populations from Cooke Islands, Fiji, Kiribati, Marshall Islands and Tuvalu have been collected, which were not previously available. A total of 13 accessions from four countries (the atoll countries of Tuvalu, Kiribati, Cook Islands and Marshall Islands) were collected by the CIRAD/COGENT team, embryo cultured and initially grown *in vitro* at the

laboratories of the Secretariat of the Pacific Community (SPC) in Suva, Fiji and subsequently sent to the ICG-SP in 2000/2001. These accessions, which were collected to prevent losing them from the threat of global warming and possible water rise, were grown in the embryo culture laboratory and nursery and planted in the field. Other germplasm from other Pacific countries will also be imported when funds become available. A total of 62 embryos from one accession (Fiji Tall) were provided by Fiji in March 2002. The imported germplasm are currently being maintained in the laboratory using the upgraded coconut embryo culture protocol. Most of the evaluation work on the performance of the germplasm is being carried out in the field.

Recently, a report was received from ICG-SP Papua New Guinea stating that frequent power outages, have been posing a serious threat to the operations of the embryo culture laboratory and causing damage to the embryo-derived plantlets. The PNG Cocoa and Coconut Institute (CCI) and the Secretariat of the South Pacific Commission requested the assistance of COGENT and IPGRI to enable the institute to purchase a standby generator. In response to this request, IPGRI/COGENT co-financed with CCI the purchase and installation of a standby generator. The generator is currently being used to support the air conditioners and other equipments of the embryo culture laboratories in case of power interruptions.

## The International Coconut Genebank for South Asia (India)

**Velamoor Rajagopal**

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The Central Plantation Crops Research Institute (CPCRI) hosts the International Coconut Genebank for South Asia (ICG-SA). The field genebank in Kidu Farm, Karnataka, which is the ICG-SA field genebank, is supported technically by the laboratory facilities at CPCRI, Kasaragod. CPCRI maintains the world's largest assemblage of germplasm by undertaking the planting and maintenance of the field genebank and activities on embryo culture, assessment of diversity using molecular markers and disease indexing. The National Bureau of Plant Genetic Resources (NBPGR), New Delhi, collaborates with CPCRI on cryopreservation activities.

In July 1995, the COGENT Task Force evaluated the forested area adjacent to the CPCRI Seed Farm in Kidu, the genebank site proposed by the government of India, and found it suitable. The Kidu field genebank is situated in Dakshina Kannada District of Karnataka about 90 km east of Mangalore and about 100 km east of Kasaragod. The farm lies between 12.30°N and 75.20°E at an elevation of 291 msl. The summer temperature range between 33 and 40°C and the winter temperature is between 22 and 18°C. The soil is mostly red lateritic, changing to alluvial laterite towards the riverbank. The average annual rainfall is 2900 mm with a river on the southern farm boundary as perennial source of irrigation water. Irrigation is essential as the site has a distinct dry period. Since the proposed site is within a forest without any coconut plantation nearby, the risk of disease spread from neighboring plantations is minimal. The nearest root wilt affected area is 650 km from the site and the disease is said to have moved only 100 km during the last 120 years.

The Memorandum of Agreement for the establishment of the ICG-South Asia was signed by the Government of India, IPGRI on behalf of COGENT and FAO as trustee in October 1998. The list of initial designated germplasm during the signing of the MOA is shown in Annex 1.3. To date, India has conserved a total of 46 of designated germplasm in the ICG-SA. Nearly 30 ha of forestland have been cleared and surrounded with electric fencing for planting. Furthermore, drip irrigation has been provided to 2700 seedlings that have been planted there.

IPGRI/COGENT has helped the ICG-SA in collecting germplasm from the Indian Ocean Islands of Maldives, Comoros, Madagascar, Reunion and Seychelles. To date, a total of 746 embryos were collected from Sri Lanka in February 2001. Out of these, a total of 396 embryos were damaged. A total of 401 embryos were collected from Bangladesh during November to December 2001, of which 157 embryos survived. The embryos were collected from the following varieties: Chinasukanya, Chinasukanya Dwarf Orange, Pubail Tall, Kayemkola Tall, Bagharpara Tall, Rupdia Tall, Khairtala Tall and BARI Narikel-I, BARI Narikel-II, Uzirpur Tall and Agailjhara.

In 2003, 34 collections were added to the existing collection. These included five collections from Goa, six from Maharashtra, eight from Assam, four from Sri Lanka and 11 from Bangladesh. The collections from Sri Lanka and Bangladesh were in the form of zygotic embryos. These embryos cultured *in vitro*, were rooted and later planted in pots. Conservation of coconut germplasm in the form of *in vitro* culture is being attempted at CPCRI, Kasaragod. Furthermore, a total of 4962 inter-crossed nuts from 31 accessions were also sown to generate planting materials at the ICG-SA. To produce additional seednuts, a total of 3004 female flowers were pollinated from eight accessions for regeneration.

From 1995 to the present, seven coconut specialists visited India to help identify a

suitable site for the International Coconut Genebank for South Asia (ICG-SA); evaluate COGENT's collecting strategies; and conduct a pest-risk assessment for the ICG-SA. In 2000, a regional training course on *In Vitro* Conservation and Cryopreservation on PGR was conducted by NBPGR with seven participants from five COGENT member countries. Another eight local staff from various collaborating institutions and NARS were trained on various topics including use of the STANTECH manual, *in vitro* embryo culture and cryopreservation techniques as well as the use of the microsatellite kit and dedicated statistical software.

## **The International Coconut Genebank for Southeast and East Asia (Indonesia)**

***Hengky Novarianto***

*Director, Indonesian Coconut and Palm Research Institute, Manado, Indonesia*

The International Coconut Genebank for Southeast and East Asia (ICG-SEEA) is hosted by the Indonesian Agency for Agricultural Research and Development (AARD) using the field genebank in Pekanbaru, Riau Province; experimental gardens in Manado, North Sulawesi; and AARD laboratory facilities in Bogor, West Java and in Manado, North Sulawesi.

In July 1995, the COGENT Task Force evaluated the proposed site at Sikijang Mati, Pekanbaru, Riau Province in Central Sumatra and found it to be generally suitable and made some suggestions for improvement. The site is located 20 km from the city of Pekanbaru, the capital of Riau. Pekanbaru has regular flights from Jakarta (1.5 hours) and Singapore (30 minutes), as well as other cities in Sumatra. The annual rainfall is about 2000 mm, well distributed over the year. The topography of the area is undulating and most of the land is covered by secondary forest, with small rivers. The soils are yellow to yellow-red podzolic, low in organic matter and with pH of around 5.0. The soils are generally very poor and unsaturated but they make a good substratum for the crop to grow and respond well to the application of fertilizers, which are readily absorbed by the crop. Since the area was not very uniform, the Task Force recommended that a detailed survey be undertaken to select only those areas where soils are generally good and more than one meter deep to avoid the hard pan. About 1000 ha of secondary forest has been offered by the Government of Indonesia which could be used for the ICG (200 ha) and the rest for production area to generate income for the maintenance cost of the ICG.

The Memorandum of Agreement for the establishment of the ICG-Southeast and East Asia was signed by the Government of Indonesia, IPGRI on behalf of COGENT and FAO as trustee in May 1999. The function of the coconut collection at Sikijang was not only for germplasm conservation and collection, but also for genetic evaluation and utilization. To date, Indonesia has conserved a total of 29 of designated germplasm in the International Coconut Genebank for Southeast and East Asia at Sikijang. The list of initial designated germplasm during the signing of the MOA is shown in Annex 1.4.

Due to the financial crisis in 1997 and the resulting lack of government budget, there was slow development of the Sikijang area, resulting in the squatting of the remaining areas by surrounding inhabitants and migrants. Two extension ICG areas have therefore been identified: the Paniki Experimental Garden (100 ha) located beside the Indonesian Coconut and Other Palmae Research Institute (ICOPRI) office in Manado, and the Pandu Experimental Garden (80 ha) which is about 18 km from the ICOPRI office and belonging to the Balai Pengkajian Teknologi Pertanian (BPTP). The soil and climate there are very suitable for coconut growing. Therefore, it was recommended that the main part of the ICG-SEEA be moved from Sikijang to North Sulawesi. However, the 29 accessions which have been collected will remain in Sikijang and maintained by the Indonesian Government.

To date, a total of four accessions have been received from Malaysia, six from China and 10 each from the Philippines, Thailand and Vietnam, respectively. A total of at least 100 accessions have been conserved from Indonesia from 1996 to 2001. Twenty-nine of these 100 accessions have been planted at Sikijang, Pekanbaru, Riau, which was the initial identified site for the ICG-SEEA. In addition, a total of 460

embryos of Malayan Tall and 469 embryos of Malayan Green Dwarfs were received from Malaysia and successfully cultured *in vitro*.

IPGRI/COGENT has supported the ICG-SEEA in collecting germplasm from the Moluccas Island, East Timor, West Nusa Tenggara, Sangir Talaud Islands, Salibabu Island, Buol District, Central Sulawesi, Sangir Talaud district and North Sulawesi.

From 1995 to 2000, 11 specialists have visited Indonesia to help the country in its coconut PGR activities. These include identifying a suitable site for the ICG-SEEA, collecting leaf samples for electron microscopy detection of mycoplasma, identifying marketable alternative products for coconut as well as suitable varieties for these products, evaluating COGENT's collecting and conservation strategies, assessing pest risk and evaluating the progress of the ICG and assisting in the installation of equipment for feasibility studies.

Four training courses were held in the country, whereby 52 researchers from nine countries attended. The training courses, which were funded by the Asian Development Bank (ADB), were hosted by ICOPRI (formerly the Research Institute for Coconut and Palmae or RICP). IPGRI/COGENT has also sponsored 20 local researchers and specialists for staff development training in coconut data analysis, coconut collecting and conservation, embryo culture, technical writing/seminar presentation and proposal writing, the use of the microsatellite kit and others which are related to the poverty reduction project.



## **The International Coconut Genebank for Africa and Indian Ocean (Côte d'Ivoire)**

**Jean Louis Konan**

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The ICG-AIO is hosted by the Marc Delorme Coconut Research Station in Côte d'Ivoire which has a total area of 1200 ha. The soil of the station is composed of alluvial deposits of tertiary sands with 8-10% clay, poor in organic matter and minerals. The climate is characterized by two dry seasons of different lengths, one from December to April and the other in August to September which alternate with two rainy seasons. The mean of annual rainfall is 1800 mm.

The MOA for the establishment of the ICG-AIO was signed in October 1999 by the Government of Côte d'Ivoire, IPGRI on behalf of COGENT and FAO as trustee. At the time of the signing of the MOA, the coconut genebank of the Marc Delorme Coconut Research Station was converted into the ICG-AIO. The list of initial designated germplasm during the signing of the MOA is shown in Annex 1.5.

To date, ICG-AIO has a total of 99 accessions. Furthermore, five tall varieties from Sri Lanka, Tonga, Vanuatu, Tagnanan and Rotuma were received and planted on eight hectares for their renewal. A total of 3,400 embryos were provided to CIRAD/IRD Montpellier for in vitro-culture-technique development. Seed nuts were also provided to participating countries in the CFC-funded multilocation hybrid trials of IPGRI/COGENT. A researcher from the Centre National Agronomique (CNRA) has visited the five others participating countries (Benin, Tanzania, Brazil, Mexico and Jamaica) to help the project trial implementation.

Two researchers from Marc DELORME visited the western region of Ghana as part of its collaborative research activity on lethal yellowing disease. Twelve kilograms of VTT (Vanuatu Tall) pollen were also provided to the Ghana coconut programme per year to produce lethal yellowing-tolerant hybrids. For 2004, nine dwarf varieties from the ICG-AIO were selected to be tested also against the disease. Marc Delorme Station also received two research teams, from Senegal and Mayotte Island to help them in coconut development. A total of 70 800 seednuts of improved varieties were produced for smallholder farmers and the industrial sectors in the country. For Nicaragua (Coconut Research Institute, CRI), 1200 grams of Panama Tall Monagre pollen have been provided per year to allow appropriate hybrids production. About 9500 seedlings of improved Mawa (PB121) hybrids were provided to Guinea in 2000 and 2001 for commercial planting.

In 1999, one COGENT-commissioned expert visited Côte d'Ivoire to conduct a pest risk analysis of the ICG-AIO. Two training courses were hosted by the Centre National de Recherche Agronomique (CNRA) in the Côte d'Ivoire up to 2002. Researchers representing 11 countries participated in the training courses. Furthermore, another two local staff underwent IPGRI/COGENT-sponsored staff development training at CIRAD in Montpellier, France on the use of molecular markers (microsatellite kit and associated statistical software), and on cryopreservation. Currently, the application of microsatellite analysis is ongoing at the central biotechnology laboratory of the Centre National de Recherche Agronomique. Leaf samples of important varieties from Ghana will also be collected and analyzed. These activities are funded by IPGRI/COGENT.

Leaf samples of Cameroon Red Dwarf x Rennell Island Tall were provided to CIRAD in France and Max Planck Institute in Germany for coconut map

construction, in collaboration with the Mikocheni Agricultural Research Institute in Tanzania (MARI), Philippines Coconut Authority (PCA), Philippines), NEIKER in Spain. For this hybrid, agronomic evaluation is being undertaken in Marc Delorme. These activities are realized

About 7200 seednuts of Tall (seven varieties), Dwarf (nine varieties) and hybrids (seven crossings) were produced by assisted and controlled pollination for Mozambique for coconut seedgarden establishment. For germplasm exchanging, the Coconut Research Institute of Sri Lanka is sending a research team to Marc Delorme Station in August 2004. Embryos of three varieties (Nawasi Tall, King coconut and Ran Thambili) will be brought to the ICG-AIO. Embryos of seven varieties will be collected and brought to Sri Lanka for conservation.

## **Proposal for the establishment of the International Coconut Genebank for Latin America and the Caribbean (Brazil)**

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During the COGENT Steering Committee meeting in November 1998, the representative of Empresa Brasileira de Pesquisa Agropecuaria (EMPRAPA) presented Brazil's proposal to host the International Coconut Genebank for Latin America and the Caribbean (ICG-LAC). Subsequently, a site suitability and pest risk assessment survey was undertaken in April 1999 to evaluate the suitability and pest risk of the ICG if situated in Itaporanga, west of Aracaju; the Neopolis plateau, northeast of Aracaju; and Betume, located between Neopolis and Ilha das Flores. Due to ownership problems, the Neopolis Plateau was dropped as a prospective site. Likewise, due to distance problem, the Betume Station was also not found suitable. Thus, the Itaporanga station was subsequently identified as the proposed ICG- LAC.

Itaporanga is 20 km to the City of Aracaju, located 10° 55' South Latitude and 37° 03' West longitude, with an elevation of only one meter above sea level. Its predominant soil is ferric with good drainage. The climate is generally warm with the coldest month higher than 15°C. The average annual rainfall is 1643 mm. The area is flat and about 100 ha is available for establishing Tall accessions. Additional areas to plant additional accessions should be identified

In 1999, COGENT commissioned one expert to go to Brazil to conduct a pest risk analysis of the proposed site of the ICG-LAC. Two local staff were sponsored by COGENT to attend staff development training course on the use of the standardized techniques in coconut breeding (STANTECH), microsatellite kit (molecular marker), dedicated statistical software, technical writing/ seminar presentation and proposal writing. Several meetings and communications were conducted between EMBRAPA and COGENT to discuss issues related to the hosting of the ICG-LAC which includes the issues of derivatives, compliance to Brazil's legislation on intellectual property rights and funding. The discussions are continuing to date.

## CHAPTER 2

# Germplasm Health Management

- Germplasm Health Management
- Pest Risk Analysis (PRA)
- Regional and Country Transfer and Exchange, and Identification of Pests



## Germplasm health management <sup>1</sup>

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This chapter aims to identify the procedures that need to be undertaken in order to identify the phytosanitary risks that are posed by the exchange of germplasm between countries. This is considered initially at a generic level, where the international context of the need for the technical justification of phytosanitary requirements is examined. Then the focus is on the management and operational considerations for risk mitigation of the movement from specific countries to the COGENT's multi-site International Coconut Genebank (ICG) where the emphasis is on the application of pest control measures to the task of coconut germplasm exchange so that the risk of the transfer of pests into new environments is addressed.

In essence, the process identifies the coconut pests that are recorded in the country of origin, determines if these pests pose a threat to the importing country and then adopts control measures, either at export or after import, to address these threats.

### Steps in the process of exchanging germplasm

	Output	Activity
<b>Step 1</b>	List of coconut pests (and diseases) that are recorded in the exporting country	Obtain information from pest lists
<b>Step 2</b>	List of coconut pests that could be carried in plants, seednuts and embryos	Compile a datasheet for each pest and identify associations
<b>Step 3</b>	List of quarantine pests	Decide if the pest can enter, establish and spread and if it has economic impact
<b>Step 4</b>	Description of level of pest risk	Determine the level of risk the pest poses in each pathway
<b>Step 5</b>	Recommended treatments against each quarantine pest.	Identify how to eliminate the pest from the pathway by inspection or treatment
<b>Step 6</b>	Effective treatments to be applied to the commodity at the effective point in the pathway (preferably before export) identified	Identify what facilities for treatment are available in exporting and importing countries
<b>Step 7</b>	A report that identifies the risk quarantine pests and the conditions for import for distribution to the exporting country	Determine the import conditions for plants, seednuts and embryos

### **Background to germplasm exchange**

The risks that the exchange of germplasm posed through the transfer of pests and diseases (hereafter referred to as pests) were examined in the pioneering publication by Hewitt and Chiarappa (1978). At that time, the pest risks that were inherent in the movement of genetic material, particularly from areas where significant economic pests were endemic, was recognized. The importance of the need to move germplasm from one country to another in order for countries to benefit from plant

<sup>1</sup> The definitions of key words/ terms used in this chapter are found in Annex 2.1.

breeding and selection elsewhere meant that systems need to be developed so that pest risks would be minimized. Nowadays, the process of exchange of germplasm is facilitated by the capacity to use different parts of the plant (particularly excised sterile tissues) and the capability to test for pests using non-destructive molecular techniques.

Throughout the 1980s, a number of technical meetings were held at which these issues were discussed, culminating in the suggestion that a global framework should be considered. In particular within the Consultative Group on International Agricultural Research (CGIAR) system, which is responsible for the collecting, curation and evaluation of the major agricultural economic crops, the movement of pests from areas where they had probably evolved was of major concern.

As organizations with global responsibilities for both plant health and germplasm resource preservation, the United Nations Food and Agriculture Organization (FAO) and the International Bureau of Plant Genetic Resources (IBPGR), which at that time, was a programme of FAO and now known as the International Plant Genetic Resources Institute (IPGRI), undertook to develop international guidelines for the safe exchange of germplasm for a range of economically important agricultural crops (Frison and Putter 1988). In 1969, FAO published a monograph on the pests of coconut as the first in a series on crops of economic importance (Lever 1969) with the emphasis on the control of pests in the field. These new guidelines consider the pest risk of collecting germplasm from all types of sources and make technical recommendations on the measures that would reduce the risk of the parallel movement of pests with the materials being exchanged. In particular for crops such as coconuts that could not be readily maintained in a protected environment, the need for appropriate control measures, particularly for genetic material collected in the field, is seen as higher than those materials kept under some form of protection, such as in a genebank.

In the past, country quarantine authorities or National Plant Protection Organizations (NPPOs) have recognized that the importation of planting materials have, as a pathway, presented the highest level of pest risk. In some countries, this risk has been considered as unacceptable and such imports have been prohibited. However, countries that have instigated such prohibitions have excluded themselves from the development of improved germplasm overseas, with obvious economic consequences. In some cases, the imposition of a total prohibition has been totally counter-productive and has precipitated the illegal importation of germplasm of unknown phytosanitary status. Countries now recognize that the controlled import of germplasm, under conditions that address the risk that each of the types of material pose (plants, seed, cuttings or tissue cultures), is the only way in which new materials can be made available to farmers. The task nowadays is to identify pest risk, use the recommended type of germplasm, test or treat this risk through an effective regime and provide sufficient facilities, equipment and staff to be able to facilitate exchange with minimal but justified import restrictions. For many developing countries, this process requires guidelines and technical advice because of the lack of local resources.

### ***FAO/IBPGR Guidelines***

In a cooperative programme between the FAO and IBPGR (now IPGRI) and a number of important commodity-specific international organizations primarily those of the CGIAR, experts from around the globe contributed to the development of germplasm exchange guidelines. The aim was to provide a standardized format for considering the risks involved in the exchange procedure by identifying the pests of concern, discussion of the types of risk that they individually posed (particularly as it

relates to the type of material being exchanged) and then the measures that would be required to be applied so that the pests are not transferred with the germplasm. Another objective is to provide information on the methods of detection of the pests that could be used to determine healthy material prior to export or as a test in post-entry-quarantine (PEQ), where required.

The FAO/IBPGR Technical Guidelines for the safe movement of coconut germplasm (Frison, Putter and Diekmann 1993) were published following a meeting hosted by the Central Research Institute for Industrial crops, Indonesia in October 1991. The experts compiled information in terms of the causes of the coconut diseases, their symptoms, their natural host ranges, geographical distributions, transmissions (particularly if vectored), virus therapy and indexing procedures for viruses and other systemic diseases, and recommendations for quarantine measures, on the following pests<sup>2</sup>:

1. Viral disease
  - Coconut foliar decay virus (CFDV)
2. Viroid disease
  - Coconut cadang-cadang (CCCVd)
  - Coconut tinangaja (CtiVd)
  - Other viroid-like sequences (VLS)
3. Mollicute diseases
  - Blast
  - Lethal yellowing (LY) and similar diseases
  - Root wilt or Kerala wilt
  - Tatipaka disease
4. Fungal diseases
  - Bole rot, shoot rot and other *Marasmiellus* diseases
  - *Phomopsis* leaf spot
  - *Bipolaris* leaf blight
  - Bud rot and fruit rot (*Phytophthora* spp)
  - Leaf blight (*lixia pequena*, *lixia grande*)
  - Stem bleeding
  - Leaf spots and blights
  - Ganoderma butt and root rots
5. Protozoan disease
  - Hartrot, fatal wilt, Cedros wilt or Marchitez (*Phytomonas* spp)
  - Nematodes
  - Red ring disease (*Bursaphelenchus cocophilus* (Cobb) Goodey.)

The guidelines made recommendations on the movement of pollen, embryo cultures and seednuts with special consideration towards the pests identified. The guidelines also made general recommendations on the procedures that should be undertaken at an operational level to facilitate safe germplasm exchange.

As a general recommendation, it was felt that germplasm should be moved as embryo cultures or pollen from trees that appeared healthy and from sites where pests of unknown etiology were known to be present.

Due to the uncertainty at that time of the role of viroid-like sequences (VLS), it was recommended that until information of the significance and distribution of the VLS are known, all germplasm from countries where the sequences are known to

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<sup>2</sup> The term 'Pests', as used in this manual and based on the International Plant Protection Convention (IPPC) Glossary, is not restricted to arthropods but also includes diseases caused by fungi, bacteria, viruses, MLOs, viroids, nematodes, weeds, etc.



occur to those where they have not been reported should be indexed, and material for which tests are positive should be rejected.

### **1997 revision**

The recommendations of the guidelines were considered at a Pacific regional meeting in 1993 (Foale and Lynch 1994), and resulted in the endorsement of the need for testing of suspect material before movement to other countries. However, this resulted in the almost complete cessation of movement of germplasm within and outside the South Pacific because the precise location of the VLS were not identified in the paper by Hanold and Randles (1991), nor were countries able to identify infested areas at this forum because of issues of national confidentiality.

Regrettably, the issue of the need for transparency of national surveys, and the international treaty obligations of countries who are parties to the International Plant Protection Convention (IPPC) were not considered at this meeting. Under Article VII of the 1979 IPPC text, countries agree to cooperate at an international level by reporting “on the existence, outbreak and spread of economically important pests of plants and plant products, which may be of immediate or potential danger.”

Clearly, the failure of countries to abide by these obligations to report the presence of the VLS was constructing barriers to trade in germplasm, and furthermore was jeopardizing the establishment of the COGENT's multi-site ICG.

During this period following the conclusion of the GATT Uruguay Round of negotiations and the formation of the World Trade Organization (WTO), the IPPC was mandated by the WTO as the technical agency to develop international phytosanitary standards<sup>3</sup>. The purpose of these standards was to harmonise the administration of phytosanitary measures at all national levels and between NPPOs with the aim of providing a platform for free trade. Under the Sanitary and Phytosanitary (SPS) Agreement of the WTO, phytosanitary measures, for the first time, would have to be based on technically justified principles, key amongst them being the acceptance of levels of risk and the implementation of the process of Pest Risk Analysis (PRA) on the measurement of the pest risk, and the determination of appropriate control measures.

In 1999, a meeting was convened by IPGRI, with support from the Australian Centre for International Agricultural Research (ACIAR), with parties interested in the revision of the coconut germplasm guidelines. The decisions made in 1991 within the SPS framework of international standards, particularly the International Standards for Phytosanitary Measures (ISPM) No. 2 on “Guidelines for Pest Risk Analysis” (FAO 1996) were specifically reviewed.

The meeting evaluated in detail the state of knowledge on the VLS since the production of the coconut germplasm guidelines and critically assessed the issues of:

1. The identification of sequences as those relating to known symptoms of a disease and whether Koch's postulates had been completed so that a relationship between cause and effect could be clearly established;
2. The characterization of the disease/disorder as meeting the criteria of a quarantine pest as specified in the IPPC through application of the international standard definition in terms of presence or absence in an area;
3. Official control if not widely distributed; and
4. Economic effect on the infected crop.

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<sup>3</sup> The International Standards for Phytosanitary Measures or ISPM. For more information, visit the IPPC Portal at [http://www.ippc.int/cds\\_ippc\\_prod/IPP/En/standards.htm](http://www.ippc.int/cds_ippc_prod/IPP/En/standards.htm).

PRA, in accordance with ISPM No. 2, was undertaken for both CCCVd and the VLS, and discussed at the meeting. It concluded that for CCCVd, restrictions on the movement of material (plants, seednuts and pollen) from the infected areas in the Philippines were justified, and that phytosanitary measures as outlined in the Guidelines would be required to ensure safe movement of germplasm (Ikin 1997).

**Quarantine pest** - pest of potential economic importance to the endangered area but not yet present there, or present but not widely distributed and being officially controlled.

However, in the case of the VLS there were clear indications that the information on the linkage of the detection of the sequences and any specific and consistent disease symptoms would question whether action would be justified, or even possible. For example, if consistent symptoms were not related to a causal organism, how would anyone determine when to require tests for presence to be conducted? Consistent with the definition of the term quarantine pest, the widespread detection of the VLS throughout the Asia-Pacific region indicates that the pest cannot be considered as such, because it is widespread in all the countries surveyed, and that as a consequence, phytosanitary measures would not be justifiable. On the economic criteria of the definition, the lack of information on a clear association between the detection of VLS, the expression of symptoms and economic loss would also suggest that classifying VLS as a quarantine pest is tenuous.

The meeting concluded that the restriction on the movement of germplasm because of the presence of VLS was not technically justified. It accordingly amended the Germplasm Guideline.

#### **Addendum to the FAO/IBPGR Technical Guidelines**

The footnote on page 6 and the section on page 23 of the Technical Guidelines under the heading "Other viroid-like sequences" should be modified as follows:

*"Several viroid-like nucleic acid sequences related to cadang-cadang viroid are widely distributed in coconuts and understorey plants. They are not proven disease-causing agents and should therefore not be considered to be of quarantine significance."*

Nevertheless, recognizing that there was a lack of information on the linkages between CCCVd and seed transmission, the relationship between the detection of VLS and symptoms and economic impact on coconuts, further work would be required on detection/indexing, pathogenicity, transmission and the host range of VLS. Regrettably, much of this work has yet to be undertaken.

#### **Pest Risk Analysis (PRA)**

The first international standard developed by the IPPC Secretariat, with the aim of facilitating the process of developing and implementing international standards, was the ISPM Pub. No. 1: *Principles of plant quarantine as related to international trade* (FAO 1995). One of these principles, risk analysis applies to the process of determining whether pests are quarantine pests and the measures that could be applied to reduce the phytosanitary risk in trade.

International standards have now been developed and adopted that describe the risk analysis process in general (ISPM No. 2) (FAO 1996), the specific pest risk analysis process to determine if pests are quarantine pests and the strength of the

**Risk analysis** - Determines which pests are quarantine pests and the strength of the measures to be taken against them. Countries should use pest risk analysis methods based on biological and economic evidence and, wherever possible, follow procedures developed within the framework of the IPPC.

measures that should be applied through pest risk management for those identified as quarantine pests (ISPM No. 11) (FAO 2002). These standards are used to determine the measures that should be applied to the movement of germplasm between countries. Germplasm movement recommendations are procedures that address the phytosanitary risks posed

by quarantine pests identified in the risk assessment phase of the PRA. The task is to devise guidelines for the exchange of seednuts, embryo cultures and pollen. Those conducting PRA follow a process defined by three stages:

- **Stage 1 (initiating the process)** involves identifying the pest(s) and pathways that are of quarantine concern and should be considered for risk analysis in relation to the identified PRA area;
- **Stage 2 (risk assessment)** begins with the categorization of individual pests to determine whether the criteria for a quarantine pest are satisfied. Risk assessment continues with an evaluation of the probability of pest entry, establishment and spread, and of their potential economic consequences; and
- **Stage 3 (risk management)** involves identifying management options for reducing the risks identified at Stage 2. These are evaluated for efficacy, feasibility and impact in order to select those that are appropriate.

The issue of risk communication has been recognized as a key to the transparency of the process, and what used to be a sub-set of risk management has developed an importance of its own through the publication of PRA reports by NPPOs. As new international standards have been adopted, the linkage of the PRA standards to these newer standards has become critical to the understanding of the role of the PRA process and its application at an operation level within the responsibilities of NPPOs. Table 1 identifies these linkages between the standards. In addition, the key principles of plant quarantine are included in Table 2 together with their relevance to the current exercise as a guide to the underlying philosophy that underpins the risk analysis process.

As noted before, the requirements for an NPPO to technically justify the phytosanitary measures that are required of imports (import conditions on the import permit) are for some a new obligation, and one which may be difficult to undertake. In some cases, the response to high risk quarantine pests has been to impose prohibitions that in fact may be counter-productive, encouraging bypassing of the quarantine system.

Although Table 1 lists a very complex matrix of interactions between the current process of Pest Risk Analysis and those international standards that have been adopted as of 2003, there are just a few key issues that underpin the process. These key principles are:

1. **Transparency** – all technical information should be available and the decisions made should be documented;
2. **Technical justification** – any decision should be supported by appropriate technical references and information; and
3. **Managed risk** – where risk is identified, the imposition of a measure is justified and the measure should not be unrealistic.

**Table 1.** Linkages of the International Standards for Phytosanitary Measures with the International Standards for Pest Risk Analysis (ISPM Nos. 2 and 11)

ISPM No.	Title	Linkage(s) to the PRA standards
1	Principles	<ul style="list-style-type: none"> <li>• Minimal impact</li> <li>• Transparency</li> <li>• Equivalence</li> <li>• Risk analysis (the process for the technical justification of measures)</li> <li>• Managed risk</li> <li>• Pest free areas</li> </ul>
4	Pest-free Areas	<ul style="list-style-type: none"> <li>• Treatment (PR Management)</li> <li>• Links to No. 14 (Systems Approach)</li> </ul>
5	Glossary of Terms	<ul style="list-style-type: none"> <li>• Dictionary of the terminology for all the standards</li> </ul>
6	Surveillance	<ul style="list-style-type: none"> <li>• Collection, storage and retrieval of information <ul style="list-style-type: none"> <li>○ Specific surveys</li> <li>○ Pest surveys</li> <li>○ Commodity or host surveys</li> </ul> </li> </ul>
7	Export Certification	<ul style="list-style-type: none"> <li>• Legal authority</li> <li>• Management responsibility</li> <li>• Staff</li> <li>• Equipment</li> <li>• Phytosanitary certificates</li> <li>• Procedures</li> <li>• Records</li> <li>• Tracing</li> <li>• Communication (within and outside the country)</li> <li>• Review mechanisms</li> </ul>
8	Pest Status	<ul style="list-style-type: none"> <li>• Pest records</li> <li>• Pest status in an area</li> <li>• Reporting practices</li> </ul>
9	Pest Eradication	<ul style="list-style-type: none"> <li>• Evaluation of pest reports</li> </ul>
10	Pest-free Places	<ul style="list-style-type: none"> <li>• Treatment (PR Management)</li> <li>• Links to No. 14 (Systems Approach)</li> </ul>
12	Phytosanitary Certificates	<ul style="list-style-type: none"> <li>• Certification issuance</li> </ul>
13	Non-compliance and Emergency Action	<ul style="list-style-type: none"> <li>• Notification of non compliance</li> <li>• Basis for notification</li> <li>• Information included in notification</li> <li>• Communication</li> </ul>
14	Systems Approach	<ul style="list-style-type: none"> <li>• Treatment (PR Management)</li> </ul>
17	Pest Reporting	<ul style="list-style-type: none"> <li>• Pest lists (PR Initiation)</li> </ul>
19	Regulated Pest Lists	<ul style="list-style-type: none"> <li>• Pest lists (PR Initiation)</li> </ul>

**Table 2.** Key principles of plant quarantine as they relate to germplasm exchange (from ISPM No. 1)**General Principles**Sovereignty

With the aim of preventing the introduction of quarantine pests into their territories, it is recognized that countries may exercise the sovereign right to utilize phytosanitary measures to regulate the entry of plants and plant products and other materials capable of harbouring plant pests.

In any bilateral situation, a country can request specific conditions for any import, but it should recognize that the other principles should also be considered as applying, and in particular if the conditions are not technically based they can be challenged under the IPPC and WTO. However, it is unlikely that this will be the case for germplasm as the process is not normally a commercial operation.

Transparency

Countries shall publish and disseminate phytosanitary prohibitions, restrictions and requirements and, on request, make available the rationale for such measures.

If requirements are changed, other countries must be informed, particularly those who are directly affected by the changed import conditions. Exporting countries may not be able to meet them immediately and trade will have to be halted for the time being.

Equivalence

Countries shall recognize as being equivalent those phytosanitary measures that are not identical but which have the same effect.

As diagnostic tests for pests are developed they tend to be applicable in specific purposes. Nevertheless, different procedures are often available for the detection of the same pest with the same reliability and specificity. In this case, this should be recognized and accepted in providing detection options. Treatments for pests are not generally very specific and a single treatment may be acceptable for a number of pests. For the same pest, treatment with different chemicals may give the same efficacy and should be equally acceptable, and a non-chemical treatment may also have the same effect.

Risk analysis

To determine which pests are quarantine pests and the strength of the measures to be taken against them, countries shall use pest risk analysis methods based on biological and economic evidence and, wherever possible, follow procedures developed within the framework of the IPPC.

Pest Risk Analysis is the key process for phytosanitary decision-making and ensures that decisions are based on sound science and the operational import conditions are those that reflect the risks that are identified and can be undertaken with minimal impact on trade. If PRA is not used then decisions become arbitrary and inconsistent.

Managed risk

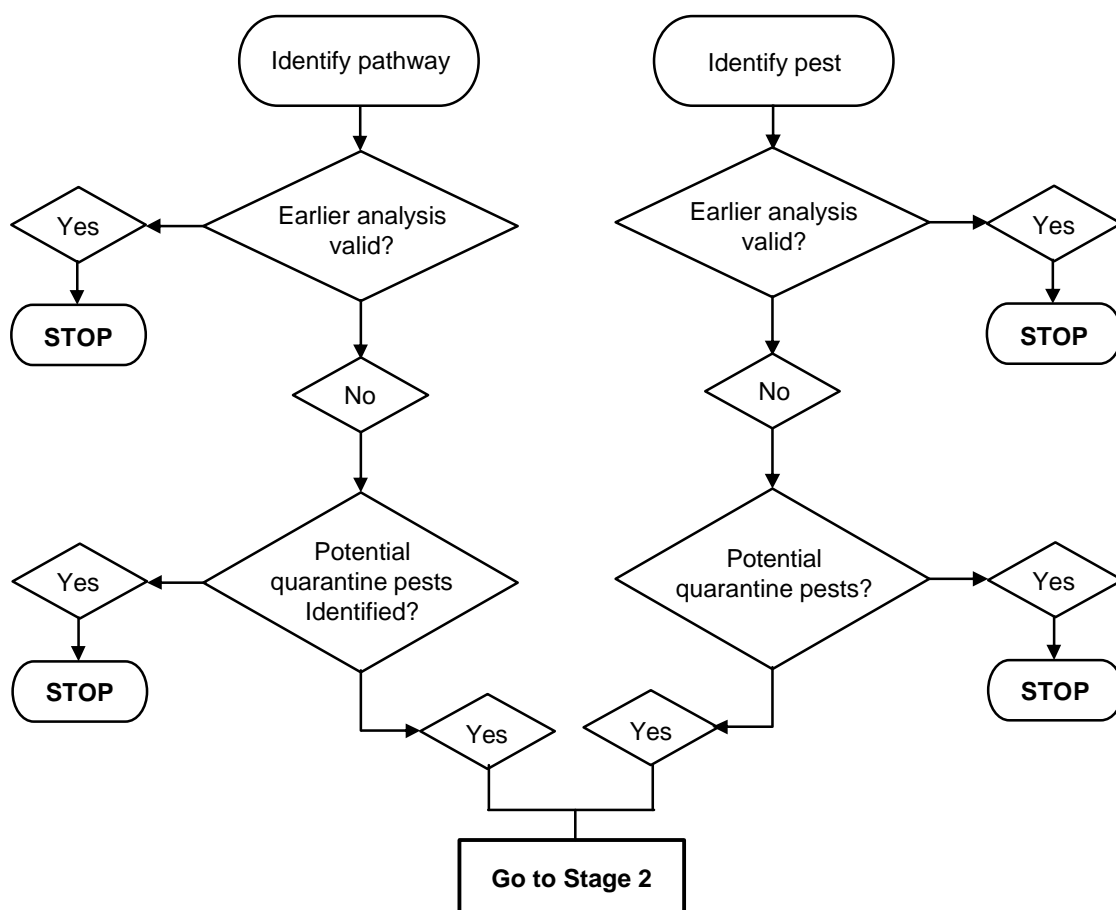
Because some risk of the introduction of a quarantine pest always exists, countries shall agree to a policy of risk management when formulating phytosanitary measures.

Risk always exists because there are always persons who are capable of smuggling material into a country who will attempt to avoid detection and treatment methodologies are based on scientific methods that have been developed by a statistical methodology that accepts less than 100% accuracy. NPPOs have to accept this position when developing import conditions for commodities.

### Stage 1: Initiating the PRA

Initiating the PRA process (Figure 1) involves the identification of the range of pests that are likely to be in the commodity pathway. In this phase, a pest is identified as having the potential to be in the pathway of a particular commodity. In the case of movement within and between the host countries of the multi-site ICG, information on national pest status are taken from the available international technical literatures. International pest data for this particular exercise were taken from sources such as the Guidelines for the Exchange of Coconut Germplasm (Frison, Putter and Diekmann 1993), the Commonwealth Agricultural Bureau International's (CABI) Crop Protection Compendium (CABI 2003), as well as other sources listed in the bibliography. Such a literature search compiles information on all pests associated with the coconut crop worldwide, irrespective of the type of material that is to be moved as germplasm. In the case of *Cocos nucifera* L., CABI listed 267 coconut pests worldwide, including weeds that are associated with the crop in the field. The list of regulated/ quarantined pests, as compiled from this initial general search of technical sources, is presented in Annex 2.2.

Coconut germplasm is nowadays exchanged as extracted embryos in sterile media, as seednuts and as pollen. In accordance with the FAO/IBPGR Guidelines, the movement of growing plants (plants for planting) is not recommended as a safe activity, and therefore is not considered in this study.



**Figure 1.** PRA Stage 1: Initiating the process

Crucial to the correct progression of the PRA process is the determination of the pest status of the respective countries in accordance with ISPM No. 8 – “Pest status in an area” (FAO 1998). In this exercise, the status of many pests is uncertain and without conducting extensive in-country surveys, will have to rely upon the literature citations presently available. In the Asia-Pacific region, there have been a number of useful compilations of pests and diseases of economic importance. These include data on coconut but these have been obtained in consultation with agencies that have not provided primary technical references as is generally the case of other compendia (APPPC 1987; Waterhouse 1993 and 1997; Li, Wang and Waterhouse 1997). This information have been included at face value in the CABI Compendium, but it has not been possible to further investigate the specific impact these pests have on the coconut plant, particularly the plant part affected, or to completely validate the records by further cross references.

Therefore, it is essential that the NPPO of the country of import, or the relevant agency checks the lists of pests compiled from literature sources to verify pest records. The recommendations of this publication must therefore be considered only as a guideline needing pest list verification prior to adoption. Indeed, pest distribution changes day-by-day and all data must be verified in view of the timeframe of technical discovery and technical publication.

For each of the pests identified in the primary pest list (Annex 2.2) the technical data for pests of potential quarantine concern are compiled in a pest datasheet. The datasheet includes information on pest biology, in particular that which relates to the capacity of the pest to be in the pathway and to enter, establish and spread in the importing area. When available, information on the economic importance of the pest is also gathered in order to support the classification of the pest as a quarantine pest in accordance with the International Plant Protection Convention and Stage 2 of the PRA process.

There is no international standard for the range of information that is required of a pest datasheet, but as a default that serves to answer the requirements of Stage 2 of the PRA process, the following data fields have emerged as those used by a number of NPPOs:

- Scientific name (including strain and biotype);
- Synonyms;
- Hosts (primary and secondary);
- Plant parts infested/infected;
- Worldwide distribution;
- Biology – aspects of the life stages that could be used to determine entry potential, establishment, spread potential and vector role;
- Economic importance; and
- Major reference sources.

In some cases, information is not specifically available and some assumptions have had to be made by the author, with assistance from other experts. In particular, if it has not been possible through searches of further literature to identify specific economic damage caused by a potential quarantine pest, then it is assumed not to meet the IPPC criteria of a quarantine pest and has to be eliminated from further consideration in the analysis. Datasheets of key quarantine pests are in Annex 2.3.

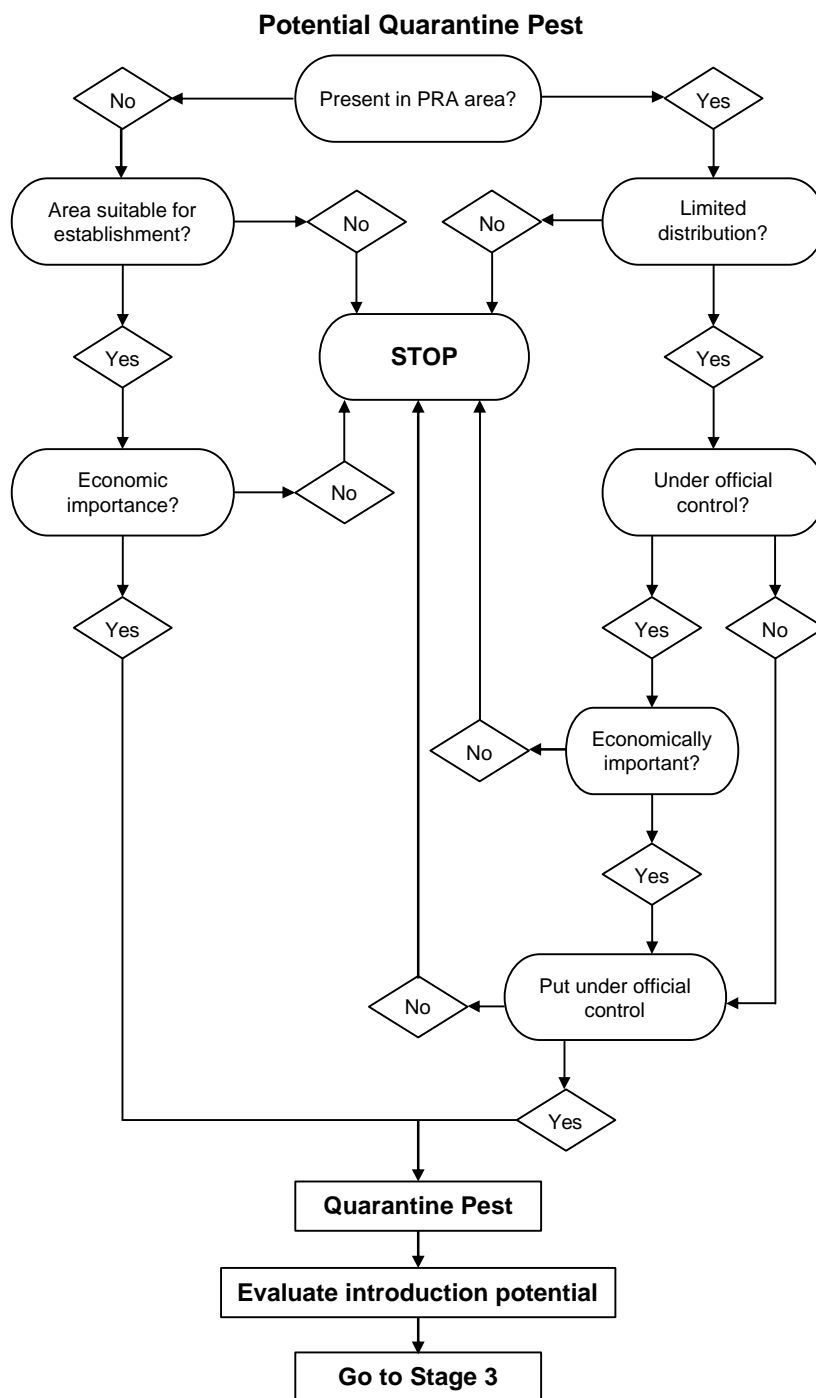
At the conclusion of the Pest Risk Initiation stage for the exchange of germplasm, a list of potential quarantine pests has been compiled by subtraction of the pests in the country of import from the country of export. Pests that are under official control in the country of import may be included in the list, in accordance with the

international standard. At this stage the compilation of the pest datasheet has begun, to be completed during Stage 2 of the PRA process.

### Pathway analysis for quarantine pests

#### Stage 2. Assessing the risk

The second stage (Figure 2) of the PRA process determines if the potential quarantine pests identified at the end of Stage 1 have the criteria to meet the specific requirements of a quarantine pest as defined by the IPPC and in the application of the term throughout the other international standards as specified in the Glossary of Phytosanitary Terms ISPM No. 5 (FAO 2003).



**Figure 2.** PRA Stage 2: Assessing the risk



The process for pest risk assessment can be broadly divided into three interrelated steps:

- Pest categorization;
- Assessment of the probability of introduction and spread; and
- Assessment of potential economic consequences (including environmental impact).

In most cases, these steps will be applied sequentially in a PRA but it is not essential to follow a particular sequence. Pest risk assessment needs to be only as complex as is technically justified by the circumstances. This standard allows a specific PRA to be judged against the principles of necessity, minimal impact, transparency, equivalence, risk analysis, managed risk and non-discrimination set out in ISPM Publication No. 1: Principles of plant quarantine as related to international trade (FAO 1995).

In the evaluation of a pathway associated with a commodity, a number of individual PRA activities may be necessary for the various pests potentially associated with the pathway. The opportunity to eliminate an organism or organisms from consideration, before an in-depth examination is undertaken, is a valuable characteristic of the categorization process.

The categorization of a pest as a quarantine pest includes the following primary elements:

- Identity of the pest;
- Presence or absence of the pest in the PRA area;
- Regulatory status;
- Potential for establishment and spread in PRA area; and
- Potential for economic and environmental consequences in the PRA area.

Pest introduction is comprised of both entry and establishment. Assessing the probability of introduction requires an analysis of each of the pathways with which a pest may be associated from its origin to its establishment in the PRA area. In a PRA initiated by a specific pathway (usually an imported commodity, in this case coconut seednuts, embryos or pollen), the probability of pest entry is evaluated for each of the pathways in turn.

### **Risk assessment based on datasheets**

Currently, the majority of assessments undertaken are qualitative, because for complex biological systems the data do not facilitate quantitative considerations. The assessments are also as objective as possible based upon the information that can be compiled into the pest data sheet, but as value judgements are made they reflect the individual views of those conducting the assessment, be they risk takers or risk averse. The international standard (FAO 2002) identifies in detail the types of information that are needed to enable a decision to be made on the status of a potential pest, but does not specify any particular questions that might be suitable for any pest or group of pests.

A number of systems for PRA have been developed with various degrees of success. The European and Mediterranean Plant Protection Organization's PRA system (EPPO 2003) provides a framework for analyzing the data within the pest datasheet, and allocates numerical values to the responses. The CABI CPC Phytosanitary Decision Support module (now called PRA) (CABI 2003) also asks specific queries of particular pest data sheet information and allows a rating of high, medium or low to be allocated to the responses (with an overall level of response also permitted at the different stages of introduction and spread). A selection of

suitable topics and questions that would be asked for an individual potential pest would be:

**1. Entry**

- What is the likelihood of the pest being associated with the host plant part at the origin? In the case of diseases and the seed pathway, is it seed transmitted?
- Is the life stage of the pest on the pathway likely to survive transport, and is it a stage that is capable of multiplication?
- Does the pest require a minimum population to survive?

**2. Establishment**

- What is the likelihood of hosts being available? Are vectors required for transmission to hosts? Are the vectors present in the country of import?
- Is the environment suitable for establishment? Are alternate hosts required and are they available?
- If the pest has a vector, can the vector alone introduce it, and what is the biology of the vector?

**3. Spread**

- What is the risk of the pest spreading to an area where the crop is of economic importance?
- Are the required vectors present?
- Can vectors be controlled by pest management systems?

**4. Economic impact**

- What is the economic impact of the pest?
- Is this information provided from assessments in the field or experimentally?
- Is the economic impact under conditions similar to the importing country on varieties currently in use and under similar management regimes?
- What is the potential loss to the environment and agriculture?

In terms of economic assessments, because much of the data on the impact of pests have been compiled to be able to demonstrate the effect of the application of control procedures, the conditions under which they were obtained are distorted to favour the pest and hinder the crop. If possible, use data from field work rather than experimental plots.

In conducting a PRA for the transfer of germplasm between countries, a number of assumptions must be made:

1. That every effort is made to ensure that the part of the plant that is being transferred could be propagated, and that as a consequence without any management of pests being implemented (unrestricted access), the pests in the pathway are likely to be present and the likelihood that they might survive is high;
2. That hosts of the pests in the pathway are likely to be present in the PRA area;
3. That only small quantities of material are likely to be moved from one centre to another;
4. That the conditions for the establishment of pests are similar to those conducive to the survival of the germplasm at the point of post entry quarantine (e.g. climate); and
5. That transfer of germplasm from the ICGs has a known specific status in terms of pests known to be present or absent, but that in the transfer from countries to the ICGs the collection could be from the field with little or no knowledge of the phytosanitary status.

As an example of the type of information that is used in determining the status of the various pests that could be associated with the various methods of germplasm exchange the decisions concerning the pest *Aceria guerreronis* is given below. The full datasheet of the pest could be found in Annex 2.3.1.

### PRA of *Aceria guerreronis*

	Seednuts	Embryo culture	Pollen
<b>Entry</b>	<i>A. guerreronis</i> occurs under the perianth of young nuts of <i>Cocos nucifera</i> L. On more mature fruits (10-13 months), coconut mites are rarely found and in small numbers (Hall and Espinoza 1981; Moore and Alexander 1987a). Coconut seedlings can be infested and it is theoretically possible for dispersal to occur by movement of seedlings but this has yet to be reported	Unlikely	Coconut mites can walk between touching inflorescences, and, being negatively geotactic, tend to move from older to younger inflorescences (Moore and Alexander 1987a). Some dispersal may take place by phoresy, either on animals/ insects directly attracted to the inflorescences (for example, pollinating insects such as bees; rodents which feed on the fruits).
<b>Establishment</b>	The coconut mite is found in tropical and subtropical climates		
<b>Spread</b>	The principal method by which coconut mites spread and colonize new palms, particularly over long distances, is almost certainly through aerial dispersal of inseminated female mites.		
<b>Economic</b>	Accurate crop loss assessments are rarely done, but estimates range from 7.5% (Julia and Mariau 1979) and 30% (Hernández 1977) to 60% (Griffith 1984) and some attacks may be so bad that farmers stop harvesting.		
<b>Level of risk</b>	Low/unlikely	Very low	Low
<b>Pest status*</b>	QP	QP?	QP
<b>Management</b>	Management of the pest on seednuts is by fumigation with methyl bromide. The pest in tissue cultures and in pollen is inspected using a binocular microscope of at least 30x magnification. Infested shipments should be refused entry and destroyed.		

**Note:** QP – quarantine pest ; NQP- Non-quarantine pest

At the conclusion of PRA Stage 2, the quarantine status for each pest for the exchange of seednuts, embryo culture and pollen should be recorded on each datasheet. If possible, the level of risk that the pest presents for each pathway is noted. In most cases, the level of risk is indicated by basic subjective ratings of High, Medium or Low.

A number of pests can be immediately eliminated from the analysis because of obvious characteristics that are not conducive to being in the pathway. These include larger mammals (vertebrates), nematodes and weeds.

### Vertebrate pests

Rats (*Rattus* sp) and the plantain squirrel (*Callsciurus notatus*) are too large to be in any pathway considered for germplasm exchange.

**Nematodes**

Nematodes can be serious pests of coconuts, but are root pests and would not be in the pathway and, therefore, are not considered further in this analysis. If nuts were harvested from the ground and could be contaminated with soil nematodes, then they could be in the pathway.

**Weeds**

A large number of weed species are recorded in association with the cultivation of coconuts as an economic crop. Many are economically significant. However, none would be considered in the pathway, as it would be expected that only seednuts from the tree would be used for germplasm exchange and they would be cleaned of any material prior to partial de-husking. Weeds would only be a problem if nuts were harvested from the ground and could be contaminated with soil.

**Risk management options**

As with the FAO/IBPGR Guidelines, the management of the quarantine pests follows a systematic recommendation that can address the risk identified. As such, these are standard recommendations and there may be alternatives available that are more acceptable or the only ones possible for particular points of entry. Again, these should be considered as guidelines, not as rules or mandatory requirements that can be adopted at national level depending upon the facilities, equipment and personnel availability.

As a general rule, the measures against a pest should be taken before export. Imagine the risk that an import would pose if a whole consignment of germplasm were to arrive that is infested with a number of pests. Action would have to be immediate, and it is possible that the pests have already escaped before the consignment is received by the NPPO.

**Selection management options**

	<b>Output</b>	<b>Activity</b>
<b>Step 1</b>	Management measures for pests identified	Examine literature and NPPO treatment manuals for effective treatments
<b>Step 2</b>	Management options assessed as suitable for onshore application	NPPO determines if the facilities, equipment and staff in importing country can undertake measures
<b>Step 3</b>	Options that must be applied before export identified	Any measures that cannot be undertaken in importing country because of operational constraints are identified for application before export*
<b>Step 4</b>	Exporting country advised of need for certain treatments at export	Liase with the exporting country NPPO to determine if measures that cannot be undertaken at import can be done at export
<b>Step 5</b>	Agreement on export/import conditions	Method and content of import permits agreed between NPPOs
<b>Step 6</b>	Post-entry quarantine conditions set	If pests cannot be identified or treated at point of export or import, then a period in Post Entry Quarantine (PEQ) will be required for the conduct of specific pest tests or observations

*\*It is preferable for all treatments, other than PEQ, to be done at exporting country*

## Arthropod pests

### **Seednuts**

The accepted method of managing arthropod pests has been fumigation with an appropriate broad-spectrum chemical (Frison *et al.* 1993). Currently, the practice is to remove part of the husk of the coconut, thereby removing some of the pests, and to fumigate with methyl bromide (MeBr) at the rate of 32g per cubic metre for three hours at 21°C. This treatment will effectively deal with all arthropod pests such as leaf feeders that have casually moved to the coconut fruit, as well as pests that reside on the surface such as scales, thrips, bugs and mites.

Methyl bromide is known to have some phototoxic effect on coconuts and care should be exercised in undertaking the treatment. The treatment at 32g per cubic metre for 24 hours at 20°C is used for devitalisation treatment into Australia (treatment A7.b. in FAO 1984). Temperatures for the treatment should not be high. Water should be placed in the chamber in trays before the fumigation begins to increase humidity. The dehusked nuts should be removed from the chamber as soon as the treatment is completed and placed in a cool, ventilated area to allow the fumigant to disperse from around the coir.

If methyl bromide (MeBr) is not available as a fumigant, then aluminium phosphide could be used as an alternative at the rate of 225 ppm of phosphine gas for 120 hrs at 20°C (treatment B4h.(5)(e) in FAO 1984), or 2-3 tablets per cubic metre for 24-72 hours (treatment C13 (30) in FAO 1984).

### **Embryo cultures**

Arthropod pests are not considered to be in the pathway when germplasm movement, in the form of tissue cultures, is correctly undertaken. However, there have been instances where small mite pests have contaminated cultures, so all tissue cultures should be carefully inspected for these pests on arrival by examination under a binocular microscope (30x magnification), or an illuminated magnifier lamp.

### **Pollen**

Established methods for collecting pollen have been described (Balingasa and Santos 1978; Frison *et al.* 1993). These methods, if carefully applied, would prevent pollen contamination from neighbouring palms and also prevent contamination by airborne pests. In particular, there are mite pests found in flowers that may also be in pollen.

Treatment of pollen is not possible, other than sieving out the larger contaminating pests; so all consignments should be carefully visually inspected using a low power microscope, before despatch and again at point of entry.

## Diseases

### **Seednuts**

A number of fungal diseases have been recorded on seednuts and flower clusters and therefore have the potential to be in the pathway. Nevertheless, whether all of these are seed borne has not been determined, although the risk exists. Invoking the precautionary principle, it is recommended that where these diseases are identified by the PRA as of concern, the nuts should be grown in post-entry quarantine (PEQ). Where a disease is generally known to occur in an area, only healthy nuts should be selected for exchange. Where diseases are not widespread and do not occur in specific and defined areas, then nuts should be sourced from these pest-free areas.

Seednuts should be treated with an acceptable and registered fungicide before sowing in PEQ.

**Embryo cultures**

Embryo cultures free of contamination would not present a pathway for the introduction of fungal diseases. Samples should be examined under a binocular microscope for contamination.

**Pollen**

Pollen should be visually inspected after gathering for fungal spores, and also again at point of entry. Pollen found infected should be destroyed.

**Viruses, viroids, mollicutes and phytoplasmas**

These systemic diseases have to be managed either through material sourcing from pest-free areas, or by active testing where the diseases occur generally and are not controlled. The causal organisms for some of these diseases have not been determined and hence, the precautionary principle is invoked to ensure that risk of incursions with exchange is negligible.

As a general principle for these diseases, material should only be collected from trees showing no symptoms. Although this in no way guarantees freedom from these diseases, it does reduce the possibility of the disease being in the pathway.

**Seednuts**

Seednuts should never be moved directly from areas where non-cultivable mollicutes or *Phytomonas* occur, to areas not affected with these pathogens (Frison *et al.* 1993). This is recommended despite the fact that there is no firm evidence that any of these systemic diseases are transmitted through seed.

The research on cadang-cadang in controlled non-infected areas has not been completed so material from the infected area should not be exchanged, or if really necessary, only made from trees indexed free of the viroid.

**Embryo cultures**

The presence of some systemic diseases has been detected in the embryo of coconuts. Therefore, material must only be taken from plants that are known to be free of these diseases. The material taken as tissue must be indexed before release for growing in a propagation nursery.

**Pollen**

Cadang-cadang has been detected in pollen, and there are reports that 4% of F<sub>1</sub> progenies exhibiting infection when healthy mother palms were pollinated with fresh pollen from infected palms (Manalo *et al.* 2000). Therefore, pollen should only be sourced from palms tested negative for cadang-cadang or from areas free of the pest.

**Regional and country transfer and exchange, and identification of pests**

In an extensive review of the phytosanitary risk involved in the exchange of germplasm among COGENT's Asia-Pacific member countries, a set of recommendations were devised based on pest risk analysis (Ikin 1999, unpublished). The recommendations from that report are included in this section. The identification of the pests is based on a PRA of the pests in the pathway and consideration of the country of export and import. In some cases, the pest status of countries is considered as the same and is dealt with in accordance with the principle of non-discrimination. As a general rule, if information is lacking on the association of a

pest with a pathway, it has been classified as a quarantine pest with the precautionary principle applied.

The specific phytosanitary measures for the following movement of coconut germplasm are presented below:

- Movement among all the ICG Centres
- PRA of the ICG-Southeast and East Asia – from India Centre
- PRA of the ICG-Southeast and East Asia – to India Centre
- PRA of the ICG-Southeast and East Asia – from Indonesia Centre
- PRA of the ICG-Southeast and East Asia – to Indonesia Centre
- PRA of the Pacific Region – from PNG
- PRA of the Pacific Region – to PNG
- PRA of the Africa Region – from Côte d'Ivoire
- PRA of the Africa Region – to Côte d'Ivoire
- PRA of the Americas Region – from Brazil
- PRA of the Americas Region – to Brazil

The information provided are in considerable detail, with the particular quarantine pests identified for which management action is required. The purpose of this listing is to provide technical justification for the phytosanitary measures. Note that many of the treatment actions deal with pests as groups and not as individuals. However, in the case of inspection, the particular pests are those that should be sought during the examination, and treatment or rejection should be based on their detection. The detection of non-quarantine (non-regulated) pests does not warrant any action. These pests are usually already present in the importing country, or are not of economic significance.

#### Specific phytosanitary measures for the movement of coconut germplasm between and among the ICG host countries (India, Indonesia, Papua New Guinea, Côte d'Ivoire and Brazil)

From\To	India	Indonesia	PNG	Côte d'Ivoire	Brazil
India		*	*	*	*
Indonesia	*		*	*	*
PNG	*	*		*	*
Côte d'Ivoire	*	*	*		*
Brazil	*	*	*	*	

#### India to Indonesia

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Coccus hesperidum</i> (brown soft scale)</li> <li>• <i>Nipaecoccus nipae</i> (spiked mealybug)</li> <li>• <i>Oligonychus biharensis</i></li> <li>• <i>Raoiella indica</i></li> <li>• <i>Tetranychus ludeni</i> (red spider mite)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Hypocrea rufa</i> (fruit rot: <i>Citrus</i> spp.)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<ul style="list-style-type: none"> <li>• Kerala wilt (root wilt)</li> </ul>	Area freedom*	Area freedom	Area freedom

**Note:** \*Pest-free Area

#### India to Papua New Guinea

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Coccus hesperidum</i> (brown soft scale)</li> <li>• <i>Nipaecoccus nipae</i> (spiked mealybug)</li> <li>• <i>Oligonychus biharensis</i></li> <li>• <i>Raoiella indica</i></li> </ul>	Fumigation	Not applicable	Inspection
<u>Mites</u> <ul style="list-style-type: none"> <li>• <i>Tetranychus cinnabarinus</i> (carmine spider mite)</li> <li>• <i>Tetranychus ludeni</i> (red spider mite)</li> </ul>			
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Hypocrea rufa</i> (fruit rot: <i>Citrus</i> spp.)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Kerala wilt (root wilt)</li> </ul>	Area freedom	Area freedom	Area freedom

#### India to Côte d'Ivoire

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Coccus hesperidum</i> (brown soft scale)</li> <li>• <i>Heliorthrips haemorrhoidalis</i> (black tea thrips)</li> <li>• <i>Icerya seychellarum</i> (Seychelles scale)</li> <li>• <i>Nipaecoccus nipae</i> (spiked mealybug)</li> <li>• <i>Oligonychus biharensis</i></li> <li>• <i>Raoiella indica</i></li> <li>• <i>Tetranychus cinnabarinus</i> (carmine spider mite)</li> <li>• <i>Tetranychus ludeni</i> (red spider mite)</li> <li>• <i>Brevipalpus phoenicis</i> (false spider mite)</li> <li>• <i>Chrysomphalus aonidum</i> (black scale)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Botryodiplodia theobromae</i> (diplodia pod rot of cocoa)</li> <li>• <i>Ceratocystis paradoxa</i> (dry basal rot of coconut)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> <li>• <i>Hypocrea rufa</i> (fruit rot: <i>Citrus</i> spp.)</li> <li>• <i>Phytophthora palmivora</i> (coconut budrot)</li> </ul>			
<ul style="list-style-type: none"> <li>• Kerala wilt (root wilt)</li> </ul>	Area freedom	Area freedom	Area freedom



**India to Brazil**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Coccus hesperidum</i> (brown soft scale)</li> <li>• <i>Ferrisia virgata</i> (striped mealybug)</li> <li>• <i>Icerya seychellarum</i> (Seychelles scale)</li> <li>• <i>Leptoglossus gonagra</i> (squash bug)</li> <li>• <i>Oligonychus biharensis</i></li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Botryodiplodia theobromae</i> (diplodia pod rot of cocoa)</li> <li>• <i>Hypocrea rufa</i> (fruit rot: Citrus spp.)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> <li>• <i>Phytophthora palmivora</i> (coconut budrot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Kerala wilt (root wilt)</li> </ul>	Area freedom	Area freedom	Area freedom

**Indonesia to India**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Hidari irava</i> (coconut skipper)</li> <li>• <i>Mahasena corbetti</i> (coconut case caterpillar)</li> <li>• <i>Rhabdoscelus obscurus</i> (New Guinea sugarcane weevil)</li> <li>• <i>Unaspis citri</i> (citrus snow scale)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Disease</u> <ul style="list-style-type: none"> <li>• Natuna wilt</li> </ul>	Area freedom	Area freedom	Area freedom

**Indonesia to PNG**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Hidari irava</i> (coconut skipper)</li> <li>• <i>Icerya pulchra</i></li> <li>• <i>Tetranychus cinnabarinus</i> (carmine spider mite)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Disease</u> <ul style="list-style-type: none"> <li>• Natuna wilt</li> </ul>	Area freedom	Area freedom	Area freedom

## Indonesia to Côte d'Ivoire

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Tetranychus cinnabarinus</i> (carmine spider mite)</li> <li>• <i>Heliethrips haemorrhoidalis</i> (black tea thrips)</li> <li>• <i>Hidari irava</i> (coconut skipper)</li> <li>• <i>Icerya pulchra</i></li> <li>• <i>Icerya seychellarum</i> (Seychelles scale)</li> <li>• <i>Mahasena corbetti</i> (coconut case caterpillar)</li> <li>• <i>Rhabdoscelus obscurus</i> (New Guinea sugarcane weevil)</li> <li>• <i>Tetranychus cinnabarinus</i> (carmine spider mite)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Botryodiplodia theobromae</i> (diplodia pod rot of cocoa)</li> <li>• <i>Ceratocystis paradoxa</i> (dry basal rot of coconut)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> <li>• <i>Phytophthora palmivora</i> (coconut budrot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Natuna wilt</li> </ul>	Area freedom	Area freedom	Area freedom

## Indonesia to Brazil

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Ferrisia virgata</i> (striped mealybug)</li> <li>• <i>Icerya pulchra</i></li> <li>• <i>Icerya seychellarum</i> (Seychelles scale)</li> <li>• <i>Leptoglossus gonagra</i> (squash bug)</li> <li>• <i>Mahasena corbetti</i> (coconut case caterpillar)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Botryodiplodia theobromae</i> (diplodia pod rot of cocoa)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Natuna wilt</li> </ul>	Area freedom	Area freedom	Area freedom

**Papua New Guinea to India**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Amblyopelta cocophaga</i> (coconut bug)</li> <li>• <i>Amblyopelta theobromae</i> (coconut bug)</li> <li>• <i>Axiagastus cambelli</i></li> <li>• <i>Mahasena corbetti</i> (coconut case caterpillar)</li> <li>• <i>Rhabdoscelus obscurus</i> (New Guinea sugarcane weevil)</li> <li>• <i>Unaspis citri</i> (citrus snow scale)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Phytophthora katsurae</i> (chestnut downy mildew)</li> </ul>	Fungicide & PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Finschhafen disease</li> </ul>	Area freedom	Area freedom	Area freedom

**Papua New Guinea to Indonesia**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Amblyopelta cocophaga</i> (coconut bug)</li> <li>• <i>Amblyopelta theobromae</i> (coconut bug)</li> <li>• <i>Axiagastus cambelli</i></li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Phytophthora katsurae</i> (chestnut downy mildew)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Finschhafen disease</li> </ul>	Area freedom	Area freedom	Area freedom

**Papua New Guinea to Côte d'Ivoire**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Heliophris haemorrhoidalis</i> (black tea thrips)</li> <li>• <i>Amblyopelta cocophaga</i> (coconut bug)</li> <li>• <i>Amblyopelta theobromae</i> (coconut bug)</li> <li>• <i>Axiagastus cambelli</i></li> <li>• <i>Icerya seychellarum</i> (Seychelles scale)</li> <li>• <i>Mahasena corbetti</i> (coconut case caterpillar)</li> <li>• <i>Rhabdoscelus obscurus</i> (New Guinea sugarcane weevil)</li> <li>• <i>Chrysomphalus aonidum</i> (black scale)</li> </ul>	Fumigation	Not applicable	Inspection

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Botryodiplodia theobromae</i> (diplodia pod rot of cocoa)</li> <li>• <i>Ceratocystis paradoxa</i> (dry basal rot of coconut)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> <li>• <i>Phytophthora katsurae</i> (chestnut downy mildew)</li> <li>• <i>Phytophthora palmivora</i> (coconut budrot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Finschhafen disease</li> </ul>	Area freedom	Area freedom	Area freedom

### Papua New Guinea to Brazil

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Amblypelta cocophaga</i> (coconut bug)</li> <li>• <i>Amblypelta theobromae</i> (coconut bug)</li> <li>• <i>Axiagastus cambelli</i></li> <li>• <i>Ferrisia virgata</i> (striped mealybug)</li> <li>• <i>Icerya seychellarum</i> (Seychelles scale)</li> <li>• <i>Leptoglossus gonagra</i> (squash bug)</li> <li>• <i>Mahasena corbetti</i> (coconut case caterpillar)</li> <li>• <i>Rhabdoscelus obscurus</i> (New Guinea sugarcane weevil)</li> <li>• <i>Unaspis citri</i> (citrus snow scale)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Botryodiplodia theobromae</i> (diplodia pod rot of cocoa)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> <li>• <i>Phytophthora katsurae</i> (chestnut downy mildew)</li> </ul>	Fungicide & PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Finschhafen disease</li> </ul>	Area freedom	Area freedom	Area freedom

### Côte d'Ivoire to India

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Pinnaspis strachani</i> (Hibiscus snow scale)</li> <li>• <i>Pseudococcus longispinus</i> (long-tailed mealybug)</li> <li>• <i>Unaspis citri</i> (citrus snow scale)</li> </ul>	Fumigation	Not applicable	Inspection

**Côte d'Ivoire to Indonesia and Papua New Guinea**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Aceria guerreronis</i> (coconut mite)</li> <li>• <i>Pinnaspis strachani</i> (Hibiscus snow scale)</li> <li>• <i>Pseudococcus longispinus</i> (longtailed mealybug)</li> </ul>	Fumigation	Not applicable	Inspection

**Côte d'Ivoire to Brazil**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Leptoglossus gonagra</i> (squash bug)</li> <li>• <i>Pseudococcus longispinus</i> (long-tailed mealybug)</li> <li>• <i>Unaspis citri</i> (citrus snow scale)</li> </ul>	Fumigation	Not applicable	Inspection

**Brazil to India**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Pinnaspis strachani</i> (Hibiscus snow scale)</li> </ul>	Fumigation	Not applicable	Inspection

**Brazil to Indonesia**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Aceria guerreronis</i> (coconut mite)</li> <li>• <i>Pinnaspis strachani</i> (Hibiscus snow scale)</li> </ul>	Fumigation	Not applicable	Inspection

**Brazil to Papua New Guinea**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Aceria guerreronis</i> (coconut mite)</li> <li>• <i>Nipaecoccus nipae</i> (spiked mealybug)</li> <li>• <i>Pinnaspis strachani</i> (Hibiscus snow scale)</li> <li>• <i>Brevipalpus phoenicis</i> (false spider mite)</li> </ul>	Fumigation	Not applicable	Inspection

**Brazil to Côte d'Ivoire**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Heliothrips haemorrhoidalis</i> (black tea thrips)</li> <li>• <i>Nipaecoccus nipae</i> (spiked mealybug)</li> <li>• <i>Brevipalpus phoenicis</i> (false spider mite)</li> <li>• <i>Chrysomphalus aonidum</i> (black scale)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Phytophthora palmivora</i> (coconut budrot)</li> <li>• <i>Ceratocystis paradoxa</i> (dry basal rot of coconut)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable

**PRA of the ICG for South Asia****Movement from the India Centre****India to Bangladesh**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Aspidiotus destructor</i></li> <li>• <i>Heliothrips haemorrhoidalis</i> (black tea thrips)</li> <li>• <i>Icerya seychellarum</i> (Seychelles scale)</li> <li>• <i>Leptoglossus gonagra</i> (squash bug)</li> <li>• <i>Oligonychus biharensis</i></li> <li>• <i>Raoiella indica</i></li> <li>• <i>Tetranychus cinnabarinus</i> (carmine spider mite)</li> <li>• <i>Tetranychus ludeni</i> (red spider mite)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Hypocrea rufa</i> (fruit rot: <i>Citrus</i> spp.)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> <li>• <i>Phytophthora palmivora</i> (coconut budrot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Kerala wilt (root wilt)</li> </ul>	Area freedom	Area freedom	Area freedom

**India to Pakistan**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Coccus hesperidum</i> (brown soft scale)</li> <li>• <i>Heliothrips haemorrhoidalis</i> (black tea thrips)</li> <li>• <i>Oligonychus biharensis</i></li> <li>• <i>Pseudococcus longispinus</i> (long-tailed mealybug)</li> <li>• <i>Tetranychus cinnabarinus</i> (carmine spider mite)</li> <li>• <i>Tetranychus ludeni</i> (red spider mite)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Hypocrea rufa</i> (fruit rot: <i>Citrus</i> spp.)</li> <li>• <i>Phytophthora palmivora</i> (coconut budrot)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Kerala wilt (root wilt)</li> </ul>	Area freedom	Area freedom	Area freedom

**India to Sri Lanka**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Coccus hesperidum</i> (brown soft scale)</li> <li>• <i>Oligonychus biharensis</i></li> <li>• <i>Pseudococcus longispinus</i> (long-tailed mealybug)</li> <li>• <i>Tetranychus ludeni</i> (red spider mite)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Hypocrea rufa</i> (fruit rot: <i>Citrus</i> spp.)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Kerala wilt (root wilt)</li> </ul>	Area freedom	Area freedom	Area freedom

**Movement to the India Centre**

All pests of quarantine concern that occur in Bangladesh, Pakistan and Sri Lanka also occur in India, therefore there are no specific phytosanitary measures that are required. Nevertheless, it is recommended that material being transferred be examined by the NPPOs of both importing and exporting countries for any sign of new pests.

## PRA of the ICG for Southeast and East Asia

### Movement from Indonesia Centre

#### Indonesia to China, Myanmar, Thailand, Vietnam and the Philippines

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Dysmicoccus brevipes</i> (pineapple mealybug) (excluding Vietnam and the Philippines)</li> <li>• <i>Hidari irava</i> (coconut skipper) – (excluding Thailand)</li> <li>• <i>Icerya pulchra</i></li> <li>• <i>Leptoglossus gonagra</i> (squash bug) - (Philippines and Vietnam only)</li> <li>• <i>Rhabdoscelus obscurus</i> (New Guinea sugarcane weevil)</li> <li>• <i>Tetranychus cinnabarinus</i> (carmine spider mite) - (excluding China and Thailand)</li> <li>• <i>Unaspis citri</i> (citrus snow scale) - (excluding China and Vietnam)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Botryodiplodia theobromae</i> (diplodia pod rot of cocoa) (excluding China and the Philippines)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> <li>• <i>Phytophthora palmivora</i> (coconut budrot) (only China and Vietnam)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Natuna wilt</li> </ul>	Area freedom	Area freedom	Area freedom

#### Indonesia to Malaysia

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropod</u> <ul style="list-style-type: none"> <li>• <i>Rhabdoscelus obscurus</i> (New Guinea sugarcane weevil)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Disease</u> <ul style="list-style-type: none"> <li>• Natuna wilt</li> </ul>	Area freedom	Area freedom	Area freedom



**Movement to the Indonesia Centre****From China**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropod</u> <ul style="list-style-type: none"> <li><i>Nipaecoccus nipae</i> (spiked mealybug)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Disease</u> <ul style="list-style-type: none"> <li><i>Hypocrea rufa</i> (fruit rot: <i>Citrus</i> spp.)</li> </ul>	Area freedom	Area freedom	Area freedom

**From Malaysia**

Quarantine Pest	Management Options		
	Nuts	Embryo	Pollen
<u>Disease</u> <ul style="list-style-type: none"> <li>Malaysia wilt</li> </ul>	Area freedom	Area freedom	Area freedom

**From Thailand**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li><i>Nipaecoccus nipae</i> (spiked mealybug)</li> <li><i>Oligonychus biharensis</i></li> </ul>	Fumigation	Not applicable	Not applicable

**From Vietnam**

Quarantine Pest	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropod</u> <ul style="list-style-type: none"> <li><i>Nipaecoccus nipae</i> (spiked mealybug)</li> </ul>	Fumigation	Not applicable	Not applicable

**From Myanmar - no quarantine pests**

## PRA of the ICG- South Pacific

### Movement from PNG Centre

#### PNG to Cooke Islands, Kiribati, Samoa, Tonga and Vanuatu

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Amblypelta cocophaga</i> (coconut bug)</li> <li>• <i>Amblypelta theobromae</i> (coconut bug)</li> <li>• <i>Aspidiotus destructor</i> (coconut scale) – (excluding Vanuatu and Samoa)</li> <li>• <i>Axiagastus cambelli</i></li> <li>• <i>Heliethrips haemorrhoidalis</i> (black tea thrips) – (Samoa only)</li> <li>• <i>Leptoglossus gonagra</i> (squash bug)</li> <li>• <i>Mahasena corbetti</i> (coconut case caterpillar)</li> <li>• <i>Rhabdoscelus obscurus</i> (New Guinea sugarcane weevil) – (PNG only)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Botryodiplodia theobromae</i> (diplodia pod rot of cocoa)</li> <li>• <i>Ceratocystis paradoxa</i> (dry basal rot of coconut) – (excluding Vanuatu)</li> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> <li>• <i>Phytophthora katsurae</i> (chestnut downy mildew)</li> <li>• <i>Phytophthora palmivora</i> (coconut budrot) – (excluding Samoa, Tonga and Vanuatu)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Finschhafen disease</li> </ul>	Area freedom	Area freedom	Area freedom

#### PNG to Fiji

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Amblypelta theobromae</i> (coconut bug)</li> <li>• <i>Axiagastus cambelli</i></li> <li>• <i>Mahasena corbetti</i> (coconut case caterpillar)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Ceratocystis paradoxa</i> (dry basal rot of coconut)</li> <li>• <i>Phytophthora katsurae</i> (chestnut downy mildew)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>• Finschhafen disease</li> </ul>	Area freedom	Area freedom	Area freedom

**PNG to Solomon Islands**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li><i>Amblypelta theobromae</i> (coconut bug)</li> <li><i>Heliothrips haemorrhoidalis</i> (black tea thrips)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Diseases</u> <ul style="list-style-type: none"> <li><i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> <li><i>Phytophthora katsurae</i> (chestnut downy mildew)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>Finschhafen disease</li> </ul>	Area freedom	Area freedom	Area freedom

**Movement to the PNG Centre****From Solomon Islands to PNG**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li><i>Coccus hesperidum</i> (brown soft scale)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Disease</u> <ul style="list-style-type: none"> <li>Coconut cadang-cadang viroid</li> </ul>	Area freedom	Area freedom	Area freedom

**From Tonga to PNG**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li><i>Coccus hesperidum</i> (brown soft scale)</li> <li><i>Dysmicoccus cocotis</i></li> <li><i>Ischnaspis longirostris</i> (black thread scale)</li> <li><i>Pinnaspis strachani</i> (Hibiscus snow scale)</li> </ul>	Fumigation	Not applicable	Not applicable

**From Vanuatu to PNG**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li><i>Myndus taffini</i></li> <li><i>Pinnaspis strachani</i> (Hibiscus snow scale)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Disease</u> <ul style="list-style-type: none"> <li>Coconut foliar decay nanavirus</li> </ul>	Area freedom	Area freedom	Area freedom

**From Fiji to PNG**

Quarantine Pest	Management options		
	Nuts	Embryo	Pollen
<u>Arthropod</u> <ul style="list-style-type: none"> <li><i>Pinnaspis strachani</i> (Hibiscus snow scale)</li> </ul>	Fumigation	Not applicable	Not applicable

**From Samoa to PNG - no quarantine pests****PRA of the ICG- Africa and the Indian Ocean<sup>4</sup>****Movement from the Côte d'Ivoire Centre****Côte d'Ivoire to Ghana, Kenya, Madagascar, Nigeria, Seychelles and Tanzania**

Quarantine Pest	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropod</u> <ul style="list-style-type: none"> <li><i>Unaspis citri</i> (citrus snow scale)</li> </ul>	Fumigation	Not applicable	Not applicable

**Côte d'Ivoire to Benin**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li><i>Ferrisia virgata</i> (striped mealybug)</li> <li><i>Pseudococcus longispinus</i> (long-tailed mealybug)</li> </ul>	Fumigation	Not applicable	Not applicable

**Côte d'Ivoire to Mozambique**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li><i>Dysmicoccus brevipes</i> (pineapple mealybug)</li> <li><i>Ferrisia virgata</i> (striped mealybug)</li> <li><i>Leptoglossus gonagra</i> (squash bug)</li> <li><i>Pinnaspis strachani</i> (Hibiscus snow scale)</li> <li><i>Pseudococcus longispinus</i> (long-tailed mealybug)</li> <li><i>Unaspis citri</i> (citrus snow scale)</li> </ul>	Fumigation	Not applicable	Not applicable

<sup>4</sup> The information for the pest risk analysis of ICG-AIO was obtained from the IPGRI consultancy report of Dr Hubert de Franqueville of CIRAD

**Movement to the Cote d'Ivoire Centre****Ghana to Côte d'Ivoire**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li><i>Heliothrips haemorrhoidalis</i> (black tea thrips)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Diseases</u> <ul style="list-style-type: none"> <li><i>Phytophthora palmivora</i> (coconut budrot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>Lethal yellowing</li> </ul>	Area freedom	Area freedom	Area freedom

**Kenya, Seychelles and Tanzania to Côte d'Ivoire**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li><i>Heliothrips haemorrhoidalis</i> (black tea thrips)</li> <li><i>Icerya seychellarum</i> (Seychelles scale)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Diseases</u> <ul style="list-style-type: none"> <li><i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> <li><i>Phytophthora palmivora</i> (coconut budrot) – only in Tanzania and Seychelles</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable

**Nigeria to Côte d'Ivoire**

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Diseases</u> <ul style="list-style-type: none"> <li><i>Ceratocystis paradoxa</i> (dry basal rot of coconut)</li> <li><i>Phytophthora palmivora</i> (coconut budrot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable
<ul style="list-style-type: none"> <li>Lethal yellowing</li> </ul>	Area freedom	Area freedom	Area freedom

**Benin to Côte d'Ivoire**

Quarantine Pest	Management Options		
	Nuts	Embryo	Pollen
<u>Disease</u> <ul style="list-style-type: none"> <li>Lethal yellowing</li> </ul>	Area freedom	Area freedom	Area freedom

## PRA of the proposed ICG for Latin America and the Caribbean<sup>5</sup>

### Movement from the Brazil Centre

#### Brazil to Colombia, Costa Rica, Guyana, Cuba and Mexico

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Aceria guerreronis</i> (coconut mite)</li> <li>• <i>Brevipalpus phoenicis</i> (false spider mite)</li> <li>• <i>Heliethrips haemorrhoidalis</i> (black tea thrips) – only in Costa Rica and Guyana</li> <li>• <i>Nipaecoccus nipae</i> (spiked mealybug)- only in Guyana and Cuba</li> </ul>	Fumigation	Not applicable	Inspection

#### Brazil to Haiti and Honduras

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Aceria guerreronis</i> (coconut mite)</li> <li>• <i>Heliethrips haemorrhoidalis</i> (black tea thrips)</li> <li>• <i>Nipaecoccus nipae</i> (spiked mealybug)</li> <li>• <i>Brevipalpus phoenicis</i> (false spider mite)</li> <li>• <i>Chrysomphalus dictyospermi</i> (Spanish red scale)</li> </ul>	Fumigation	Not applicable	Inspection
<u>Disease</u> <ul style="list-style-type: none"> <li>• <i>Phytophthora palmivora</i> (coconut budrot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable

#### Brazil to Jamaica and Trinidad - no identified quarantine pests

### Movement to the Brazil Centre

#### Cuba, Jamaica, Haiti, Mexico, Honduras and Trinidad (countries with lethal yellowing disease) to Brazil

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Ferrisia virgata</i> (striped mealybug)</li> <li>• <i>Ischnaspis longirostris</i> (black thread scale)</li> <li>• <i>Leptoglossus gonagra</i> (squash bug)</li> <li>• <i>Pseudococcus longispinus</i> (long-tailed mealybug)</li> <li>• <i>Unaspis citri</i> (citrus snow scale)</li> <li>• <i>Vinsonia stellifera</i> (star scale)</li> </ul>	Fumigation	Not applicable	Not applicable

<sup>5</sup> The information for the pest risk analysis of the proposed ICG-LAC was obtained from the IPGRI consultancy report of Dr Hubert de Franqueville of CIRAD

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Diseases</u> <ul style="list-style-type: none"> <li>• <i>Myndus crudus</i> (as a vector)</li> <li>• Lethal yellowing</li> </ul>	Area freedom	Area freedom	Area freedom

### Colombia to Brazil

Quarantine pest	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Unaspis citri</i> (citrus snow scale)</li> <li>• <i>Ferrisia virgata</i> (striped mealybug)</li> <li>• <i>Pseudococcus longispinus</i> (long-tailed mealybug)</li> </ul>	Fumigation	Not applicable	Not applicable
<u>Disease</u> <ul style="list-style-type: none"> <li>• <i>Marasmius (Marasmiellus) palmivorus</i> (oil palm bunch rot)</li> </ul>	Fungicide and PEQ	Not applicable	Not applicable

### Costa Rica to Brazil

Quarantine Pest	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropod</u> <ul style="list-style-type: none"> <li>• <i>Ferrisia virgata</i> (striped mealybug)</li> </ul>	Fumigation	Not applicable	Not applicable

### Guyana to Brazil

Quarantine Pests	Management Options		
	Nuts	Embryo	Pollen
<u>Arthropods</u> <ul style="list-style-type: none"> <li>• <i>Ferrisia virgata</i> (striped mealybug)</li> <li>• <i>Ischnaspis longirostris</i> (black thread scale)</li> <li>• <i>Leptoglossus gonagra</i> (squash bug)</li> <li>• <i>Pseudococcus longispinus</i> (long-tailed mealybug)</li> <li>• <i>Myndus crudus</i> (but not lethal yellowing disease)</li> </ul>	Fumigation	Not applicable	Not applicable

As can be deduced from the complex combination of recommendations, these can be simplified (on the basis that the management of pests is not always restricted to individual pests) to groups of pests with similar biology and in particular life stages, as shown below.

Pest type	Management option
Arthropods	Fumigation with methyl bromide
Fungal diseases	Fungicide treatment of nuts and post-entry quarantine screening
Systemic virus, viroid and MLOs	Indexing to ensure area of plant freedom
Pests of unknown cause	Visual certification of area freedom
Contaminating pests in pollen	Visual inspection, particularly of mites

### Conclusion

PRA is a subjective process at the moment. The lack of information on many pests, especially those found only in developing countries, is a constraint to making any reliable judgement on their biology and behaviour. In the case of absence of information, it has been assumed that the pest is of minor importance and therefore is not of economic significance. This approach may not be acceptable to those who wish to apply a "precautionary principle" to all decision-making, but this is the system that was previously adopted by the FAO/IBPGR Guidelines. Therefore, many pests that may emerge as quarantine pests in the future could have been omitted from this analysis. Nevertheless, this work is undertaken as benchmarks of the application of PRA on the clear understanding that as more information becomes available on the biology of pests so will the conclusions of the analysis need to be modified.

Fortunately, the management options that have been identified to address pest risk are broad spectrum and generally applicable to pest groups rather than individual pests. Only when new quarantine pests that substantially differ in their biology from those already considered will the level of phytosanitary risk increase.

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## CHAPTER 3

# Operational Management of Germplasm Movement

- Importing Seednuts
- Importing and Growing Embryos for the Coconut Genebank
- Preparing Pollen for Export



## Importing seednuts

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### **General phytosanitary measures for the movement of coconut germplasm**

Along with the compliance with countries' phytosanitary requirements, the collecting and exchange of germplasm should be undertaken with the full participation of the stakeholders, which could be the collectors, breeders, other scientists and farmers. In the case of exchange between national and regional centres, it can be assumed that formal approval is sought for the movement at a bilateral level. Nevertheless, with the possibility of the movement from national sources outside the collections into other centres, compliance with good collecting practices should be reiterated, particularly if a standard procedure is being developed and adopted worldwide, such as the International Code of Conduct for Plant Germplasm Collecting and Transfer.

This Code "aims to promote the rational collection and sustainable use of genetic resources, to prevent genetic erosion, and to protect the interests of both donors and collectors of germplasm". The Code, a voluntary one, has been developed by FAO and negotiated by its member nations through the Organization's Commission on Plant Genetic Resources. It is based on the principle of national sovereignty over plant genetic resources and sets out standards and principles to be observed by those countries and institutions that adhere to it.

The Code proposes procedures to request and/or to issue licences for collecting missions, provides guidelines for collectors themselves, and extends responsibilities and obligations to the sponsors of missions, the curators of genebanks, and the users of genetic material (FAO 1993).

The Code outlines the arrangements that should be made prior to germplasm movement/ exchange. In particular, import permits should be requested and obtained clearly indicating the phytosanitary conditions that must be met prior to the material being exported. With the increasing reliance on the concept of 'area freedom' (i.e., pest-free area) and the indexing of source plants these requirements must be fulfilled, otherwise, the material will most likely be destroyed on arrival at destination.

Specifically, the Code requires collectors or curators of collections to – "(c) make arrangements with quarantine officials, seed storage managers and curators to ensure that the samples are transferred as quickly as possible to conditions which optimise their viability; and (d) obtain, in accordance with the importing countries' requirements, the phytosanitary certificate(s) and other documentation needed for transferring the material collected."

### **Importing coconut germplasm as seednuts**

The following are the steps recommended in importing coconut germplasm as seednuts:

<b>Step 1</b>	Determine import conditions with reference to guidelines and pest risk analysis	Check pest status of exporting country and modify PRA as required
<b>Step 2</b>	Issue import permit to importing authority	Two copies to importer; one copy to Inspectorate

<b>Step 3</b>	Prepare post entry quarantine area for growing of nuts	Ensure area is clean and supplied with necessary materials and staff
<b>Step 4</b>	Check compliance of import phytosanitary certification and conditions specified on permit	If non-compliant, reject shipment. In particular, check certification of area freedom and treatments undertaken.
<b>Step 5</b>	Inspect material for pests	Treat if pests are found
<b>Step 6</b>	Once import conditions are met, transfer material to post-entry-quarantine site	
<b>Step 7</b>	Complete post entry quarantine	Destroy any material with disease like symptoms

### ***Post-entry quarantine***

The purpose of post-entry quarantine (PEQ) is to grow plants for a specified period of time in some form of isolation in order for visual examination or specific tests to be conducted to detect pests (generally diseases). This procedure is used when symptoms of the disease are only visible under specific environmental conditions, or at particular times of the year, or where there is the possibility of symptomless infection requiring testing (referred to as disease indexing).

The primary responsibility of the PEQ system is to successfully establish plants so that appropriate tests can be conducted to verify the general health of the plant material. Its key activities are:

1. Preparation of a PEQ area where the plants will grow and secured from local pests nor from transmitting exotic pests; and
2. Preparation of growing media that will support the growth of the plants through at least one season while not being infested by local pests.

### **PEQ facilities**

Much has been written about the need for secure plant quarantine areas, and in some cases the complexity of the facilities recommended do not take into account the limited resources of developing countries and the particular maintenance needs of complex machinery such as air conditioners and other electrical equipment. The recommendations below are for basic facilities that mitigate the pest risk identified.

The basic containment facility recommended is a screenhouse that is constructed in a manner that could eliminate the possibility of the transfer of insect pests into and outside the facility. This involves screening the wall structure. This will aid the prevention of spread of insects and insect borne pathogens that are vectored by *Myndus* spp. Mites will not be excluded by this facility, but these arthropods are not known to be vectors of coconut pests.

If insect-transmitted pests and diseases are not of significance, an isolated field plot may be all that is necessary. The area should have benches on a concrete floor, that will ensure general cleanliness of the area, but it is possible that an isolated plot could suffice as long as there is no way in which local pests and diseases can infect the imports. A raised bed on a soil surface covered with strong polythene sheeting and with adequate drainage to the edges would be sufficient for growing seednuts in polythene bags.

For seednuts and transferred embryo cultures, these should be grown in pasteurised compost in polythene bags.

Procedures for the successful reception and propagation of seednuts have been dealt with in detail by Bourdeix (1999) and will not be repeated here.

**Propagation (growing) media**

Soil propagation media should:

- Be of a structure that permits air exchange and moisture retention, whilst having good drainage;
- Be free from harmful organisms and toxins;
- Provide adequate nutrition to the plants at all stages of the PEQ period;
- Consist of standardized local material ingredients to obtain uniform and reproducible growth; and
- Be pasteurised at 60°C for 30 minutes before use.

**Pest control**

It is important that local pests and diseases are controlled while the plants are being grown in PEQ. Specific recommendations will not be made here because the use of pest control chemicals is regulated by national legislation, which indicates the use of chemicals on particular crops and their rates of application. In the past, Whitehead (1968) has recommended the use of DDT, aldrin and dieldrin, but these are now considered unsafe chemicals for use in agriculture, although in some cases specific exemptions may be obtained for use in PEQ. But as a general rule, usage of such chemicals should follow what is permitted by the country's rules and regulations because of health and safety considerations.

If diseased plants are detected in PEQ, it is recommended that the affected plant be removed and destroyed as well as all adjacent plants (normally four around). The remainder of the consignment should then be sprayed with a broad spectrum fungicide/bactericide and kept under close observation for at least a week.

The time period for PEQ depends upon the growing conditions at the facility. The recommended period for plants that are in for visual observation of pests is a minimum of six months or at least one growing season (in the tropics, the duration of a wet season and at least three months of a dry season) before release. When tests are required, the material may be released only after the tests have been completed.

**Record keeping**

It is important that detailed records of the time period in PEQ be kept. In particular, this information provides the basis for the release of the germplasm and a corporate memory of the performance of germplasm, the facilities, procedures and the staff.

Records of each import should include the following:

- Date of import/ importer;
- Variety of germplasm;
- Number of imported plants or seeds;
- Treatment before import;
- Type of soil mix;
- Pesticide treatments, rate, type of chemical and date of application;
- Fertilizer applications;
- Symptoms on plants/pest responsible/action taken; and
- Release date and number of plants released to importer.

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## Importing and growing embryos for the coconut genebank

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### **Introduction**

Coconut is one of the plant species with the biggest seed with no dormancy (i.e., it germinates right after maturity). This characteristic presents a problem in coconut germplasm collecting, conservation and exchange. With embryo culture, it is now possible to send coconut embryos in culture tubes to various coconut-growing countries. Significant savings on freight costs is realized aside from avoiding inadvertent transfer of contaminating propagules.

However, there are some factors/considerations necessary for a successful *in vitro* culture of coconut embryos. Some of the more important ones are: the right age of the embryo, which should be about 10 to 11 months old (a good indication of this in the nut is the colour break (Fig 1a) of the husk from green to brown). Injury to the embryo during excision should be avoided. It was also observed that embryos from fertilized palms perform better *in vitro*. Moreover, shorter duration from collecting to actual inoculation in the laboratory is better. Of course, these are all in addition to the conditions in the laboratory as well as the expertise of the operator.

### **Extraction, packaging and dispatch of the embryos**

Ideally, 10- to 11-month old coconuts are used as younger or older nuts do not respond favourably to *in vitro* conditions. With the use of a pointed but blunt instrument mounted on a sturdy wooden base, dehusk the nuts by prying the husk off the nut. Split the nuts into halves by striking it with the blunt side of a machete. The nut has a prominent longitudinal vein that runs from the active eye to the stigmatic end. Striking this vein with sufficient force will easily split the nut crosswise. The embryo embedded in the solid endosperm is located under one of the three 'eyes' of the coconut. This active eye is usually depressed due to the non-lignification of the cells thereby making it soft and allowing for easier germination of the embryos.

With the use of a cork borer (No. 10 or bigger), extract the embryo which should still be intact in the solid endosperm of the split nut (Fig 1b). Push the endosperm cylinders out of the cork borer using a clean stick. Place the endosperm cylinders in a clean container (Fig 1c) using coconut water or plain water as medium.

After all of the endosperm cylinders are extracted, wash them in tap water and quickly rinse in 95% ethanol to remove the fats. Afterwards, disinfect with 100% commercial bleach (Zonrox™ which is 5.25% NaOCl) for 20 minutes. Wash the endosperm cylinders three or more times with sterile water to remove the bleach. This step is best done in a clean room with still air to minimize contamination.

For transporting, sterilized cylinders are then transferred in sealed sterile plastic bags with moist cotton inside to keep them damp (Rillo and Paloma 1991). To keep the embryo cylinders cold during local accompanied land transport, the embryos should be transported in a Styrofoam box with ice inside. However, for farther destinations or transport by plane, a fast courier service will be necessary. The embryos should reach the destination within 4-5 days, otherwise germination will be significantly affected. Since airlines do not allow ice on board, a Styrofoam box is the best choice for a container as the material will allow the temperature to be kept at a minimum. The box for dispatch should be kept refrigerated overnight and should be

stored in a cool place or kept away from direct sunlight. The box with the embryos should be consigned to a reliable courier without much delay with the necessary information regarding storage during transport and phytosanitary certificates from the originating and receiving countries.

Upon arrival in the point of destination, the embryo cylinders are again washed in 5% NaOCl for two minutes and then washed three or more times with sterile water inside the laminar flow cabinet. The embryos are then excised carefully, avoiding any injury, from the endosperm cylinders and collected in a clean beaker. Once all embryos have been extracted, these are finally sterilized in 10% commercial bleach or 1% NaOCl for one minute, washed three or more times in distilled water and then blotted dry on sterile filter paper. Good (plump and not deformed) embryos are then selected, which are then individually inoculated into the culture medium in tubes or vials.

An alternative is to have the embryos extracted, selected, sterilized and inoculated into the medium before transport. A disadvantage of this method is that the package is bigger and heavier as the embryos are already in individual vials (the screw type is best), although they could be inoculated in 3's or 5's depending on the size of the vial. The vials are further sealed with Parafilm or Nesco film. Another constraint in this method is that if some embryos get contaminated during transport, then more than one embryo is thrown away. If this alternative is to be followed, it is best to inoculate them individually, which will not necessitate immediate transfer after arrival.

### ***Growing the embryos in vitro***

The success of plant tissue culture as a means of plant propagation is greatly influenced by the nature of the culture medium used. Plant tissue culture media should provide not only the major (macro-) and (micro-) nutrients, but also a carbohydrates source, usually sucrose, to replace the carbon, which the plant normally fixes from the atmosphere by photosynthesis. Organic nitrogen compounds and undefined complex substances such as coconut milk, banana, tomatoes, etc are added depending on the crop to be cultured. A basal medium is generally referred to as one without any growth regulator. Modified media contain growth regulators at various concentrations, which are determined mainly by the requirements of the explants.

Dr CJ Eeuwens of Wye College, University of London, formulated a Y3 medium especially for coconut tissues (Eeuwens 1976). It was the third formulation that worked satisfactorily with coconut tissues hence, the number '3' affixed to the letter 'Y' to designate the medium. The formulation, without the hormones, was compared with White's and Murashige and Skoog's media (Rillo and Paloma 1990) to culture coconut embryos. Results showed that Y3 was significantly better in supporting growth and development of coconut embryos. With funds from COGENT, an optimised embryo culture protocol was developed in 2002, which is described below. Table 1 shows the components of the Y3 medium formulation for coconut embryo culture.

### **Preparation of one litre Y3 medium**

Weigh the needed salts one by one and dissolve in 250 ml distilled water. Alternatively stock solutions could be prepared. Weigh 60 gm table sugar and dissolve in the above solution. Using a volumetric flask, make up the volume to one liter using distilled water. Adjust the pH to 5.6 using 0.1-0.5M NaOH or 0.1-0.5M HCl. Add the activated charcoal (AC) and stir.

While stirring to distribute the AC, dispense 10 ml of the liquid medium into 25 x



150 mm test tubes (for initiation), 15-20 ml for semi-solid medium and 80-100 ml of the liquid medium for the last sub culture in bigger bottles. For semi-solid medium, add 7-8g agar. Cover appropriately depending on the type of vessels used. Autoclave the medium at 121°C at 15 psi for 15 minutes for the 10 ml, and 20 minutes for the rest. Cool completely before use. (Note: Use 60 g/l table sugar until the seedling has developed shoot and roots (1-4 months) then lower to 45 g/l thereafter. The medium is liquid during initiation, while the first and second subculture is solid and liquid thereafter.

**Table 1.** Components of the modified Y3 mineral formulation for coconut embryo culture

	Medium	Components	1x (mg/l)	1x (g/l)	10x (g/l)
Macro	Eeuwens (Y3)	NH <sub>4</sub> Cl	535.00	0.535	5.35
		KNO <sub>3</sub>	2020.00	2.020	20.20
		MgSO <sub>4</sub> .7H <sub>2</sub> O	247.00	0.247	2.47
		CaCl <sub>2</sub> .2H <sub>2</sub> O	294.00	0.294	2.94
		KCl	1492.00	1.492	14.92
		NaH <sub>2</sub> PO <sub>4</sub> .2H <sub>2</sub> O	312.00	0.312	3.12
Micro	Eeuwens (Y3)		<b>1x (mg/l)</b>	<b>1x (g/l)</b>	<b>100x (g/l)</b>
		KI	8.30	0.00830	0.830
		H <sub>3</sub> BO <sub>3</sub>	3.10	0.00310	0.310
		MnSO <sub>4</sub> .4H <sub>2</sub> O	11.20	0.01120	1.120
		ZnSO <sub>4</sub> .7H <sub>2</sub> O	7.20	0.00720	0.720
		CuSO <sub>4</sub> .5H <sub>2</sub> O	0.250	0.000250	0.0250
		CoCl <sub>2</sub> .6H <sub>2</sub> O	0.240	0.000240	0.0240
		NaMoO <sub>4</sub> .H <sub>2</sub> O	0.240	0.000240	0.0240
		NiCl <sub>2</sub> .6H <sub>2</sub> O	0.024	0.000024	0.0024
EDTA	UPLB		<b>1x (mg/l)</b>	<b>1x (g/l)</b>	<b>100x (g/l)</b>
		Fe <sub>2</sub> SO <sub>4</sub> .7H <sub>2</sub> O	41.70	0.04170	4.170
		Na <sub>2</sub> EDTA	55.80	0.05580	5.580
Vitamins	UPLB + ARC		<b>1x (mg/l)</b>	<b>1x (g/l)</b>	<b>100x (g/l)</b>
		Pyridoxine HCl	0.05	0.00005	0.005
		Thiamine HCl	0.05	0.00005	0.005

	Medium	Components	1x (mg/l)	1x (g/l)	10x (g/l)
		Nicotinic acid	0.05	0.00005	0.005
		Ca-D-pantothenate	0.05	0.00005	0.005
		Biotin	0.05	0.00005	0.005
		Folic acid	0.05	0.00005	0.005
		Glycine	1.00	0.001	0.1
Inositol	0				
Table-grade sugar	60 g/l	60 g/l from culture initiation until seedlings have developed shoots and roots (until the 3 <sup>rd</sup> to 4 <sup>th</sup> month)			
	45 g/l	45 g/l for maintenance prior to transplanting to the soil			
Activated charcoal (acid washed)	1 g/l				
Gelling agent (Sigma agar)	7 g/l				
State of the medium	Liquid/solid/ solid/liquid	Culture initiation (So) = <b>liquid</b> 1 <sup>st</sup> Subculture (S1) = <b>solid</b> 2 <sup>nd</sup> Subculture (S2) = <b>solid</b> 3 <sup>rd</sup> Subculture (S3) = <b>liquid</b> until the seedlings are ready to be transplanted to the soil			
pH	5.6				

As in any laboratory-based technology, the vigorous exclusion of contaminating microorganism is an absolute necessity in tissue culture. Nutrient media, culture vessels and instruments used in manipulating the tissue and the plant material itself must be sterile. Cleanliness, efficient organization and routine sterilization of all materials will reduce the risk of contamination.

If the embryos are to be collected, prepared and inoculated on site, the following procedures should be followed:

1. Wash the solid endosperm cylinders with tap water several times;
2. Quickly rinse in 95% ethanol;
3. Decant;
4. Immerse in 100% commercial bleach or 5.25% NaOCl for 20 minutes in a clean, sterilized beaker;
5. Decant bleach inside the laminar flow cabinet after sterilization; and
6. Decant bleach and rinse with sterile water at least three or more times.

Sterilize forceps, blades, and flasks either in the autoclave (121°C at 15 psi for 15 minutes) or in the oven (160-170°C for 1 hour). Petri dishes lined with sterile filter should also be autoclaved. Inside the laminar flow cabinet, frequently dip the forceps and scalpel, scissors, etc. in 80% ethanol and sterilize them in the glass bead sterilizer or flame in an alcohol lamp for 20 seconds. Let cool on an aluminium instrument rack.

Using these sterile instruments, excise embryos (Fig. 1d) out from the solid endosperm in the sterile Petri dishes lined with filter papers. Transfer embryos to sterile flask or beaker (Figure 1e) and disinfect them again in 10% commercial bleach for one minute. Rinse with sterile distilled water for 3-5 times. Decant. Blot dry the embryos on sterile Petri dishes lined with filter paper. Inoculate singly (Fig. 1f) onto test tubes containing Y3 liquid medium.

### Culture conditions

Incubate cultures at 28-30°C with approximately 4000-5000 lux at 9 hr photoperiod (15 hours dark and 9 hours light). Sub culture to fresh medium at monthly interval. The first sub culture is in solid medium (Fig. 1g). Sub culture again in solid medium until the formation of the shoot and root (Fig. 1h). Large haustorium may or may not be removed, although they are preferably removed for ease in subsequent transfers (Fig. 1i). Check the cultures periodically for contamination.



**Figure 1.** Early stages of the coconut embryo culture protocol: (a) 11 month-old nuts as source of embryos. Colour of husk breaks from green to brown; (b) extraction of embryos using cork borer; (c) disinfection of embryos with 100% bleach; (d) excision of embryo; (e) disinfection of exposed embryos with 10% bleach; (f) inoculation of embryo in liquid medium; (g) first subculture of embryos in solid medium with 60 g/l sugar for 2-3 transfers; (h) solid medium is used until the formation of green shoot and about one inch long root; (i) primary root and large haustorium are cut at early stage.

Embryos grow at different rates. Generally, 6-8 weeks after the shoot and the roots are formed. The earliest recorded time to transfer *ex vitro* is four months. To economize on the medium used and orient the seedlings to grow properly upright, these are cultured in long test tubes (Fig. 2a). When they have 2-3 expanded leaves and secondary roots are already formed, these are transferred to bigger bottles (Fig. 2b) and extended with autoclavable plastic bags so that the leaves will have enough spaces to grow normally upright. Seedlings with 3 to 4 leaves and enough secondary and tertiary roots (Fig. 2c) are ready for potting. The whole culture period could take about a year or more.

To improve germination, Benzylaminopurine (BAP) at 20 ppm is added to the medium during the first transfer in solid medium for one month. In cases of cultures with very few roots, naphthalene acetic acid (NAA) is added at 10 ppm for one month on the 4<sup>th</sup> sub culture. In addition, freeing the base of the seedling of the brown old tissues and making 2-3 pricks on the site where the root initials are located could facilitate root initiation. The primary root is also cut to enhance secondary and tertiary root formation on the cultures on the 3<sup>rd</sup> to the 4<sup>th</sup> transfer. The tertiary roots are also enhanced when the secondary roots are trimmed.

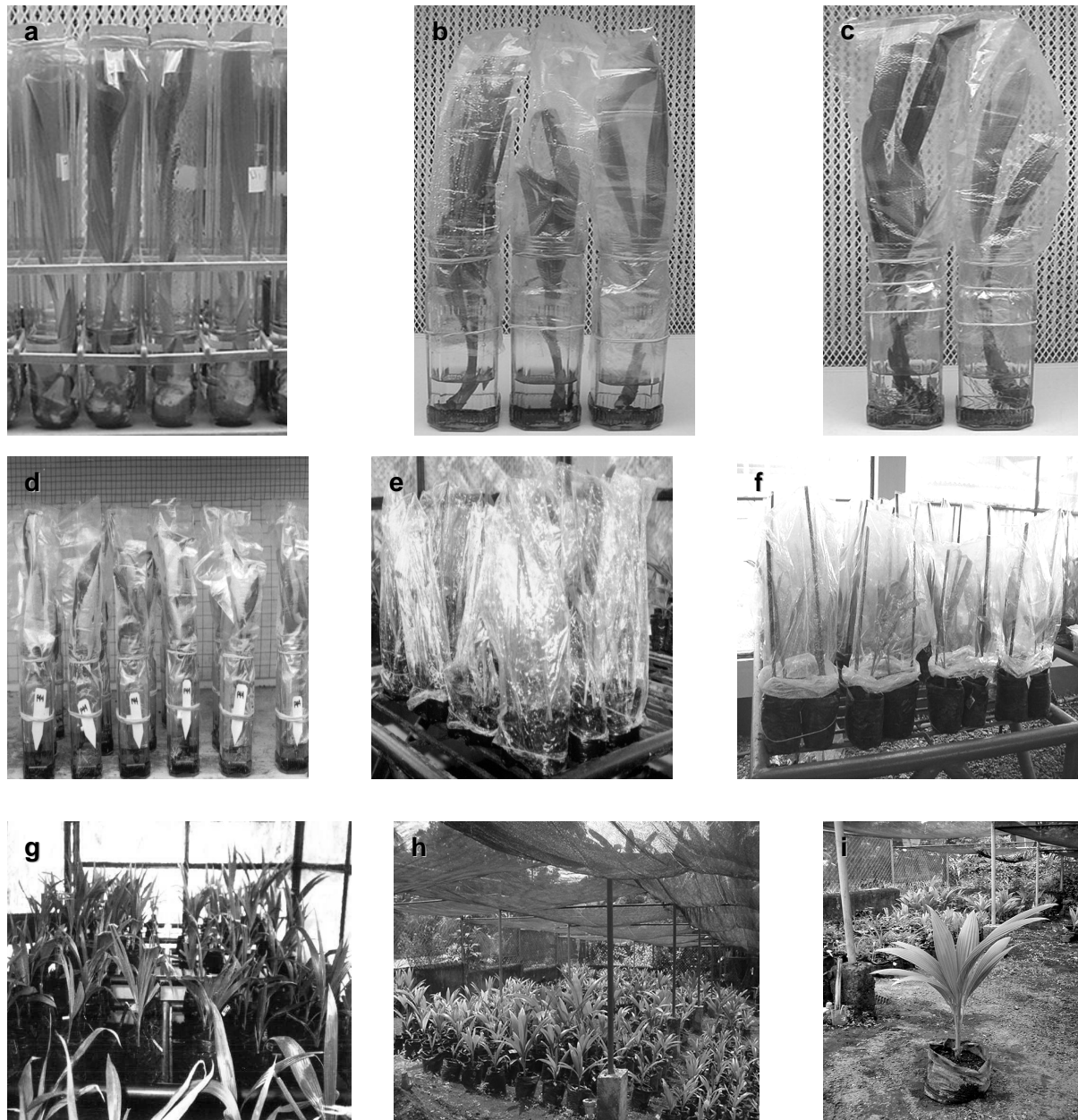
### **Transplanting in the nursery and in the field**

Transplanting has to be done carefully, otherwise a significant number of plants may be lost when transferred from aseptic tissue culture conditions to grow in an external environment. The internal anatomy and ultrastructure of seedlings propagated *in vitro* are different from those of greenhouse- or field-grown plants. Seedlings growing on a sugar-supplemented medium *in vitro* produce only a small amount of their carbohydrate requirement through CO<sub>2</sub> fixation.

When they are taken out of these culture conditions, they have to adapt to the new environment and grow autotrophically. Therefore, hardening of the seedlings is started while still *in vitro* in the screenhouse (Fig. 2d) for about a week. After one week, take out hardened seedling and wash off the medium completely (the liquid medium contains sugar that will attract ants if not completely washed). Dip the seedlings in 2.5 g/l fungicide solutions (e.g. Daconil) then plant in sterilized sand, vermiculite or similar materials. To maintain high relative humidity, cover the seedlings (Fig. 2e) with plastic bags. Support the plastics with bamboo pegs so that they will not sag on the leaves of the seedlings. Keep them covered for three to four weeks or until the seedlings have manifested complete recovery from *in vitro* conditions.

After this period, gradually expose the seedlings to screenhouse conditions by partially lifting (Fig. 2f) the plastic cover for a week. Thereafter, the plants can be fully exposed to screenhouse conditions (Fig. 2g). Water the plants as needed and apply dilute foliar fertilizer solution weekly. Apply necessary control measures against pests and diseases once noticed (Rillo 1995).

After three months, transfer the plants to bigger polyethylene bags with non-sterilized soil and transfer to the nursery under partial shade (Fig. 2h). After another 3-5 months, the plants can be transferred to the field. The plants should have 4-6 leaves (Fig. 2i) by then. Field transplanting should be done during the cooler months to avoid the harsh dry weather during summer. Temporary shade of the plants will have to be provided (i.e., using coconut fronds) to avoid/prevent transfer shock after field planting. Provide the seedlings with the necessary cultural practices for optimum growth response especially during the first three years.



**Figure 2.** Late stages of the coconut embryo culture protocol: (a) slender seedlings are maintained in long test tubes; (b) seedlings with expanded leaves and finer roots are transferred to bigger bottles, covered with autoclavable plastic bag; (c) seedlings with 3-4 expanded leaves, at least one primary root and profuse secondary and tertiary roots could be potted-out; (d) seedlings are hardened in the screenhouse for one week before potting-out; (e) newly potted-out seedlings are covered with plastic bag for 3-4 weeks or until completely recovered from *in vitro* transfer; (f) plastic cover is partially lifted for a couple of weeks; (g) the plants are fully exposed to screen house conditions; (h) after three months, the plants are transferred to bigger polyethylene bags with non-sterilized soil in the nursery; (i) seedlings ready for field planting have 5-6 expanded leaves.

**Possible constraints of the transfer of the technology**

Since the protocol was developed under optimized conditions, its application in other laboratories may not result in the desired number of *ex vitro* transplanted seedlings. Factors such as the age of the embryos, climate and conditions in the receiving laboratory have to be considered aside from following the protocol closely.

It should be emphasized that the right culture vessels and closures should be used. This is a very important aspect of the protocol which is easily overlooked. Coconut is a crop that grows upright only; therefore, it should be provided with enough headspace for it to grow properly.

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## Preparing pollen for export

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### **Introduction**

Coconut germplasm exchange in the form of pollen has a direct advantage on varietal improvement. While it represents only one of the parents of a hybrid, the use of pollen makes it possible to 'pre-select' the variety or varieties to be introduced into a recipient country. In particular, pollen exchange makes it possible to check or test the general combining ability (GCA) of a foreign variety against that of local ones.

Phytosanitary-wise, germplasm exchange using pollen is relatively safer as compared to embryos, more so with seednuts or seedlings. Mites, small insects and other minute organisms, which are associated with the coconut inflorescence either leave or perish during the process of pollen preparation and storage. It is highly unlikely for such organisms to survive the rigors of pollen processing such as freeze drying and vacuum sealing. Trials conducted at the Davao Research Centre of the Philippine Coconut Authority (PCA-DRC) in the early 1990's proved that the spores of *Phytophthora palmivora* that may have been accidentally mixed with coconut pollen during male flower collecting could not survive the extremely dry condition of processed coconut pollen (Concibido 1992, personal communications). Despite this finding, however, common sense dictates that inclusion of varieties susceptible to this disease must be carefully considered in the breeding programme. Palms to be used as pollen sources must be checked against diseases and pests of coconut.

Due to its light weight and minute size, pollen is also less cumbersome and cheaper to transport. In fact, several agricultural development programmes done in the past in major coconut producing countries in the early 1970s (e.g. Philippines and Indonesia) started with the importation of pollen of 'pre-tested' varieties for the mass production of hybrids like the MAWA. This strategy allowed these countries to initiate their coconut development programmes while locally established pollen source varieties are not yet productive.

### **Extracting, processing, packaging and dispatching pollen**

The primordium of the coconut inflorescence, located at the axil of each developing frond, takes around three years to mature into a full-grown spathe. The first to form are thousands of male flowers lined along 30 or so spikelets. It takes three to four months before an emerging inflorescence becomes fully mature at which time the aging male flowers start shedding pollen, which could be two or three days before the natural opening of the spathe. The pattern of male flower maturation is basipetal, and is collectively called the male phase, often lasting for more or less 24 days depending on the variety. A male flower has a natural longevity of one day after opening. It usually begins early in the morning and ends by evening.

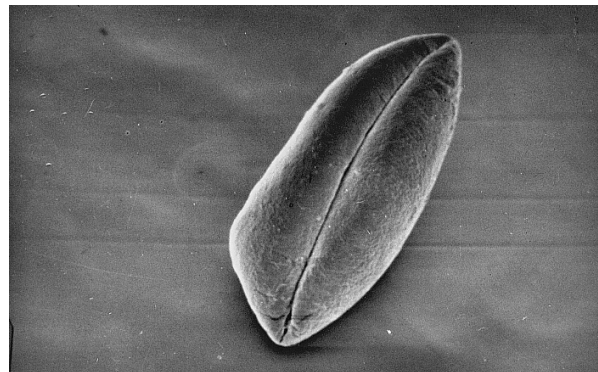
Before male flower collecting, the inflorescence is allowed to mature a little to ensure normal and optimum male flower maturation and also to optimize the collection of more viable pollen (Fig. 2). Apart from time of gathering, the quantity and quality of coconut pollen are influenced by variety. Tall coconut varieties generally produce more pollen than a Dwarf one due to more male flowers and bigger and more robust inflorescences in the former. Coconut inflorescence of a Tall palm can yield as much as nine grams of pollen with a viability of about 45% when

flowers are collected six days after spathe opening and following the bagging technique of male flower collection (Santos and Baliñgasa 1977).



**Figure 1.** An inflorescence showing mature male flower

Coconut pollen is monocolpate with a tough exine and measures  $30\mu$  in diameter (Fig. 2). Coconut pollen can be dispersed by wind up to 200 m in most cases and up to 315 m in some (Child 1964). It can remain viable for several days at ambient temperature. Pollen succumbs easily to exposure to alcohol and to a temperature of  $150^{\circ}\text{C}$ . Naturally shed pollen is considered no longer viable after six days owing to alternating wet and dry regimes, and alternating low night and high day temperatures.



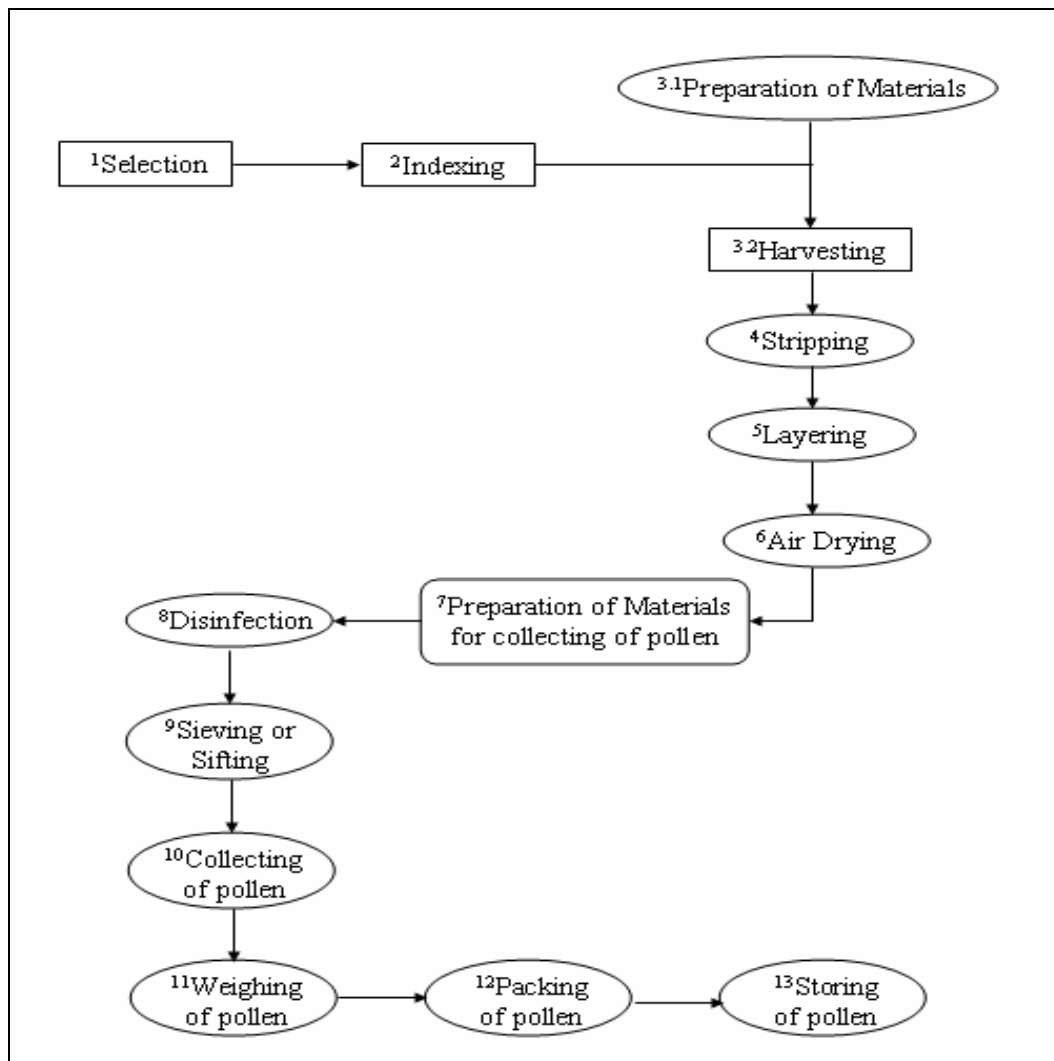
**Figure 2.** A magnified close-up of a coconut pollen

Coconut pollen is easy to view under an ordinary field microscope at low magnification (100x) and even without staining. Past observations revealed that the presence of other pollen grains at the immediate vicinity of a germinating pollen grain induces the pollen tube of the latter to elongate faster and develop more quickly than solitary ones. This phenomenon is called 'population effect'. A trace amount of  $\text{CaCl}_2$  in the medium prevents this from occurring. Thus, to get a good estimate of pollen viability through the germination test, sowing of pollen grains in the germinating medium should be done carefully ensuring that grains are evenly and equally sown on the medium.



### Harvesting/collecting male flowers

Figure 3 below summarizes the flow of activities related to harvesting/ collecting pollen.



**Figure 3.** Process flowchart for harvesting/ collecting pollen: (1) Selection of palms; (2) Indexing of palm for inflorescence opening; (3) Preparation of materials (e.g., sacks, ladder, harvesting pole/scythe, pruning shears, plastic twine, marking pen, metal drying trays, plastic basins, plastic cans, leather gloves, rolling pin) and harvesting of male flowers; (4) Stripping of male flowers from spikelets; (5) Layering male flowers in the metal drying trays, cracking of male flowers; (6) Air drying of male flowers; (7) Preparation of materials for collecting of pollen (e.g., pollen manipulation box (PMB), plastic bags, 5" x 10" and 2" x 4", plastic sealer, marking pen/labels, brush, brass sieve (200 mesh), rubber band, alcohol); (8) Disinfection of PMB and brass sieves with alcohol; (9) Sieving or Sifting of dried male flowers; (10) Collecting of pollen in 5" x 10" plastic bags; (11) Weighing of pollen; (12) Packing of pollen in smaller, e.g., 2" x 5" plastic bags; and (13) Storing of pollen in a freezer

Harvesting of spikelets bearing the mature male flowers is done six to seven days after the natural opening of the spathe. For palms that are to be temporarily used as male parents, the spikelets bearing the male flowers are cut two to three inches from the main rachis of the bunch (if there is no female flower) or two inches above the female flower, if there is any. This is to allow the subsequent natural pollination of the female flowers within the bunch. In case the palms are to be used solely as a source of pollen for a long time, the whole inflorescence is cut when about 1/4 of the spikelets in the inflorescence are already showing mature male flowers. The spikelets bearing the male flowers are then cut and placed in canvas or nylon bags, and these are brought to the pollen laboratory where the male flowers are detached. Immature male flowers are usually creamy-yellow and smaller than mature male flowers, which are almost green with bluish tips.

**Stripping male flowers.** The coconut male flower has a tough and sturdy structure. To avoid injury, personnel must wear leather gloves (Fig. 3) when stripping them from the spikelets. They are collected in plastic basins and weighed prior to cracking and drying.



**Figure 4.** Stripping male flowers (note use of gloves)

**Cracking male flowers.** The separated male flowers are cracked and placed thinly over galvanized or aluminium trays. Cracking of male flowers before drying facilitates the subsequent exposure of the anthers to the atmosphere inside the drying room. This technique promotes the release of the pollen from the anther sacs. Cracking is usually done by rolling a baker's pin over the flowers in the tray (Fig. 4).



**Figure 5.** Cracking-open male flowers using a rolling pin

**Drying male flowers.** Drying is done in a dehumidified and artificially heated room with a relative humidity of about 10% and a temperature of about 40°C for 24 hours. Drying is longer without a dehumidifier, usually taking about 36 hours. Exposure to a temperature of 40°C promotes the early maturation of the male flowers and anther dehiscence. A label bearing the information of the variety, time, and date of collecting and processing, as well as the name of the pollen processor, is placed on the door of the pollen drying room.

**Sifting and storing pollen.** When the flowers are sufficiently dry, the dried flowers are sieved to separate the pollen grains from male floral parts (Fig. 5) using a 200 mm mesh brass sieve. The pollen grains are collected in the bottom pan and transferred to plastic bags or any suitable container at five grams per pack for sealing and subsequent storage in a freezer (-20 to -25°C) (Fig. 6). The pollen source, date of collection and processing are indicated in each pollen packet. A small sample from each pollen batch is set aside for viability and moisture content determination.



**Figure 6.** Sieving the male flowers to separate pollen grains from other floral parts



**Figure 7.** Bagged and sealed pollen grains stored in a freezer

**Pollen viability.** Dusting pollen samples in an agar-sugar medium (0.5% agar plus 10% sugar plus 0.001-PPM Boric acid) tests pollen viability (Fig. 7). After an incubation period of two hours at room temperature (28°C-30°C), pollen germination percentage is taken by counting the number of germinated pollen grains against the total pollen grains observed from 10 to 15 microscopic fields or until about 100 grains are counted. For best results, counting of germinated pollen grains should be done after six hours of incubation.



**Figure 8.** Testing pollen viability using an agar-sugar medium

This ensures that even pollen tubes that are inverted can be counted. For a certain batch of pollen to be considered viable, percentage germination should be at least 25% (Santos *et al.* 1996). Periodic inventory and testing of viability of stored pollen is advised. All pollen batches with poor viability must be properly disposed.

#### Preparing pollen for shipment

**Packing in ampoules** - The pollen is collected in quantities of 0.4-0.5 g per glass ampoule, plugged with sterile cotton wad for freeze-drying purposes and labelled properly using a paper tag indicating the pollen identity and the date of collection. The pollen must be lightly packed in the ampoule to facilitate freeze-drying. In case no glass ampoules are available, the pollen may be collected in glass vials and similarly sealed.

**Constriction of ampoules** - This is necessary to facilitate the sealing of the ampoules using a blowtorch after freeze-drying. Using a special kind of equipment, called ampoule constrictor, the ampoules containing the pollen are constricted at about two centimetres from its open end (Fig. 8).



**Figure 9.** Constricting ampoules containing pollen using an ampoule constrictor

**Freeze-drying** - After constriction, the ampoules are tightly fitted one by one to the rubber teats in the freeze-drying apparatus (Fig. 9). Freeze-drying preserves the viability of the pollen longer because it is eventually dried with minimum residual moisture content while fully retaining its original properties (e.g. protein, viability, enzymes, etc). A vacuum is created inside the ampoule. Thus, the pollen retains its full quality despite long storage.



**Figure 10.** Freeze-drying the pollen ampoules in preparation for storage

**Desiccating the pollen** - If a freeze-dryer is not available, coconut pollen can be put in vials with porous covers and placed in a desiccator. A bag of silica gel (amount depends on the size of the desiccator), which is colour indicative may be used to maintain the desired relative humidity in the desiccator.

The desiccated pollen can be kept viable for hand-pollination of short duration (2-3 months). It is advisable, however, to provide a device to isolate each kind of pollen in the desiccator to prevent pollen contamination.



**Figure 11.** The pollen ampoules being tested for vacuum leak using a spark tester

**Sealing of ampoules and testing for vacuum** - After running the freeze-dryer for 15-20 minutes at 10-1 torr, the ampoules are sealed by heating them at the constricted end with an air-gas sealing torch (Fig. 10). The sealed ampoules are then tested for vacuum by means of a spark tester. The spark is seen to concentrate and pass through any crack or small hole on the glass ampoule to give a clear indication of a leak. In any case, improperly sealed ampoules are the first ones that should be used.

**Storing the pollen** - The sealed ampoules are kept in the freezer (Fig. 11) for a period of up to six months or longer. Experience at the Philippine Coconut Authority (PCA) - Zamboanga Research Centre has shown that properly prepared and conditioned pollen, which is stored in a freezer, can retain its viability even after five years. Nonetheless, it is necessary to regularly test pollen viability before using, particularly if the age of the pollen is over six months.

Pollen should be taken out of the freezer only when it is needed because frequent alternate freezing and thawing are detrimental to pollen. All stored pollen must be recorded in a pollen registry book to facilitate withdrawal and identification.



**Figure 12.** The pollen ampoules stored in a freezer

**Moisture content of pollen** - Drying the freshly collected pollen (after sieving) in an oven at 105°C for 24 hours reduces moisture content to 4-8%. If moisture content is more than 8%, further drying or desiccation of pollen is needed.

**Dispatching pollen**

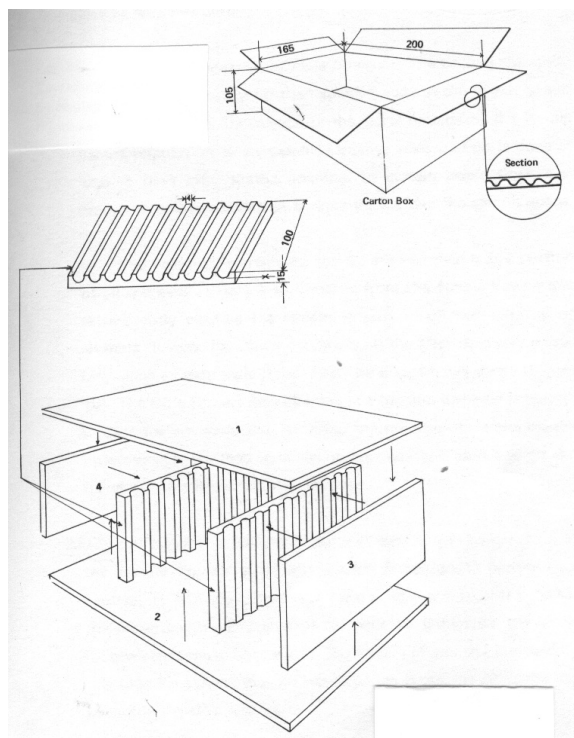
With the increasing interest in inter-agency and international coconut pollen exchange programmes, a standard form and system of packing pollen must be provided and followed. A copy of pollen dispatch (PD) inside an enveloped sealed with plastic to prevent from getting wet must always go together with the pollen container. The PD format is illustrated in Form 1 below:

**Form 1**

Origin: _____	PD No.: _____
Sender: _____	Series of: _____
Destination: _____	Shipment to: _____
	Date of Shipment: _____
Kind of Pollen: _____	Quantity: _____

Population	Collection Date	No. of Packs/ Ampoules	Weight (grams)	Percent Germination

The system of packing vacuum-sealed ampoules of pollen in specially designed styropor/styrofoam packaging material is illustrated in Figure 12. Pollen in plastic packs can use the same packaging material except for the corrugated separator. The boxes may vary in size depending upon the volume of pollen to be sent.



**Figure 13.** Packaging material and design for transporting pollen

### Use

Only when the pollens are to be utilized should they be taken out of the freezer. Viable pollen is mixed with talcum powder at the ratio of 8 parts talcum to 1 part pollen. Mixing of the talcum powder and pollen in a puffer bottle is done inside the manipulation box (Fig. 13) or right at the center of the field where the pollination will take place. The emasculated inflorescences are pollinated as soon as the female flowers start becoming receptive and continuously up to the end of the female phase (normally for three consecutive days). Pollination is done daily from 6:00 AM to 11:00 AM since the female flowers are receptive during this period.



**Figure 14.** Mixing pollen with talcum powder inside a manipulation box in preparation for using the pollen

Collecting coconut pollen for research purposes follows a more rigorous procedure. On the other hand, collecting of pollen for hybrid seed production in a seed garden is less cumbersome as pollen is collected in bulk. Unlike in pollen collection for research purposes where bagging of the inflorescence is required prior to the male flower collection, the latter is easier and faster since harvesting of male flowers is made in the open. The choice of the variety to be exported or imported generally depends on the requirement of the breeder.

### ***Documents needed in pollen transport***

The basic documents needed in transporting pollen include an FAO Phytosanitary Certificate, which is normally issued from the point of collection in the country of origin. If pollens are to be transported via commercial couriers (i.e., DHL, FedEx, UPS, etc), import and export permits are required by countries and are likewise imposed by such international courier companies.

In the Philippines, an application for the Phytosanitary Certificate is required prior to the issuance of the FAO Phytosanitary Certificate. The document is issued by the Quarantine Office of the Department/Ministry of Agriculture in the Region where the pollen is taken from. The application form requires the applicant to provide information on source, kind and nature of plant material being applied for issuance of the certificate stating the following:

- Health/nutritional status of the source plant;
- Treatments made on the material;
- Packaging specifications and destination; and
- Other relevant information (i.e., purpose of transport, e.g. commercial use or for research).

### **Pre-arrangement/ requirements**

As in the exchange of planting materials, government to government arrangements are normally required prior to the issuance of import/export permits for coconut pollen. International couriers require that shipments be accompanied by such certificates/permits to facilitate issuance of exit (from country of origin) to entry permits and customs/duty certificates in the destination.

Per PCA's experience, government to government arrangements made through diplomatic channels (i.e., embassies or consulates) could facilitate faster germplasm exchange.

### **References**

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# ANNEXES

- **ANNEX 1.1:** MOA for the establishment of the International Coconut Genebank for the South Pacific
- **ANNEX 1.2:** List of designated germplasm for the International Coconut Genebank for the South Pacific
- **ANNEX 1.3:** List of designated germplasm for the International Coconut Genebank for South Asia
- **ANNEX 1.4:** List of designated germplasm for the International Coconut Genebank for Southeast and East Asia
- **ANNEX 1.5:** List of designated germplasm for the International Coconut Genebank for Africa and Indian Ocean
- **ANNEX 2.1:** Key terms and their definitions as used in Chapter 2
- **ANNEX 2.2:** Distribution table of regulated/ quarantined coconut pests
- **ANNEX 2.3:** Datasheets of major quarantined pests



***Annex 1.1. MOA for the establishment of the International Coconut Genebank for the South Pacific***

**AGREEMENT BETWEEN THE GOVERNMENT OF PAPUA NEW GUINEA, THE INTERNATIONAL PLANT GENETIC RESOURCES INSTITUTE (IPGRI) AND THE FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS (FAO) PLACING COCONUT GERMPLASM COLLECTIONS UNDER THE AUSPICES OF FAO**

**PREAMBLE**

The Government of Papua New Guinea (hereinafter referred to as "Host Country"), hosting the International Coconut Genebank for the South Pacific, the International Plant Genetic Resource Institute (hereinafter referred to as "IPGRI", one of the Centres of the Consultative Group on International Agricultural Research), acting on behalf of the International Coconut Genetic Resources Network (COGENT), as described in the attachment "Background to the Agreements") and the Food and Agriculture Organization of the United Nations (hereinafter referred to as "FAO");

Considering the importance to humanity of protecting and conserving coconut germplasm for future generations;

Considering the International Undertaking on Genetic Resources adopted by the FAO Conference at its 22nd Session in 1983 (Resolution 8/83) and in particular Article 7 thereof; and the Annexes of the Undertaking adopted by the FAO Conference in 1989 and 1991;

Considering that the FAO Commission on Genetic Resources for Food and Agriculture (hereinafter referred to as the "Commission"), as the relevant intergovernmental body in this field, has the responsibility for monitoring the implementation of Article 7 of the International Undertaking on Plant Genetic Resources;

Considering the Memorandum of Understanding Between the Food and Agriculture Organization of the United Nations and the International Board for Plant Genetic Resources (IBPGRI) legally succeeded by IPGRI, dated September 21, 1990, on the respective roles of the two organizations in establishing, maintaining and managing germplasm collections and setting standards for these collections;

Considering the importance of the International Coconut Genebank held by the Government of Papua New Guinea within COGENT and supported by IPGRI, as part of a global strategy for germplasm conservation;

Considering that the Coconut germplasm accessions have been donated to the International Coconut Genebank for the South Pacific on the understanding that these accessions will remain freely available;

Considering that any country that so desires may participate in COGENT;

Considering that the Government of Papua New Guinea has expressed the wish that the designated coconut germplasm accessions, kept in the International Coconut

Genebank for the South Pacific, be recognized as part of the International Network of *Ex Situ* Collections (as per the International Undertaking on Plant Genetic Resources) under the Auspices of FAO;

- Taking note of the provisions of the Convention on Biological Diversity, particularly those pertaining to affirmation of sovereign rights of nations over their biological resources and access and benefit sharing mechanisms.
- Also taking note of the ongoing process of harmonisation of the International Undertaking on Plant Genetic Resources with the CBD, and the request of the Conference of the Parties to the Convention on Biological Diversity to the governments to speed up this process.

Have agreed as follows:

### **Article 1**

#### **APPLICATION OF THIS AGREEMENT**

This Agreement shall be construed and applied in a manner consistent with the provisions of the Convention on Biological Diversity and the International Undertaking on Plant Genetic Resources.

### **Article 2**

#### **BASIC UNDERTAKING**

The Government of Papua New Guinea hereby places under the auspices of FAO, as part of the International Network of *Ex Situ* Collections provided for in Article 7 of the International Undertaking on Plant Genetic Resources, the accessions of coconut genetic resources listed in the Appendix hereto (hereinafter referred to as the "designated germplasm"), in accordance with the terms and conditions set forth in this Agreement. The List of designated germplasm will be updated every two years as new accessions are added to the collection.

### **Article 3**

#### **STATUS OF DESIGNATED GERMPLOSM**

- a) The Government of Papua New Guinea shall hold the designated germplasm in trust for the benefit of all countries in accordance with the International Undertaking on Plant Genetic Resources and the terms and conditions set out in this Agreement.
- b) The Government of Papua New Guinea shall not claim legal ownership over the designated germplasm, nor shall it seek any intellectual property rights over that germplasm or related information.

**Article 4****PREMISES**

- a) The premises, i.e., land and/or laboratories, in which the designated germplasm is conserved, shall remain in the charge of the Government of Papua New Guinea.
- b) FAO shall have a right of access to the premises at any time and the right to inspect all activities performed therein directly related to the conservation and exchange of the designated germplasm.

**Article 5****MANAGEMENT AND ADMINISTRATION**

- a) The Government of Papua New Guinea undertakes to manage and administer the designated germplasm in accordance with Internationally Accepted Standards, including standards as agreed upon by COGENT, and the International Genebank Standards, endorsed by the Commission, where these are applicable to coconut, and ensuring that all the designated germplasm is duplicated in order to ensure its safety.
- b) FAO may recommend action, if it considers such action to be desirable, to ensure the proper conservation of the designated germplasm.
- c) If the orderly maintenance of the designated germplasm is impeded or threatened by an event, including *force majeure*, and the Government of Papua New Guinea does not have the capacity to take appropriate preventive or curative action, FAO and IPGRI shall seek the necessary resources from the international community for action to ensure the safety of the designated germplasm, including if necessary by its evacuation and transfer.

**Article 6****POLICIES**

The Government of Papua New Guinea and IPGRI recognize the intergovernmental authority of FAO and its Commission in setting policies for the International Network of *Ex Situ* Collections referred to in Article 7 of the International Undertaking and undertake to consult with FAO and its Commission on proposed policy changes related to the conservation of, or accessibility to, the designated germplasm, subject, always to the provisions of Article 9 hereinafter. The Government of Papua New Guinea and IPGRI shall give full consideration to any policy changes proposed by the Commission.

**Article 7****STAFF**

- a) Staff responsible to manage and administer the designated germplasm shall be employed and remunerated by the Government of Papua New Guinea.
- b) As and when deemed appropriate, FAO and IPGRI shall furnish technical

backstopping on request by the Government of Papua New Guinea and COGENT.

## **Article 8**

### **FINANCES**

The Government of Papua New Guinea shall remain responsible for financing the maintenance of the designated germplasm.

## **Article 9**

### **AVAILABILITY OF DESIGNATED GERmplasm AND RELATED INFORMATION**

Subject to the provisions of Article 10 below, the Government of Papua New Guinea undertakes to make samples of the designated germplasm and related information available directly to all countries participating in COGENT, for the purpose of scientific research, plant breeding or genetic resource conservation, without restriction.

## **Article 10**

### **TRANSFER OF DESIGNATED GERmplasm AND RELATED INFORMATION**

Where samples of the designated germplasm and/ or related information are transferred to any other person or institution the Government of Papua New Guinea shall ensure that such other person or institution, and any further entity receiving samples of the designated germplasm from such person or institution, is bound by the conditions set out in Article 3 (b) and, in the case of samples duplicated for safety purposes, to the provisions of Article 5 (a).

This provision shall not apply to the repatriation of germplasm to the country that provided such germplasm.

## **Article 11**

### **DURATION**

- a) This Agreement is concluded for a period of 4 years and shall be automatically renewed for further periods of 4 years unless notice of non-renewal is given in writing by either party not less than 2 years before the end of any 4-year period.
- b) This Agreement shall be revised, if necessary, in accordance with the provisions of the revised International Undertaking.

## **Article 12**

### **TERMINATION**

- a) Either FAO or the Government of Papua New Guinea may terminate this

Agreement at any time by giving notice to the other, two years in advance of the termination date.

- b) FAO, the Government of Papua New Guinea and IPGRI, shall, in such case, take all necessary measures to wind up joint activities in an appropriate manner and, within the limits of their respective competencies, to ensure the continued conservation of and access to the designated germplasm.

### **Article 13**

#### **SETTLEMENT OF DISPUTES**

- a) Any dispute concerning the implementation of this Agreement shall be settled by mutual consent.
- b) Failing mutual consent, such dispute may be submitted, at the request of either FAO, or the Government of Papua New Guinea or IPGRI, to an arbitral tribunal composed of four members. Each party shall appoint one arbitrator. The three arbitrators thus appointed shall designate by mutual consent the fourth arbitrator, who will act as the presiding arbitrator of the tribunal. In case of equal division of votes the presiding arbitrator will have a second vote.
- c) If within two months after the receipt of a party's notification of the appointment of an arbitrator one or both of the other parties has/have not notified the first party of the arbitrators they have appointed, the first party may request the Secretary-General of the United Nations to appoint arbitrators to represent parties that have not appointed an arbitrator.
- d) If within two months after the appointment of the three arbitrators they have not agreed on the choice of the presiding arbitrator, such presiding arbitrator shall be designated by the Secretary-General of the United Nations at the request of either party.
- e) Unless the parties to the dispute decide otherwise, the tribunal shall determine its own procedure.
- f) A majority vote of the arbitrators shall be sufficient to reach a decision which shall be final and binding for the parties to the dispute.

### **Article 14**

#### **AMENDMENT**

- a) FAO, the Government of Papua New Guinea or IPGRI may propose that the Agreement be amended by so informing the other parties
- b) If there is mutual agreement in respect of a proposed amendment, the amendment shall enter into force on whatever date is set, and be reported to the next session of the Commission.



**Article 15**

**DEPOSITARY**

The Director-General of FAO shall be the Depository of this Agreement. The Depository shall:

- a) Send certified copies of this Agreement to the Member Nations of FAO and to any other Government which so requests;
- b) Arrange for the registration of this Agreement, upon its entry into force, with the Secretariat of the United Nations in accordance with Article 102 of the Charter of the United Nations;
- c) Inform FAO Members Nations of:
  - i) The signature of this Agreement in accordance with Article 16; and
  - ii) The adoption of amendments to this Agreement in accordance with Article 14.

**Article 16**

**COMING INTO FORCE**

This Agreement shall come into force upon signature by the authorized representative of FAO, the Government of Papua New Guinea and IPGRI.

The Food and Agriculture Organization  
Of the United Nations

The Government of Papua New Guinea  
Hon. Tukape Masane, M.P.  
Minister for Agriculture and Livestock

By .....  
(Signature)

Date:

By .....  
(Signature)

Date:

The International Plant Genetic  
Resources Institute (IPGRI)

By .....  
(Signature)

Date:

**APPENDIX 1**

## List of germplasm accessions covered by this Agreement

Accessions	Source	Accessions	Source
	<b>East New Britain</b>	31. Saiho	<b>Oro</b>
1. Pellavarua	- Gazelle Peninsula	32. Ajoa	
2. Raulawat		33. Kikibator	
3. Natava		34. Siagara	<b>Milne Bay</b>
4. New Massava		35. Bubuleta	
5. Natava Many		36. Baibara	<b>Central</b>
6. Fruited		37. Hisihu	
7. Gaungo	<b>West New Britain</b>	38. Poligolo	
8. Naviro		39. Miha Kavava	<b>Gulf -Vailala</b>
9. Talasea Red		40. Keakea	
	<b>New Ireland</b>	41. Iokea	- Iokea
10. Karu village	- Namatanai		
11. Kenapit		42. Severimabu	<b>Western (Kiwai Tall)</b>
12. Sohu		43. Boze	
13. Etalat	- Mussau Is.		<b>Exotic Talls</b>
14. Lawes	<b>Manus</b>	44. Rennell	- Rennell Tall
15. Lako			
16. Baluan		45. PNG Yellow	<b>Local Dwarfs</b>
17. Wutung	<b>Sandaun</b>	46. PNG Red 1	
18. Hawaii	<b>East Sepik</b>	47. PNG Red 2	
19. Yangoru		48. Rabaul Red	
20. Vokio		49. PNG Brown	
21. Marineberg		50. Iokea Red	
22. Guanaga	<b>Madang (Karkar Tall)</b>	51. Malayan Yellow	<b>Exotic Dwarfs</b>
23. Kinim		52. Malayan Red	
24. Ulatava		53. Nias Green	
	<b>Morobe</b>	54. Nias Yellow	
25. Markham Farm	- Markham Tall	55. Nias Red	
26. Liara village			
	<b>East New Britain</b>		
27. Raulawat Yellow	- Gazelle Peninsula		
28. Raulawat Red			
29. Natava Yellow			
30. Natava Red			

## Additional list of international germplasm to be established

Ecotype	Source
56. Rotuma Tall	Fiji
57. Tonga Tall	Tonga
58. Kiribati Tall	Kiribati
59. Rangiroa Tall	Tahiti
60. Vanuatu Tall	Ivory Coast, PNG
61. Western Samoan Tall	Western Samoa

62. Samoan Yellow Dwarf	Western Samoa
63. Nui Leka Green Dwarf	Fiji
64. Fiji Tall	Fiji
65. Niu Vai	Western Samoa
66. Niu Afa	Western Samoa
67. Christmas Is. Tall	Kiribati
68. Kiribati Green Dwarf	Kiribati
69. New Caledonia Tall	New Caledonia
70. Vanikoro Tall	Solomon Island
71. Solomon Tall	Solomon Island
72. Niu-bubu, or Pine or Mami Kokonas	PNG & Solomon Islands

**Other Ecotypes**

73. Cameroon Red Dwarf	Ivory Coast
74. Salak Green Dwarf	Indonesia
75. Pilipog Green Dwarf	Philippines
76. Tacunan Green Dwarf	Philippines
77. Aromatic Green Dwarf	Thailand
78. Catigan Green Dwarf	Philippines
79. Brizilian Green Dwarf	Ivory Coast
80. West African Tall	Ivory Coast
81. Sri Lankan Tall	Sri Lanka
82. Panama Tall	Jamaica

Locations where materials are held:

- **1 – 55** Stewart Research Station; Papua New Guinea Cocoa & Coconut Research Institute Madang, Madang Province
- **56 – 82** To be established at Stewart Research Station, Madang, PNG

## APPENDIX 2

### Background to the agreements

The Coconut Genetic Resources Network (COGENT) was established in 1992 to improve coconut production on a substantial basis and to increase incomes in developing countries through improved cultivation of the coconut and efficient utilization of its products. COGENT is actively undertaking an international collaborative programme with member countries to improve the conservation and use of coconut genetic resources in the following areas:

- 1) Establishing and maintaining an International Database on existing and future collections;
- 2) Encouraging the protection and utilization of existing germplasm collections;
- 3) Identifying and securing additional threatened diversity through the development and adoption of suitable technologies and conservation strategies;
- 4) Promotion of greater collaboration among research groups in producer countries and advanced technology sources in the exchange of germplasm and the development of new techniques; and
- 5) Appropriate training, information dissemination and securing the necessary funding.

COGENT operates through a steering committee comprised of two members from each of the 5 sub-networks namely Southeast Asia, South Asia, Pacific, Africa and Latin America/Caribbean, and a full time coordinator based in the Asia, Pacific and Oceania Regional Office of the International Plant Genetic Resources Institute (IPGRI-APO) in Serdang, Malaysia.

COGENT's membership has now grown to 35 coconut-producing countries, with each country having to agree to provide access to its coconut germplasm and data as one of the conditions for membership. The member countries are shown in the table below.

Southeast and East Asia	South Asia	South Pacific	Africa/Indian Ocean	Latin America/Caribbean
1. China 2. Indonesia 3. Malaysia 4. Myanmar 5. Philippines 6. Thailand 7. Vietnam	1. Bangladesh 2. India 3. Pakistan 4. Sri Lanka	1. Cook Is. 2. Fiji 3. Kiribati 4. Papua New Guinea 5. Solomon Is. 6. Tonga 7. Vanuatu 8. Samoa	1. Benin 2. Cote d'Ivoire 3. Ghana 4. Kenya 5. Mozambique 6. Nigeria 7. Seychelles 8. Tanzania	1. Brazil 2. Costa Rica 3. Cuba 4. Guyana 5. Haiti 6. Jamaica 7. Mexico 8. Trinidad-Tobago

Under the mandate of the CGIAR, the IPGRI established COGENT with the endorsement of the Technical Advisory Committee. IPGRI functions as the executing institution for COGENT and provides administration and technical support and advice.

An essential component for sustainable production and improvement in coconut is the availability of a wide diversity of germplasm from around the world for use as introductions or in coconut breeding programmes to develop improved coconut varieties and hybrids for coconut producing countries.

To further ensure the security of germplasm in national collections which are

important to each region and to provide member countries with germplasm for developing better varieties and hybrids, COGENT will establish **an international multi-site genebank** consisting of a regional genebank in each of the five COGENT regions. The host country will benefit from the use of the entire germplasm collection, and duplicates supplied from the other regional genebanks, in its breeding programme to develop high-yielding and adapted coconut varieties. The host countries have agreed to a 10-point criterion which includes, among others, access of member countries to the held germplasm and commitment to gather and submit data and to maintain the collection.

The Convention on Biological Diversity (CBD) is a legally binding international agreement that sets out the sovereign rights of countries over their genetic resources as well as the responsibilities of states to conserve and to share these resources and benefits arising from their use.

The Food and Agriculture Organization (FAO) is in the process of establishing Global Network of *Ex Situ* Collections. In December 1994, close to half a million germplasm accessions of food crops held by 12 International Agricultural Research Centres under the CGIAR were placed under FAO trusteeship through a series of agreements signed by FAO and the chairman of the CGIAR acting on behalf of each of the 12 Centres. These agreements were developed in accordance with the CBD.

During a COGENT workshop held on 26-28 February 1996 at Pekanbaru, Riau, Indonesia, representatives of IPGRI, CIRAD and World Bank participated with COGENT members in developing a series of legal agreements, seven-year workplans and proposed budgets for each of the initial four genebanks to be hosted by India for South Asia, Indonesia for Southeast Asia, Papua New Guinea for the Pacific and Côte d'Ivoire for Africa.

The following three agreements, which are considered consistent with the CBD and necessary to facilitate access to coconut genetic resources of which individual countries agree to designate to the international genebanks, are enclosed. These agreements follow closely those agreed to by FAO and the CGIAR centres, with two important changes. First, each host country holding the designated accessions is to be a party in signing the tripartite agreement, and IPGRI is the second party, acting on behalf of COGENT.

- (a) The tripartite agreement [Agreement between {Name of Host Country}, the International Plant Genetic Resources Institute (IPGRI) and the Food and Agriculture Organization of the United Nations (FAO) Placing Coconut Germplasm Collections under the Auspices of FAO], provides a list of designated accessions for each genebank, and spells out the rights and obligations of the parties to the agreement.
- (b) The Germplasm Acquisition Agreement sets out the terms and conditions of movement of coconut germplasm accessions from the providing country to each of the international genebanks.
- (c) A standard Material Transfer Agreement specifies that the recipient agrees not to claim legal ownership over the designated germplasm or take out any intellectual property rights over that germplasm or related information. Furthermore, the recipient also undertakes to pass the same obligations to all future recipients of designated germplasm. The MTA will be used designated germplasm and related information is made available.

**Annex 1.2 List of designated germplasm for the International Coconut Genebank for the South Pacific**

Accessions	Source	Accessions	Source
	<b>East New Britain</b>	31. Saiho	<b>Oro</b>
1. Pellavarua	- Gazelle Peninsula	32. Ajoa	
2. Raulawat		33. Kikibator	
3. Natava		34. Siagara	<b>Milne Bay</b>
4. New Massava		35. Bubuleta	
5. Natava Many		36. Baibara	<b>Central</b>
6. Fruited		37. Hisihu	
7. Gaungo	<b>West New Britain</b>	38. Poligolo	
8. Naviro		39. Miha Kavava	<b>Gulf -Vailala</b>
9. Talasea Red		40. Keakea	
	<b>New Ireland</b>	41. lokea	- lokea
10. Karu village	- Namatanai		
11. Kenapit		42. Severimabu	<b>Western (Kiwai Tall)</b>
12. Sohu		43. Boze	
13. Etalat	- Mussau Is.		<b>Exotic Talls</b>
14. Lawes	<b>Manus</b>	44. Rennell	- Rennell Tall
15. Lako			
16. Baluan		45. PNG Yellow	<b>Local Dwarfs</b>
17. Wutung	<b>Sandaun</b>	46. PNG Red 1	
18. Hawaii	<b>East Sepik</b>	47. PNG Red 2	
19. Yangoru		48. Rabaul Red	
20. Vokio		49. PNG Brown	
21. Marineberg		50. lokea Red	
22. Guanaga	<b>Madang (Karkar Tall)</b>	51. Malayan Yellow	<b>Exotic Dwarfs</b>
23. Kinim		52. Malayan Red	
24. Ulatava		53. Nias Green	
	<b>Morobe</b>	54. Nias Yellow	
25. Markham Farm	- Markham Tall	55. Nias Red	
26. Liara village			
	<b>East New Britain</b>		
27. Raulawat Yellow	- Gazelle Peninsula		
28. Raulawat Red			
29. Natava Yellow			
30. Natava Red			

**Additional list of international germplasm to be established**

Ecotype	Source	Other Ecotypes	Source
56. Rotuma Tall	Fiji	73. Cameroon Red Dwarf	Ivory Coast
57. Tonga Tall	Tonga	74. Salak Green Dwarf	Indonesia
58. Kiribati Tall	Kiribati	75. Pilipog Green Dwarf	Philippines
59. Rangiroa Tall	Tahiti	76. Tacunan Green Dwarf	Philippines
60. Vanuatu Tall	Ivory Coast, PNG	77. Aromatic Green Dwarf	Thailand

61. Western Samoan Tall	Western Samoa	78. Catigan Green Dwarf	Philippines
62. Samoan Yellow Dwarf	Western Samoa	79. Brizilian Green Dwarf	Ivory Coast
63. Nui Leka Green Dwarf	Fiji	80. West African Tall	Ivory Coast
64. Fiji Tall	Fiji	81. Sri Lankan Tall	Sri Lanka
65. Niu Vai	Western Samoa	82. Panama Tall	Jamaica
66. Niu Afa	Western Samoa		
67. Christmas Is. Tall	Kiribati		
68. Kiribati Green Dwarf	Kiribati		
69. New Caledonia Tall	New Caledonia		
70. Vanikoro Tall	Solomon Island		
71. Solomon Tall	Solomon Island		
72. Niu-bubu, or Pine or Mami Kokonas	PNG & Solomon Islands		

**Note:**

Locations where materials are held:

- **1 – 55** Stewart Research Station; Papua New Guinea Cocoa & Coconut Research Institute Madang, Madang Province
- **56 – 82** To be established at Stewart Research Station, Madang, PNG

**Annex 1.3 List of designated germplasm for the International Coconut Genebank for South Asia**

<b>KASARAGOD</b>	Ayiramkachi
Borneo	Kulasekharam Green Dwarf
Standard Kudat	Kulasekharam Yellow Dwarf
Java	Kulasekharam Orange Dwarf
Malayan Orange Dwarf	Calangute
Malayan Green Dwarf	Nadora Tall
F.M.S.	Andaman Giant
S.S. Green	Andaman Ranguchan
S.S . Apricot	Car Nicobar
Philippines Lono	Auck Chung
San Ramon	Tamaloo
Cochin China	Kimos
Lifou Tall	Kimmai
British Solomon Islands	Katchal
Jamaica Sanblas	Campbell Bay
St. Vincent	Lakshdweep Micro
Blanchissuse	
Kenya Tall	<b>KIDU</b>
Cameroon Dwarf	West Coast Tall
West African Tall	Andaman Ordinary
Mawa Hybrid (PB 121	Benaulim
Zanzibar Tall	Tiptur Tall
Ceylon Tall	East Coast Tall
King Coconut	Chowghat Green Dwarf
Kappadam	Malayan Yellow Dwarf
Spicata	Philippines Ordinary

**Annex 1.4 List of designated germplasm for the International Coconut Genebank for Southeast and East Asia**

	<b>Cultivars</b>	<b>Code</b>	<b>Source</b>
1	Malayan Tall	MLT	Malaysia
2	Malayan Yellow Dwarf	MYD	Malaysia
3	Malayan Red Dwarf	MRD	Malaysia
4	Malayan Green Dwarf	MGD	Malaysia
5	Eo Brown Dwarf	EOD	Vietnam
6	Xiem Green Dwarf	XGD	Vietnam
7	Tam Quan Yellow Dwarf	TYD	Vietnam
8	Ta Tall	TAAT	Vietnam
9	Dau Tall	DAUT	Vietnam
10	Bung Tall (Bi Tall)	BIT	Vietnam
11	Giay Tall	GIT	Vietnam
12	Pluak Wan (Edible husk)	PKWT	Thailand
13	Pak Chok Tall	PCKT	Thailand
14	Maphrao So Tall	SOXT	Thailand



15	Kalok Thailand Tall	KLKT	Thailand
16	Thalai Roi Thailand Tall	TLRT	Thailand
17	Nalike Dwarf	NKED	Thailand
18	Maphrao Fai	FAID	Thailand
19	Bali Tall	BAT	Indonesia
20	Tenga Tall	TAT	Indonesia
21	Palu Tall	PUT	Indonesia
22	Sawarna Tall	SAT	Indonesia
23	Riau Tall	RUT	Indonesia
24	Mapanget Tall	MTT	Indonesia
25	Takome Tall	TET	Indonesia
26	Nias Yellow Dwarf	NYD	Indonesia
27	Bali Yellow Dwarf	BYD	Indonesia
28	Bali Green Dwarf	BYD	Indonesia
29	Jombang Green Dwarf	JGD	Indonesia
30	Sagerat Orange Dwarf	SOD	Indonesia
31	Salak Green Dwarf	SGD	Indonesia
32	Raja Brown Dwarf	RBD	Indonesia
33	Taganan Tall	TAGT	Philippines
34	Macapuno Tall	MACT	Philippines
35	Laguna Tall	LAGT	Philippines
36	Baybay Tall	BAYT	Philippines
37	Bago-Oshiro Tall	BAOT	Philippines
38	San Ramon Tall	SNRT	Philippines
39	Catigan Green Dwarf	CATD	Philippines
40	Pilipog Green Dwarf	PILD	Philippines
41	Aromatic Dwarf	AROD	Thailand
42	Hainan Tall	HAT	China
43	Cambodia tall	KAT	Côte d'Ivoire
44	West African Tall	W AT	Indonesia
45	Rennel Island Tall	RIT	Indonesia
46	Cameroon Red Dwarf	CRD	Indonesia
47	Tahiti Tall	TAT	Indonesia
48	Panama Tall	PNT	Côte d'Ivoire
49	Niu Leka Dwarf	NLAD	Côte d'Ivoire
50	Vanuatu Tall	VTT	Côte d'Ivoire
51	Indian West Coast Tall	WCT	India
52	Sri Lanka Tall	SLT	Sri Lanka

**Annex 1.5 List of designated germplasm for the International Coconut Genbank for Africa and Indian Ocean**

	<b>Cultivars</b>	<b>Code</b>	<b>Source</b>
1.	Andaman Giant Tall	AGT	India
2.	Andaman Ordinary Tall	ADOT	India
3.	Baybay Tall	BAYT	Philippines
4.	Cambodia Battambang Tall	KAT09	Cambodia
5.	Cambodia Koh Rong Tall	KAT10	Cambodia
6.	Cambodia Ream Tall	KAT07	Cambodia
7.	Cambodia Sre Cham Tall	KAT08	Cambodia

8.	Cambodia Tuk Sap Tall	KAT02	Cameroon
9.	Cameroon Kribi Tall	CKT	Cameroon
10.	Cameroon Red Dwarf	CRD	Cameroon
11.	Catigan Green Dwarf	CATD	Philippines
12.	Comoro Moheli Tall	CMT	Comoro
13.	Equatorial Guinea Green Dwarf	EGD	Equatorial Guinea
14.	Gazelle Peninsula Tall	GPT	Papua New Guinea
15.	Kappadam Tall	KPDT	India
16.	Karkar Tall	KKT	Papua New Guinea
17.	Kinabalan Green Dwarf	KIND	Philippines
18.	Laccadive Micro Tall	LMT	India
19.	Laccadive Ordinary Tall	LCT	India
20.	Madang Brown Dwarf	MBD	Papua New Guinea
21.	Malayan Green Dwarf	MGD	Malaysia
22.	Malayan Red Dwarf	MRD	Malaysia
23.	Malayan Tall	MLT	Malaysia
24.	Malayan Yellow Dwarf	MYD	Malaysia
25.	Markham Valley Tall	MVT	Papua New Guinea
26.	Mozambique Tall	MZT	Mozambique
27.	Niu Leka Dwarf	NLAD	Fiji
28.	Palu Tall	PUT	Indonesia
29.	Pilipog Green Dwarf	PILD	Philippines
30.	Rangiroa Tall	RGT	French Polynesia
31.	Rennell Island Tall	RIT	Solomon Islands
32.	Rotuman Tall	RTMT	Fiji
33.	Solomon Island Tall	SIT	Solomon
34.	Sri Lanka Green Dwarf	PGD	Sri Lanka
35.	Sri Lanka Tall Ambakelle	SLT02	Sri Lanka
36.	Tacunan Green Dwarf	TACD	Philippines
37.	Tagnanan Tall	TAGT	Philippines
38.	Tahitian Red Dwarf	TRD	French Polynesia
39.	Tahitian Tall	TAT	French Polynesia
40.	Takome Tall	TKT	Indonesia
41.	Tenga Tall	TGT	Indonesia
42.	Ternate Brown Dwarf	TBD	Indonesia
43.	Thailand Green Dwarf	THD	Thailand
44.	Thailand Tall Ko Samui	THT04	Thailand
45.	Thailand Tall Sawi	THT01	Thailand
46.	Tonga Tall	TONT	Tonga
47.	West African Tall Akabo	WAT03	Côte d'Ivoire
48.	West African Tall Mensah	WAT04	Côte d'Ivoire
49.	West African Tall Quidah	WAT06	Benin

**Annex 2.1. Key terms and their definitions as used in Chapter 2**

<b>ACIAR</b>	Australian Centre for International Agricultural Research
<b>Area</b>	An officially defined country, part of a country or all or parts of several countries (FAO 1990; revised FAO 1995; CEPM 1999; based on the World Trade Organization Agreement on the Application of Sanitary and Phytosanitary Measures)
<b>Area Freedom</b>	Pest-free area
<b>CABI</b>	Commonwealth Agriculture Bureau International
<b>CGIAR</b>	Consultative Group on International Agricultural Research
<b>COGENT</b>	International Coconut Genetic Resources Network
<b>Commodity</b>	A type of plant, plant product, or other article being moved for trade or other purposes (FAO 1990; revised ICPM 2001)
<b>Commodity Class</b>	A category of similar commodities that can be considered together in phytosanitary regulations (FAO 1990)
<b>Commodity Pest List</b>	A list of pests occurring in an area which may be associated with a specific commodity (CEPM 1996)
<b>Containment</b>	Application of phytosanitary measures in and around an infested area to prevent spread of a pest (FAO 1995)
<b>Delimiting Survey</b>	Survey conducted to establish the boundaries of an area considered to be infested by or free from a pest (FAO 1990)
<b>Detection Survey</b>	Survey conducted in an area to determine if pests are present (FAO 1990; revised FAO 1995)
<b>Entry (of a pest)</b>	Movement of a pest into an area where it is not yet present, or present but not widely distributed and being officially controlled (FAO 1995)
<b>EPPO</b>	European and Mediterranean Plant Protection Organization
<b>Establishment</b>	Perpetuation, for the foreseeable future, of a pest within an area after entry (FAO 1990; revised FAO 1995; IPPC 1997)
<b>Exotic</b>	Not native to a particular country, ecosystem or eco-area (applied to organisms intentionally or accidentally introduced as a result of human activities). As the Code is directed at the introduction of biological control agents from one country to another, the term "exotic" is used for organisms not native to a country (ISPM Pub. No. 3 1996)

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<b>FAO</b>	Food and Agriculture Organization of the United Nations
<b>Fumigation</b>	Treatment with a chemical agent that reaches the commodity wholly or primarily in a gaseous state (FAO 1990; revised FAO 1995)
<b>GATT</b>	General Agreement on Tariffs and Trade
<b>Germplasm</b>	Plants intended for use in breeding or conservation programmes (FAO 1990)
<b>Growing season</b>	Period of the year when plants are grown in an area
<b>Harmonization</b>	The establishment, recognition and application by different countries of phytosanitary measures based on common standards
<b>Host Pest List</b>	A list of pests that infest a plant species, globally or in an area
<b>Host Range</b>	Species of plants capable, under natural conditions, of sustaining a specific pest
<b>Infestation (of a commodity)</b>	Presence in a commodity of a living pest of the plant or plant product concerned. Infestation includes infection
<b>Inspection</b>	Official visual examination of plants, plant products or other regulated articles to determine if pests are present and/or to determine compliance with phytosanitary regulations
<b>International Standard for Phytosanitary Measures</b>	An international standard adopted by the Conference of FAO, the Interim Commission on phytosanitary measures or the Commission on phytosanitary measures, established under the IPPC
<b>International Standards</b>	International standards established in accordance with Article X, Paragraphs 1 and 2 of the IPPC
<b>Introduction</b>	The entry of a pest resulting in its establishment
<b>IPPC</b>	International Plant Protection Convention, as deposited in 1951 with FAO in Rome and as subsequently amended
<b>IPGRI</b>	International Plant Genetic Resources Institute
<b>ISPM</b>	International Standard for Phytosanitary Measures
<b>Monitoring Survey</b>	Ongoing survey to verify the characteristics of a pest population

<b>Non-Quarantine Pest</b>	Pest that is of no economic importance to the area and not present in the pathway; or present but does not cause serious infestation
<b>NPPO</b>	National Plant Protection Organization
<b>Pathway</b>	Any means that allows the entry or spread of a pest
<b>Pest</b>	Any species, strain or biotype of plant, animal or pathogenic agent injurious to plants or plant products
<b>Pest Categorization</b>	The process for determining whether a pest has or has not the characteristics of a quarantine pest or those of a regulated non-quarantine pest
<b>Pest Record</b>	A document providing information concerning the presence or absence of a specific pest at a particular location at a certain time, within an area (usually a country) under described circumstances
<b>Pest Risk Analysis (PRA)</b>	The process of evaluating biological or other scientific and economic evidence to determine whether a pest should be regulated and the strength of any phytosanitary measures to be taken against it
<b>Pest Risk Assessment (for quarantine pests)</b>	Evaluation of the probability of the introduction and spread of a pest and of the associated potential economic consequences
<b>Pest Risk Management (for quarantine pests)</b>	Evaluation and selection of options to reduce the risk of introduction and spread of a pest
<b>Phytosanitary Certificate</b>	Certificate patterned after the model certificates of the IPPC
<b>Phytosanitary Measure</b>	Any legislation, regulation or official procedure having the purpose to prevent the introduction and/or spread of quarantine pests, or to limit the economic impact of regulated non-quarantine pests
<b>Planting (including replanting)</b>	Any operation for the placing of plants in a growing medium, or by grafting or similar operations, to ensure their subsequent growth, reproduction or propagation
<b>Plants</b>	Living plants and parts thereof, including seeds and germplasm
<b>Plants for planting</b>	Plants intended to remain planted, to be planted or replanted

<b>Plants <i>in vitro</i></b>	A commodity class for plants growing in an aseptic medium in a closed container
<b>Point of entry</b>	Airport, seaport or land border point officially designated for the importation of consignments, and/or entrance of passengers
<b>Post-Entry Quarantine</b>	Quarantine applied to a consignment after entry
<b>Quarantine Pest</b>	A pest of potential economic importance to the area endangered thereby and not yet present there, or present but not widely distributed and being officially controlled
<b>Regional Plant Protection Organization (RPPO)</b>	An intergovernmental organization with the functions laid down by Article IX of the IPPC
<b>Spread</b>	Expansion of the geographical distribution of a pest within an area
<b>SPS</b>	Sanitary and Phytosanitary Agreement of the World Trade Organization
<b>Surveillance</b>	An official process which collects and records data on pest occurrence or absence by survey, monitoring or other procedures
<b>Survey</b>	An official procedure conducted over a defined period of time to determine the characteristics of a pest population or to determine which species occur in an area
<b>Technically justified</b>	Justified on the basis of conclusions reached by using an appropriate pest risk analysis or, where applicable, another comparable examination and evaluation of available scientific information
<b>Test</b>	Official examination, other than visual, to determine if pests are present or to identify pests
<b>Transparency</b>	The principle of making available, at the international level, phytosanitary measures and their rationale
<b>Treatment</b>	Officially authorized procedure for the killing or removal of pests or rendering pests infertile
<b>WTO</b>	World Trade Organization









### **Annex 2.3. Datasheets of major quarantine coconut pests**

It is not possible to publish the pest datasheets for all the pests that have the potential to be in the pathway with the exchange of germplasm as seednuts, embryos or pollen. Therefore model pest datasheets have been selected from the general pest groups as examples of the decisions that are made and the conclusions drawn. Much of the material has been taken from the datasheets that accompany the CABI Crop Protection Compendium.

#### **Annex 2.3.1 Data sheet of *Aceria guerreronis***

##### **Names and Taxonomy**

**Preferred Name:** *Aceria guerreronis* Keifer

##### **Taxonomic Position**

Domain : Eukaryota  
 Kingdom : Metazoa  
 Phylum : Arthropoda  
 Class : Arachnida  
 Subclass : Acari  
 Suborder : Prostigmata  
 Family : Eriophyidae

**Other name used** : *Eriophyes guerreronis* (Keifer)

**Bayer Code** : ACEIGU

##### **Common names**

English : coconut mite  
 Spanish : acaró del cocotero  
 French : acarien du cocotier; ravageur du cocotier (dahomey)  
 German : ilbe; Kokosblüten  
 Portuguese: Acaro da necrose do olho do coqueiro

##### **Host range**

*A. guerreronis* is the only species of eriophyoid mite considered to be a serious pest of coconuts, *Cocos nucifera*. It was first described in 1965 from specimens from Guerrero State, Mexico (Keifer 1965). Until reported from *Lytocaryum weddellianum*, a cocosoid palm species, it was only known from the coconut (Flechtmann 1989).

##### **Habitat**

*A. guerreronis* occurs under the perianth of young nutlets of *Cocos nucifera*, largely from a few weeks to 7-8 months after fertilization of the female flower. Migrating individuals may be found on the nut surface and populations have been recorded on seedlings.

**Primary hosts** : *Cocos nucifera* L. (coconut)

**Secondary hosts** : *Lytocaryum weddellianum*

**Infested plant stages** : Seedling and fruiting stages

**Infested plant parts** : Fruits/pods

**Geographic distribution**

Since first reported from Mexico, *A. guerreronis* has been reported from many coconut-growing regions of the Americas, in West Africa from Côte d'Ivoire to Nigeria (Hall and Espinosa 1981) and Gambia (Howard *et al.* 2001), Tanzania, India and Sri Lanka (Sathiyama *et al.* 1998; CRI/UNDP 2000).

**List of countries**

**Asia**

- India
  - Kerala: present, no further details (Sathiyamma *et al.* 1998)
  - Tamil Nadu: present, no further details (Muthiah and Bhaskaran 1999)
- Sri Lanka: present, no further details (CRI/UNDP 2000; Moore 2000)

**Africa**

- Benin: widespread (EPPO 2003)
- Cameroon: widespread (EPPO 2003)
- Côte d'Ivoire: widespread (EPPO 2003)
- Gambia: present, no further details (Howard *et al.* 2001)
- Nigeria: widespread (EPPO 2003)
- Sao Tome and Principe : widespread (EPPO 2003)
- Tanzania: widespread (CRI/UNDP 2000; EPPO 2003)
- Togo: widespread (EPPO 2003)

**Western Hemisphere**

- Anguilla: present, no further details (EPPO 2003)
- Bahamas: widespread (EPPO 2003)
- Brazil: widespread (EPPO 2003)
- Colombia: widespread (EPPO 2003)
- Cuba: widespread (EPPO 2003)
- Dominica: present, no further details (EPPO 2003)
- Dominican Republic: present, no further details (EPPO 2003)
- Grenada: widespread (EPPO 2003)
- Guadeloupe: present, no further details (EPPO 2003)
- Haiti: widespread (EPPO 2003)
- Jamaica: widespread (EPPO 2003)
- Martinique: present, no further details (EPPO 2003)
- Mexico: widespread (EPPO 2003)
- Puerto Rico: widespread (Howard *et al.* 1990)
- Saint Lucia : restricted distribution (EPPO 2003)
- Saint Vincent and the Grenadines: widespread (EPPO 2003)
- Trinidad and Tobago: widespread (EPPO 2003)
- USA
  - Florida: present, no further details (Howard *et al.* 1990)
  - Venezuela: widespread (EPPO 2003)

### **Biology and ecology**

Relatively little is known of the biology of *A. guerreronis*. A review of eriophyoid mites of coconuts by Moore and Howard (1996) focused on this species and much of the data are derived from that work.

The adult female coconut mite is 36-52  $\mu\text{m}$  wide and 205-255  $\mu\text{m}$  long (Keifer 1965). It can pass between the upper and lower sepals to reach the fruit surface covered by the perianth within a few weeks to a month after fertilization of the flower (Ortega *et al.* 1965; Mariau and Julia 1970; Hall and Espinosa 1981; Moore and Alexander 1987a; Howard and Abreu-Rodríguez 1991). The perianth almost completely covers the young fruit, providing protection against many hazards. During the first month of development the petals are tightly pressed to the fruit (Howard and Abreu-Rodríguez 1991), so that the perianth gives maximal protection. As the fruit develops, it becomes increasingly larger in relation to the perianth, and within about a month spaces develop between the coconut surface and the perianth which are sufficiently large to permit the entry of coconut mites. With a development cycle from egg to adult of about 10 days (Mariau 1977), mite numbers can build up rapidly. Spermatophores associated with coconut mite colonies have been observed underneath the perianth, showing that reproductive activities take place there. The fruits remain susceptible to mite attack almost throughout the whole development, but decreasingly so after the nut reaches full size. On more mature fruits (10-13 months), coconut mites are found rarely and in small numbers (Hall and Espinosa 1981; Moore and Alexander 1987a).

The coconut mite is found in tropical and subtropical climates, but populations can survive both short periods of freezing temperatures and periods of cool temperatures more prolonged than those normally encountered where coconut palms are grown (Howard *et al.* 1990). Some workers claim that coconut mite attacks are more severe in relatively dry climates or during the dry season of wetter climates (Zuluaga and Sanchez 1971; Griffith 1984). However, in other localities there is no detectable relationship between coconut mite populations and wet and dry weather (Doreste 1968; Mariau 1969 1977; Howard *et al.* 1990).

### **Means of movement and dispersal**

#### **Natural dispersal (non-biotic)**

The principal method by which coconut mites spread and colonize new palms, particularly over long distances, is almost certainly through aerial dispersal of inseminated female mites. The coconut palm provides a large target for aerially dispersed organisms, and air currents may carry the mites to racemes or to the more vertical leaves in the crown from which they may drop to inflorescences. Coconut mites can walk between touching inflorescences, and being negatively geotactic, tend to move from older to younger inflorescences (Moore and Alexander 1987a). Coconut mites walk at a rate of 20-100  $\mu\text{m}$  per second but are probably inefficient in finding sites to colonize. A high reproductive rate and rapid development compensate for inefficient dispersal and host-finding.

#### **Vector transmission**

Some dispersal may take place by phoresy, either on animals directly attracted to the inflorescences (for example, pollinating insects such as bees; rodents which feed on the fruits), or on those attracted by such animals (i.e., predatory lizards, birds, predaceous insects).

### **Seedborne spread**

This is unlikely as the mature nut is not infested by mites.

### **Agricultural practices**

Coconut seedlings can be infested and it is theoretically possible for dispersal to occur by movement of seedlings; this has not been reported.

### **Movement in trade**

This is unlikely as the mature nut is not infested by mites.

### **Plant parts liable to carry the pest in trade/transport:**

Fruits (including pods): Eggs, larvae, nymphs, adults; borne externally; visible under light microscope.

### **Plant parts not known to carry the pest in trade/transport:**

- Bark
- Bulbs/tubers/corms/rhizomes
- Growing medium accompanying plants
- Leaves
- Roots
- Stems (above ground)/shoots/trunks/branches
- True seeds (including grain)
- Wood

### **Economic impact**

Accurate crop loss assessments are rarely done, but estimates range from 7.5% (Julia and Mariau 1979) and 30% (Hernández 1977) to 60% (Griffith 1984) and some attacks may be so bad that farmers stop harvesting. Yield losses depend on cultivar, age, health and general maintenance of the crop, climate etc, but average copra losses may be 20-30% with premature nut fall and increased difficulty in dehusking (leading to greater labour requirements for this job) also contributing to economic loss.

### **Morphology**

Keifer (1965) first described *A. guerreronis*. The adult female coconut mite is vermiform, 36-52 µm wide and 205-255 µm long with two pairs of legs and a finely ringed body with several long setae. The genital opening of both sexes is positioned proximally, closely behind the legs.

### **Detection and inspection methods**

The scarring and distortion of nutlets can be observed from the ground, although with taller trees the use of binoculars may be necessary. Harvested nuts also bear the marks, although few, if any, mites will be found on these.

### **Diagnostic methods**

Eriophyoid mites can be easily distinguished by removing the sepals and examining them under a microscope. Full confirmation requires mounting and careful taxonomic study (Amrine and Manson 1996).

## Risk assessment of pest

	Seednuts	Embryo Cultures	Pollen
<b>Entry</b>	<i>A. guerreronis</i> occurs under the perianth of young nutlets of <i>Cocos nucifera</i> L. On more mature fruits (10-13 months), coconut mites are found rarely and in small numbers (Hall and Espinoza 1981; Moore and Alexander 1987a). Coconut seedlings can be infested and it is theoretically possible for dispersal to occur by movement of seedlings; this has not been reported.	Unlikely	Coconut mites can walk between touching inflorescences, and, being negatively geotactic, tend to move from older to younger inflorescences (Moore and Alexander 1987a). Some dispersal may take place by phoresy, either on animals directly attracted to the inflorescences (for example, pollinating insects such as bees; rodents which feed on the fruits).
<b>Establishment</b>	The coconut mite is found in tropical and subtropical climates.		
<b>Spread</b>	The principal method by which coconut mites spread and colonize new palms, particularly over long distances, is almost certainly through aerial dispersal of inseminated female mites.		
<b>Economic</b>	Accurate crop loss assessments are rarely done, but estimates range from 7.5% (Julia and Mariau 1979) and 30% (Hernández 1977) to 60% (Griffith 1984) and some attacks may be so bad that farmers stop harvesting.		
<b>Level of risk</b>	Low/unlikely	Very low	Low
<b>Pest status</b>	QP	QP?	QP
<b>Pest management</b>	Management of the pest on coconut nuts is by fumigation with MeBr. The pest in tissue cultures and in pollen is managed through inspection using a binocular microscope of at least 30x magnification. Infested shipments should be refused entry and destroyed.		

**Note :** *QP* – quarantine pest ; *NQP*- Non-quarantine pest

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### Annex 2.3.2. Data sheet of *Aspidiotus destructor*

#### Names and taxonomy

**Preferred name :** *Aspidiotus destructor* (Signoret 1869)

#### **Taxonomic Position**

Domain : Eukaryota  
 Kingdom : Metazoa  
 Phylum : Arthropoda  
 Class : Insecta  
 Order : Hemiptera  
 Suborder : Sternorrhyncha  
 Superfamily : Coccoidea  
 Family : Diaspididae

#### **Other names used**

*Aspidiotus cocotis* (Newstead 1893)  
*Aspidiotus lataniae* (Green 1896)  
*Aspidiotus simillimus translucens* Fernald  
*Aspidiotus translucens* (Cockerell and Robinson 1915)  
*Aspidiotus transparentis* (Green 1980)  
*Aspidiotus vastatrix* Leroy  
*Aspidiotus watanabei* (Takagi 1969)  
*Temnaspidotus destructor* (Signoret) Borchsenius

**Bayar Codes :** ASPDDE ; ASPDTR

#### **Common names**

English : coconut scale; transparent scale; bourbon scale; bourbon aspidiotus  
 Spanish : escama transparente; cochinita del cocotero; escama blanca del cocotero; escama del cocotero; escama del pino; cochinita blanca-amarilla del coco  
 French : cochenille du cocotier  
 German : Schildlaus; Kokospalmen  
 Italian : Cocciniglia del cocco

#### **Notes on taxonomy and nomenclature**

*Aspidiotus destructor* was first described by Signoret in 1869. Williams and Watson (1988) list synonyms and discuss nomenclature.

#### **Host range**

*A. destructor* is a highly polyphagous species. Davidson and Miller (1990) recorded it from hosts belonging to 75 genera in 44 plant families, but its host range is probably wider than this. Its hosts are typically perennial species and include many species of fruit trees, such as avocado, breadfruit, mango, guava and papaya.

Coconut is its favourite host; the undersurface of the leaves is mainly attacked but frond stalks, flower clusters and young fruit can also be affected. Older trees (over four years) or trees on well-drained soil are seldom seriously infested.



**Primary hosts:** *Cocos nucifera* L. (coconut), *Musa* (banana), *Actinidia*, *Elaeis guineensis* (African oil palm), *Mangifera indica* (mango).

**Infested plant stages:** Flowering stage, fruiting stage, seedling stage, and vegetative growing stage.

**Infested plant parts:** Fruits/pods, leaves, and stems.

#### **Geographic distribution**

*A. destructor* apparently originated in the Pacific Islands (Burger and Ulenberg 1990) but is now recorded in tropical and subtropical regions worldwide. It is present in nearly all countries where coconuts are grown. In the northern parts of its range, it is found only under glass (Danzig and Pellizzari 1998). It has been recorded under glass at a few botanic gardens in the UK (C Malumphy, Central Science Laboratory, UK, personal communication).

#### **List of countries**

##### **Europe**

- Germany: present, no further details (Danzig and Pellizzari 1998)
- Italy: present, no further details (Longo *et al.* 1995)
- Portugal
  - Madeira: present, no further details (CIE 1966)
- Russian Federation : present, no further details (CIE 1966; Danzig and Pellizzari 1998)
- Spain
  - Canary Islands : present, no further details (CIE 1966)

##### **Asia**

- Azerbaijan : present, no further details (CIE 1966; Danzig and Pellizzari 1998)
- Bangladesh: present, no further details (APPPC 1987)
- Bhutan: present, no further details (NHM 1985)
- Brunei Darussalam : present, no further details (Waterhouse 1993)
- Cambodia: present, no further details (CIE 1966; Waterhouse 1993)
- China
  - Fujian: present, no further details (CIE 1966; Tao 1999)
  - Guangdong : present, no further details (CIE 1966; Tao 1999)
  - Guangxi: present, no further details (CIE 1966; Tao 1999)
  - Hainan : present, no further details (CIE 1966; Tao 1999)
  - Hong Kong: present, no further details (CIE 1966)
  - Hubei: present, no further details (Tao 1999)
  - Hunan: present, no further details (Zhou *et al.* 1993)
  - Jiangsu: present, no further details (CIE 1966)
  - Jiangxi: present, no further details (Tao 1999)
  - Shandong: present, no further details (CIE 1966; Tao 1999)
  - Sichuan: present, no further details (Tao 1999)
  - Taiwan : present, no further details (NHM 1930; Takagi 1969; Wong *et al.* 1999)
  - Zhejiang: present, no further details (CIE 1966; Tao 1999)
- Georgia (Republic): present, no further details (CIE 1966; Danzig and Pellizzari 1998)

- India
  - Andaman and Nicobar Islands: present, no further details (NHM 1992)
  - Andhra Pradesh : present, no further details (CIE 1966)
  - Assam: present, no further details (CIE 1966)
  - Bihar: present, no further details (CIE 1966)
  - Gujarat : present, no further details (CIE 1966)
  - Indian Punjab : present, no further details (CIE 1966)
  - Jammu and Kashmir : present, no further details (CIE 1966)
  - Karnataka: present, no further details (CIE 1966)
  - Kerala: present, no further details (CIE 1966)
  - Lakshadweep : present, no further details (NHM 1940; CIE 1966)
  - Madhya Pradesh : present, no further details (CIE 1966)
  - Maharashtra : present, no further details (CIE 1966)
  - Orissa: present, no further details (CIE 1966)
  - Sikkim: present, no further details (CIE 1966)
  - Tamil Nadu : present, no further details (CIE 1966)
  - Uttar Pradesh : present, no further details (CIE 1966)
  - West Bengal: present, no further details (CIE 1966)
- Indonesia: present, no further details (Waterhouse 1993)
  - Irian Jaya: present, no further details (Williams and Watson 1988)
  - Java: present, no further details (CIE 1966)
  - Nusa Tenggara : present, no further details (CIE 1966)
  - Sumatra: present, no further details (CIE 1966)
- Iran: present, no further details (CIE 1966; Danzig and Pellizzari 1998)
- Japan: present, no further details (Kawai 1980)
  - Bonin Island : present, no further details (Nakahara 1982)
  - Honshu: present, no further details (CIE 1966)
- Korea, DPR: present, no further details (Danzig and Pellizzari 1998)
- Malaysia: present, no further details (Waterhouse 1993)
  - Peninsular Malaysia: present, no further details (CIE 1966)
  - Sabah: present, no further details (CIE 1966)
  - Sarawak: present, no further details (CIE 1966)
- Maldives: present, no further details (Watson *et al.* 1995)
- Myanmar: present, no further details (CIE 1966; Waterhouse 1993)
- Nepal: present, no further details (NHM 1967)
- Oman: widespread (Kinawy 1991)
- Pakistan: present, no further details (CIE 1966)
- Philippines: present, no further details (CIE 1966; Velasquez 1971; Waterhouse 1993)
- Saudi Arabia: present, no further details (NHM 1969; Danzig and Pellizzari 1998)
- Singapore: present, no further details (APPPC 1987; Waterhouse 1993)
- Sri Lanka: present, no further details (CIE 1966)
- Thailand: present, no further details (APPPC 1987; Waterhouse 1993)
- Vietnam: present, no further details (CIE 1966; Waterhouse 1993)
- Yemen: present, no further details (NHM 1958)

**Africa**

- Angola: present, no further details (CIE 1966)
- Benin: present, no further details (CIE 1966)
- British Indian Ocean Territory : present, no further details (CIE 1966)
- Burundi: present, no further details (CIE 1966)
- Cameroon: present, no further details (CIE 1966)
- Cape Verde: present, no further details (CIE 1966)
- Congo Democratic Republic:: present, no further details (Buyckx 1962)
- Congo: present, no further details (CIE 1966)
- Côte d'Ivoire: present, no further details (CIE 1966)
- Egypt: present, no further details (CIE 1966, Danzig and Pellizzari 1998)
- Eritrea: present, no further details (CIE 1966)
- Ethiopia: present, no further details (CIE 1966)
- Ghana: present, no further details (CIE 1966)
- Guinea-Bissau: present, no further details (CIE 1966)
- Guinea: present, no further details (CIE 1966)
- Kenya: present, no further details (CIE 1966)
- Madagascar: present, no further details (CIE 1966)
- Mauritania : present, no further details (CIE 1966)
- Mauritius: present, no further details (CIE 1966; Williams and Williams 1988)
- Mozambique: present, no further details (CIE 1966)
- Nigeria: present, no further details (CIE 1966)
- Rwanda: present, no further details (CIE 1966)
- Réunion: present, no further details (CIE 1966)
- Sao Tome and Principe : present, no further details (CIE 1966; Fernandes 1974)
- Senegal : present, no further details (CIE 1966)
- Seychelles: present, no further details (CIE 1966)
- Sierra Leone: present, no further details (CIE 1966)
- Somalia: present, no further details (CIE 1966)
- South Africa: present, no further details (CIE 1966)
- Sudan: present, no further details (CIE 1966)
- Tanzania: present, no further details (CIE 1966)
- Zanzibar : present, no further details (CIE 1966)
  - Togo: present, no further details (CIE 1966)
  - Uganda: present, no further details (CIE, 1966)
- Zambia: present, no further details (CIE 1966)
- Zimbabwe: present, no further details (NHM 1957)

**Western Hemisphere**

- Antigua and Barbuda: present, no further details (CIE 1966)
- Bahamas: present, no further details (NHM 1968)
- Barbados : present, no further details (CIE 1966; Bennett and Alam 1985)
- Belize : present, no further details (CIE 1966)
- Brazil
  - Alagoas : present, no further details (CIE 1966; Foldi 1988)
  - Amazonas : present, no further details (Foldi 1988; Claps *et al.* 2001)
  - Bahia : present, no further details (CIE 1966; Claps *et al.* 2001)
  - Ceara : present, no further details (CIE 1966; Claps *et al.* 2001)
  - Fernando de Noronha : present, no further details (Foldi 1988; Claps *et al.* 2001)
  - Maranhao : present, no further details (Foldi 1988; Claps *et al.* 2001)
  - Paraiba : present, no further details (CIE 1966; Foldi 1988; Claps *et al.* 2001)

- Pará: present, no further details (Foldi 1988; Claps *et al.* 2001)
- Pernambuco: present, no further details (CIE 1966; Foldi 1988; Claps *et al.* 2001)
- Piauí: present, no further details (CIE 1966; Foldi 1988; Claps *et al.* 2001)
- Rio Grande do Norte: present, no further details (CIE 1966; Foldi 1988; Claps *et al.* 2001)
- Rio de Janeiro: present, no further details (CIE 1966; Foldi 1988; Claps *et al.* 2001)
- Santa Catarina: present, no further details (Foldi 1988; Claps *et al.* 2001)
- Sao Paulo: present, no further details (CIE 1966; Foldi 1988; Claps *et al.* 2001)
- Sergipe: present, no further details (CIE 1966)
- Cayman Islands: present, no further details (CIE 1966)
- Chile
  - Easter Island: present, no further details (Charlin 1973; Claps *et al.* 2001)
- Colombia: present, no further details (CIE 1966; Kondo 2001)
- Costa Rica: present, no further details (CIE 1966)
- Cuba: present, no further details (CIE 1966)
- Dominica: present, no further details (CIE 1966)
- Dominican Republic: present, no further details (CIE 1966)
- Ecuador: present, no further details (CIE 1966; Kondo 2001)
- Galapagos Islands: present, no further details (CIE 1996)
- Grenada: present, no further details (CIE 1966)
- Guadeloupe: present, no further details (CIE 1966)
- Guatemala: present, no further details (CIE 1966)
- Guyana: present, no further details (CIE 1966)
- Haiti: present, no further details (CIE 1966)
- Honduras: present, no further details (CIE 1966)
- Jamaica: present, no further details (CIE 1966)
- Martinique: present, no further details (CIE 1966)
- Mexico: present, no further details (CIE 1966; Miller 1996)
- Montserrat: present, no further details (CIE 1966)
- Netherlands Antilles: present, no further details (CIE 1966)
- Nicaragua: present, no further details (CIE 1966)
- Panama: present, no further details (CIE 1966)
- Peru: present, no further details (CIE 1966)
- Puerto Rico: present, no further details (CIE 1966)
- Saint Kitts and Nevis: present, no further details (CIE 1966)
- Santa Lucia: present, no further details (CIE 1966)
- Saint Vincent and the Grenadines: present, no further details (CIE 1966)
- Suriname: present, no further details (CIE 1966)
- Trinidad and Tobago: present, no further details (CIE 1966)
- USA
  - Connecticut: present, no further details (Nakahara 1982)
  - Florida: present, no further details (CIE 1966)
  - Georgia (USA): present, no further details (Nakahara 1982)
  - Hawaii: present, no further details (Heu 2002)
  - Pennsylvania: present, no further details (Nakahara 1982)
  - Virgin Islands: present, no further details (CIE 1966)
- Venezuela: present, no further details (CIE 1966)

## Oceania

- American Samoa: present, no further details (CIE 1966; Williams and Watson, 1988; NAPPO 15:2)
- Australia
  - Australian Northern Territory: present, no further details (CIE 1966; CSIRO 2001)
  - Queensland: present, no further details (CSIRO 2001)
- Belau: present, no further details (CIE 1966; NAPPO 15:2)
- Federated states of Micronesia: present, no further details (Suta and Esguerra 1993; NAPPO 15:2)
  - Caroline Islands: present, no further details (CIE 1966)
- Fiji: widespread (CIE 1966; Williams and Watson 1988; NAPPO 15:2)
- French Polynesia: widespread (CIE 1966; Williams and Watson, 1988; NAPPO 15:2)
- Guam: present, no further details (CIE 1966; NAPPO 15:2)
- Marshall Islands: present, no further details (CIE 1966; NAPPO 15:2)
- New Caledonia: present, no further details (CIE 1966; NAPPO 15:2)
- Northern Mariana Islands: present, no further details (CIE 1966; NAPPO 15:2)
- Papua New Guinea: present, no further details (CIE 1966; NAPPO 15:2)
- Samoa: widespread (CIE 1966; Williams and Watson 1988)
- Solomon Islands: present, no further details (CIE 1966; NAPPO 15:2)
- Tuvalu: present, no further details (NAPPO 15:2)
- Vanuatu: present, no further details (NAPPO 15:2)
  - Wallis and Futuna: present, no further details (CIE 1966; NAPPO 15:2)

## Biology and ecology

*A. destructor* reproduces sexually. Males locate unmated females by following pheromones released by them. The life cycle of *A. destructor* typically lasts for 32-34 days. In one study the life cycle was found to be 32 days for females and 27 days for males (Tabibullah and Gabriel 1973; Taylor 1935).

Each female deposit 20 to 50 eggs under her scale cover over a few days. In China on *Actinidia*, the average number of eggs laid by one female was 32-42 (Zhou *et al.* 1993). At room temperature (26-28°C), the egg stage lasted for five days, the larval stage lasted 17 days, the pre-oviposition stage in adult females lasted 25 days, the female generation lasted 44 days and the male generation lasted 38 days (Zhou *et al.* 1993). In the Philippines, on coconut, the egg stage lasted for eight days in both sexes (Tabibullah and Gabriel 1973). After hatching, the nymphs crawl under the scale edge out into the open and colonize the undersides of leaves and tender shoots. They drop off the leaves easily, so mortality is high during heavy rain.

In China, *A. destructor* produced three generations annually, with the fertilized females overwintering on the stems of *Actinidia* trees (Zhou *et al.* 1993). In Japan on tea plants, *A. destructor* had only one generation per year (Murakami 1970). However, in tropical conditions in Trinidad reproduction is continuous (Goberdhan 1962).

The dispersal phase of *A. destructor* is the first instar, or crawler, which has legs. Crawlers can walk up to perhaps 1 m, but can be distributed across much greater distances by wind, flying insects and birds and transport of infested plant material by man.

## Economic impact

*A. destructor* is potentially the most destructive pest species on coconut, wherever it occurs in the world (Chua and Wood 1990). Before the introduction of successful

biological control in 1955, copra production in Principe fell from 1400 to 500 tons per year owing to an invasion of *A. destructor* (Rosen 1990a). After a heavy attack by *A. destructor* on coconuts in Côte d'Ivoire, yield was reduced by at least 25% over the next 2-3 years, although some heavily infested trees were able to catch up production in the two years after elimination of the infestation (Mariau and Julia 1977).

This species is highly polyphagous and therefore can easily be re-introduced, even if it is successfully controlled on the primary host crop.

### **Symptoms**

On leaves, *A. destructor* causes yellow spots to develop beneath the insects, due to the toxicity of saliva injected in to plant tissues while feeding. Entire leaves may turn yellow to brown and fall, and fruits may be discoloured, stunted or fall prematurely. The bright yellow colour of affected coconut palms is clearly visible from a great distance. The undersurface of the leaves is mainly attacked, but frond stalks, flower clusters and young fruit can also be affected. In extreme cases, the leaves dry out, entire fronds drop off and the crown dies.

### **Descriptors**

Fruits/pods	:	black or brown lesions; external feeding; discoloration
Leaves	:	necrotic areas; abnormal colours; abnormal leaf fall
Stems	:	external feeding

### **Morphology**

Jalaluddin and Mohanasundaram (1992) describe the morphology of different instars and the adult female and male of *A. destructor*.

### **Egg**

The eggs are yellow and very small. They are laid under the scale around the body of the female.

### **Larva and pupa**

Females have two nymphal stages, while males have two feeding nymphal stages, followed by non-feeding pre-pupal and pupal stages (four immature stages altogether) (Tabibullah and Gabriel 1973).

The first-instar larvae are about 1mm long, yellowish-brown, oval and translucent. Second-instar females become immobile and secrete a translucent wax scale cover. The second-instar males are smaller than the females. They group together, secrete a filamentous waxy material and become immobile. The male pre-pupal and pupal stages are spent under the scale produced by the second instar stage.

### **Adults**

The scale cover of the adult female is oval to circular, 1.5-2.0 mm across, fairly flat, very thin and translucent. The pale yellow exuviae are more or less central on the scale (Williams and Watson 1988). The yellow adult female under the scale is 0.6-1.1 mm long.

The adult male scale cover is redder than the female's, smaller and more oval (Williams and Watson 1988). The male has one pair of wings and is motile.

### **Similarities to other species**

*Aspidiotus excisus* looks very similar to *A. destructor* in life but can be distinguished

when slide-mounted adult females are examined microscopically; the median lobes on the pygidium of *A. excisus* are recessed into the margin, whereas in *A. destructor* they are not recessed (Williams and Watson 1988).

### Control

#### **Introduction**

*A. destructor* is highly polyphagous and therefore can easily be re-introduced, even if it is successfully controlled on the primary host crop.

#### **Phytosanitary Measures**

Dharmaraju and Laird (1984) describe the transport of *A. destructor* around Oceania, mainly through human agency. They emphasize the importance of rigid quarantine procedures.

#### **Risk assessment of pest**

	Seednuts	Embryo cultures	Pollen
<b>Entry</b>	The dispersal phase of <i>A. destructor</i> is the first instar, or crawler, which has legs. Crawlers can walk up to perhaps 1 m, but can be distributed across much greater distances by wind, flying insects and birds and transport of infested plant material by man.	Not an internal pest	Flower clusters and young fruit can also be affected, but would be visible to naked eye in pollen. The scale cover of the adult female is oval to circular, 1.5-2.0 mm across. Would be eliminated by sieving pollen after collection.
<b>Establishment</b>	It is present in nearly all countries where coconuts are grown.		
<b>Spread</b>	It is present in nearly all countries where coconuts are grown.		
<b>Economic</b>	<i>A. destructor</i> is potentially the most destructive pest species on coconut, wherever it occurs in the world (Chua and Wood 1990).		
<b>Level of risk</b>	Very low	Minimal	Minimal
<b>Pest status</b>	QP	NQP	NQP
<b>Pest management</b>	Management of pest on nuts is through fumigation with MeBr.		

**Note :** *QP* – quarantine pest ; *NQP*- Non-quarantine pest

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### Annex 2.3.3. Data sheet of Coconut cadang-cadang viroid

#### Names and taxonomy

**Preferred name :** Coconut cadang-cadang viroid

#### **Taxonomic position**

Virus group : Viroids  
Family : Pospiviroidae  
Genus : Cocadviroid

**Other name used:** Palm cadang-cadang viroid

#### **Common names**

English : cadang cadang disease; yellow mottling disease

#### **Notes on taxonomy and nomenclature**

Classification schemes for viroids (Flores 1995) are still evolving. The Viroid Study Group of the International Committee for the Taxonomy of Viruses is developing a classification system for the viroids. Family (Viroidae), subfamily, genus, species and variant taxa are proposed.

#### Host range

There is natural infection of all commercial cultivars of *Cocos nucifera* L. (coconut palm) in the Philippines, *Elaeis guineensis* (oil palm), *Corypha elata* (buri palm), and *Livistona rotundifolia*. Experimental hosts susceptible to mechanical inoculation with partially purified CCCVd include a wide range of Arecaceae (Imperial *et al.* 1985; Randles 1987), and possibly Maranta species. Related molecules are also found naturally in members of the Arecaceae and Pandanaceae, as well as herbaceous monocotyledons including Zingiberaceae, Marantaceae and Commelinaceae (Hanold and Randles, 1991a, b).

**Primary hosts:** *Cocos nucifera* (coconut), *Arenga pinnata* (sugar palm), *Areca catechu* (betelnut palm), *Borassus*, *Chloris* (fingergrasses), *Elaeis guineensis* (African oil palm), *Phoenix dactylifera* (date-palm).

**Infected Plant Stages:** All growing stages

**Infected Plant Parts:** Whole plant, leaves, inflorescence, and fruits/pods

#### Geographic distribution

CCCVd is found in the central Philippines (see Hanold and Randles 1991a and Randles and Rodriguez 2003 for distribution maps) and is spreading slowly but continuously north and south from its site of first appearance in the 1930s in Albay province (see also CABI/EPPO 1998, No. 312).

#### List of countries

##### **Asia**

- Philippines: restricted distribution (Randles and Imperial 1984; EPPO 2003)

**Oceania**

- Guam: absent, unreliable record (EPPO 2003)
- Solomon Islands: restricted distribution (EPPO 2003)

**Biology and ecology**

The lethal cadang-cadang disease was first recorded in the central Philippines. However, since its first appearance in the 1930s it has been spreading with no means of control and it is thus regarded as a threat to the whole region. The disease was shown to have viroid aetiology (Randles *et al.* 1976), and the pathogen was named coconut cadang-cadang viroid (CCCVd). Its epidemiology has been studied, but the natural mode of spread is still unknown.

The disease occurs rarely in palms of pre-bearing age, but its incidence increases with age thereafter. Transmission in the field may occur by several means (Randles *et al.* 1992). CCCVd can be detected in coconut husk, embryo and pollen by molecular diagnostic methods and is seed-transmitted at the low rate of about one in 300 (Randles and Imperial 1984; Hanold and Randles 1991a; Pacumbaba *et al.* 1994). A low rate of transmission by artificial pollination has been demonstrated (Pacumbaba *et al.* 1994).

Over a period of about 7-15 years (depending on their age), infected palms progress through three well-defined disease stages which were shown to be associated with distinct changes in the molecular structure of the viroid (Haseloff *et al.* 1982). This is a unique feature of CCCVd. In its 246-nucleotide form, CCCVd is the smallest known viroid, as well as being the smallest known pathogen. The closely related coconut tinangaja viroid (CTiVd) causes a similar disease in Guam, and together CCCVd and CTiVd are the only known viroids to affect monocotyledons and to kill their host.

Using a mechanical inoculation technique and purified CCCVd a large number of varieties and hybrids on coconut seedlings were screened for possible resistance or tolerance to the pathogen, but so far none has been identified. Recently, mutants of CCCVd associated with a 'brooming' effect of the crown (by reduction of leaf lamina), have been identified. The increased severity of pathogenicity was shown to be due to only a few point mutations (Rodriguez 1993; Rodriguez and Randles 1993). A range of molecules related to CCCVd in structure and nucleotide sequence (one of which is suspected to be a viroid agent associated with the well-known 'genetic' orange spotting syndrome (GOS) of oil palm) has been detected in coconut and other tropical monocotyledons in many locations both inside and outside the Philippines, causing quarantine concerns (Hanold and Randles 1991a, b). Their exact degree of relatedness to CCCVd and possible economic risks posed by them still need to be investigated.

**Seedborne aspects****Incidence**

CCCVd can be detected in coconut husks, embryos and pollen (Hanold and Randles 1991). A low rate of transmission to seeds by artificial pollination has been demonstrated (Pacumbaba *et al.* 1994).

**Effect on seed quality**

Nuts of infected palms show characteristic rounding and scarifications (Hanold and

Randles 1991).

### **Pathogen transmission**

CCCVd is seed-transmitted at a low rate of about 1 in 300 (Randles and Imperial 1984; Hanold and Randles, 1991; Pacumbaba *et al.*, 1994), but the presence of CCCVd can only be determined by molecular diagnostic methods.

### **Seed health tests**

None developed, but CCCVd can be detected in coconut husk and embryo by molecular DNA hybridization (Hanold and Randles 1991).

### **Economic impact**

It was estimated that, since cadang-cadang was first recorded, it has killed more than 30 million coconut palms. This would amount to a loss of produce worth US\$ 2400-3000 million at the rate of US\$ 80-100 (depending on copra prices) for each planting (palm) site. Annual yield loss of about 22 000 t of copra has been attributed to this disease in the Philippines (Zelazny *et al.* 1982). Coconut is both a vital subsistence and major cash crop in developing countries, and most coconut products are supplied by smallholders. Therefore, CCCVd must be considered a serious economic threat (Hanold and Randles 1991a).

### **Morphology**

CCCVd (246-nucleotide form) is the smallest known viroid, as well as the smallest known pathogen. It is a member of the potato spindle tuber viroid (PSTVd) group with the same central conserved region (Keese and Symons 1987; Koltunow and Rezaian 1989). Physical properties and melting profile are similar to those of other viroids (Randles *et al.* 1982). CCCVd is the only viroid known to show molecular changes during disease progression. These include a 247-nucleotide form (by addition of one cytosine residue), and 296- and 297-nucleotide forms (by duplication of the right hand terminus of 246- and 247-nucleotide forms, respectively; Haseloff *et al.* 1982). The order of appearance of forms is normally 246, 247, 296, 297 nucleotides (Imperial and Rodriguez 1983; Randles 1987). Associated dimeric forms are also usually present. This is a unique feature of CCCVd and CTiVd. The presence of dimers as a major component of viroid preparations is only known for CCCVd and CTiVd (Randles 1985).

### **Similarities to other species**

The only known close relative is coconut tinangaja viroid (CTiVd) which causes a similar disease in Guam (Boccardo 1985). CTiVd has 256 nucleotides with 64% sequence homology to CCCVd (Keese *et al.* 1988). It has dimeric forms, but does not show changes in size during disease progression.

### **Detection and inspection methods**

Symptoms must be considered unreliable for detection of CCCVd (Hanold and Randles 1991a) therefore quarantine inspection will not detect it. Symptoms can resemble physiological changes due to poor nutrition and water stress in the field, typhoon damage, and sterility. Leaf spots resemble insect feeding damage and microbial infection. In practice, early-stage infection is difficult to diagnose even for experienced workers, whereas mid- and late-stage disease may not be specific. Symptomatic diagnosis usually depends on repeated observations of the disease on the same tree. Therefore, viroid isolation is essential to confirm diagnosis, which is done by extracting from leaf or root tips (Randles and Rodrigues 2003).

Growth in quarantine confinement is unsuitable, since symptom development is slow and unreliable, and no indicator species are available. Thus, strict certification of origin is essential (Anonymous 1987). Reliable diagnosis is achievable only by molecular methods (see Diagnostic Methods).

#### **Diagnostic methods**

Only molecular methods are applicable. It is essential that diagnosis be carried out in suitably equipped facilities and by personnel experienced in working with RNA due to its special requirements (Hanold 1993). Reliable methods for extraction of CCCVd and related nucleic acids from tissues of coconut, other palms and herbaceous monocotyledons have been developed (Hanold and Randles 1991b; Imperial *et al.* 1985). Analysis of extracts may be carried out by polyacrylamide gel electrophoresis (PAGE) followed by silver staining (Imperial *et al.* 1985). Gels of 5-20% can be used in one- or two-dimensional assays, but "Return" PAGE is generally unsuitable (Hanold 1993). For higher sensitivity and accuracy, radioactively labelled probes may be used (Haseloff *et al.* 1982; Hanold 1993) either in dot-blot or Northern blot hybridization assays at varying stringencies (Hanold and Randles 1991a, b). Non-radioactive labelling methods have generally been found to be insufficiently sensitive or specific (Hanold 1993). The most conclusive results are obtained by Northern blot hybridization, as similarities in both molecular structure and nucleotide sequence can be assessed (Hanold and Randles 1991a).

#### **Control**

No methods, apart from exclusion, are known to control CCCVd. No genetically resistant or tolerant coconut cultivars have been identified (Randles 1985). Eradication campaigns have been unsuccessful; replacing infected palms does not decrease disease incidence, but has allowed reduced production to continue in affected areas (Randles 1987). Vector control is not applicable because the means of spread are unknown. Mild-strain protection may be possible, but will need investigation (Hanold and Randles 1991a).

#### **Regulatory Control**

Movement of living coconut tissue, such as seedlings, nuts for germination and pollen out of the general cadang-cadang region in the central Philippines has been stopped, both to unaffected areas of the country and internationally.

Growth in quarantine confinement is unsuitable, as symptom development is slow and unreliable, and no indicator species are available. Thus, strict certification of origin is essential (Anon. 1987; Frison, Putter and Diekmann 1993).

#### **Risk assessment of pest**

	<b>Seednuts</b>	<b>Embryo cultures</b>	<b>Pollen</b>
<b>Entry</b>	CCCVd can be detected in coconut husk, embryo and pollen by molecular diagnostic methods and is seed-transmitted at the low rate of about one in 300 (Randles and Imperial 1984;	CCCVd can be detected in coconut husk, embryo	A low rate of transmission by artificial pollination has been demonstrated (Pacumbaba <i>et al.</i> 1994)

	Hanold and Randles 1991a; Pacumbaba <i>et al.</i> 1994).		
<b>Establishment</b>	Transmission in the field may occur by several means (Randles <i>et al.</i> 1992).		
<b>Spread</b>	No methods, apart from exclusion, are known to control CCCVd. Its epidemiology has been studied, but the natural mode of spread is still unknown.		
<b>Economic</b>	CCCVd must be considered a serious economic threat (Hanold and Randles 1991a).		
<b>Level of risk</b>	Low	High	Low
<b>Pest status</b>	QP	QP	QP
<b>Pest management</b>	Growth in quarantine confinement is unsuitable, since symptom development is slow and unreliable, and no indicator species are available. Thus strict certification of origin is essential (Anonymous 1987). Reliable diagnosis is achievable only by molecular methods (see Diagnostic Methods).		

**Note :** QP – quarantine pest ; NQP- Non-quarantine pest

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### Annex 2.3.4. Data sheet of *Ferrisia virgata*

#### Names and taxonomy

**Preferred name** : *Ferrisia virgata* Cock

#### **Taxonomic position**

Domain : Eukaryota  
 Kingdom : Metazoa  
 Phylum : Arthropoda  
 Class : Insecta  
 Order : Hemiptera  
 Suborder : Sternorrhyncha  
 Superfamily: Coccoidea  
 Family : Pseudococcidae

#### **Other names used**

*Dactylopius ceriferus* Newstead  
*Dactylopius dasyliirii* (Cockerell 1896)  
*Dactylopius magnolicida* (King 1902)  
*Dactylopius segregatus* (Cockerell 1893)  
*Dactylopius setosus*  
*Dactylopius taplini* (Green 1896)  
*Dactylopius virgatus* Cockerell  
*Ferrisiana virgata* (Cockerell)  
*Heliococcus malvastrus* (McDaniel 1962)  
*Pseudococcus bicaudatus* Keuchenius  
*Pseudococcus ceriferus*  
*Pseudococcus marchali* Vayssière  
*Pseudococcus setosus*  
*Pseudococcus talina*  
*Pseudococcus virgatus* Cockerell

**Bayer codes** : FERRVI; PSECVI

#### **Common names**

English : guava mealybug; lamtoro luis; spotted mealybug; striped mealybug;  
 Tailed coffee mealybug; tailed mealybug; white-tailed mealybug  
 Spanish : cochinilla embandada  
 French : cochenille rayée  
 German : weisse lamtoro-laus  
 Netherlands: witte lamtoro-luis  
 South African: gestrepte witluis

#### **Notes on taxonomy and nomenclature**

It has long been known that specimens normally identified as *Ferrisia virgata* were either biparental or parthenogenetic (Williams 1996). Nur (1977), using electrophoretic techniques, indicated that there was one uniparental species and at least two biparental species, while Miller and Kostarab (1979) mentioned seven species. The type species of *F. virgata* is biparental and Williams (1985) illustrated the



uniparental form, showing morphological differences between the two forms. The uniparental form was later described as *F. consobrina* (Williams and Watson 1988) and subsequently synonymized with *F. malvastra* by Williams (1996). Early records of *F. virgata* from all regions of its distribution need to be verified due to confusion with *F. malvastra* (Ben-Dov, 1994).

#### **Host range**

*F. virgata* is one of the most highly polyphagous mealybugs known, attacking plant species belonging to some 150 genera in 68 families. Many of the host species belong to the Leguminosae and Euphorbiaceae. Among the hosts of economic importance are avocado, banana, betel vine, black pepper, cassava, cashew, cauliflower, citrus, cocoa, coffee, cotton, custard apple, eggplant, grapevine, guava, jute, lantana, *Leucaena*, litchi, mango, oil palm, pigeon pea, pineapple, soybean and tomato.

**Primary hosts:** *Leucaena leucocephala* (horse tamarind), *Coffea* (coffee), *Abelmoschus esculentus* (okra), *Acalypha* (Copperleaf), *Anacardium occidentale* (cashew nut), *Ananas comosus* (pineapple), *Annona*, *Cajanus cajan* (pigeon pea), *Cocos nucifera* L. (coconut)

**Affected Plant Stages:** Flowering stage, fruiting stage, post-harvest, and vegetative growing stage

**Affected Plant Parts:** Leaves, stems, growing points, and fruits/pods

#### **Geographic distribution**

The genus *Ferrisia* is apparently of New World origin (Williams 1996). However, *F. virgata* has spread to all zoogeographical regions, mainly in the tropics, but often extends well into the temperate regions (see CIE distribution map No. 219 (1966) and Ben-Dov (1994)).

Early geographical records of *F. virgata* need to be verified due to confusion with *F. malvastra* (Ben-Dov 1994).

#### **List of countries**

##### **US (minor outlying islands)**

- Johnston Island: present, no further details (CIE 1966)
- Wake Island: present, no further details (Ben-Dov 1994)

##### **Asia**

- Bangladesh: present, no further details (APPPC 1987; CIE 1966)
- Brunei Darussalam: present, no further details (Waterhouse 1993)
- Cambodia: present, no further details (CIE 1966; Waterhouse 1993; Ben-Dov 1994)
- China
  - Guangdong: present, no further details (CIE 1966)
  - Hong Kong: present, no further details (CIE 1966)
  - Taiwan: present, no further details (CIE 1966; Ben-Dov 1994)
- India
  - Andhra Pradesh: present, no further details (CIE 1966)
  - Assam: present, no further details (Mustafee 1970)
  - Bihar: present, no further details (CIE 1966)
  - Goa: present, no further details (Ali 1972)

- Indian Punjab: present, no further details (CIE 1966)
- Karnataka: present, no further details (CIE 1966)
- Kerala: present, no further details (CIE 1966)
- Madhya Pradesh: present, no further details (CIE 1966)
- Maharashtra: present, no further details (CIE 1966)
- Orissa: present, no further details (CIE 1966)
- Rajasthan: present, no further details (CIE 1966)
- Tamil Nadu: present, no further details (CIE 1966)
- Tripura: present, no further details (CIE 1966)
- Uttar Pradesh: present, no further details (CIE 1966)
- West Bengal: present, no further details (CIE 1966)
- Indonesia: present, no further details (Waterhouse 1993)
  - Irian Jaya: present, no further details (Williams and Watson 1988)
  - Java: present, no further details (CIE 1966; Ben-Dov 1994)
  - Sumatra: present, no further details (CIE 1966)
- Japan: present, no further details (Ben-Dov 1994)
- Laos: present, no further details (Waterhouse 1993)
- Malaysia: present, no further details (Waterhouse 1993)
  - Peninsular Malaysia: present, no further details (CIE 1966)
  - Sabah: present, no further details (CIE 1966)
  - Sarawak: present, no further details (CIE 1966)
- Myanmar: present, no further details (CIE 1966; Waterhouse 1993)
- Pakistan: present, no further details (CIE 1966)
- Philippines: present, no further details (CIE 1966; Waterhouse 1993)
- Saudi Arabia: present, no further details (Ben-Dov 1994)
- Singapore: present, no further details (Waterhouse 1993)
- Sri Lanka: present, no further details (CIE 1966; Ben-Dov 1994)
- Thailand: present, no further details (CIE 1966; Waterhouse 1993; APPPC 1987; Ben-Dov 1994)
- United Arab Emirates: present, no further details (CIE 1966)
- Vietnam: present, no further details (Ben-Dov 1994)
- Yemen: present, no further details (CIE 1966)

### **Africa**

- Angola: present, no further details (CIE 1966)
- British Indian Ocean Territory: present, no further details (Ben-Dov 1994)
- Cameroon: present, no further details (CIE 1966)
- Comoros: present, no further details (Ben-Dov 1994)
- Congo Democratic Republic: widespread (Buickx)
- Congo: present, no further details (CIE 1966; Ben-Dov 1994)
- Côte d'Ivoire: present, no further details (CIE 1966; Ben-Dov 1994)
- Egypt: present, no further details (CIE 1966)
- Ethiopia: present, no further details (CIE 1966)
- Ghana: present, no further details (CIE 1966; Ben-Dov 1994)
- Guinea: present, no further details (Ben-Dov 1994)
- Kenya: present, no further details (CIE 1966; Ben-Dov 1994)
- Madagascar: present, no further details (CIE 1966; Ben-Dov 1994)
- Malawi: present, no further details (CIE 1966)
- Mauritius: present, no further details (CIE 1966; Ben-Dov 1994)
- Mozambique: present, no further details (CIE 1966)
- Nigeria: present, no further details (CIE 1966)
- Sao Tome and Principe: present, no further details (CIE 1966)
- Senegal: present, no further details (CIE 1966)

- Seychelles: present, no further details (CIE 1966; Ben-Dov 1994)
- Sierra Leone: present, no further details (CIE 1966)
- Somalia: present, no further details (CIE 1966)
- South Africa: present, no further details (CIE 1966; Ben-Dov 1994)
- Sudan: present, no further details (CIE 1966; Ben-Dov 1994)
- Tanzania: present, no further details (CIE 1966; Bohlen 1973; Ben-Dov 1994)
- Togo: present, no further details (CIE 1966)
- Uganda: present, no further details (CIE 1966; Ben-Dov 1994)
- Zambia: present, no further details (CIE 1966)
- Zimbabwe: present, no further details (CIE 1966)

### **Western Hemisphere**

- Argentina: present, no further details (CIE 1966; Ben-Dov 1994)
- Bahamas: present, no further details (Ben-Dov 1994)
- Barbados: present, no further details (CIE 1966; Ben-Dov 1994)
- Belize: present, no further details (Ben-Dov 1994)
- Bermuda: present, no further details (CIE 1966; Ben-Dov 1994)
- Bolivia: present, no further details (Ben-Dov 1994)
- Brazil: present, no further details (Ben-Dov 1994)
  - Paraiba: present, no further details (CIE 1966)
  - Rio Grande do Norte: present, no further details (CIE 1966)
  - Rio de Janeiro: present, no further details (CIE 1966)
  - Sao Paulo: present, no further details (CIE 1966)
- Cayman Islands: present, no further details (Ben-Dov 1994)
- Colombia: present, no further details (CIE 1966; Ben-Dov 1994)
- Costa Rica: present, no further details (CIE, 1966; Ben-Dov, 1994)
- Cuba: present, no further details (CIE 1966; Ben-Dov 1994)
- Dominica: present, no further details (Ben-Dov 1994)
- Ecuador: present, no further details (Ben-Dov 1994)
- Guatemala: present, no further details (Ben-Dov 1994)
- Guyana: present, no further details (CIE 1966; Ben-Dov 1994)
- Haiti: present, no further details (CIE 1966)
- Honduras: present, no further details (CIE 1966; Ben-Dov 1994)
- Jamaica: present, no further details (CIE 1966; Ben-Dov 1994)
- Martinique: present, no further details (CIE 1966; Ben-Dov 1994)
- Mexico: present, no further details (CIE 1966; Ben-Dov 1994)
- Netherlands Antilles: present, no further details (CIE 1966)
- Nicaragua: present, no further details (CIE 1966; Ben-Dov 1994)
- Panama: present, no further details (CIE 1966; Ben-Dov 1994)
- Paraguay: present, no further details (Ben-Dov 1994)
- Peru: present, no further details (Ben-Dov 1994)
- Puerto Rico: present, no further details (CIE 1966; Ben-Dov 1994)
- Saint Kitts and Nevis: present, no further details (Ben-Dov 1994)
- Suriname: present, no further details (CIE 1966; Ben-Dov 1994)
- Trinidad and Tobago: present, no further details (CIE 1966; Ben-Dov 1994)
- USA
  - Alabama: present, no further details (CIE 1966)
  - California: present, no further details (CIE 1966; Ben-Dov 1994)
  - Florida: present, no further details (CIE 1966; Ben-Dov 1994)
  - Hawaii: present, no further details (CIE 1966)
  - Louisiana: present, no further details (CIE 1966; Ben-Dov 1994)
  - Maryland: present, no further details (CIE 1966)
  - Mississippi: present, no further details (CIE 1966)

- New Mexico: present, no further details (CIE 1966; Ben-Dov 1994)
- New York: present, no further details (Ben-Dov 1994)
- Pennsylvania: present, no further details (CIE 1966)
- Texas: present, no further details (CIE 1966; Ben-Dov 1994)
- United States Virgin Islands: present, no further details (Ben-Dov 1994)
- Venezuela: present, no further details (CIE 1966; Ben-Dov 1994)

### **Oceania**

- Australia
  - Australian Northern Territory: present, no further details (CIE 1966; Ben-Dov 1994)
  - Queensland: present, no further details (CIE 1966; Ben-Dov, 1994)
- Belau: present, no further details (Ben-Dov 1994)
- Cook Islands: present, no further details (Ben-Dov 1994)
- Federated states of Micronesia
  - Caroline Islands: present, no further details (CIE 1966)
- Fiji: present, no further details (CIE 1966; Ben-Dov 1994)
- French Polynesia: present, no further details (Ben-Dov 1994)
- Kiribati: present, no further details (CIE 1966; Ben-Dov 1994)
- Marshall Islands: present, no further details (CIE 1966; Ben-Dov 1994)
- New Caledonia: present, no further details (CIE 1966; Ben-Dov 1994)
- Northern Mariana Islands: present, no further details (Ben-Dov 1994)
- Papua New Guinea: present, no further details (CIE 1966; Ben-Dov 1994)
- Samoa: present, no further details (Ben-Dov 1994)
- Solomon Islands: present, no further details (Ben-Dov 1994)
- Tonga: present, no further details (CIE 1966; Ben-Dov 1994)
- Tuvalu: present, no further details (Ben-Dov 1994)
- Vanuatu: present, no further details (CIE 1966; Ben-Dov 1994)
- Wallis and Futuna: present, no further details (CIE 1966)

### **Biology and ecology**

There are several papers on the biology of *F. virgata* but these needs to be verified due to the confusion with *F. malvastra*, particularly in India where both species occur. *F. virgata* is biparental and *F. malvastra* is parthenogenetic.

In India, *F. virgata* can produce several overlapping generations a year (Nayar *et al.* 1976). Mating took place only once and lasted for about 12-23 minutes. The female lays eggs in groups beneath her body in a loose ovisac of waxy fibres. Fecundity (egg number) ranged from 109 to 185 per generation and may exceed 500 (Schmutterer 1969). The oviposition period lasted 20-29 days. The incubation period lasted about 3-4 hours. Female and male nymphs moulted 3 and 4 times, respectively, and the development period varied from 26-47 and 31-57 days, respectively. Longevity of the adult female was 36-53 days and for the male, 1-3 days.

In Egypt, *F. virgata* produced three generations a year on *Acalypha macrophylla* (Ammar *et al.* 1979); the first in early June, the second in early July and the third in August. The population increased in size with each generation. *F. virgata* overwintered, probably as adult females, in cracks and junctions of trunks and larger branches and on fallen leaves. In the laboratory, females migrated to the soil in winter. The preferred direction of distribution of the pest was south-east in the first generation and south-west in the second and third generations. A significant positive correlation was found between population density and daily maximum and minimum temperatures, but not between population density and relative humidity.

Like other mealybugs, the main dispersal stage of *F. virgata* is the first instar

which may be naturally dispersed by wind and animals. The females are active and mobile throughout their life, until they start to produce an ovisac and lay eggs. All life stages may be carried on consignments of plant material and fruit.

#### **Economic impact**

*F. virgata* is a known vector of cocoa swollen shoot virus (CSSV) in West Africa and cocoa Trinidad virus (CTV, Diego Martin valley isolate) in Trinidad (Thorold 1975). Le Pelley (1968) gave a general discussion of the pest status of *F. virgata* on coffee and Keuchenius (1915) discussed it as a pest of coffee in Java, Indonesia. Schmutterer (1969) stated it was a major pest of irrigated guava trees in the drier areas of the Sudan where it is common on many other crops, shade, ornamental and wild plants. In Tanzania it is a pest of cashew and in some parts of the world it is a pest of cotton (Williams 1996). In India, it has been recorded as a pest of a range of crops including coffee (Chacko and Bhat 1976), custard apple (Mani and Krishnamoorthy 1989), betel vine (Patil *et al.* 1987), black pepper (Sarma *et al.* 1987), pigeon pea (Gautam and Saxena, 1986) and milk tree (*Manilkara hexandra*), and a rootstock for sapodilla (*Manilkara achras*) (Jhala *et al.* 1988). It is also recorded as a pest of kenaf (*Hibiscus cannabinus*) and mesta (*H. sabdariffa*) in Bangladesh (Jalil 1971), of *Leucaena leucocephala* in Taiwan (Chang and Sun 1985) and of glasshouse ornamental plants in Egypt (Nada 1986).

#### **Symptoms**

Infestations of *F. virgata* remain clustered around the terminal shoots, leaves and fruit, sucking the sap which results in yellowing, withering and drying of plants and shedding of leaves and fruit. The foliage and fruit also become covered with large quantities of sticky honeydew which serves as a medium for the growth of black sooty moulds. The sooty moulds and waxy deposits result in a reduction of photosynthetic area, and ornamental plants and products lose their market value due to the cosmetic effect of the infestation on quality.

#### **Descriptors**

**Leaves:** abnormal colours; honeydew or sooty mould; wilting

**Stems:** discoloration; external feeding; honeydew or sooty mould

**Growing points:** dead heart; external feeding

**Fruits/pods:** discoloration; external feeding; honeydew or sooty mould; honeydew or sooty mould

#### **Morphology**

The adult females are oval, greyish-yellow, with two longitudinal, submedian, dark stripes on the dorsum showing through the waxy secretion, hence the common name 'striped mealybug'. The dorsum also bears numerous straight, glassy threads of wax. They attain 4-4.5 mm in length.

Authoritative identification involves detailed microscopic examination of teneral adult females by a competent taxonomist. Adult female *Ferrisia* is fairly easy to recognise by the presence of enlarged dorsal ducts, each with the orifice surrounded by a circular, sclerotized area associated with one or more short setae. *F. virgata* may be recognised by the anal lobe cerarii, each with two conical setae and the multilocular disc pores present on abdominal segment VI in a distinct row, always numbering at least eight. Williams (1996) provides a useful synopsis of the genus *Ferrisia* and includes a morphological key to the world species. For detailed morphological descriptions, illustrations and keys to the species of *Ferrisia* that occur in North America, see Ferris (1950; 1953) and McKenzie (1967); for Central and South

America, see Williams and Granara de Willink (1992); for the Tropical South Pacific Region, see Williams and Watson (1988); and for Australia, see Williams (1985).

#### **Similarities to other species**

*F. virgata* should be distinguished from *F. malvastra* (until recently known as *F. consobrina*) which is also highly polyphagous and recorded from many parts of the world. They are morphologically very similar and cannot be distinguished in the field by simple superficial features. Slide-mounted preparations are needed for examination. Other species closely related to *F. virgata* have been described in recent years from South America and should be separated using the keys given by Williams (1996) and Williams and Granara de Willink (1992).

#### **Risk assessment of pest**

	<b>Seednuts</b>	<b>Embryo cultures</b>	<b>Pollen</b>
<b>Entry</b>	All life stages may be carried on consignments of plant material and fruit.	Not an internal pest	Adults not likely in sieved pollen. They attain lengths of 4 to 4.5 mm.
<b>Establishment</b>	<i>F. virgata</i> has spread to all zoogeographical regions, mainly in the tropics, but often extends well into the temperate regions (see CIE distribution map No. 219 (1966) and Ben-Dov (1994)).		
<b>Spread</b>			
<b>Economic</b>	Among the hosts of economic importance are avocado, banana, betel vine, black pepper, cassava, cashew, cauliflower, citrus, cocoa, coffee, cotton, custard apple, eggplant, grapevine, guava, jute, lantana, <i>Leucaena</i> , litchi, mango, oil palm, pigeon pea, pineapple, soybean and tomato.		
<b>Level of risk</b>	Low		
<b>Pest status</b>	QP	NQP	NQP
<b>Pest management</b>	Pest with seed nuts managed by fumigation with MeBr.		

**Note :** QP – quarantine pest ; NQP- Non-quarantine pest

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### Annex 2.3.5. Data sheet of *Heliothrips haemorrhoidalis*

#### Names and taxonomy

**Preferred name** : *Heliothrips haemorrhoidalis* Bouché

#### **Taxonomic position**

Domain : Eukaryota  
 Kingdom : Metazoa  
 Phylum : Arthropoda  
 Class : Insecta  
 Order : Thysanoptera  
 Family : Thripidae

#### **Other names used**

*Dinurothrips rufiventris* Girault  
*Heliothrips adonium* Haliday  
*Heliothrips ceylonicus*  
*Heliothrips haemorrhoidalis* var. *abdominalis* Reuter  
*Heliothrips haemorrhoidalis* var. *andustior* Priesner  
*Heliothrips haemorrhoidalis* var. *ceylonicus* Schmutz  
*Heliothrips semiaureus* Girault  
*Heterothrips haemorrhoidalis*  
*Thrips haemorrhoidalis* Bouché

**Bayer Code** : HELTHA

#### **Common names**

English : black glasshouse thrips; black tea thrips; greenhouse thrips  
 Spanish : piojillo de los invernaderos; piojillo negro de los jardines; piojito del aguacate, trípido negro; trips de los cítricos (Mexico); trips de los invernaderos (Mexico); trips del aguacate (Mexico); trips o cuerudo del palto  
 French : thrips des serres ; thrips noir de l'avocatier  
 German : Rotschwaenziger Blasenfuss; Schwarze Fliege Schwarzer Gewaechshaus-Blasenfuss  
 Italian : Eliotripide emorroidale ; Tripide delle lantane ; Tripide delle serre  
 Dutch : gewone trips; Kastrips

#### **Notes on taxonomy and nomenclature**

*H. haemorrhoidalis* was first described by Bouché from Berlin, Germany in 1833. Although it has some synonyms, its taxonomic situation has now been clarified by Wilson (1975) and Mound (1976).

#### **Host range**

*H. haemorrhoidalis* is highly polyphagous, and has been recorded with certainty from more than 100 plant species, including some ferns (Daniel and Chandrasekar 1986) and conifers (Ananthakrishnan 1971; Zondag 1977; Cerda 1980). Seriously damaged cultivated crops include avocado, kiwifruit, persimmon, citrus, tea, croton, pine and cyclamen.



**Primary hosts:** *Persea americana* (avocado), *Actinidia chinensis* (Chinese gooseberry), Citrus, *Diospyros kaki* (oriental persimmon), *Camellia sinensis* (tea), *Acalypha* (Copperleaf), *Annona*, *Begonia*, *Cocos nucifera* L. (coconut), *Codiaeum variegatum* (croton), *Cinchona*, *Cinnamomum*, *Cymbidium*, *Cryptomeria japonica* (Japanese cedar), *Coffea* (coffee), *Carya illinoensis* (pecan), *Diospyros* (malabar ebony), *Dracaena*, *Erica* (heaths), *Gossypium* (cotton), *Ilex* (Holly), *Juniperus chinensis* (Chinese juniper), *Lagerstroemia indica*, *Ludwigia*, *Manihot esculenta* (cassava), *Macadamia*, *Mangifera indica* (mango), *Passiflora quadrangularis* (giant granadilla), *Pinus* (pines), *Platanus* (planes), *Primula* (Primrose), *Prunus* (stone fruit), *Psidium guajava* (common guava), *Sapium*, *Schinus*, *Syzygium jambos* (rose apple), *Terminalia catappa* (beach almond), *Theobroma cacao* (cocoa) and *Viburnum*.

**Infested plant stages:** Flowering stage, fruiting stage, seedling stage, vegetative growing stage, and post-harvest

**Infested plant parts:** Leaves and fruits/pods

### Geographic distribution

*H. haemorrhoidalis* is widely distributed in the tropics and subtropics; it also occurs in greenhouses of temperate areas as it was first described from specimens collected in a greenhouse in Berlin. *H. haemorrhoidalis* is presumed to have originated in tropical America, probably Brazil. It has been introduced to various parts of the world by man and has become naturalized in those areas.

### List of countries

#### **Europe**

- Austria: present, no further details (CIE 1961)
- Belgium: present, no further details (CIE 1961)
- Bulgaria: present, no further details (Batalova 1975)
- Czechoslovakia (former ): present, no further details (CIE 1961; Pelikán 1977)
- Denmark: present, no further details (CIE 1961)
- Finland: present, no further details (CIE 1961)
- France: present, no further details (CIE 1961; Bournier 1983; Brun 1992)
- Germany: present, no further details (CIE 1961; Schliephake 1984)
- Greece: present, no further details (CIE 1961)
- Hungary: present, no further details (CIE 1961; Gabor 1982)
- Italy: present, no further details (CIE, 1961; Ragusa and Russo 1989)
- Netherlands: present, no further details (CIE 1961)
- Norway: present, no further details (CIE 1961)
- Poland: present, no further details (CIE 1961; Seczkowska 1974)
- Portugal: present, no further details (CIE 1961)
  - Azores: widespread (zur Strassen 1973)
- Romania: present, no further details (CIE 1961)
- Russian Federation: present, no further details (CIE 1961)
- Spain: present, no further details (CIE 1961; Melia 1976; Mansilla and Puerto, 1984)
  - Canary Islands: widespread (zur Strassen 1983)
- Sweden: present, no further details (CIE 1961)
- Switzerland: present, no further details (CIE 1961)
- United Kingdom: present, no further details (CIE 1961; Mound *et al.* 1976)

- Channel Islands: present, no further details (CIE 1961)

## Asia

- Azerbaijan: present, no further details (CIE 1961)
- Cambodia: present, no further details (Waterhouse 1993)
- China: present, no further details (CIE 1961; Zhang and Tong 1993)
  - Fujian: present, no further details (Zhang and Tong 1993)
  - Guangdong: present, no further details (Zhang and Tong 1993)
  - Guizhou: present, no further details (Zhang and Tong 1993)
  - Hainan: present, no further details (Zhang and Tong 1993)
  - Hong Kong: present, no further details (Kudô 1980)
  - Sichuan: present, no further details (Zhang and Tong 1993)
  - Taiwan: widespread (CIE 1961; Kudô 1980; Chen 1981)
  - Yunnan: present, no further details (Zhang and Tong 1993)
- Georgia (Republic): present, no further details (CIE 1961)
- India: present, no further details (CIE 1961; Wilson 1975; Bhatti 1990)
  - Andaman and Nicobar Islands: present, no further details (Bhatti 1990)
  - Andhra Pradesh: present, no further details (Wilson 1975)
  - Karnataka: present, no further details (Bhatti 1990)
  - Kerala: present, no further details (CIE 1961; Bhatti 1990)
  - Maharashtra: present, no further details (Wilson 1975)
  - Tamil Nadu: present, no further details (Wilson 1975; Bhatti 1990)
- Indonesia: present, no further details (CIE 1961; Waterhouse 1993; zur Strassen 1994)
  - Java: present, no further details (CIE 1961; zur Strassen 1994)
  - Kalimantan: present, no further details (zur Strassen 1994)
  - Nusa Tenggara: present, no further details (zur Strassen 1994)
  - Sumatra: present, no further details (CIE, 1961; zur Strassen 1994)
- Israel: present, no further details (CIE 1961; Halperin 1990)
- Japan: present, no further details (CIE 1961; Kudô 1992)
  - Honshu: present, no further details (CIE 1961; Kudô 1992)
  - Kyushu: present, no further details (CIE 1961; Kudô 1992)
  - Ryukyu Archipelago: widespread (CIE1961)
  - Shikoku: present, no further details (Kudô 1992)
- Korea, Republic of: present, no further details (APPPC 1987)
- Malaysia: widespread (Kudô 1995)
  - Peninsular Malaysia: widespread (CIE 1961; Kudô 1995)
  - Sarawak: present, no further details (Kudô 1995)
- Myanmar: present, no further details (Kudô 1995)
- Nepal: present, no further details (Kudô 1995)
- Philippines: widespread (Reyes 1994; Kudô 1995)
- Sri Lanka: widespread (CIE 1961; Wilson 1975)
- Thailand: present, no further details (Kudô 1980; Waterhouse 1993)
- Turkey: widespread (CIE 1961; Zumreoglu 1986)
- Vietnam: present, no further details (Waterhouse 1993)

## Africa

- Cape Verde: present, no further details (zur Strassen 1980)
- Congo: present, no further details (CIE 1961)
- Egypt: present, no further details (CIE 1961)
- Ghana: present, no further details (CIE 1961)
- Kenya: present, no further details (CIE 1961; Layock and Templer 1973)
- Malawi: present, no further details (CIE 1961)
- Mauritius: present, no further details (CIE 1961)

- Morocco: present, no further details (CIE 1961)
- Saint Helena: present, no further details (CIE 1961)
- Seychelles: present, no further details (Bhatti 1990)
- Sierra Leone: present, no further details (CIE 1961; Pitkin and Mound 1973)
- South Africa: widespread (CIE 1961; Dennill 1992)
- Tanzania: present, no further details (CIE 1961)
- Uganda: present, no further details (CIE 1961)
- Zimbabwe: present, no further details (CIE 1961)

### **Western Hemisphere**

- Argentina: present, no further details (CIE 1961)
- Bahamas: present, no further details (CIE 1961; Bennett and Baranowski 1982)
- Barbados: present, no further details (CIE 1961)
- Bermuda: present, no further details (CIE 1961)
- Bolivia: present, no further details (CIE 1961)
- Brazil: present, no further details (CIE 1961; Wilson 1975)
  - Bahia: present, no further details (CIE 1961)
  - Rio Grande do Sul: present, no further details (CIE 1961)
  - Rio de Janeiro: present, no further details (CIE 1961)
  - Sao Paulo: present, no further details (CIE 1961)
- Canada: present, no further details (CIE 1961; Chiasson 1986)
  - Alberta: present, no further details (Steiner and Elliott 1983)
  - British Columbia: present, no further details (CIE 1961; Steiner and Elliott 1983)
- Chile: present, no further details (CIE 1961; Gonzalez 1986; Prado 1988)
- Colombia: present, no further details (Wilson 1975; Escobar *et al.* 1985)
- Cuba: present, no further details (CIE 1961)
- Dominica: present, no further details (Wilson 1975)
- Dominican Republic: present, no further details (CIE 1961)
- Ecuador: present, no further details (CIE 1961)
- Grenada: present, no further details (Wilson 1975)
- Guadeloupe: present, no further details (Wilson 1975)
- Honduras: present, no further details (CIE 1961; Wilson 1975)
- Jamaica: present, no further details (CIE 1961; Sakimura 1986)
- Mexico: present, no further details (CIE 1961; Johansen 1976)
- Panama: present, no further details (Wilson 1975)
- Peru: present, no further details (CIE 1961)
- Puerto Rico: present, no further details (CIE 1961)
- Saint Vincent and the Grenadines: present, no further details (CIE 1961)
- Suriname: present, no further details (CIE 1961; Wilson 1975)
- Trinidad and Tobago: present, no further details (CIE 1961; Wilson 1975)
- USA: present, no further details (CIE 1961)
  - Alabama: present, no further details (CIE 1961)
  - California: present, no further details (CIE 1961; Hessein and McMurtry 1988)
  - Connecticut: present, no further details (CIE 1961)
  - Delaware: present, no further details (CIE 1961)
  - Florida: present, no further details (CIE 1961; Denmark 1985)
  - Georgia (USA): present, no further details (CIE 1961; Beshear 1983)
  - Hawaii: present, no further details (CIE 1961)
  - Illinois: present, no further details (CIE 1961; Stannard 1968)
  - Indiana: present, no further details (CIE 1961)
  - Iowa: present, no further details (CIE 1961)

- Kansas: present, no further details (CIE 1961)
- Louisiana: present, no further details (CIE 1961)
- Maryland: present, no further details (CIE 1961)
- Massachusetts: present, no further details (CIE 1961)
- Michigan: present, no further details (CIE 1961)
- Mississippi: present, no further details (CIE 1961)
- Missouri: present, no further details (CIE 1961)
- Nebraska: present, no further details (CIE 1961)
- New Hampshire: present, no further details (CIE 1961)
- New Jersey: present, no further details (CIE 1961)
- New York: present, no further details (CIE 1961)
- North Dakota: present, no further details (Huntsinger *et al.* 1982)
- Ohio: present, no further details (CIE 1961)
- Oregon: present, no further details (CIE 1961)
- Pennsylvania: present, no further details (CIE 1961)
- Rhode Island: present, no further details (CIE 1961)
- South Carolina: present, no further details (CIE, 1961)
- South Dakota: present, no further details (CIE 1961)
- Texas: present, no further details (CIE 1961)
- Washington: present, no further details (CIE 1961)
- West Virginia: present, no further details (CIE 1961)
- Uruguay: present, no further details (CIE 1961)
- Venezuela: present, no further details (CIE 1961)

### **Oceania**

- Australia: present, no further details (CIE 1961; Mound and Houston 1987)
  - Australian Northern Territory: present, no further details (CIE 1961)
  - New South Wales: present, no further details (CIE 1961; Mound and Houston 1987)
  - Queensland: widespread (CIE 1961; Mound and Houston 1987)
  - South Australia: present, no further details (CIE 1961; Mound and Houston 1987)
  - Tasmania: present, no further details (CIE 1961; Anon. 1971)
  - Victoria: present, no further details (CIE 1961; Mound and Houston 1987)
  - Western Australia: present, no further details (CIE 1961; Mound and Houston, 1987)
- Cook Islands: present, no further details (Mound and Walker 1987)
- Fiji: present, no further details (CIE 1961; Mound and Walker 1987)
- Kiribati: present, no further details (Mound and Walker 1987)
- New Zealand: present, no further details (APPPC 1987; CIE 1961; Mound and Walker 1987)
- Papua New Guinea: present, no further details (CIE 1961)
- Tonga: present, no further details (APPPC 1987; CIE 1961; Mound and Walker, 1987)
- Vanuatu: present, no further details (Mound and Walker 1987)

### **Biology and ecology**

There have only been a few brief accounts of the biology and ecology of *H. haemorrhoidalis* in recent years, although some detailed studies were carried out in earlier years (Russell 1909; Rivnay 1934, 1935a, b).

Males of *H. haemorrhoidalis* are rare, chiefly known from Brazil, and reproduction

is parthenogenetic and thelytokus<sup>1</sup>. The female takes 4-6 days to start oviposition after emergence and produces up to 47 eggs on average at 21-28°C during her lifetime of about one month. The eggs are laid singly in the epidermis of the under surface of the leaf and each egg is covered with an excretory droplet. The larvae emerge in about 14-15 days at an optimal temperature of 26-28°C, and 16-22 days at 21-25°C. They carry a large excretory droplet between the anal setae at the end of the abdomen, which is raised and lowered at intervals to deposit the droplet. The first and second instars occupy 9-11 days at 26-28°C, and 10-16 days at 21-25°C, followed by the prepupal and pupal stages lasting for 3-4 days at 26-28°C and 4-6 days at 21-25°C, respectively. The adult lives for up to 35 days at 25-27°C. The complete life cycle of *H. haemorrhoidalis* occurs on the leaves of the host. This thrip may produce about seven generations under temperate weather conditions and more than 12 under tropical conditions.

Both adults and larvae feed mostly on leaves and fruits in concentrated colonies; the youngest or oldest leaves are rarely preferred. The colonies gather mainly on the under surfaces of the leaves; they do not live on the tops of the leaves and fruits until the tissue becomes unsuitable for feeding and oviposition.

The biology of *H. haemorrhoidalis* on the fern *Polypodium phegopteris* was studied in India (Daniel and Chandrasekar 1986). The thrip was mostly restricted to mature fronds of the fern and mating was evident, in contrast to parthenogenetic reproduction on coffee leaves. The male:female ratio was 3:25, and the life cycle ranged from 20-30 days. This thrips-fern association was observed only at altitudes above 1900 m. The mature fronds preferred by *H. haemorrhoidalis* had 1.5 times the lipid content of young fronds. There was little variation in other chemical compounds between the young and mature fronds. A higher concentration of protein and nitrogen in *P. phegopteris*, compared with other fern hosts, appeared to attract and enhance the survival and growth of *H. haemorrhoidalis*.

### **Economic impact**

Crop losses caused by *H. haemorrhoidalis* are difficult to assess and there have been few critical studies despite the importance of this pest on many crops.

*H. haemorrhoidalis* was found to be one of five important pests on avocado fruits in South Africa (the others were *Pseudotheraptus wayi*, *Selenothrips rubrocinctus*, *Pterandrus rosa* [*Ceratitis rosa*] and *Nezara viridula*) in a packhouse survey. *H. haemorrhoidalis*, together with *S. rubrocinctus*, caused 2.1% (potentially up to 80%) cull of the fruits by lesions and crack (Dennill and Erasmus 1992a). *H. haemorrhoidalis* caused considerable mortality in up to 3-year-old *Pinus radiata* on warm sites, both in the nursery and in the forest in New Zealand (Zondag 1977).

### **Symptoms**

Symptoms of attack by *H. haemorrhoidalis* result from feeding by adults and/or larvae on the leaves and pods; the feeding punctures cause the development of chlorotic spots. Severely infested leaves become papery and wilted, and soon die. Serious infestation usually results in defoliation. Brown patches occur on the surfaces of fruits and, if injured during growing, cracks often appear.

Small brown patches of excretory droplets, typical of thrips infestation, are also an obvious means of identifying infestation.

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<sup>1</sup> The mode of parthenogenic reproduction in which unmated females produce only female progeny, males being unknown or very rare and without apparent function. Thus, a thelytokus population consists of genetically-identical females. This mode of reproduction is found in insects, sometimes as a phase that alternates with sexual reproduction (e.g. aphids) (CABI-CPC Glossary).

**Descriptors****Leaves:** abnormal colours; abnormal leaf fall; necrotic areas**Fruits/pods:** discoloration; lesions: black or brown**Morphology****Larvae**

Full grown second-instar larvae are about 1.1 mm long. The body is yellow, with ninth and tenth abdominal segments brown. The antennae, except the first segment, are pale grey; the terminal segment is long, slender and needle-like. The pterothorax and abdomen have many thin and longitudinal plaques. Dorsal body setae are small; three pairs of anal setae are short, about as long as the tenth abdominal segment.

**Adults**

Wilson (1975), Mound (1976), Mound and Walker (1982), Kudô (1992) and Reyes (1994) provide illustrated descriptions of the adults of *H. haemorrhoidalis*.

The body is dark brown with the apex of the abdomen paler, being 1.4-1.7 mm long in the female and 1.1-1.2 mm in the male. The legs are entirely white or yellow. Teneral individuals have orange abdomen. The body and legs are strongly polygonally reticulate. The head is clearly constricted and neck-like. The antennae are 8-segmented; the second to fifth, seventh and eighth segments are yellow; the third and fourth segments have a simple sense cone; the eighth is long, slender, and needle-like. The tarsi are single-segmented. The forewings are white with a longitudinal brown line medially, parallel-sided but swollen basally and rounded apically, and have minute setae on veins. Abdominal terga are weakly reticulated medially, and the sternal marginal setae are minute. The male has three pairs of thorn-like setae on ninth abdominal tergum; the anterior pair of setae is stoutest. Male third to seventh abdominal sterna each have a transverse oblong glandular area.

**Detection and inspection methods**

Both adult and immature stages are detected by examining the under surfaces of leaves and the surfaces of the pods and fruits. Chlorotic spots, brown patches and necrotic lesions are apparent. Microscope preparations should be made for identification; it is necessary to examine thrips under a compound microscope.

**Risk assessment of pest**

	Seednuts	Embryo cultures	Pollen
<b>Entry</b>	Both adults and larvae feed mostly on leaves and fruits in concentrated colonies	No	No
<b>Establishment</b>	<i>H. haemorrhoidalis</i> is highly polyphagous, and has been recorded with certainty from more than 100 plant species, including some ferns (Daniel and Chandrasekar 1986) and conifers (Ananthakrishnan 1971; Zondag 1977; Cerda 1980). Seriously damaged cultivated crops include avocado, kiwifruit, persimmon, citrus, tea, croton, pine and cyclamen.		
<b>Spread</b>			
<b>Economic</b>	Crop losses, caused by <i>H. haemorrhoidalis</i> , are difficult to assess and there have been few critical studies despite the importance of this pest on many crops.		
<b>Level of risk</b>	Very Low	Minimal	Minimal

Pest status	QP?	NQP	NQP
Pest management	Management of pests likely as hitchhikers on seednuts by fumigation with MeBr.		

Note : **QP** – quarantine pest ; **NQP**- Non-quarantine pest

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**Annex 2.3.6. Data sheet of *Marasmiellus cocophilus*****Names and taxonomy****Preferred name:** *Marasmiellus cocophilus* Pegler**Taxonomic position**

Domain : Eukaryota  
Kingdom : Fungi  
Phylum : Basidiomycota  
Class : Basidiomycetes  
Subclass : Agaricomycetidae  
Order : Agaricales  
Family : Marasmiaceae

**Bayer Code** : MARLCO**Common names**

English : basal stem break; lethal bole rot of coconut

**Notes on taxonomy and nomenclature**

*M. cocophilus* was first described from Kenya and Tanzania by Pegler (1969) from coconut palms with lethal bole rot disease.

**Host range**

In East Africa, *M. cocophilus* has only been recorded from coconuts; in the Solomon Islands, in addition to coconuts, the fungus has also been found growing from roots and leaves of several species of grass around bagged coconut seedlings in a nursery (Jackson and Firman 1979).

**Primary host:** *Cocos nucifera* L. (coconut)**Infected plant stages:** Seedling and vegetative growing stages**Infected plant parts:** Whole plant**Geographic distribution****List of countries****Africa**

- Kenya: present, no further details (Bock *et al.* 1970; EPPO 2003)
- Madagascar: absent, unreliable record (EPPO 2003)
- Tanzania: present, no further details (Bock *et al.* 1970; EPPO 2003)

**Oceania**

- Solomon Islands: present, few occurrences (Jackson and Firman 1979)

### **Biology and ecology**

In East Africa, *M. cocophilus* causes death of palms up to eight years old, with seedlings being highly susceptible upon transplanting to the field. Spread occurs through soil, root contact between palms, infected coconut debris and probably by airborne basidiospores. Infection also occurs via wounds. Basidiomata occur on exposed roots, on leaf bases of seedlings, exposed tops of seednuts and on the soil surfaces (growing from coconut debris) (Bock *et al.* 1970; Frison and Putter 1994). Little is known about the methods of infection and spread of the fungus in the Solomon Islands, although it is assumed that similar methods probably operate.

### **Means of movement and dispersal**

#### **Plant parts liable to carry the pest in trade/transport:**

- Fruits (including pods): Hyphae, fruit bodies
- Leaves: Hyphae, fruit bodies; borne internally; visible to naked eye
- Roots: Hyphae, fruit bodies; borne internally; visible to naked eye
- Stems (above ground)/shoots/trunks/branches: Hyphae, fruit bodies; borne internally; invisible
- Wood: Hyphae; borne internally; invisible

#### **Plant parts not known to carry the pest in trade/transport:**

- Bark
- Bulbs/tubers/corms/rhizomes
- Growing medium accompanying plants
- Seedlings/micropropagated plants.

### **Seedborne aspects**

Conditions within the coconut husk are thought to be ideal for basidiomycetous fungi, and some marasmioid fungi are abundant during germination and early seedling development (Jackson and Firman 1982). Basidiomata have been detected on seednuts taken from the outbreak area in the Solomon Islands to other parts of the country, supporting the notion that the fungus is seedborne (Jackson and McKenzie 1988).

### **Economic impact**

Bock *et al.* (1970) reported that the disease was serious along the East African coasts of Kenya and Tanzania, and surveys in the late 1960s detected several discontinuous outbreaks and other isolated occurrences of one to a few palms. Losses of over 90% were recorded, and in one 40 ha block, half the palms died over a 5-year period.

In the Solomon Islands, an outbreak of the disease occurred in 1978/79 in a commercial estate on one island. Several thousand seedlings broke at the junction of the leaves and seednut and were discarded. Since that time, the disease has not recurred, although the fungus can still be found on grasses growing in the coconut nursery. The seedlings planted at the time of the outbreak have not shown signs of disease (McKenzie 1986). Surveys in other parts of the country have not detected the presence of the fungus.

### **Symptoms**

In East Africa, symptoms are most often seen on seedlings after transplanting to the field (Bock *et al.* 1970). Root infections occur, leading to decay of basal tissues and finally a rotting spear leaf. On older palms, the first symptoms are a general wilt of the fronds, which remain as a 'skirt' around the trunk. The spear leaf dies and a foul-

smelling soft rot develops at the base of the leaves. A dry, reddish-brown rot with a yellow margin is typically present at the base of the bole. Cavities within these areas of rot are lined with mycelium in young palms, 2-4 years old, but rare in 4-6-year-old palms, and absent in mature palms. Basidiomata commonly occur on exposed roots, leaf bases of seedlings and on the soil surface around holes where diseased palms had been removed two years previously. On average, there are only eight weeks from the time of onset of symptoms till the death of the palm; this interval depended on the extent of fungal decay in the bole.

Symptoms in the Solomon Islands are considerably different. Symptoms have been described on seedlings infected in the nursery and on young palms after transfer to the field (Jackson and Firman 1979; Jackson and McKenzie 1988). The first sign of disease in the nursery is the premature death of the oldest two or three leaves. White mycelium and carpophores of the fungus are present at the base of the petioles and the top of the seednut. Younger leaves are infected successively as the fungus colonizes the leaf bases, producing a brown rot. Cracks are common in the leaf bases, and isolated rots, 1-1.5 cm deep, with shallow, reddish-brown margins, extend into the bole. In the Solomon Islands, root decay was not extensive, although new roots which had penetrated decayed leaf bases were often colonized by the fungus and the root tips destroyed. Affected seedlings were prone to break at the junction of the petioles and the seednut.

Normal seedlings transplanted to the field develop symptoms similar to those in the nursery. The most conspicuous symptom is the development of small leaves which start to unfurl before they are fully emerged. Rots are also present in the bole tissues, leaf bases are swollen, roots decayed and mycelium and basidiomata may be present at the base of the petioles. Few of the transplanted palms die. Most begin to recover 5-6 months after planting when new uninfected roots are produced and new leaves develop which are progressively more normal. Wilts, crown rots and bole rots have not been seen in the Solomon Islands.

Attempts to reproduce symptoms have been successful in East Africa. Root infections using artificial inoculum showed that the fungus moved rapidly inside the roots, into the stem and killed the seedlings (Bock *et al.* 1970). Less success was reported in the Solomon Islands (Jackson and McKenzie 1988). *M. cocophilus* was shown to kill root tips, but movement in the roots was slow; even after three months, lesions were only 2-3 cm long. Inoculating seedlings by placing artificially inoculated petiole fibre between the leaf bases or by planting nuts in *M. cocophilus*-infested nursery soil was equally inconclusive. Rots did occur in the leaf bases and roots were infected, but the number of successful inoculations was low and few of the plants showed sufficient damage to arrest growth or lead to basal stem break.

### Descriptors

**Whole plant:** dwarfing; unusual odour

**Leaves:** abnormal colours; abnormal forms; wilting; fungal growth

**Stems:** stunting or rosetting; mould growth on lesion; mycelium present

**Roots:** soft rot of cortex

**Fruits/pods:** extensive mould

### Morphology

Pegler (1969) gives a diagnosis of *M. cocophilus*. The species is recognized by its marasmioid habit, the lack of pigmentation in the basidiomata, small habit, its association with coconut, and large lacrymoid spores (10-17.5 x 3-4.5 µm).

**Similarities to other species**

Several seedborne marasmioid fungi have been detected on coconuts (Jackson and Firman 1982). In addition to *M. cocophilus*, basidiomata of *M. inoderma* commonly grow on seednuts in coconut nurseries. The fungus infects seednuts through the calyx end, colonizes the fibrous husk tissues and grows beneath the operculum as it is raised by the emerging shoot. Infections can even be found when coconuts are still on the palm. *Marasmiellus albofuscus* infects roots and causes shallow rots at the base of the trunk of mature palms. *Maramius crinisequi* is not known to infect coconuts, but causes a thread blight of cocoa. Distinctions between these species are made on the morphology of the basidiomata and spores (Pegler 1969; Pegler 1977).

*Marasmiellus dealbatus* from tropical America most resembles *M. cocophilus*, but is known only as a saprophyte on forest litter and the spores are smaller: 8-9 x 4.5-5 µm.

**Detection and inspection methods**

*M. cocophilus* basidiomata develop most commonly at the base of the petioles at the junction with the seedling husk. Detection of *M. cocophilus* in the seednut is not possible by visual observation, although occasionally, infection of the germinating embryo by *M. inoderma* can be observed, even on coconuts taken from the palm.

**Control****Regulatory control**

International movement of coconut germplasm should follow the technical guidelines recommended by FAO/IBPGR (Frison and Putter 1994). Seednuts should not be transferred directly from countries in East Africa where *M. cocophilus* infections are known to occur, to areas not affected by such pathogens.

**Chemical control**

Seed treatment should always be considered as a precautionary measure whenever seednuts are being moved between countries, or between areas within countries where, as in the case of the Solomon Islands, coconut pathogens have a restricted distribution. Seednuts are taken directly from the mother palm, partially dehusked by trimming at the top and three sides and dipped in an appropriate fungicide for 15 minutes. The addition of a wetting agent is considered beneficial.

**Risk assessment of pest**

	Seednuts	Embryo cultures	Pollen
Entry	Basidiomata have been detected on seednuts taken from the outbreak area in the Solomon Islands to other parts of the country, supporting the notion that the fungus is seedborne (Jackson and McKenzie 1988).	Parts of micropropagated plants are not known to carry the pest in trade/transport.	No information
Establishment Spread			

<b>Economic</b>	Bock <i>et al.</i> (1970) reported that the disease was serious along the East African coasts of Kenya and Tanzania. Losses of over 90% have been recorded.		
<b>Level of risk</b>	Low	Minimal	Minimal
<b>Pest status</b>	QP?	NQP	NQP
<b>Pest management</b>	Management of seed nut transmission by treatment with fungicide and growing in PEQ and observing seedling growth.		

**Note :** *QP* – quarantine pest ; *NQP*- Non-quarantine pest

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**Annex 2.3.7. Data sheet of *Radopholus similis*****Names and taxonomy****Preferred name:** *Radopholus similis* (Cobb 1893; Thorne 1949)**Taxonomic position**

Domain : Eukaryota  
 Kingdom : Metazoa  
 Phylum : Nematoda  
 Family : Pratylenchidae

**Other names used**

*Anguillulina acutocaudatus* (Zimmermann 1898; Goodey 1932)  
*Anguillulina biformis* (Cobb 1909; Goodey 1932)  
*Anguillulina granulosa* (Cobb 1893; Goodey 1932)  
*Radopholus acutocaudatus* (Zimmermann 1898; Siddiqi 1986)  
*Radopholus biformis* (Cobb 1909; Siddiqi 1986)  
*Radopholus citrophilus* Huettel (Dickson and Kaplan 1984)  
*Radopholus granulatus* (Cobb 1893; Siddiqi 1986)  
*Radopholus similis citrophilus* Huettel (Dickson and Kaplan 1984)  
*Tetylenchus granulatus* (Cobb 1893; Filipjev 1936)  
*Tylenchorhynchus acutocaudatus* (Zimmermann 1898; Filipjev 1934)  
*Tylenchus biformis* (Cobb 1909)  
*Tylenchus similis*  
*Tylenchus granulatus*  
*Rotylenchus similis*  
*Anguillulina similis*

**Common names**

English : burrowing nematode; citrus burrowing nematode; pepper yellows nematode; slow wilt nematode; spreading decline of citrus; nematode root rot; black head disease of banana; banana burrowing nematode  
 Spanish : declinación propagante de los cítricos; nematodo coco; nematodo del banano (Argentina); nematodo del plátano (Mexico)  
 French : anguillule mineuse du bananier  
 Brazilian : Nematode cavernicola  
 Sri Lankan : mid-country species of nematode

**Notes on taxonomy and nomenclature**

*Radopholus citrophilus*, previously regarded, at least in some quarters, as a separate species from *Radopholus similis* is now accepted as being synonymous (Valette *et al.* 1998a; Elbadri *et al.* 1999; Kornobis 1999).

**Host range**

*Radopholus similis* (sensu lato) is very polyphagous, attacking hundreds of plant species notably those belonging to the *Rustaceae* (Citrus and related genera) but also many other families including *Arecaceae*, *Musaceae*, *Poaceae*, *Brassicaceae*, *Rubiaceae* and *Solanaceae* to name a few.

It is a serious pest on commercial citrus in Florida and on banana, plantain, black pepper, ginger, coffee, tea, coconut, arecanut and other such crops in tropical and

subtropical areas worldwide, with only a few exceptions.

**Primary hosts:** *Musa* (banana), *Musa x paradisiaca* (plantain), *Citrus*, *Piper nigrum* (black pepper), *Coffea* (coffee), *Zea mays* (maize), *Saccharum officinarum* (sugarcane), *Ananas comosus* (pineapple), *Arachis hypogaea* (groundnut), *Areca catechu* (betelnut palm), *Anthurium andreaeanum*, *Camellia sinensis* (tea), *Cocos nucifera* (coconut), *Coffea arabica* (arabica coffee), *Coffea canephora* (robusta coffee), *Curcuma longa* (turmeric), *Daucus carota* (carrot), *Dioscorea* (yam), *Lycopersicon esculentum* (tomato), *Musa textilis* (manila hemp), *Persea americana* (avocado), *Piper betle* (betel pepper), *Pyrus* (pears), *Zingiber officinale* (ginger).

**Infested plant stages:** Seedling stage, vegetative growing stage, flowering stage, and fruiting stage

**Infested plant parts:** roots, and underground vegetative organs

### Geographic distribution

Records under the name *R. citrophilus* are included in the distribution (CABI/EPPO 1999).

### List of countries

#### **Europe**

- Belgium: restricted distribution (CABI/EPPO 1999; EPPO 2003)
- Croatia: absent, never occurred (EPPO 2003)
- Denmark: eradicated (CABI/EPPO 1999; EPPO 2003)
- France: restricted distribution (CABI/EPPO 1999; EPPO 2003)
- Germany: restricted distribution (CABI/EPPO 1999; EPPO 2003)
- Italy: restricted distribution (CABI/EPPO 1999; EPPO 2003)
- Netherlands: restricted distribution (CABI/EPPO 1999; EPPO 2003)
- Poland: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Portugal: eradicated (CABI/EPPO 1999)
  - Madeira: eradicated (CABI/EPPO 1999; EPPO 2003)
- Slovenia: restricted distribution (CABI/EPPO 1999; EPPO 2003)
- Sweden: eradicated (CABI/EPPO 1999; EPPO 2003)
- Switzerland: absent, intercepted only (CABI/EPPO 1999; EPPO 2003)
- United Kingdom: eradicated (CABI/EPPO 1999; EPPO 2003)

#### **Asia**

- Brunei Darussalam: present, few occurrences (Bridge 1993; CABI/EPPO 1999; EPPO 2003)
- China: eradicated (CABI/EPPO 1999)
  - Fujian: eradicated (CABI/EPPO 1999; EPPO 2003)
  - Taiwan: eradicated (CABI/EPPO 1999; EPPO 2003)
- India: restricted distribution (CABI/EPPO 1999)
  - Arunachal Pradesh: present, no further details (CABI/EPPO 1999; EPPO 2003)
  - Assam: present, no further details (Khan 1999; EPPO 2003)
  - Bihar: present, no further details (Khan 1999; EPPO 2003)
  - Goa: present, no further details (CABI/EPPO 1999; EPPO 2003)
  - Jammu and Kashmir: present, no further details (CABI/EPPO 1999; EPPO 2003)



- Karnataka: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Kerala: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Madhya Pradesh: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Maharashtra: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Manipur: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Nagaland: present, no further details (Khan 1999; EPPO 2003)
- Orissa: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Tamil Nadu: widespread (CABI/EPPO 1999; EPPO 2003)
- Uttar Pradesh: present, no further details (Khan 1999; EPPO 2003)
- West Bengal: present, no further details (Khan 1999; EPPO 2003)
- Indonesia: present, no further details (CABI/EPPO 1999)
  - Sumatra: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Israel: present, few occurrences (CABI/EPPO 1999; EPPO 2003)
- Japan: eradicated (CABI/EPPO 1999; EPPO 2003)
- Lebanon: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Malaysia: restricted distribution (CABI/EPPO 1999)
  - Peninsular Malaysia: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Oman: present, no further details (Waller and Bridge 1978; CABI/EPPO 1999; EPPO, 2003)
- Pakistan: present, no further details (Shahina and Maqbool 1992; CABI/EPPO 1999; EPPO, 2003)
- Philippines: present, no further details (Timm 1965; CABI/EPPO 1999; EPPO 2003)
- Singapore: present, no further details (AVA 2001)
- Sri Lanka: present, no further details (Sivapalan 1968; Gnanapragasam *et al.* 1991; CABI/EPPO, 1999; EPPO, 2003)
- Thailand: present, no further details (Timm 1965; CABI/EPPO 1999; EPPO 2003)
- Turkey: absent, never occurred (EPPO 2003)
- Yemen: present, few occurrences (CABI/EPPO 1999; EPPO 2003)

#### **Africa**

- Benin: widespread (EPPO 2003)
- Burundi: present, no further details (Bridge 1988a; CABI/EPPO 1999; EPPO 2003)
- Cameroon: present, no further details (Bridge *et al.* 1995; CABI/EPPO 1999; EPPO, 2003)
- Central African Republic: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Congo Democratic Republic: present, no further details (Elmiligy and Geraert 1971; CABI/EPPO 1999; EPPO 2003)
- Congo: present, no further details (Luc *et al.* 1964; CABI/EPPO 1999; EPPO 2003)
- Côte d'Ivoire: widespread (Adiko 1988; CABI/EPPO 1999; EPPO 2003)
- Egypt: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Ethiopia: present, no further details (O'Bannon 1975; CABI/EPPO 1999; EPPO 2003)
- Gabon: present, no further details (O'Bannon 1977; CABI/EPPO 1999; EPPO 2003)
- Gambia: present, no further details (Bridge 1993; CABI/EPPO 1999; EPPO 2003)

- Ghana: present, no further details (Addoh 1971; CABI/EPPO 1999; EPPO 2003)
- Guinea-Bissau: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Guinea: present, no further details (Luc 1968; CABI/EPPO 1999; EPPO 2003)
- Kenya: present, no further details (Ngundo and Taylor 1973; CABI/EPPO 1999; EPPO 2003)
- Madagascar: present, no further details (Luc 1968; CABI/EPPO 1999; EPPO 2003)
- Malawi: present, no further details (Saka and Siddiqi 1979; CABI/EPPO 1999; EPPO 2003)
- Mauritius: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Morocco: present, no further details (Sarah 1989; CABI/EPPO 1999; EPPO 2003)
- Mozambique: present, no further details (Evaristo 1969; CABI/EPPO 1999; EPPO 2003)
- Nigeria: present, no further details (Caveness 1965; CABI/EPPO 1999; EPPO 2003)
- Réunion: present, no further details (Vilardebó and Guerout 1976; CABI/EPPO 1999; EPPO 2003)
- Senegal: present, no further details (Luc 1968; CABI/EPPO 1999; EPPO 2003)
- Seychelles: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Somalia: present, no further details (Beccari and Scavazzon 1966; CABI/EPPO 1999; EPPO 2003)
- South Africa: restricted distribution (Jones and Milne 1982; CABI/EPPO 1999; EPPO 2003)
- Sudan: present, no further details (Decker *et al.* 1980; CABI/EPPO 1999; EPPO 2003)
- Tanzania: restricted distribution (Ngundo and Taylor 1973; CABI/EPPO 1999; EPPO 2003)
  - Zanzibar: present, no further details (Sebasigari and Stover 1987)
- Uganda: present, no further details (Ngundo and Taylor 1973; CABI/EPPO 1999; EPPO 2003)
- Zambia: present, no further details (Raemaekers and Patel 1973; CABI/EPPO 1999; EPPO 2003)
- Zimbabwe: present, no further details (Martin 1969; CABI/EPPO 1999; EPPO 2003)

### **Western Hemisphere**

- Argentina: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Barbados: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Belize: present, no further details (Pinochet and Ventura 1977; CABI/EPPO 1999; EPPO 2003)
- Bolivia: present, no further details (Bridge *et al.* 1982; CABI/EPPO 1999; EPPO 2003)
- Brazil: present, no further details (Zem and Lordello 1983; CABI/EPPO 1999)
  - Bahia: present, no further details (Zem and Lordello 1983; CABI/EPPO 1999; EPPO 2003)
  - Ceara: present, no further details (Zem and Lordello 1983; CABI/EPPO 1999; EPPO 2003)
  - Espirito Santo: present, no further details (CABI/EPPO, 1999; EPPO, 2003)
  - Minas Gerais: present, no further details (CABI/EPPO 1999; EPPO 2003)
  - Rio de Janeiro: present, no further details (Zem and Lordello 1983; CABI/EPPO 1999; EPPO 2003)
  - Sao Paulo: present, no further details (Zem and Lordello 1983; CABI/EPPO

- 1999; EPPO 2003)
- Canada: present, few occurrences (CABI/EPPO 1999)
  - British Columbia: present, few occurrences (CABI/EPPO 1999; EPPO 2003)
  - Colombia: present, no further details (Loos 1961; CABI/EPPO 1999; EPPO 2003)
  - Costa Rica: widespread (CABI/EPPO 1999; EPPO 2003)
  - Cuba: present, no further details (Stoyanov 1967; CABI/EPPO 1999; EPPO 2003)
  - Dominica: present, no further details (Edmunds 1969; CABI/EPPO 1999; EPPO 2003)
  - Dominican Republic: present, no further details (CABI/EPPO 1999; EPPO 2003)
  - Ecuador: restricted distribution (Bridge 1976; CABI/EPPO 1999; EPPO 2003)
  - El Salvador: present, no further details (Wehunt and Edwards 1968; CABI/EPPO 1999; EPPO 2003)
  - French Guiana: present, no further details (CABI/EPPO 1999; EPPO 2003)
  - Grenada: widespread (Edmunds 1969; CABI/EPPO 1999; EPPO 2003)
  - Guadeloupe: present, no further details (Scotto la Massese 1969; CABI/EPPO 1999; EPPO 2003)
  - Guatemala: present, no further details (Loos 1961; CABI/EPPO 1999; EPPO 2003)
  - Guyana: present, no further details (CABI/EPPO 1999; EPPO 2003)
  - Honduras: present, no further details (Loos 1961; CABI/EPPO 1999; EPPO 2003)
  - Jamaica: present, no further details (Cobb 1915; CABI/EPPO 1999; EPPO 2003)
  - Martinique: widespread (Scotto la Massese 1969; CABI/EPPO 1999; EPPO 2003)
  - Mexico: present, no further details (Taboada and Caballero 1968; CABI/EPPO 1999; EPPO, 2003)
  - Nicaragua: present, no further details (Wehunt and Edwards 1968; CABI/EPPO 1999; EPPO 2003)
  - Panama: present, no further details (CABI/EPPO 1999; EPPO 2003)
  - Peru: present, no further details (Sasser *et al.* 1962; CABI/EPPO 1999; EPPO 2003)
  - Puerto Rico: widespread (Romàn *et al.* 1974; CABI/EPPO 1999; EPPO 2003)
  - Saint Kitts and Nevis: present, no further details (CABI/EPPO 1999; EPPO 2003)
  - Saint Lucia: present, no further details (Edmunds 1969; CABI/EPPO 1999; EPPO 2003)
  - Saint Vincent and the Grenadines: widespread (Edmunds 1969; CABI/EPPO 1999; EPPO 2003)
  - Suriname: present, no further details (Maas 1969; CABI/EPPO 1999; EPPO 2003)
  - Trinidad and Tobago: restricted distribution (Scotto la Massèse 1969; CABI/EPPO 1999; EPPO 2003)
  - USA: restricted distribution (CABI/EPPO 1999)
    - Arizona: absent, never occurred (EPPO 2003)
    - California: eradicated (CABI/EPPO 1999; EPPO 2003)
    - Florida: present, no further details (Suit and Ducharme 1953; CABI/EPPO 1999; EPPO 2003)
    - Hawaii: present, no further details (Sher 1954; CABI/EPPO 1999)
    - Louisiana: present, no further details (Suit and Ducharme 1953;

CABI/EPPO 1999; EPPO 2003)

- Texas: present, few occurrences (CABI/EPPO 1999; EPPO 2003)
- United States Virgin Islands: restricted distribution (CABI/EPPO 1999; EPPO 2003)
- Venezuela: present, no further details (Haddad *et al.* 1973; CABI/EPPO 1999; EPPO 2003)

### **Oceania**

- American Samoa: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Australia: restricted distribution (Blake 1972; CABI/EPPO 1999)
  - Australian Northern Territory: present, no further details (CABI/EPPO 1999; EPPO 2003)
  - New South Wales: present, no further details (Blake 1963; CABI/EPPO 1999; EPPO 2003)
  - Queensland: widespread (Blake 1963; CABI/EPPO 1999; EPPO 2003)
  - South Australia: present, no further details (CABI/EPPO 1999; EPPO 2003)
  - Western Australia: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Belau: restricted distribution (Bridge 1988b; CABI/EPPO 1999; EPPO 2003)
- Cook Islands: present, no further details (Grandison 1990; CABI/EPPO 1999; EPPO 2003)
- Federated states of Micronesia: present, no further details (Bridge 1988b; CABI/EPPO 1999; EPPO 2003)
- Fiji: present, no further details (Cobb 1915; CABI/EPPO 1999; EPPO 2003)
- French Polynesia: present, no further details (CABI/EPPO 1999; EPPO 2003)
- Guam: present, no further details (Bridge 1988b; CABI/EPPO 1999; EPPO 2003)
- Niue: present, no further details (Orton Williams 1980; CABI/EPPO 1999; EPPO 2003)
- Norfolk Island: present, no further details (Khair 1982; CABI/EPPO 1999; EPPO 2003)
- Papua New Guinea: present, no further details (Bridge and Page 1984; CABI/EPPO 1999; EPPO 2003)
- Samoa: present, no further details (Orton Williams 1980; Grandison 1996; CABI/EPPO 1999; EPPO 2003)
- Solomon Islands: present, no further details (Bridge 1988b; CABI/EPPO 1999; EPPO 2003)
- Tonga: present, no further details (Kirby *et al.* 1980; CABI/EPPO 1999; EPPO 2003)

### **Biology and ecology**

*R. similis* is a migratory endoparasitic species which completes its life cycle within the root cortex and tissues of corms and tubers.

In bananas, penetration occurs mostly near the root tips, but nematodes can invade along the entire length of the root. Females and all juvenile stages are infective although males, morphologically degenerate (without stylet), are probably not parasitic. After entering the roots of banana, the nematodes occupy an intercellular position in the cortical parenchyma where they feed on the cytoplasm of nearby cells, causing cavities which then coalesce to appear as tunnels. Invasion of the stele is never observed, even in heavily infected roots. The presence of lignified and suberized layers in endodermal cells of endodermal layers limits invasion of the vascular bundle by *R. similis*. Phenolic compounds play a significant role in the host plant's defence response to the nematode. High levels of lignin, flavanoids,

dopamine, caffeic esters and ferulic acids were associated with low levels of penetration in resistant cultivars (Valette *et al.* 1998b).

It is within infected tissues that females lay their eggs, with an average of four to five eggs per day for two weeks. The complete life cycle from egg to egg spans 20-25 days at a temperature range of 24-32°C, the eggs hatch after 8-10 days and the juvenile stages are completed in 10-13 days (Gowen and Quénehervé 1990; Loos 1962).

In the absence or reduced densities of competitors such as *Helicotylenchus multincinctus*, high populations of *R. similis* colonize the entire set of banana roots. The presence of competitors reduces the density of *R. similis* in the soil and roots and restricts it to the areas close to the rhizome (Queneherve 1990).

*R. similis* forms a disease complex with *Fusarium oxysporum f.sp cubense* and the damage caused by *R.similis* was greater in the presence of the fungus. The percentage of root rots caused by the fungus was 6.5% in the presence of *R. similis* and 4% with the fungus alone (Abdel-Hadi *et al.* 1987).

In coconut, *R. similis* takes about 25 days at 25-28°C to complete its life cycle. Most juveniles and adults, including gravid females, infest healthy, succulent root tips. In the field, the nematode can survive for six months in moist soil (27-36°C) and only one month in dry soil (29-39°C). Under glasshouse conditions, it survives for longer periods: 15 months in moist soil (25.5-28.5°C) and three months in dry soil (27-31°C) (Griffith and Koshy 1990).

#### **Means of movement and dispersal**

##### **Plant parts liable to carry the pest in trade/transport:**

- Bulbs/tubers/corms/rhizomes: Eggs, juveniles, adults; borne internally; visible under light microscope
- Growing medium accompanying plants: Eggs, juveniles, adults; borne internally; visible under light microscope
- Seedlings/micropropagated plants: Eggs, juveniles, adults; borne internally; visible under light microscope
- Roots: Eggs, juveniles, adults; borne internally; visible under light microscope
- Stems (above ground)/shoots/trunks/branches: Eggs, juveniles, adults; borne internally; visible under light microscope

##### **Plant parts not known to carry the pest in trade/transport:**

- Bark
- Fruits (including pods)
- Flowers/inflorescences/cones/calyx
- Leaves
- True seeds (including grain)
- Wood

##### **Transport pathways for long distance movement:**

- Conveyances (transport vehicles): with soil
- Mail: with plants
- Non-host plant material
- Containers and packing: with planting material
- Soil, gravel, water, etc.
- Travellers and baggage: with plants

**Economic impact**

*R. similis* causes non-specific general decline symptoms on coconut such as stunting, yellowing, reduction in number and size of leaves and leaflets, delay in flowering, button shedding and reduced yield (Griffith and Koshy 1990; Koshy *et al.* 1991). In pot experiments, soil population levels of 100 nematodes per seedling cause a 35% reduction in height and a 14% reduction in girth of coconut palms over a five year period (Koshy and Sosamma 1987). In large field tanks (microplots) in India after seven years, an initial inoculum level of 1000 nematodes per seedling (10 nematodes per 35 640 cmn of soil) gave reductions of 17, 14 and 35% over uninoculated control in height, number of leaves and girth of stem, respectively (Koshy *et al.* 1991).

**Phytosanitary risk**

*R. similis* is spread on infested vegetative planting material such as rootstocks, corms and tubers. It is a tropical nematode and can become a pest of any of the susceptible host crops in subtropical and tropical climates. Crops in temperate climates are not at risk.

**Symptoms**

In coconut, *R. similis* causes non-specific general decline symptoms such as stunting, yellowing, reduction in number and size of leaves and leaflets, delay in flowering, button shedding and reduced yield. *R. similis* infestation produces small, elongate, orange-coloured lesions on tender creamy-white roots. Tender roots of coconut seedlings with heavy infestation become spongy in texture. Surface cracks develop on the semi-hard, orange-coloured main roots. Lesions and rotting are confined to the tender portions of the root. Lesions are also not conspicuous on the secondary and tertiary roots as these are narrow and rot quickly on infestation (Griffith and Koshy 1990).

**Morphology** (Orton Williams and Siddiqi 1973)**Female**

The female's body is straight to slightly arcuate ventrally; cuticle distinctly annulated. Lateral field with four incisures, not areolated except towards extremities, arising from near median oesophageal bulb and ending near tail terminus; inner incisures coalescing near middle of tail. Lip region hemispherical, sometimes offset, usually with 3-4 annules; sclerotization strong; dorsal and ventral arms of framework not wider than submedians; lips six, equal. Anterior cephalids just posterior to labial sclerotization. Spear about 18 µm long, with well developed round basal knobs which are usually indented anteriorly; dorsal knob sometimes appearing larger than subventrals. Median oesophageal bulb well developed, round to oval, valvular apparatus prominent. Oesophageal glands 3, in separate lobes, overlapping intestine dorsally and dorso-laterally; dorsal gland anterior. Hemizonid 3 annules long, just anterior to excretory pore which is at or just behind the level of the oesophago-intestinal valve. Vulva prominent, just postequatorial. Reproductive organs paired, opposed, outstretched. Spermathecae spherical, usually packed with small rod-shaped sperms. Ovaries generally with a single row of oocytes. Intestine filled with spherical granules, indistinctly overlapping rectum. Tail somewhat elongate-conoid with a narrow rounded or indented terminus.

**Male**

Oesophagus and spear degenerate; median bulb and valvular apparatus indistinct, spear without distinct knobs. Lip region elevated, 4-lobed, with lateral lips

considerably reduced, not strongly sclerotized, with 3-5 annules posteriorly. Hemizonid just anterior to excretory pore which is usually 2-3 body widths behind median oesophageal bulb. Single testis, outstretched anteriorly; spermatocytes in 3 rows followed by 5; spermatozoa rod-like. Bursa coarsely crenate, enveloping about two thirds of tail. Spicules strongly cephalated, 18-22  $\mu\text{m}$  long, with pointed distal ends. Gubernaculum rod-like, protrusible, with distinct sharp claw-like titillae at distal end.

**Note:** Cobb (1893) published the descriptions of *Tylenchus granulosus n. sp.* and *Tylenchus similis n. sp.* from diseased banana plant material sent to him in New South Wales from Fiji in July, 1891. *T. granulosus* is the female and *T. similis* the male of *R. similis*, *T. granulosus* having page priority over *T. similis*. To preserve the well known name 'similis' for this widely distributed economic pest, Sher (1968) proposed its retention, regarding *T. granulosus* as a senior synonym.

SEM studies of populations of *R. similis* collected from different countries showed differences in morphological characteristics, especially in the number of anterior hypopygmata in the males and annules terminating the vulva of the females. Many of the Indonesian populations were found to have a forked tail end (Elbadri *et al.* 1999).

#### **Similarities to other species**

*R. similis* has a superficial resemblance to the genus *Pratylenchus*, but can be distinguished by having a median vulva with two genital tracts in the female as opposed to a posterior vulva with one tract. It can be most easily distinguished from other species of *Radopholus* by the length of the female tail. *R. similis* is also similar to *Hirschmanniella* spp. Molecular methodologies are being increasingly employed to investigate the diversity of *R. similis* populations.

#### **Risk assessment of pest**

	Seednuts	Embryo cultures	Pollen
<b>Entry</b>	<i>R. similis</i> is a migratory endoparasitic species which completes its life cycle within the root cortex and tissues of corms and tubers. Records of transmission in tissue cultures are not relevant for coconuts.		
<b>Establishment</b>	It is a tropical nematode and can become a pest of any of the susceptible host crops in subtropical and tropical climates.		
<b>Spread</b>			
<b>Economic</b>			
<b>Level of risk</b>	Minimal	Minimal	Minimal
<b>Pest status</b>	NQP	NQP	NQP
<b>Pest management</b>	No management is required for this pest for plant part pathways, but soil as a contaminant of nuts should not be permitted import.		

**Note :** QP – quarantine pest ; NQP- Non-quarantine pest

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- the Ivory Coast. *Revue de Nématologie*, 11:109-113.
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