

## Chapter 5

# More about delimiting surveys

### 5.1. What is different about delimiting surveys?

The International Standard for Phytosanitary Measures (ISPM 6) defines a delimiting survey as follows:

...survey conducted to establish the boundaries of an area considered to be infested by or free from a pest.

These surveys are usually carried out to determine the boundaries of an infestation rather than to define an area that is free from a pest.

The main difference between delimiting surveys and the other surveys covered in these guidelines is how sites are selected. The initial detection site is used as a starting point to determine how the pest arrived, where it originated and to where it might have spread. Determining to where the pest might have spread will determine where the surveying and resources for managing the pest need to be focused.

### 5.2. Trace-back and trace-forward techniques

Because the site where a pest is first detected might not be the initial site of the infestation, a delimiting survey can be used to identify the original source of the pest. The process of backtracking to find the original source of the pest is called 'trace-back', and the process of tracing the pest's possible spread is called 'trace-forward'. If trace-back enquiries are successful in identifying the likely original site of introduction of the pest, trace-forward activities will help to locate areas that might be infested and will need to be surveyed. The results of a delimiting survey will often have consequences for quarantine and trade and may lead to attempts to eradicate the pest. The results may be used to justify the establishment of a quarantine area around the infestation and to decide if eradication is possible.

## 5.3. The role of delimiting surveys in biosecurity plans

Given that the purpose of a delimiting survey is usually to identify where an exotic pest is present rather than identifying areas free from a pest, plant health authorities may have a generic survey plan for exotic pests as part of an incursion response plan. These plans are referred to as biosecurity<sup>14</sup> plans.

Biosecurity planning is a strategic exercise in which pest threats are identified and ranked according to the likelihood of the pests being introduced and establishing in sensitive areas where they would affect an industry. The plan would likely include strategies for preventing the entry of any exotic pests that have the potential to damage an industry, including its trading prospects. Biosecurity plans will usually identify pre-emptive actions that plant health authorities could take to reduce the impact of pests that enter and establish in a new area. These plans document how authorities would respond to a new pest incursion, such as what eradication procedures would be needed once a pest has been detected, and include instructions for performing a delimiting survey. As a result, while a delimiting survey of an exotic pest is being conducted, other processes such as eradication and sterilising of the known infested sites are often performed at the same time.

## 5.4. Who conducts delimiting surveys?

A delimiting survey will typically be undertaken by a regulatory agency, often the NPPO. However, the operational and regulatory capacity may reside with other agencies within the jurisdiction. In Australia, for example, plant health is the responsibility of the State governments and regulatory action on new pest incursions is usually in the hands of State agriculture departments.

At an early stage following the detection of a new pest incursion, the regulatory agency performing the delimiting survey will appoint staff to act as planning and logistics managers. They will be responsible for:

- designing and conducting the survey
- applying legislation giving authority to enter premises to undertake surveys and to apply other measures to contain the pest
- ensuring good hygiene and phytosanitary measures are applied during the survey
- record keeping.

---

<sup>14</sup> The term 'biosecurity' was brought into prominence with the introduction in New Zealand of the *Biosecurity Act* (1993) that sought to 'restate and reform the law relating to the exclusion or eradication and effective management of pests and unwanted organisms'. The term 'biosecurity' is not defined in the legislation, but a definition has been proposed by Penman (1998) as 'effective management of risks by a system of coordinated pre-border, border, management and sector responses aimed at preventing the establishment and spread of organisms that may have adverse effects on the economy, environment and people's health'.

It is essential that regulatory authorities can access the sites that need to be surveyed. In order to do this, they should have legislation in place to enable staff to enter these sites and, if necessary, to quarantine the movement of commodities, planting stock, farm machinery and any other means by which the pest might be dispersed.

The planning and logistics managers will need the support of technical specialists to identify pests that are not easily recognised or to confirm preliminary diagnosis by field staff. Technical specialists will also play a key role in advising on the biology of the pest, particularly its means of spread.

### 5.5. Survey design

In the absence of any biosecurity or pest incursion plans, follow the steps described in Chapters 2 and 7 with the following additional considerations and adjustments.

#### Steps 1 and 2

Complete these steps.

#### Step 3

You will need to find out as much as possible about the biology of a pest to enable you to identify all the likely sites that it might have infested. Research the epidemiology of the pest, its means of survival, reproductive rate, life span, and the effects of environmental factors.

Estimate how long the pest could have been at the site before it was detected. Even sedentary and slow-moving pests can disperse some distance if not detected early. Some pests have escaped detection for several years.

You will need to closely consider how the pest can be spread, as discussed in Box 12 on the next page.

#### Steps 4 and 5

In completing these steps, you will need to identify all the known hosts and where these are, especially those that are close to the site of detection. A list of properties and sites with host plants can be developed from a number of sources, including industry and government records and personnel, local grower groups and cooperatives, fruit packers and distributors, extension staff, researchers and property owners. If available, aerial photographs can also be useful for identifying areas that are densely populated by hosts, such as production areas. For cultivated hosts, the most susceptible varieties are targeted if these are known.

Consider all alternative hosts, as well as the susceptibility of endemic flora in remnant forest, parklands, gardens and other areas close to the detection site. Keep in mind that the symptoms might be masked or subtle on resistant hosts. Viruses might be latent in some cultivars with the host expressing symptoms only in response to particular environmental conditions, or when in mixed infections.

#### Step 6

Complete this step. Access any biosecurity or pest incursion plans as a priority.

## Box 12. Human-assisted and natural dispersal of pests

### Human-assisted dispersal

A priority for survey managers will be to determine the likelihood that the pest may have been dispersed with commodities from packing houses and planting stock from commercial nurseries or seed suppliers. These commodities have the potential to disperse an exotic pest over great distances and to many sites very quickly. The immediate destinations of bulk commodities and planting stock will likely be documented, but sales from retailers may not be so readily traced.

Consider also the following:

- Is the pest likely to have been carried by workers and their equipment to other properties, including their homes?
- Is harvesting equipment shared between properties?
- Are packing boxes and other storage materials re-used?
- What vehicles could have come in contact with the pest and where have these travelled?
- How are agricultural wastes, such as manure, disposed of?
- Has infected produce or plant material entered the market?

### Natural dispersal

Consider the following:

- Is the pest spread by wind or rain?
- What is the prevailing wind direction and what have been recent weather conditions in the area?
- Can the pest be dispersed by waterways, irrigation channels or by flood events?
- Over what distance is the pest likely to spread by natural means considering recent weather conditions, life stage of the pest, flight and survival characteristics?
- Is the pest vectored by insects, mites, fungi, nematodes or other organisms?
- Are vectors present in the area and, if so, at what density?
- How effective are vectors at transmitting the pest? Does the pest replicate within the vector?
- Do endemic close relatives of known vectors also transmit the pest?
- Is there dropped fruit on the ground that could harbour the pest?
- For weed seeds and the seeds of parasitic plants, are they dispersed by birds?
- Are there geographical features, such as a sea border, that limit the direction in which a pest can travel?

### Steps 7 to 9

Determining the sites will be largely based on the nature and dispersal of the pest and the nature and distribution of hosts (steps 3 to 5). The sites would need to include distribution points of infested host material, such as produce or root stocks that may have entered the market.

You may need to develop a questionnaire. Read Box 13, Using a questionnaire to identify survey sites (page 108), which includes information on designing a questionnaire.

As a result, the survey may need to include all production places within an area, district or place and to target all susceptible species in orchards, nurseries, areas of natural vegetation, residential, public and commercial properties.

### Step 10

The survey design is simply a delimiting survey as detailed in these steps.

### Step 11

While statistics will not be needed to calculate how many sampling points are required, you may have to choose a sampling structure, such as a grid of traps, that is statistically sound. A random element could be added by inspecting a few sites where it is thought that the pest is unlikely to be present or that is between sampling points, such as weeds on road verges.

### Step 12

Not applicable as the timing will be set by the date that the first detection was observed and how quickly a survey can be planned and organised.

### Steps 13 and 14

Specimens should be collected together with details in accordance with ISPM 8, and submitted to an official collection to confirm identification and to provide a pest record. See also Chapter 3.

### Steps 15 and 16

Complete these steps.

### Step 17

While a NPPO may have legislation in place to access sites, you may still need to approach people to gain access to sites such as domestic gardens or farms.

### Step 18

It is unlikely that there would be sufficient time to perform a pilot survey, unless the plan is designed and practised before the target pest is actually detected.

### Step 19

Complete your survey.

## Box 13. Using questionnaires to identify survey sites

An important component of the delimiting survey is a trace-back and trace-forward analysis to determine the source of the outbreak and to identify other premises that might have been exposed because of closeness to or contact with the infested property, through movement of infected planting material or fruit, or by sharing employees and equipment.

Face-to-face interviews or questionnaires distributed to property owners can be a useful way of determining whether hosts are present on a property. The questionnaire will be useful for determining production details, the ownership of properties where hosts occur, and for collecting preliminary trace-forward and trace-back information, including the source of planting material, movements of equipment and staff, and propagation practices. This information will allow you to develop a risk profile for each property.

See also step 4 in Chapter 2.

Questionnaires are particularly useful in trace-forward and trace-back investigations where seed or other forms of planting material are suspected as the source of the pest, especially if you can verify any reported instances of the pest. A questionnaire may be of less use in trace-forward and trace-back investigations when the pest has blown or flown in.

Where the new pest is associated with purchased seed or planting material it will be necessary to go to the supplier and repeat the survey questionnaire to seek out the original source of the planting materials and to identify the places to which the planting material, and therefore the pest, might have been distributed. The same set of questions should be put to farmers, seed suppliers and nursery owners who can be traced from answers to the questionnaire.

### Designing a questionnaire

The questions could be written to identify:

- the source of the planting material
- the destination of plants and plant products which may have been moved from the property, be it a farm, an orchard or a nursery
- the location of properties that share equipment or have labour that moves from property to property such as when harvesting fruits and vegetables
- adjacent fields/properties owned by the farmer on whose land the pest was detected
- the movement of commercial apiarists (if applicable)
- the movement of other visitors that may have been on the property
- climatic conditions or weather events that might favour the establishment and spread of the pest.

- Enquire about people who may have been travelling, especially internationally, as they may have brought pests back with them. There are reports based on circumstantial evidence that some rusts, smuts and ergots might have been introduced into new areas on the clothing of returning travellers.

If the responses to the questionnaire are to be entered into a database or other computer program, a simple way of saving a lot of time is to design your questionnaire and database so that they match in design layout. This will speed up the process of entering the data into the storage system. See Step 15, Chapter 2.

Once results are received from questionnaires and interviews with workers, property owners etc. these are used identify other sites and locations that must be surveyed.

## Step 20

As the purpose of the survey is to identify where the pest has spread, a map of its distribution would be useful.

## Step 21

Complete this step.

# 5.6. Example delimiting survey case studies

The following case studies are in Chapter 8.

- Case study T: Delimiting survey of papaya ringspot virus
- Case study U: Delimiting survey for Huanglongbing disease of citrus and its vector the Asian citrus psyllid in Papua New Guinea
- Case study V: Delimiting survey for red-banded mango caterpillar in northern Queensland
- Case study W: Delimiting survey of the Queensland fruit fly in Rarotonga, Cook Islands.



## Reference

Penman, D.R. 1998. Managing a leaky border: towards a biosecurity research strategy. Wellington, New Zealand, Ministry of Research, Science and Technology, 61p.





## Chapter 6

# More about general surveillance

ISPM 6 briefly covers what is *required* under the term ‘general surveillance’. The requirements fall into two categories of activities. The first is collecting information about a pest. The second is to develop clear communication between NPPOs and other people who have information about pests.

## 6.1. Collecting information about a pest

ISPM 6 refers first to the process of collecting information about a pest. This is covered in detail at step 3 in Chapter 2 (see Section 2.4.1). The standard then requires that the information collected from the various sources be compiled and verified. The information should be both stored and retrievable. Verifying the information sources on pests is also covered under step 3 in Chapter 2 (see Section 2.4.2).

The ISPM states that this information about the pest can be used to:

- support NPPO declarations of pest freedom
- aid early detection of new pests
- report to other organisations such as RPPOs and FAO
- compile host and commodity pest lists and distribution records.

In other words, the information collated can be used as one part of designing specific surveys as described in Chapter 2, or it may be adequate on its own to develop a report of the pest status of an area which can be used for other purposes. If the information compiled is inadequate for these other purposes, then specific surveys can be performed to provide extra information about the pest.

This process of compiling information on pests is also necessary when developing target pest lists. See Box 14, Developing target pest lists, on the next page.

## Box 14. Developing target pest lists

Pest lists are an inventory of pests in an area. Target pest lists are an inventory of pests in the surrounding regions and countries that threaten to enter the area.

Target pest lists are used to focus surveillance activities and incursion management planning on high-priority threats. Depending on their purpose, target lists vary in their scope. For example, they could include all high-priority exotic pests that threaten an industry on all pathways from all sources, or simply focus on pests of concern from one source along one pathway.

The development of target lists is based on identifying pests that could arrive, then performing a risk assessment for each pest.

If the list of pests is developed as part of a market-access application, the list is restricted to the range of pests associated with commodity host plants and materials in the exporting country or region. If the list is being developed to create a list of quarantine pests, then all pests from all neighbouring countries need to be included, as well as from countries from which people and cargo arrive.

To identify pests associated with a host or a neighbouring area, a pest list needs to be developed. This process is detailed in Section 3.1.

### Assessing the threat posed by the listed pests

The aim is to assign an overall level of risk for each pest, based on the probability of its entry, establishment, spread and consequences. The overall risk rating is usually expressed in qualitative terms (e.g. on a scale of 1 to 5, or in terms of 'low', 'moderate' or 'high') and can be used to decide which pests pose a higher priority and warrant inclusion on a target list.

Guidance on the process and considerations involved in assessing pest risk are provided in detail in ISPM 11, Pest risk analysis for quarantine pests.

## 6.2. Open communication channels with NPPOs

The standard requires that there be *communication channels to transfer information from the sources* [of pest information] *to the NPPO*. It proposes that, if necessary, the communication could be improved by introducing incentives to people to report information about a pest. The incentives suggested are:

- legislative obligations (for the general public or specific agencies)
- cooperative agreements (between the NPPO and specific agencies)
- use of contact personnel to enhance communication channels to and from NPPOs
- public education/awareness programs.

There are no further details about these suggestions in the ISPMs. The next section contains information about public education/awareness programs.

Another incentive that has been used successfully<sup>15</sup> is to provide a free pest identification service, which encourages people to become involved and to send in unusual specimens.

## 6.3. Developing awareness campaigns

Awareness campaigns are often initiated to alert farmers and the public to the detection of a pest new to an area, the likely arrival of an exotic pest or when an endemic pest increases in significance in response to changing environmental conditions or cropping practices. The awareness material has two main thrusts: to inform the audience about the targeted pests and to provide the audience with instructions on how they can assist.

### 6.3.1. Providing information about pests

Providing information on the pest can be achieved using a range of media. These fall into the categories of handouts and public announcements.

#### 6.3.1.1. Preparing handouts

Raising awareness through the preparation and distribution of booklets or fact sheets (sometimes called ‘pest alerts’) is a common approach. Postcards, posters, calendars and bookmarks are other easily distributed materials. The information could be included in existing newsletters.

Effective material allows the audience to readily recognise the pest or pest symptoms. Therefore handouts should include:

- information about the name of the pest and why it is important
- descriptions of the pest and/or pest symptoms
- colour photographs of the pest and/or symptoms
- description of the host or environment where the pest might be found
- when the pest might be found, for example, in terms of seasons or host growth stage
- who you are—the group preparing the handout and why it is your topic.

Other attributes of the handout should be that:

- the material is easy to read and understand
- the material captures the reader’s attention
- the information is presented on a single sheet of paper—single or double-sided.

If you make computer files of the handouts available (such as on the Internet), remember that the handouts are most likely to be printed out in black and white, which may affect the information that you have provided. Print it out and look at what other people would see and make any improvements possible.

---

<sup>15</sup> By NAQS, as part of an integrated system to protect Australia’s northern borders from exotic pests

### 6.3.1.2. Pest alerts

Pest alerts can be structured as follows: introduction (circumstances in which the pest has been found), identification of the pest, biology of the pest, distribution and hosts, symptoms on hosts, further reading, and how to report a sighting of the pest. There are many pest alert examples available on the Internet.

Example: SPC

Pest alerts released by the SPC can be viewed at <[http://www.spc.int/pps/pest\\_alerts.htm](http://www.spc.int/pps/pest_alerts.htm)>.

### 6.3.1.3. Booklets

Small flip charts can be made that describe a series of pests that people can look out for. Typically, these are small, sturdy, ring-bound booklets with water-resistant pages. They are small enough (e.g. page size 11 by 15 cm) to fit easily into a car glove box or to be carried. The pages on each pest would have colour photos of the pest and/or symptoms, then brief details of the pest's name, characteristics, hosts, known distribution, potential impact and any other useful information such as other organisms it could be confused with.



Example: WEEDeck

The WEEDeck series targeted selected weeds exotic to Australia; see <<http://www.weeds.org.au/weeddeck.htm>>, with examples of weed cards provided on the publisher's website at <<http://www.sainty.com.au/weeddeckpg1/weeddeckpg1.html>>.

Example: Forests and timber: a field guide to exotic pests and diseases

This booklet of Australian forest pests is available free from the Australian quarantine website at <<http://www.aqis.gov.au>> by selecting 'Publications' then choosing 'Timber—a field guide to exotic pests and diseases'.

### 6.3.1.4. Public announcements

Awareness activities may also include public seminars, talks with small groups of locals, information stalls at community events, road signage, and radio and television announcements or press releases. If your organisation has a website, information—including copies of pamphlets and pest alerts—can be made available to anyone with access to the Internet.

The timing of campaigns can be important to the success of the campaign, as demonstrated in the example on the next page, describing involvement of the public in Siam weed (*Chromolaena odorata*) eradication in Queensland, Australia.

#### Example: 'Lord of the weeds' school competition

The Cooperative Research Centre (CRC) for Weed Management has run a competition in schools in which students design a strategy to manage weeds that are in the school, or in the local area. The winning school was awarded \$1000 to be used as they wished. The CRC provided suggested lesson activities, student proformas to guide students through writing the report and, for teachers, a marking guide and appropriate contacts. These materials and further information are available on the Internet at <[http://www.weeds.crc.org.au/education\\_training/school\\_resources.html](http://www.weeds.crc.org.au/education_training/school_resources.html)>.



#### Example: Involving the public in an eradication campaign for Siam weed

The Northern Australia Quarantine Strategy (NAQS) undertakes vigorous public-awareness campaigns to bring potential pests of quarantine concern to the attention of the general public. This includes talks and demonstrations to schoolchildren, landholders and interest groups (e.g. Landcare groups); media articles and radio broadcasts (in regional areas); preparation and distribution of awareness material including newsletters, booklets and calendars; and inviting landholders and government officials to submit specimens of unknown pests or weeds for identification.

Effective public-awareness campaigns can assist in mapping the distribution of weeds. As part of a campaign to eradicate Siam weed, the Queensland Department of Natural Resources and Mines undertook a vigorous advertising campaign timed to coincide with this species' flowering season in May to August, when it is usually most conspicuous. Television advertising and newspaper articles showed the plant in flower and advised concerned residents who to contact if they thought they had seen the weed. This publicity campaign led to the reporting and confirmation of four previously unknown infestations. Strategic advertising for this and other species that are eradication targets will continue to play an important role in the success of the eradication efforts.

Television and newspaper advertising timed to coincide with the flowering season of a weed can be valuable in identifying new infestations. While television is probably the most effective tool, the charges for production and advertising are likely to be prohibitive and thus may not be the most affordable means. In the case of the Siam weed eradication campaign in Queensland, the cost was reduced by television stations agreeing to broadcast the advertisements at little or no cost as part of community service obligations. Posters, photographs, talks accompanied by slide presentations, live specimens (where legislation permits such use) and laminated herbarium specimens also provide useful tools to illustrate weeds of concern to the general public. In far north Queensland, NAQS illustrates several weeds of concern in the annual Torres Strait and Cape York calendars, with photographs appearing on the month when that species is most likely to be flowering. In all cases, it is vital that members of the public be informed about who they should contact if they think they have found a weed of concern. Timely identification and feedback is provided for all reports made or specimens submitted.



### 6.3.2. Targeting audiences

People who work regularly among host crops or in targeted areas are likely to be aware of the pests that are normally present and so be most likely to notice a new pest or anything unusual. Such groups of people include farmers and farm personnel, extension staff, field technicians and interested community groups. The general public can also be very helpful in increasing the area covered and the number of people looking for the pest, as can experts in taxonomy and plant health. Projects could be set up to involve students and staff in schools and universities to both increase their knowledge of entomology or plant pathology and to help with finding pests. Domestic pest controllers and staff at garden nurseries can also be useful groups to assist with reporting new insect pests.

It is important to identify and inform any groups who may already be conducting pest surveys or control programs of pest issues, as they may be unaware of other pest surveillance programs operating in their region.

Example: Forests and timber: a field guide to exotic pests and diseases

This booklet (see page 114) targets people who work with timber—wharf workers, container depot staff, timber handlers, timber-yard workers, forest workers and forest technical staff.

### 6.3.3. Reporting networks—how the audience can report pests

Once you have informed the people who may be able to assist in detecting a pest, you need to have a way for people to let you know, and a system in place to keep track of pest reports. This will enable you to manage a series of pests and provide you with the information to assess if the campaign is effective.

Some of the options that have been used are free-call telephone services, text messaging to a central database, and providing a direct contact number, fax and email of a plant protection officer on handouts.

Example: Pestex early warning system for pests of corn

The Philippines Department of Agriculture has developed a corn (maize) pest surveillance system called Pestex to help prevent pest outbreaks and reduce economic losses caused by plant pests. One of the objectives of the program is to establish a farmer-based surveillance network in order to determine pest status, generate forecasting data and provide information to assist pest-management decisions. Farmers and agricultural technicians report pest data to a central authority (the Bureau of Primary Industries) by mobile phone text message. Information is added to a database and verified by technicians who either visit the areas reported to be affected, or seek samples from more remote areas. The appropriate response plan is then implemented.

Example: Telephone hotline for pest reporting

The exotic plant pest hotline is a freecall telephone service provided primarily for members of Australia's plant production sectors and plant health services to report suspect exotic plant pests. The hotline is staffed during business hours. Callers are directed to government staff in their State who have expertise on the pest and who are able to determine what course of action should be taken.

The hotline number is promoted by the Australian Government Department of Agriculture, Fisheries and Forestry's public relations section, through awareness campaigns that distribute the number on pamphlets and bookmarks, and by including the number on all pest booklets produced.

For more information go to <<http://www.outbreak.gov.au>>.

Example: GrainGuard

In Western Australia, the Department of Agriculture operates GrainGuard, a program targeting both specific and general surveillance of grain threats. The program involves growers and agribusiness, integrating grain pest response activities in Western Australia. It includes distribution of information on exotic pest threats of grain crops and collection kits to promote submission of suspect exotic pests to the Department of Agriculture. For more information go to <<http://www.agric.wa.gov.au>>. Choose 'Crops' on the menu then 'GrainGuard'.





## Chapter 7

# Step 21. Reporting the results

### 7.1. To whom should you report?

If you were funded to do a survey, it is likely that your funding body will require a report of the survey. If the survey was designed for trade-related purposes, NPPOs will need to be given a copy of the report. If you are representing the NPPO, there are obligations as to whom notifications of trade-related pest detections must be reported. More information is given at Sections 7.7 and 7.8. If you belong to an academic institution, you may need to produce a report for your head of department, or you may intend to submit the findings to a journal.

### 7.2. Writing a summary

It can be useful to have a simple summary that can be provided as follow-up information for the people who were involved in the survey; from the team members to the local farmers, rangers and community leaders. This will acknowledge their assistance and show that their involvement was appreciated. This is particularly important if you need to continue to return to the sites, such as for monitoring of pests, because you need to keep the communication channels open with everyone involved.

Reports or summaries that are to be given to people involved in the survey can be much simpler than a full report and may be reduced to a handout or pamphlet. In this instance, less detail is required, but content such as photos and anecdotes are encouraged.

In this situation, the pamphlet text might include:

- the survey title and team members
- the aim of the survey, including which pests, hosts and sites were targeted and why
- what was found
- what it means to the people who will read the pamphlet

More information is provided about pamphlets and educational material in Chapter 6.

## 7.3. Press releases

A summary may be sufficient for press releases. If you need to write a press release, you will be working for an organisation that will likely have public relations staff who can help you with the structure and appropriate content of the release, and with its distribution. Some organisations, such as the SPC, include press releases on their websites; go to <http://www.spc.org.nc/> and choose 'Press releases' from the menu.

## 7.4. Newsletter articles

Newsletters are typically a way of informing a select group, such as fruit growers, as to what is current news in the field. Depending on the newsletter, a simple summary and contact details may be sufficient. Others may want submissions that are more detailed and more like a journal article.

## 7.5. Writing a basic report

A basic report would include material from a number of the steps in the survey plan, so most of the work is done already, and the writing will be a shortened version of this accompanied by results and an interpretation of the survey findings.

### 7.5.1. Components of a basic report

A basic report should provide at least the following information:

- the survey title and team members, from step 1
- the reason for surveying, from step 2
- background information on the pest, host and sites of interest, including discussion of any earlier, related surveys, from steps 3–6
- the survey design methods in detail—this would include site selection from steps 7 to 11, timing of the survey from step 12, the type of data and specimens collected, from steps 13 and 14
- how the data were analysed and interpreted, from step 20
- conclusions that can be drawn about the survey findings, and how these relate back to the purpose of surveying.

The report could also have a brief abstract near the beginning and could include a glossary of terms and acknowledgments such as from whom permission and funding were received.

## 7.6. Formal reports with set formatting

For reports that are to be submitted to funding bodies, NPPOs or journals, there will be information provided by the organisations as to how the report should be written and formatted. This information needs to be sought from the specific organisation.

For situations involving trading partners, there can be obligations as to the format and content of pest reports. These are covered in ISPMs 13 and 17. The reporting obligations are reproduced in part in Sections 7.7 and 7.8.

## 7.7. ISPM 13—Reporting of pests in imported consignments

This standard describes as follows the actions to be taken by NPPOs about the notification of:

- failure to comply with phytosanitary requirements
- detection of regulated pests
- failure to comply with documentary requirements, including:
  - absence of phytosanitary certificates
  - uncertified alterations or erasures to phytosanitary certificates
  - serious deficiencies in information on phytosanitary certificates
  - fraudulent phytosanitary certificates
- prohibited consignments
- prohibited articles in consignments (e.g. soil)
- evidence of failure of specified treatments
- repeated instances of prohibited articles in small, non-commercial quantities carried by passengers or sent by mail...
- an emergency action taken on the detection in an imported consignment of a regulated pest not listed as being associated with the commodity from the exporting country
- an emergency action taken on the detection in an imported consignment of organisms posing a potential phytosanitary threat.

The importing contracting party is required to notify the exporting contracting party as soon as possible about significant instances of non-compliance and emergency actions applied to imported consignments. The notification should identify the nature of non-compliance in such a way that the exporting contracting party may investigate and make the necessary corrections.

Notification should be timely and follow a consistent format. Where there is a significant delay in confirming the reason for the notification (e.g. identification of an organism), a preliminary notification may be provided.

### 7.7.1. Format of notifications

Notifications should include the following information:

- *Reference number*—the reporting country should have a means of tracing the communication sent to an exporting country. This could be a unique reference number or the number of the phytosanitary certificate associated with the consignment
- *Date*—the date on which notification is sent should be noted
- *Identity of the NPPO* of the importing country
- *Identity of the NPPO* of the exporting country
- *Identity of consignment*—consignments should be identified by the phytosanitary certificate number if appropriate or by references to other documentation and including commodity class and scientific name (at least plant genus) for plants or plant products
- *Identity of consignee and consignor*
- *Date of first action* on the consignment
- *Specific information regarding the nature of the non-compliance and emergency action* including:
  - identity of pest
  - whether part or all of the consignment is affected
  - problems with documentation
  - phytosanitary requirements to which the non-compliance applies
- *Phytosanitary actions taken*—the phytosanitary actions should be specifically described and the parts of the consignment affected by the actions identified
- *Authentication marks*—the notifying authority should have a means for authenticating valid notifications (e.g. stamp, seal, letterhead, authorised signature).

For more information, refer to ISPM 13.

## 7.8. ISPM 17—Pest reporting

This standard describes the responsibilities of and requirements for contracting parties in reporting the occurrence, outbreak and spread of pests in areas for which they are responsible.

It also provides guidance on reporting successful eradication of pests and establishment of pest free areas. These reports are termed ‘pest reports’.

### 7.8.1. Content of reports

A pest report should clearly indicate:

- the identity of the pest with scientific name (where possible, to the species level, and below species level, if known and relevant)
- the date of the report
- host(s) or articles concerned (as appropriate)
- the status of the pest under ISPM 8

- the geographical distribution of the pest (including a map, if appropriate)—the nature of the immediate or potential danger, or other reason for reporting. It may also indicate the phytosanitary measures applied or required, their purpose, and any other information as indicated for pest records in ISPM 8 (Determination of pest status in an area).

If all the required information is not available on the pest situation then a preliminary report should be made with updates as further information becomes available.

## 7.8.2. How to submit reports

Pest reports which are obligations under the IPPC should be made by NPPOs using at least one of the following three systems:

- direct communication to official contact points (mail, facsimile, or email)—countries are encouraged to use electronic means of pest reporting to facilitate wide and prompt distribution of information
- publication on an openly available, official national website (such a website may be designated as part of an official contact point)—precise information on the website access address to the pest reports should be made available to other countries, or at least to the Secretariat of the IPPC
- the International Phytosanitary Portal.

In addition, for pests of known and immediate danger to other countries, direct communication to those countries, by mail or email, is recommended. Countries may also address pest reports to RPPOs, to privately contracted reporting systems, through bilaterally agreed reporting systems, or in any other manner acceptable to the countries involved. Whatever reporting system is used, the NPPO should retain responsibility for the reports.

Publication of pest reports in a scientific journal, or in an official journal or gazette that typically has limited distribution, does not meet the requirements of this standard.

## 7.8.3. Timing of reporting

Reports on occurrence, outbreak and spread should be provided without undue delay. This is especially important when the risk of immediate spread is high. It is recognised that the operation of the national systems for surveillance and reporting, and in particular the processes of verification and analysis, requires a certain time, but this should be kept to a minimum.

Reports should be updated, as new and more complete information becomes available. For more information, see ISPM 17.



## Chapter 8

# Case studies

### 8.1. Case study attributes

Case study	Survey type	Pest type	Host common name	Vegetation	Country	Site selection method
A	Pest list	Plant pathogen	Commercial and wild sugarcane cultivars	Community gardens, domestic gardens and road verges	Papua New Guinea, Indonesia, northern Australia	Targeted
B	Early detection, pest lists	Plant pathogen	Range including: bananas, citrus trees, sugarcane	Urban, agroforestry, orchard and fields	Pacific islands, northern Australia, Torres Strait, Papua New Guinea, Indonesia	Targeted, convenience
C	Pest status, early detection	Insect	Mahogany and cedar trees	Plantations and amenity trees	Fiji, Vanuatu, Tonga, Samoa	Targeted, drive-by
D	Pest status	Plant pathogen	Target list included mango, citrus, bananas, cucurbits, grapes and other Vitaceae and Malvaceae species, solanaceous crops	Urban, domestic gardens, high-risk sites, public parks, feral commercial plants	Northern Australia	Targeted
E	Area freedom	Insect	Stored grain, including wheat, barley, oats, rye, maize and rice	Commodity	Western Australia	Targeted, trapping
F	Area freedom	Insect	Apple, pear, apricot, nectarine, peach, citrus	Orchard	Southern Australia	Systematic trapping
G	Area freedom	Weed	Amongst Niger seed, sorghum, pearl millet	Field	Northern Australia	Targeted, convenience
H	Area freedom	Insect	Mango	Orchard and urban	Guimaras Islands, Philippines	Randomised

Case study	Survey type	Pest type	Host common name	Vegetation	Country	Site selection method
I	Early detection	Insect	Thirteen target list food plant groups	Domestic gardens	Northern Australia	Targeted, convenience
J	Early detection	Plant pathogen	Sugarcane	Field	Northern Australia	Targeted, randomised
K	Early detection	Plant pathogen	Rice	Field	Thailand	Systematic, transects
L	Monitoring	Insect	Rose gum, Dunn's white gum, forest red gum, river red gum	Plantations	Southern Australia	Stratified, transects
M	Monitoring	Plant pathogen	Any seedlings	Garden nurseries and greenhouses	Any	Targeted, full sampling
N	Monitoring	Plant pathogen	Hardwood plantations including hoop pine	Plantations	Any	Targeted
O	Monitoring	Plant pathogen	Gum tree	Plantations	Southern Australia	Stratified
P	Monitoring	Plant pathogen and insects	Shining gum	Natural forest	Southern Australia	Targeted, randomised
Q	Monitoring, pest status	Plant pathogen and insects	Pine tree	Plantations	Southern Australia	Vantage point
R	Monitoring, pest status	Insect	Crucifers including cabbage, brussels sprouts, radish, cauliflower, tobacco	Field	Vietnam	Convenience, systematic
S	Monitoring	Insect	Stored grain including wheat, barley, oats, rye, maize, rice	Commodity	Western Australia	Targeted, trapping
T	Delimiting	Plant pathogen	Papaya	Orchard and domestic gardens	Cook Islands	Targeted
U	Delimiting	Plant pathogen and vector	Citrus trees	Orchard and urban	Papua New Guinea	Targeted
V	Delimiting	Insect	Mango	Feral trees, urban and orchards	Northern Australia	Targeted
W	Delimiting	Insect	Fruit fly hosts	All types	Cook Islands	Targeted, trapping



## 8.2. Case study A. Sugarcane pests in Papua New Guinea, Indonesia and northern Australia

### Step 1. Purpose of the survey

New Guinea is the centre of diversity for *Saccharum officinarum*, the source species for high-sucrose genes in commercial sugarcane cultivars, and *Saccharum* species are widely cultivated and naturally distributed through eastern Indonesia and Papua New Guinea (PNG). There are many exotic pests and diseases, some of which occur in Indonesia and PNG, that have the potential to reduce the productivity and profitability of the Australian sugar industry.

The focus of the surveys was to determine the distribution of known insect pests and diseases of *Saccharum* spp. in the PNG–Indonesian–Australian region. This would allow the development of quarantine strategies to limit the spread of these pests.

### Step 2. Targeted pest name/s and diagnostic characteristics

All insects and diseases (fungi, bacteria, viruses, phytoplasmas) encountered were collected. In PNG and Indonesia these are mainly endemic, in Australia they are exotic.

Insect specimens were allotted preliminary identification in the field, based on the experience of the surveyors. Some stemborers from the PNG survey were reared through to adults at Ramu Sugar (PNG). Specimens (pinned or in ethanol) were later sent to a specialist for confirmatory identification.

Disease specimens were photographed and allotted preliminary identification in the field, based on the experience of the surveyors. Where the identification was tentative, leaf and/or stem specimens were dried in a plant press or in bottles containing calcium chloride. Fungi were later identified on morphological characters; viruses, bacteria and phytoplasmas were identified using DNA technology.

### Step 3. Target host

Cultivated *Saccharum* spp. (*officinarum*, *edule* and commercial hybrids).

### Step 4. Alternative hosts

Wild *Saccharum* spp. (*spontaneum* and *robustum*).

### Step 7. The area

Four surveys were undertaken in Papua New Guinea, eastern Indonesia and northern Australia and the Torres Strait/Cape York Peninsula region. In PNG, areas visited were Daru, Morehead, Tabubil, Vanimo, Wewak, Manus, New Ireland, New Britain, Lae, Ramu, Popondetta, Alotau and Port Moresby; these covered the extremities of PNG. In Indonesia, Sumba, Flores, Sumbawa, Lombok and Bali were visited. In northern Australia, 19 major coastal and near-coastal settlements from Normanton to Broome were visited. A number of Torres Strait Islands were visited (Mabuiag, Boigu, Saibai, Dauan, York,

Murray, Darnley, Thursday, Horne) as well as a number of Cape York communities. Areas such as West Papua (Indonesia) and the Highlands and Bougainville areas of PNG could not be visited because of security concerns.

Most pests and diseases are more active or more obvious towards the end of the wet season when humidity is high and there has been time for the development of populations.

## Steps 10 and 11. Site choice and sample size

In all areas, traditional and household gardens containing plantings of *Saccharum officinarum* and commercial hybrid cultivars were targeted. Traditional gardens are planted in community gardens in and around villages. In addition, wild canes growing on roadsides were also inspected.

Due to limited time, villages (3–5 per day) and roadsides within about 20–50 km of airports that were accessible by road were surveyed. In northern Australia, townships were surveyed.

The entire area of each village was searched for *Saccharum* plants. This was usually about 1 ha.

All *Saccharum* plants in the community gardens and communities were surveyed—usually 5–15 stools of sugarcane.

## Step 12. Timing of survey

Most sugarcane pests and diseases are more active or obvious towards the end of the wet season—this and the requirements for road and air transport dictated surveys in May–June.

## Step 14. Samples collected

The location of each collecting site was determined by GPS and the host species noted.

Insect specimens were collected either as adults or when immature. Most were placed in >95% ethanol (suitable for later DNA analysis) in labelled tubes, while some were pinned after killing. In PNG, stem borers were kept alive and placed in tubes with a food source for rearing and identification at Ramu Sugar. Specimens were taken to Australia (under AQIS permit) for further identification (often by specialists in Australia or overseas). Some samples were duplicated and retained in Indonesia or PNG as reference samples.

Disease specimens were collected as leaf or stem samples. Samples were pressed between sheets of newspaper in a plant press or were cut into small (2 mm by 2 mm) squares and dried in sealed McCartney bottles containing calcium chloride. Material was imported into Australia under AQIS permit (fumigated where necessary). Pressed leaves were deposited in the herbarium of the Queensland Department of Primary Industries and Fisheries and dried leaf samples were sent to BSES Limited's Indooroopilly laboratory for DNA identification of causal organisms.

## Comments

It was essential in all surveys to interact with local personnel, usually staff from the national quarantine service or agricultural extension service. These people provided knowledge on local conditions and acted as go-betweens in securing permission to enter villages and collect material. It also helped in technology transfer between the survey team and the local staff.

In many places it was difficult to obtain sufficient newspaper to dry plant material—ample supplies should be taken on all such trips.

Airline regulations meant that tubes containing ethanol had to be packed in a certain way—make sure that this is checked before setting out.

Transport from place to place was by chartered aircraft—this gave much better flexibility and use of time than relying on commercial flights.

## References

Magarey R.C., Suma, S., Irawan, Kuniata, L.S. and Allsopp, P.G. 2002. Sik na binatang bilong suka—Diseases and pests encountered during a survey of *Saccharum* germplasm ‘in the wild’ in Papua New Guinea. Proceedings of the Australian Society of Sugar Cane Technologists, 24, 219–227.

Magarey, R.C., Kuniata, L.S., Croft, B.J., Chandler, K.J., Irawan, Kristini, A., Spall, V.E., Samson, P.R. and Allsopp, P.G. 2003. International activities to minimise industry losses from exotic pests and diseases. Proceedings of the Australian Society of Sugar Cane Technologists, 25 (CD-ROM).

## 8.3. Case study B. NAQS and SPC early detection and pest list survey design for plant pathogens

### Step 1. Purpose of the survey

A broad pest survey to determine baseline data for host and pathogen lists, including organisms of quarantine concern.

### Step 2. Targeted pest names and diagnostic characteristics

A wide range of pest species is targeted during these surveys. In general, the pests are identified by surveying all plants showing disease symptoms. For quarantine surveys, target pest lists are developed either through consultants, consultations with stakeholders or literature searches. A quarantine pest was defined as a pest of potential economic importance to the area endangered and not yet present there, or present but not widely distributed and being officially controlled.

The main pests targeted by both NAQS and SPC are citrus canker (*Xanthomonas axonopodis* pv *citri*), banana bunchy top virus, sugarcane smut (*Ustilago scitaminea*), blood disease bacterium, Panama wilt (*Fusarium oxysporum* f. sp. *cubense*) and Huanglongbing (‘*Candidatus* Liberibacter asiaticus’).

### Step 3. Target hosts

A wide range of hosts is targeted, but surveys generally concentrate on economically or culturally important species. The main crops targeted are sugarcane, bananas and citrus plants.

## Step 4. Alternative hosts

Weeds are surveyed in the areas visited to detect potential biological control organisms and alternative hosts.

## Step 7. The area

This survey is applied to the entire Pacific islands when performed by the SPC and to northern Australia, the Torres Strait islands, Papua New Guinea and Indonesia when performed by the NAQS.

## Steps 10 and 11. Site choice and sample size

A wide range of habitats is surveyed. Particular attention is paid to agricultural areas, including broad-scale agriculture and village and house gardens.

As these surveys are detection surveys, time is the factor limiting the number of sites surveyed. The aim is to cover as much as possible of the growing region in each area.

Sites are sometimes chosen on the basis that there are multiple target hosts present in the one location or when farmers or extension officers have reported something new or unusual.

## Step 12. Timing of survey

In seasonally wet and dry climates, the surveys are generally timed to coincide with the end of the wet season as the sites were easier to access then and host plants were still growing rapidly. In areas with less-variable seasons it is best to time surveys for when host species are most abundant and crops are growing. The growth of phytoplasmas seems to be favoured in the drier periods of the year.

## Step 14. Samples collected

Samples are collected from any surveyed plants showing pests or pest symptoms. Samples can be treated in one or other of three ways. Samples with definite disease signs, i.e. fruiting bodies, are dried and pressed as herbarium specimens. Samples with symptoms are isolated onto fungal growth media or, in the case of inter- and intracellular pathogens, dried over calcium chloride for later analysis.

## Comments

Good quality photographs are taken of all the samples, particularly those suspected of harbouring viruses or phytoplasmas. It is useful to take photographs of specimens sent for identification so that there is an image of exactly what has been identified and is held as a voucher specimen. The photographs are also useful in publications.

## 8.4. Case study C. Pest status and early detection survey for shoot borers in mahogany and cedar trees

### Step 1. Purpose of the survey

Pest status surveys and early detection monitoring for shoot borers of mahogany and cedar in forest plantations, woodlots and amenity plantings.

### Step 2. Targeted pest name and diagnostic characteristics

*Hypsipyla robusta* (Moore) (Lepidoptera: Pyralidae)

Common names: mahogany borer, cedar shoot caterpillar

*Hypsipyla robusta* is indigenous or established in some countries in the Pacific and exotic to others.

Damage symptoms: Insect tunnelling in terminal and lateral shoots of tree causing shoot death and dieback of leaders and branches, and multi-stemming. Early symptoms include wilting of tips and small amounts of frass in a leaf junction. A web containing plant particles and frass usually covers entrance holes. Early instar larvae are brownish-red in colour, and late instar larvae are a distinctive blue with black spots. Adult moths are very rarely seen. The fruits of some hosts are also attacked, damage symptoms being frass and webbing the fruit together in a clump.

### Step 3. Target host

Tree species of the subfamily Swietenioideae, family Meliaceae; for example, species of *Toona* (red cedar), *Swietenia* (American mahogany), *Cedrela* (Mexican cedar), *Chukrasia* (Asian mahogany), *Khaya* (African mahogany).

### Step 4. Alternative hosts

*Xylocarpus* spp. (mangroves)

### Step 7. The area

Fiji, Vanuatu, Samoa and Tonga

### Steps 10 and 11. Site choice and sample size

Sub-areas were defined as forest plantations, woodlots and amenity plantings of Swietenioideae. These were identified by consultation with forestry organisations in each country to determine their location, age and area of plantings.

Surveys were structured to cover the range of susceptible tree species (e.g. *Toona*, *Swietenia*, *Khaya*) and planting types (e.g. plantation, agroforestry, urban) in each country at several geographic locations as resources permitted.

Young plantings (less than 5 years old) of susceptible tree species were chosen because damage symptoms are easier to detect and insect samples more readily obtained. Amenity trees adjacent to air- and seaports receiving international freight were targeted in the surveys because these are considered high-risk sites for incursions by exotic pests. Surveillance was usually concentrated within 1 km of high-risk sites, although plantations of susceptible host species within several kilometres of ports were also generally inspected. Visual inspections of trees were conducted by both roadside cruise ('drive by') and ground transect. If the damage symptoms described above were detected then the tree was examined more closely and attacked shoots were dissected to determine the causal agent. If moth caterpillars were found and their appearance was consistent with that of *H. robusta* larvae, then samples were collected for rearing to adulthood in the laboratory. The moths were then sent to a taxonomist for identification.

Roadside cruise involved driving at speeds not exceeding 15 kph, preferably in a team of two—a driver and an observer. Detection efficiency declines with increasing distance from the road (rarely reliable beyond 40 m) and increasing vegetation density. Periodically during roadside surveys, the team stopped the vehicle and conducted a ground transect of 100 trees through a plantation away from the road.

The number of trees sampled at each site varied with type of planting and survey method. Plantations with good road access were sampled in drive-by surveys allowing the scanning of large numbers of trees for signs of damage. Ground surveys were conducted in all planting types, usually transects totalling 100 trees, the number of transects varying with the size of the planting and according to the time/resources available.

## Step 12. Timing of survey

The insect may be present throughout the year but is most active in the hotter, wetter months so sampling was performed at those times.

## Step 13. Data collected

Locality, situation (e.g. plantation, amenity), host species, symptoms, incidence (number of trees affected), severity (number of shoots attacked per tree), date, observer and GPS reading.

## Step 14. Samples collected

Specimens: 15 cm of shoot containing late-instar moth caterpillar for rearing in the laboratory, additional larvae for preservation, any live pupae for rearing, plant foliage and flowers if required for identification, photographs.

## Comments

Permission was sought before entering the survey properties.

## 8.5. Case study D. Urban pest status survey in Cairns

### Step 1. Purpose of the survey

The purpose of the survey was to conduct a pest status survey for a target list of plant pests and diseases in a high-risk urban environment. Cairns city is considered 'high-risk' due to the high level of tourist and trade traffic at this port, and because of the diverse range of horticultural and alternative host flora in this area. The survey also included elements of a monitoring survey, as staff collected information to support PFA status for specific quarantine pests during the course of the survey.

### Step 2. Targeted pest names and diagnostic characteristics

Over 100 plant pests listed on the Queensland Department of Primary Industries and Fisheries biosecurity plant pest target list were targeted. The specific number of pests surveyed depended on the horticultural and alternative host plant species encountered during the survey.

Exotic ants, termites and other invertebrate pests were also targeted.

### Step 3. Target hosts

The biosecurity plant pest target list identifies approximately 20 different host groups. The main host plants targeted include: mangoes, citrus and other Rutaceae, banana and other *Musa* spp., cucurbits, Malvaceae, grapes and other Vitaceae, and solanaceous crops.

### Step 4. Alternative hosts

A range of other horticultural and ornamental host plants was surveyed when encountered by the surveillance staff.

### Step 7. The area

The area was defined as the city of Cairns and surrounding suburbs, in Queensland, Australia (Figure D1). The habitats encountered in this area are diverse and include residential backyards, rubbish dumps, industrial and port areas, creek banks, parks and feral stands of horticultural host plants.

### Steps 10 and 11. Site choice and sample size

The number of field sites corresponded to the number of contiguous suburbs in the greater Cairns area.

The resources assigned to the project determined the number of sampling sites within a suburb. A team of two scientists surveyed an average of seven sites per day: the time allocated to the project and the number of suburbs to be covered determined how many sampling sites could be completed. Approximately 2.2 sites in each of 38 suburbs were covered, totalling 84 sampling sites.



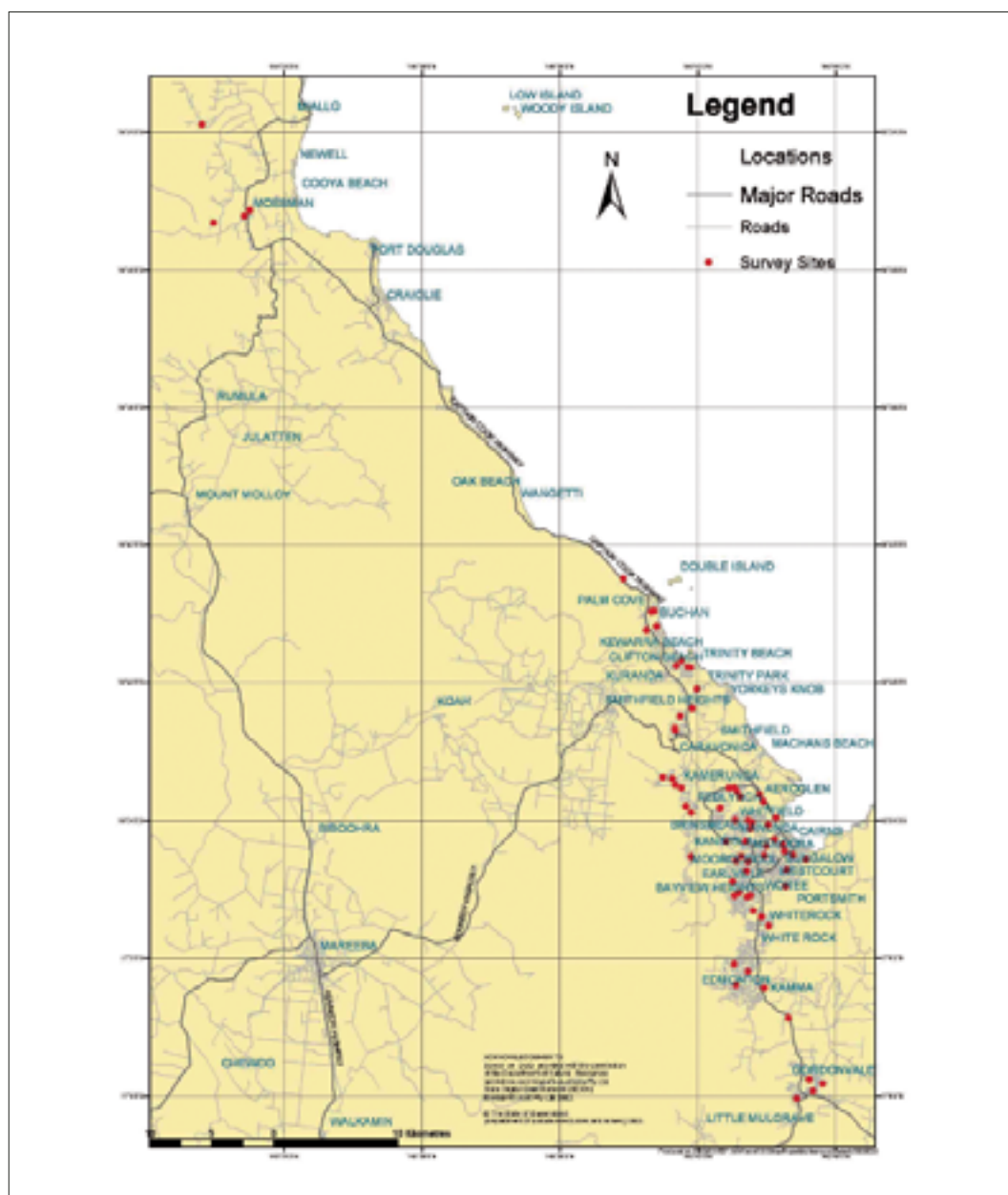


Figure D1. Map of suburbs in and around Cairns, Australia surveyed in September 2003

In order to achieve the most effective surveillance and efficient use of resources, sites were not chosen at random but instead properties with a high number of host plants and a varied host range were targeted. This approach was chosen to increase the likelihood of detecting a target pest.

At each site, all horticultural and alternative host plants were examined. The relatively small size of each site and the fact that host plants are not usually densely planted in residential gardens, often enabled staff to closely examine every host plant. Where there was a high number of plants, such as a large stand of bananas, the group as a whole was surveyed and then a few plants were examined in detail. Over 3760 host plants were surveyed in total, with an average of 11 taxa per site.



## Step 12. Timing of survey

This survey is conducted annually. Year-round access can easily be achieved in urban areas which allows scientists the luxury of varying the time of survey each year to enable staff to detect target pests that may have a seasonal life cycle.

## Step 13. Data collected

Negative data were collected for pest species as well as recording the presence of regulated and exotic pests. A range of basic host and site location information was also gathered. A sequential site number was used to identify each site. General information was recorded for each site against this number on a survey sheet. The data recorded for each site included the names of observers, date, site description, geographical coordinates, number and type of hosts present, number of hosts examined and number of samples taken. Absence data for specific pests were also recorded on this form.

## Step 14. Samples collected

Any suspect exotic species or pests unfamiliar to scientists and causing significant damage were collected for taxonomic identification using the appropriate method. Photographs were taken of collected pests and diseases *in situ* for future reference.

## Comments

Urban surveillance has a high element of community involvement, as permission is sought to enter each property. Many of the pest incursions detected in Queensland have resulted from a public enquiry about an unusual insect or sick plant. Urban surveillance and close contact with gardeners is a good opportunity to educate members of the general public about exotic pest species and quarantine awareness. The surveyors took time to talk to property owners, as they can be valuable reporters of exotic pests.

The survey was designed as a pest status survey but data collected could be used to obtain pest free area status to support interstate and international trade.

## 8.6. Case study E. Pest free area status survey for khapra beetle in stored grain

### Step 1. Purpose of the survey

To maintain Australia's pest free status for the khapra beetle

### Step 2. Targeted pest names and diagnostic characteristics

Targeted were the khapra beetle (*Trogoderma granarium*) and the warehouse beetle (*Trogoderma variabile*). The khapra beetle is the world's worst pest of stored grain. It is not found in Australia and many grain export markets would be lost overnight if this pest were found here. The warehouse beetle is present in the midlands region of Western Australia. The main significance of the warehouse beetle is that it can mask the presence of the khapra beetle.

Infestations are usually noticed through cast larval skins. Identification requires the dissection of mouthparts, and suspect beetles are sent to a taxonomist for identification.

### Step 3. Target host

Grain, cereals and products including wheat, barley, oats, rye, maize, rice, flour, malt, and noodles.

### Step 4. Alternative hosts

None were surveyed.

### Step 7. The area

Grain export terminals, storage sites and grain processors in Western Australia, where the warehouse beetle is known or at risk of infesting.

### Steps 10 and 11. Site choice and sample size

Field sites were selected in areas at risk of infestation. The number was determined by the storage sites involved, which was approximately 130 across 30 towns. The storage sites included commercial buildings containing grain and grain products, and commercial grain storage facilities.

Sticky traps (see next page) were set up within each grain store. Also, up to five baiting sites were chosen near food sources in large buildings, and one site in small buildings (such as shops).

Some sentinel pheromone traps were placed on farms on an ad hoc basis, targeting farms with poor hygiene.

## Step 12. Timing of survey

Trapping was performed during the warmer summer months, when beetle activity is at it highest (December to March). Traps are effective for 2 months and so were replaced at the end of January. In warmer climates, the beetles could possibly be active all year and so would require continuous surveillance. At seaports, the trapping was continuous.

All traps were checked every 2 weeks.

## Step 13. Data collected

Recorded were trap identifier, date, location, property name, property type, nearby food sources and comments including trap position within the storage site.

## Step 14. Samples collected

Sticky traps with pheromone lures were used. These can attract beetles from a distance of 5 km. The lures attract native *Trogoderma* beetles, warehouse beetles and khapra beetles. Because khapra beetles do not fly, traps are placed at ground level.

As the pest was not found, no results were recorded. Null records are not kept but it is intended that they will be in future surveillance.

## References:

Emery, R., Dadour, I., Lachberg, S., Szito, A. and Morrell, J. 1997. A final report prepared for the Grains Research and Development Corporation. The biology and identification of native and pest *Trogoderma* species. Project number DAW 370. South Perth, Agriculture Western Australia.

Banks, H. J. 1990. Identification keys for *Trogoderma granarium*, *T. glabrum*, *T. inclusum* and *T. variabile* (Coleoptera: Dermestidae). Black Mountain, Canberra, Australia, CSIRO Division of Entomology.

## Comments

Surveys should be more rigorous (structured rather than sentinel), ongoing, nationally coordinated and the results entered in a database.

The traps could be inspected more frequently to allow rapid action if an incursion occurs. Beetles can be carefully removed from traps without destroying the trap and sent off for identification.

## 8.7. Case study F. Pest free area status survey of Queensland fruit fly and Mediterranean fruit fly

### Step 1. Purpose of the survey

To seek PFA status in order to gain international market access.

### Step 2. Names of targeted pests and their diagnostic characteristics

Mediterranean fruit fly (Medfly)—*Ceratitis capitata* (Wiedemann); Queensland fruit fly (Qfly)—*Bactrocera tryoni* (Froggatt).

Medfly is an exotic species that has a restricted distribution in Western Australia, with permanent populations existing only in the southwestern part of that State (Figure F1). This area is over 2000 km from the Riverland, Riverina and Sunraysia PFAs.

Qfly is a native species and originally had a limited distribution around southeastern Queensland. Qfly now exists in permanent populations along coastal strips of the eastern seaboard, extending up to 300 km inland in Queensland, through New South Wales and tapering off to limited areas in northeastern parts of Victoria.

Any detections of economically damaging fruit fly species are viewed as serious.



Figure F1. Distribution map of QFly (*Bactrocera tryoni*) and Medfly (*Ceratitis capitata*)

### Step 3. Target host

Fruit trees: apple, pear, apricot, nectarine, peach and citrus.

### Step 4. Alternative hosts

None were surveyed.

### Step 7. The area

The area comprises the Riverland, Sunraysia and Riverina regions in Victoria and New South Wales, Australia (Figure F2). All three regions and the land surrounding them are geographically isolated by significant distances from areas of Australia infested with permanent populations of Medfly and Qfly.

Medfly and Qfly do not occur in the area and they are not capable of naturally dispersing to these pest free areas (PFAs) from infested areas, due to the hostile climatic conditions experienced in the PFAs and surrounding lands. These conditions are sufficiently detrimental that the risk of fruit fly establishment in these areas is marginal.

Any introductions of Medfly and Qfly from infested areas into the PFAs could only be through transport by humans. The illegal transportation of infested fruit from fruit-fly infested areas by travellers in private vehicles is believed to be the main potential source of introduction of fruit fly into the PFAs.

Transport of host fruit by humans into the PFAs is strictly controlled by State legislation. Additional phytosanitary measures are also used to prevent the introduction and spread of these fruit flies. There is therefore minimal risk of fruit flies entering and establishing in these PFAs.

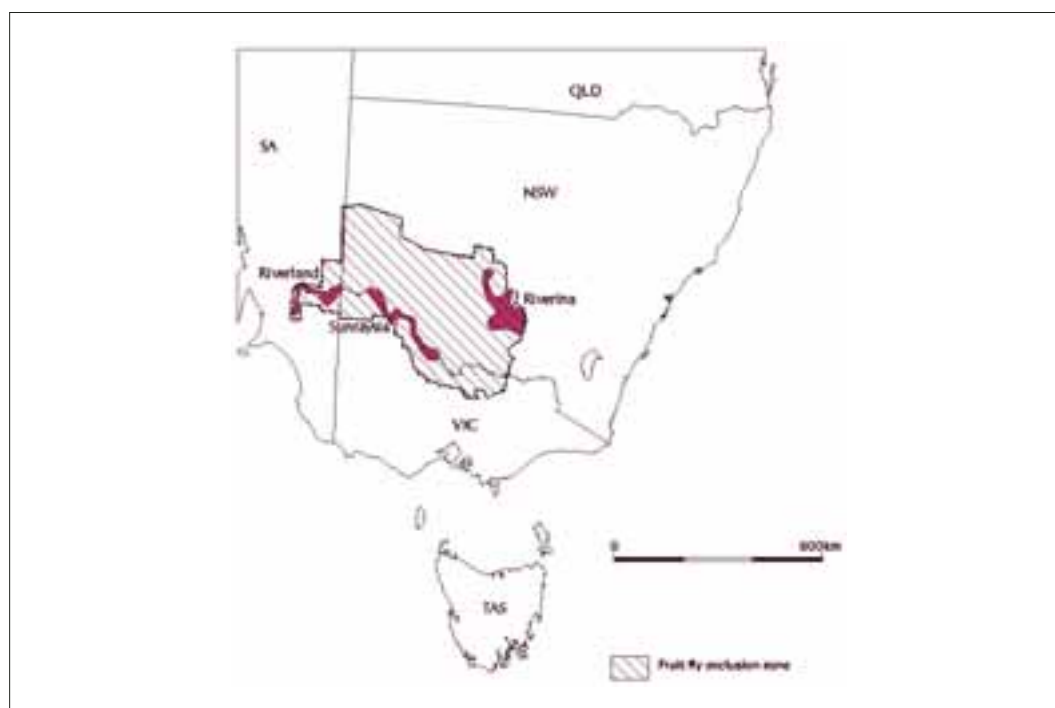


Figure F2. Regions (in red) seeking pest free area status

## Steps 10 and 11. Site choice and sample size

Permanent trapping grids are used. Traps are placed at a higher density in urban areas than in non-urban horticultural areas because urban areas are considered to carry a higher risk for the entry and establishment of fruit fly. Sentinel traps are used in major towns.

In the PFAs, the trap sites are placed in a grid with:

- 1 trap site every 400 m in urban areas
- 1 trap site every 1 km in non-urban horticultural areas where host production occurs.

## Step 12. Timing of survey

Traps are monitored throughout the year; weekly during late spring, summer and early autumn when trees are producing fruit, and fortnightly at other times.

## Step 14. Samples collected

Separate traps were maintained for Medfly and Qfly but the traps are placed at the same site. Each trapping site consists of:

- one Lynfield trap charged with *Cue-lure* plus maldison for Qfly
- one Lynfield trap charged with *Capi-lure* plus dichlorvos for Medfly

Traps were placed in the best vegetative cover available. During the southern hemisphere spring/summer, traps were placed in host trees of apple, pear, apricot, nectarine or peach wherever possible, while in autumn and winter, citrus trees were used.

Traps are located within the canopy of host trees bearing fruit, approximately half the distance from the trunk to the outer edge of the foliage, and at least 1.5 m above the ground. If no fruiting trees are available, the traps are placed in trees with similar foliage (i.e. broadleaved trees). Traps are located at least 3 m apart at each site.

*Cue-lure* traps are recharged twice per year in spring (September) and summer (January). The entire trap (base and lid) is replaced every 12 months in spring (September), unless a trap is damaged, in which case it is replaced immediately.

*Capi-lure* traps are recharged four times per year in spring, summer, autumn and winter. The entire trap (base and lid) is replaced every 12 months in spring (October).

All insects from traps are examined and suspect fruit flies forwarded to a reference entomologist for positive identification. Any specimens are placed in individual plastic vials and a label attached listing details of the trap number, date and other details. If there is doubt about the identity of suspect flies, the specimen is sent to a taxonomist for positive identification.

## Comments

AQIS is responsible for ensuring that the permanent fruit-fly monitoring program in the Riverland, Riverina and Sunraysia PFAs is conducted in accordance with trading partner requirements. The State government departments are responsible for the routine maintenance and management of the fruit fly monitoring programs in their States (e.g. establishment and servicing of traps, identification of flies, implementation of eradica-

tion campaigns). AQIS conducts audits of the fruit-fly monitoring programs to ensure compliance, and the State authorities are responsible for ongoing internal audits of their activities and procedures.

The State departments have the responsibility and legislative power for declaring outbreaks. They are required to inform AQIS of the details of any outbreak and the required suspension area.

AQIS is responsible for the subsequent notification of any outbreaks to trading partners. As for other national plant protection organisations, AQIS has the responsibility and legislative power for export certification of host produce originating from the PFAs. When outbreaks do occur, AQIS will not permit exports from those designated outbreak areas.

## 8.8. Case study G. Pest free area status for dodder weed

### Step 1. Purpose of the survey

The purpose of this survey was to demonstrate if the Ord River Irrigation Area (ORIA) in the north of Western Australia was free of weed pests belonging to the genus *Cuscuta* (dodder weed). This information was needed to support negotiations for market access to the USA market for Niger seed, which is used in caged bird seed mixes. US regulations insist that Niger seed be steam cooked to kill any weed seeds present, especially those of *Cuscuta* species. The US Quarantine Service APHIS agreed that ORIA Niger seed could be imported into the USA without steam cooking if the area was demonstrated to be free of *Cuscuta*.

All recorded locations of the pests were researched. It was unknown in the survey area. The nearest known *Cuscuta* species were 1000 km to the south and 200 km east-southeast. To the south of 18°S latitude there were scattered populations of *Cuscuta* species. Surveys in 1993 and 1994 extended the range of *Cuscuta* species in Western Australia to within 300 km of the ORIA.

### Step 2. Targeted pest name and diagnostic characteristics

*Cuscuta* species. This parasitic plant has neither leaves nor chlorophyll. Its fine stems form a tangled mass, clinging to herbaceous plants such as legumes, tomatoes and capsicums by means of small suckers. The dodder uses these suckers to draw nutrients from the hosts, which become stunted and discoloured.

### Step 3. Target hosts

Niger seed (*Guizotia abyssinica*), hybrid sorghum (*Sorghum* sp.) and pearl millet (*Pennisetum glaucum* hybrids). These annual crops are grown in the dry season. They are generally planted in April–May and harvested in August–September.

## Step 4. Alternative sites

Other places that the weed could possibly grow and that were relevant were:

- amongst any other crop (bananas, mangoes, maize, chickpeas, melons and leucaena)
- in non-cropped wet habitats (springs, drainage ditches, water-supply channels, gardens, lake margins, drainage swamps, paddock margins and river margins)
- road verges.

## Step 7. The area

The ORIA is serviced by the town of Kununurra. The area was defined for this survey as the Kununurra townsite and that area of the Ord River Irrigation Project that is used for irrigated agriculture. It covers approximately 5400 square kilometres.

The ORIA is in the semi-arid tropics. The climate is hot and wet in summer (wet season) and warm and dry in the winter (dry season). Average annual rainfall is 787 mm, falling mostly between December and March with the maximum temperature sometimes exceeding 40°C. Maximum temperatures in the dry season average 32°C and minimums 15°C.

This area consists largely of black cracking-clay plains that are used for intensive irrigated row-crop agriculture. There are some areas of red loams and sands and these are either row-cropped, or under tree crops.

## Steps 10 and 11. Site choice and sample size

The number of sites was influenced by the large area and the fact that the species are hard to see. The number of sites was designed to cover the entire area. The level of surveying intensity varied with the site type.

1. Surveying of all Niger, sorghum and pennisetum (millet) crops intended for export to the USA. These were examined at an intensity of one site per 10 ha. N = 20.
2. One site was surveyed for each type of other crops (bananas, mangoes, maize, chickpeas, melons and leucaena) and all the non-cropped wet habitats (springs, drainage ditches, water-supply channels, gardens, lake margins, drainage swamps, and paddock and river margins). N = 30.
3. When travelling in the area, all road verges were observed by a drive-by survey.

At each sampling point, surveying consisted of walking a 500 m transect in crops (or twice as far around the edges of dense vegetation such as sorghum and bananas) inspecting to 1 m to the left and right of the transect.

If the area surveyed was a low crop or consisted of uncleared bush, a 500 m transect in zigzag pattern was used to survey as much of the crop or site as possible.

Sites in the crop were targeted if there were areas that were uneven or showing yellowing, especially near irrigation supply and drainage sites of crops.

## Step 12. Timing of survey

The area was surveyed for *Cuscuta* species once only in the wet season (March–April) and once during each subsequent dry season when Niger and the hybrid pennisetums and sorghums are growing under irrigation.



The frequency of surveying was specified by the trading partners. Export of Niger to the US has ceased for the time being, but as part of the NAQS survey for a wide range of pests in northern Australia, *Cuscuta* is searched for every 6 months in a less structured manner.

### Step 14. Samples collected

No samples were collected as the pest was not detected.

### Step 21. Reporting

The survey results were reported to AQIS after every survey.

### Comments

Legislative changes proposed to support the pest free area status:

- prohibit the importation of *Cuscuta* species into the ORIA under the *Plant Diseases Act 1914*
- declare *Cuscuta* species a noxious weed in the ORIA under the *Agricultural and Related Resources Protection Act 1976*, which will allow eradication of possible future incursions.

## 8.9. Case study H. Pest free area status for mango pulp weevil and mango seed weevil

### Step 1. Purpose of the survey

To seek market access into Australia for mangoes.

### Step 2. Targeted pest names and diagnostic characteristics

Mango pulp and seed weevils, which are similar in appearance but attack different parts of the fruit. Because there are no external signs of the weevil attack, fruit have to be cut open and examined. The pulp weevil larvae form distinctive brown chambers within the flesh.

### Step 3. Target host

Mango

### Step 4. Alternative hosts

None examined.

## Step 7. The area

Guimaras Province, Republic of the Philippines, which consists of a group of islands. Some 8% of the total land is dedicated to mango production. The islands are geographically isolated from other islands by sea straits.

## Steps 10 and 11. Site choice and sample size

A census of the province identified over 97,000 mango-bearing trees and the location of each. This permitted randomised selection of the target trees. The sampling sites were stratified by municipality, and then divided by the distribution of cultivars and the management practices.

The sample size (approved by an Australian government statistician) was such that if either the mango pulp weevil or the mango seed weevil were present with a tree infestation rate of 1% or greater and if 15% of the fruit on an infested tree were infested, there would be a greater than 95% chance of detection during the survey. The sample size required 5% of all known mango-bearing trees in the province to be surveyed and that 10 fruits be collected from each.

Trees were visited for suitability before the survey and painted with a number.

## Step 12. Timing of survey

The survey was conducted from February to May 1999. Production is throughout the year, but peaks during December–May and in March and April.

## Step 13. Data collected

The grower or agent was interviewed on aspects of cultivar, horticultural practices, tree management, yield and pest incidence.

## Step 14. Samples collected

Ten mangoes, older than 65 days after flower induction, were collected per tree. At 5%, the sample size needed was 4857 trees, collecting 48,570 mangoes for investigation. At least two mangoes were to be collected from each of the four quadrants around each tree.

Mangoes were bagged and labelled then sent to a laboratory for examination. Mangoes were examined externally then dissected to search the pulp and the seed for all pests, but with emphasis on the target pests.

Fruit collecting was considered to be more efficient and effective than sticky traps, visual observations, beating of branches and debris examination.

## 8.10. Case study I. Insect pests of food plants in Aboriginal communities in the Northern Territory

### Step 1. Purpose of the survey

The purpose was to survey the Aboriginal communities of Yirrkala and Garrthalala, the town of Nhulunbuy and nearby native vegetation for exotic insect pests.

### Step 2. Targeted pest names and diagnostic characteristics

A targeted pest list was used, based on insect pests that are not yet present in Australia but are found in neighbouring countries. The list included 56 high-priority and 24 moderate-priority species. These pests attack primarily food crops but could possibly survive on related plants.

### Step 3. Target host

A group of priority plants that are important sources of food and other resources in northern Australia was surveyed. These were sugarcane, banana, citrus, mango, cotton, grape, grain sorghum, cucurbits, maize, pasture legumes and grasses, *Eucalyptus* spp., *Acacia* spp. and palms.

### Step 4. Alternative hosts

Native plants belonging to the same genus or family as the priority plants were also targeted, especially if the targeted insect was known to be polyphagous.

Where time permitted, other food plants, especially native food-plant species important to local people, were also surveyed.

### Step 7. The area

The survey was confined to the mining town of Nhulunbuy (population 2000), and the Aboriginal communities of Yirrkala (population approximately 1000) and Garrthalala (population approximately 30). Nhulunbuy is a coastal town on the north-eastern tip of Arnhem Land in the Northern Territory of Australia. An uninhabited coastal site near Garrthalala, Murjbi, was also surveyed following reports of a foreign vessel in the area previously.

Nhulunbuy supports a wide variety of plant species in backyard settings, usually not in close contact with native vegetation. Yirrkala also has some backyard plantings, though not as substantial as those in Nhulunbuy, but native plant species growing close to commercial plant species are more common. There is a 5 ha banana plantation at Yirrkala. Garrthalala has few commercial plant species and is totally surrounded by native vegetation. Murjbi is an uninhabited and relatively undisturbed site.

Access to backyards in Nhulunbuy and Yirrkala was dependent on the homeowner being available to grant permission. Access to yards in Garrthalala was easily negotiated with the community elders. Garrthalala was a two-hour drive from Nhulunbuy and Murjbi, and a further hour on a narrow, dirt track. Access to Aboriginal lands required permission from the local land council and was greatly facilitated by the participation of local people in the survey.

## Steps 10 and 11. Site choice and sample size

Backyards growing target hosts were found by asking local people and by driving and walking around town looking into yards. Native vegetation bordering the communities was targeted.

Sites were chosen largely as they were located. Due to time restrictions, the proportion of yards surveyed was inversely proportional to the size of the community. In Garrthalala, 100% of commercial species were surveyed, while in Nhulunbuy and, to a lesser extent, Yirrkala, lower proportions were included.

The number of plants examined varied according to the site. All plants in the community gardens surveyed were examined. In the banana plantation at Yirrkala, banana plants growing around the edge of each block, and those along a transect through the middle of the plantation, were surveyed. Native vegetation growing on the edge of the communities was surveyed.

## Step 12. Timing of survey

The survey was conducted in December to coincide with the start of the wet season—before roads are cut by rainfall but after plants had started growing. It also allowed for inspection of mango fruits.

## Step 13. Data collected

At each community, a list of food plants present was compiled.

## Step 14. Samples collected

All insects collected were identified in the field as far as possible, usually to family level. Only those specimens which could not be definitively identified as not being a targeted species, or those of uncertain identity yet causing significant damage to plants, were retained. Interesting or unusual species or symptoms were photographed.

## 8.11. Case study J. Early detection survey for sugarcane smut

### Step 1. Purpose of the survey

Sugarcane smut is a serious disease of sugarcane that can cause yield losses over 30% in susceptible cultivars. The disease was found for the first time in Australia in July 1998 in the Ord River Irrigation Area of Western Australia. An initial rapid survey in eastern Australian sugarcane crops and a review of disease inspections conducted in 1998 failed to locate smut in eastern Australia.

More extensive sugarcane smut surveys throughout Queensland and New South Wales were undertaken in 1998–99 and 1999–2000 to determine the presence or absence of sugarcane smut in eastern Australia and enable appropriate quarantine or incursion management decisions to be made to reduce production losses.

### Step 2. Targeted pest name/s and diagnostic characteristics

**Pathogen:** *Ustilago scitaminea* H & P Sydow

**Disease:** Sugarcane smut

**Diagnostic symptoms:** Highly characteristic black whip-like structures (sori) are produced from the heart of the shoot. Whips can be from a few centimetres to over one metre in length (Figure J1). The disease also causes severe stunting, profuse tillering and thin grassy stalks. Experienced plant pathologists can provide reliable diagnoses of the disease.



Figure J1. Characteristic appearance of sugarcane smut whips on sugarcane

Had a suspect infected plant been found, the spores of the fungus would have been sent to the Queensland Department of Primary Industries and Fisheries Herbarium for confirmation of identification, using available DNA methods to identify the fungus.

The disease primarily infects buds and may remain dormant until the buds germinate. Because of this, it may be 6–12 months before any symptoms develop, with the disease going undetected until it reaches sufficient intensity for a survey to have a reasonable chance of detecting an infected plant.

### Step 3. Target host

Commercial sugarcane (*Saccharum* spp. hybrids)

### Step 4. Alternative hosts

None surveyed.

### Step 7. The area

Commercial sugarcane fields in eastern Australia. Generally, these areas have good access and are relatively flat.

### Steps 10 and 11. Site choice and sample size

Mill maps were used to provide details on susceptible varieties, farm numbers, block numbers, variety, and crop class. Each field of sugarcane was considered a potential sample site.

Fields were selected at random, although high-risk fields (e.g. on farms where residents had visited the Ord River area) and fields of varieties known to be highly susceptible were also targeted. Ratoon cane was preferred for smut inspections because of longer exposure to possible infection.

There were several factors that determined which parts of the crops could be accessed. The height of cane crops, the narrow laneways with overhanging crops, wet and boggy conditions during summer and the distances to be covered required specialised transport. Both two- and four-wheeled motorbikes were used. The four-wheeled motorbike had trouble fitting through interrows less than 1.5 m apart or where the cane was older than third ratoon. However, it was the best infield survey transport in most mill areas. Each bike had a cage designed to prevent face and eye injury from sugarcane leaves. In some areas, over-row spray machines were used to inspect cane from above the crop; safety harnesses and rails were put in place. Cane was also inspected from the tray of a utility along headlands and winch tracks, and by walking rows in some areas.

A target of 1% of the eastern Australian crop was set as a minimum coverage area for the first year. The potential infection area per mill was comprised of the total area harvested for milling that season, the area harvested for plants and the area left as stand-over. These three estimates were combined to produce the area under cane and referred to as the potential survey area. The required inspection rate per mill area was 1% of the area under cane. The 1% was covered by inspecting 10% of blocks in a mill area, then 10% of the rows within those blocks.

The 1998–99 season was the wettest season for many years in most districts of the Queensland and New South Wales sugar industry. This wet weather hampered the survey in many districts and so only 0.76% of the area was surveyed. A drier season in 1999–2000 and an earlier start (September) allowed a greater area to be surveyed that year.

The combined sugarcane smut surveys covered an area of 15,000 ha or 3.75% of the eastern Australian crop over 2 years. This gave approximately a greater than 95% chance of detecting a 0.1% infection (assumptions:  $100,000 \times 4$  ha units = total 400,000 ha;  $3750 \times 4$  ha units were inspected).

## Step 12. Timing of survey

November–March in the first year; September–March in the second year.

The timing follows harvest of the preceding plant (first year after planting) or ratoon (subsequent regrowth) crop. This means that the fields are accessible and there has been some time for the plants to grow and allow the smut infection to develop the ‘whip’ that is the indicator to the survey team.

## Step 13. Data collected

Inspection results were recorded in a Microsoft Excel database with the following information: mill area; farm name; farm number; inspection date; block number; area of block; cultivar; crop class; actual area inspected; diseases noted.

## Step 14. Samples collected

No samples were collected as the pest was not found. If the pest had been found, the plants were not to be touched but marked for further inspection. All inspectors had been briefed on symptoms of sugarcane smut and carried photographs of the disease.

## Reference

Croft, B.J., Magarey, R.C. and Smith, D.J. 1999. Survey of sugarcane in eastern Australia for sugarcane smut. BSES Project Report PR99003.

## Comments

As a contingency for a smut incursion, a trailer with a set of protective clothing and disinfection equipment was carried to the sites being inspected. The trailer contained:

- a Spitwater cold water petrol HP152—a high-pressure water cleaner (2000 psi; 11 litres/min)
- 200 L plastic drum and holder
- toolbox
- 20 L and 10 L jerry cans for motorbike and pressure cleaner fuel (unleaded)
- 5 L container concentrated truck wash
- box of 20 heavy duty disposable overalls.

The washing equipment was used to wash all dirt, mud and seeds from the motorbikes, utility and trailer between mill areas. If the bikes became very muddy or the blocks within a farm had heavy weed infestations, the bikes were washed thoroughly at the farmer's shed before moving to another farm.

Personal disinfection equipment was placed in a backpack as a precautionary measure in case of smut incursion. This equipment was referred to as a smut incursion kit (SIN kit) and included:

- stiff-bristled scrubbing brush to remove mud and dust
- spray bottle for application of 70% alcohol
- screwdriver for removal of mud in soles of shoes
- 1 L of 70% alcohol (diluted methylated spirits)
- spare set of clothes (pants and shirt)
- packet of heavy duty garbage bags.

## 8.12. Case study K. *Pseudomonas* in rice

### Step 1. Purpose of the survey

Early detection survey

### Step 2. Targeted pest name and diagnostic characteristics

Targeted was the bacterium *Pseudomonas*. Early symptoms can be easily confused with those of sheath blight. The lower sheath-leaf of infected seedlings turns yellow, becoming brown and then dark brown. When severe, the whole sheath becomes necrotic. The grains discolour, become deformed, or are empty. Symptoms are generally observed about 80 days after sowing.

### Step 3. Target host

Rice

### Step 4. Alternative hosts

None examined.

### Step 7. The area

Parit Buntar, northern Perak State, Malaysia. This is one of the main granary areas in Peninsular Malaysia. The area under rice cultivation in Parit Buntar is about 20,000 hectares.

### Steps 10 and 11. Site choice and sample size

One sampling block covers approximately 40–100 hectares depending on the terrain and infrastructure (e.g. irrigation canals, roads etc.) of the field. Each sampling block was divided into 10 sub-blocks. At each sub-block, 10 sampling points (15–20 tillers/each



point) are randomly chosen to monitor the status of pests and diseases. Generally, the coverage in terms of area is about 5–10%, depending on the availability of resources such as scouting staff and vehicles.

The survey was performed in farmers' fields. In each field, a diagonal line was laid down and, walking along that line, one clump of plants every 10 steps was checked. One clump generally measured 15 × 15 cm and consisted of about 20 tillers.

### Step 12. Timing of survey

There are two annual planting seasons, September–February and March–July. Sampling was carried out 70 days after planting in both seasons, when symptoms were present. Surveys usually took about a week to complete.

### Step 14. Samples collected

The lower sheaths are examined for symptoms in all 20 tillers at all the 10 points to determine the severity status. Leaf sheaths with suspect symptoms were collected for laboratory culture and identification.

### Reference

Saad, A., Jatil Aliah, T., Azmi, A.R. and Normah, I. 2003. Sheath brown rot: a potentially devastating bacterial disease of rice in Malaysia. International Rice Conference, Alor Setar, Kedah, Malaysia, 2003.

## 8.13. Case study L. Monitoring survey of giant wood moth on eucalypt and teak trees

### Step 1. Purpose of the survey

Monitoring for stem symptoms of wood moth attack in large monoculture plantations of eucalyptus to determine changes in population size or distribution and to determine damage levels. This will assist forest managers in their decision-making on the need to implement pest management measures.

### Step 2. Targeted pest name and diagnostic characteristics

- *Endoxyla cinerea* (Tepper) (Lepidoptera: Cossidae) (formerly in the genus *Xyleutes*).
- Common name: giant wood moth.
- Indigenous to Australia.

**Damage symptoms:** The insect affects trees 2 years of age and older. Tunnelling by the moth larva within the stem/trunk of a tree produces a swelling of the stem around a small entrance hole. Coarse frass or sawdust is often evident around the base of the tree. Before

the adult moths emerge in midsummer, a large, circular exit hole (3–5 cm diameter) is formed above the smaller entrance/feeding hole. When the moth emerges, its empty pupal skin is usually left protruding from the hole, another indicator of wood moth attack.

Visual inspections of trees are conducted by ground transect. If stem swelling and/or coarse frass is observed, then the tree is examined more closely to find an entrance/feeding hole (distinguishing the damage from that of other agents that can cause swelling, such as fungal canker or physical wounding). If it is necessary to confirm identification, smaller trees are usually felled and a section of stem containing the larva is collected for rearing in the laboratory to the adult moth. The moth is then sent to a taxonomist for identification. Larvae of the giant wood moth may reach 15 cm in length and 3 cm in diameter.

### Step 3. Target hosts

*Eucalyptus grandis* (rose gum) and hybrids, *Eucalyptus dunnii* (Dunn's white gum), *Eucalyptus tereticornis* (forest red gum), *Eucalyptus camaldulensis* (river red gum).

### Step 4. Alternative hosts

Several other species of native eucalypt not used in commercial plantations in Queensland and New South Wales.

### Step 7. The area

Coastal areas of Queensland and northern New South Wales, Australia.

### Steps 10 and 11. Site choice and sample size

The districts were industrial forest plantations of the host species listed. These were identified by consultation with known commercial tree growers in Queensland and New South Wales to determine location, age and area of plantings.

Surveys were structured to cover a range of susceptible tree species and age classes across the geographic range of the pest. Larger plantation areas were targeted over small ones because this was more time and cost effective.

Because the insect affects trees 2 years of age and older, younger plantings were excluded. Plantations of host tree species aged 2–3 years were sampled before the first thinning to determine the initial incidence and severity. The impact of wood moth attack is generally most severe for these age classes (tunnelled trees are susceptible to breakage by wind and cockatoo predators seeking the larvae). Sampling was carried out across the range of older age classes but was generally less intensive than for younger plantings.

Ground surveys, although much more effective than roadside cruise in detecting giant wood moth symptoms, are slower to complete and this limited the area of forest that could be sampled. There were four different approaches used: surveying lots of 100 tree transects; surveying fixed-length transects (e.g. 100 m long × 10 m wide strips) per unit area of forest; surveying every 10th row in a plantation compartment; or surveying five lots of 20 tree plots (4 rows × 5 trees) per compartment. An advantage of fixed plots was that they could be sampled over time to show population changes. Paired observers were best for these surveys as they can cover two sides of a tree.

## Step 12. Timing of survey

Surveys are commonly carried out during winter when the exit holes of moths are still visible and when new attack can be easily assessed. Where specimens are required for identification, sampling is best conducted in mid-summer when circular exit holes are visible but late-instar larvae or pupae are still present in the stem. Surveys in late summer can also be effective because pupal skins protruding from wood moth holes aid detection.

## Step 13. Data collected

Locality, plantation compartment, host species, planting date, symptoms, incidence (number of trees affected), severity (number of attacks per tree), date, observer, GPS reading.

## Step 14. Samples collected

Stem lengths of 30–50 cm containing late-instar larvae or preferably pupae for rearing in the laboratory were collected, along with additional larvae for preservation, plant foliage and flowers if required for identification, and photographs were taken.

## Comments

This technique is applicable for surveys of other stem-boring wood moths such as *Xyleutes ceramica*, the beehole borer of teak (*Tectona grandis*) in Asia, and could be combined with surveys for other borers such as *Phoracantha* spp. longicorn beetles.

# 8.14. Case study M. Monitoring survey for damping-off in garden nurseries

## 1. Purpose of the survey

The purpose was to monitor for damping-off in nurseries. Damping-off is the most serious disease affecting forest plantation nurseries in the tropics. Losses of up to 100% of a season's sowings have been reported. While the disease may destroy the entire nursery stock during one rainy season, it may not be important in other years.

This case study is written to provide guidance as to how to survey for damping-off in nurseries.

## 2. Targeted pest name and diagnostic characteristics

Damping-off fungi may be endemic in a nursery without causing damage until environmental conditions change to favour the pathogen and disease development but not early seedling growth. Such conditions are, for example, densely sown seedbeds or trays of seedlings, high soil moisture and humidity, over-watering, excessive shade, and poor ventilation. Damping-off can occur within 2 weeks of seed germination causing large-scale mortality. Viable seeds are killed before the shoot emerges from the soil. Some seedlings that emerge appear to collapse and die. Affected seedlings develop water-soaked,

constricted stem tissue at soil level causing them to fall over and die. Dead and dying plants occur in irregular spreading patches. Once the infection occurs, it can spread very quickly and kill a large number of seedlings within a few days.

Damping-off problems very often develop into root-rot problems after the stem and some of the roots have begun to become woody, so that it becomes impossible to separate the two. Root-rot symptoms are manifested as stunting of seedlings, top dieback, chlorosis and premature defoliation. Roots are discoloured and/or decayed.

## Description of pests

Many indigenous soil-borne fungi are common damping-off fungi that invade the succulent stem tissues. These include fungi in the genera *Cylindrocladium*, *Fusarium*, *Pythium*, *Phytophthora*, *Rhizoctonia* and *Sclerotium*.

Pre-emergence damping-off occurs as a result of fungus attack of the radicle before seedling emergence from the ground. Post-emergence damping-off occurs when the fungus attacks the base of the seedling stem after it emerges from the ground.

Identification of the fungus or fungi causing damping-off should be carried out by a forest pathology or plant pathology laboratory because a wide and diverse group of fungi may be involved.

## Step 3. Target host

Damping-off is not host-specific and commonly occurs throughout the world wherever seedlings grow, in greenhouses, nurseries and natural areas.

## Step 4. Alternative hosts

See Step 3.

## Step 7. The area

This survey can be applied to any garden nurseries.

## Steps 10 and 11. Site choice and sample size

A general monitoring survey should be carried out in any nursery or greenhouse where large numbers of seedlings are raised for a planting program. If there is any knowledge of the occurrence of the disease in a particular area, such locations should also be included in the survey.

Seedlings in seedbeds, nurseries and sometimes in the natural forest may be affected. Densely sown seedlings may be more susceptible, especially during the wet season or when over-watered or raised on substrates with a high organic-matter content.

If the nursery is relatively small and adequate personnel are available for the survey, then a complete survey of the newly sown beds in the nursery could be carried out. However, in large nurseries, or where personnel are limited, about 10% of the newly sown nursery beds in the nursery could be surveyed.

## Step 12. Timing of survey

The survey should be carried out about a week after sowing, just as the seedlings emerge from the soil. This is the time when symptoms will be evident if damping-off is present.

## Step 13. Data collected

In newly sown seedling beds, a visual assessment should be made of the extent of the disease in each bed, as it is impossible to count the number of individual seedlings affected. Large nursery beds can be divided into sectors, quadrants or strips for visual assessment. Assess disease incidence as follows:

Disease incidence	Symptoms	Score
Nil	Nil	0
Low	Up to 25% of seedlings affected	1
Medium	25–50 % of seedlings affected	2
Severe	More than 50% of seedlings affected	3

The disease scores as determined above and the number of seedling/nursery beds surveyed are used to calculate the disease index, which is an indication of the severity of the disease. The disease index is determined as follows:

$$\text{Disease index} = [(na \times 0 + nb \times 1 + nc \times 2 + nd \times 3) \div (N \times 3)] \times 100$$

where:

$na$  = number of beds with score 0

$nb$  = number of beds with score 1

$nc$  = number of beds with score 2

$nd$  = number of beds with score 3

$N$  = total number of beds assessed or in the nursery.

Data to be collected include the total number of seedbeds in the nursery, quantity of seeds sown per seedbed, date of sowing and emergence, frequency of watering, shading conditions, and any disease observations made by the staff of the nursery.

## Step 14. Samples collected

Diseased specimens, i.e. the entire infected seedling, should be collected for isolation and determination of the associated fungi.

## 8.15. Case Study N. Monitoring for root diseases in hardwood plantations

### Step 1. Purpose of the survey

The purpose is to monitor root and butt rot in hardwood plantations, including some conifers such as hoop pine. Root diseases are widespread in nature and require special attention at all levels of planning. Since they affect forest productivity, recreational safety, and biodiversity, monitoring is important so that appropriate planning and management measures can be undertaken.

This case study is written to provide guidance as to how to survey for root diseases in trees, using root rot as an example. The survey is described in Old et al. (1997).

### Step 2. Targeted pest name and diagnostic characteristics

In plantation forests, *Phellinus noxius* (Corner) G. Cunn. root-rot disease is characterised by slowly enlarging patches of dead and dying trees. The foliage of affected trees is usually paler green, sparse and much reduced in size. The condition of the tree crown shows a general decline and the overall growth rate is poor. Young shoots may wilt, and some of the stressed trees may flower and fruit out of season. A high incidence of wind thrown trees in a plantation is usually an indication of the presence of root-rot disease. When symptoms begin to appear in the aerial parts it is too late to save the tree. Fructifications of the fungus appear much later and often after the tree has been killed, and are therefore not helpful in the early diagnosis and control of the disease. To recognise the disease, one needs to look at the symptoms appearing on the roots.

*Phellinus noxius* causes a root-rot disease commonly known as brown root disease in which the roots are encrusted in a mass of earth, sand and stones intermingled and held together by rusty-brown velvety patches of mycelia. The fungus forms a continuous tawny brown fungal skin that darkens with age over infected roots and sometimes extends sock-like up the base of the tree. In the early stages, the rot is pale brown while in later stages brown zigzag lines appear in the wood which remains firm. When the rot is considerably advanced, the wood becomes friable, light and dry, permeated with sheets of brown fungal mycelia which form a honeycomb-like structure. The cells of the honeycomb may be hollow or filled with loose mycelia. A brown network of lines can be seen on the surface of the wood under the bark in advanced stages of the disease.

*Phellinus noxius* forms relatively small, hard fructifications, which may be pileate, effused-reflexed to resupinate. They may be solitary to imbricate. The surface of the pileus is at first finely velvety and pale ferruginous to umber in concentric zones, soon becoming glabrous in irregular sulcate zones and dark brown to black, covered with a 0.2–1 mm thick, resinous, hard crust. The margin is entire, round, often undulating and paler than the rest of the pileus. For further detailed descriptions, see Pegler and Waterston (1968) and Núñez and Ryvarden (2000).

Whether on roots or in heartwood, *Phellinus* rot can, with experience, be identified based on the characteristic pocket (honeycomb) pattern of the rotted wood.

While identification can be based on the presence of fruiting structures, these are seldom found. Instead, roots showing symptoms are collected and the fungi are isolated on artificial media and identified based on culture characteristics or after induction of fructification. For details of the technique, see Lee and Noraini Sikin Yahya (1999).

### Step 3. Target host

The fungus is an important parasite on hardwood plantations in the tropics and although less frequent on conifers is a serious pest of hoop pine (*Araucaria cunninghamii*).

### Step 4. Alternative hosts

None surveyed.

### Step 7. The area

This survey can be applied to any hardwood plantation.

### Steps 10 and 11. Site choice and sample size

Locations within a plantation where mortality due to root and/or butt rot disease has been observed in the previous crop/rotations should be targeted in subsequent surveys.

The number of plantations or field sites to visit may be determined by the range of plantations in the area of interest. Stands may be chosen according to such factors as age, provenance, soil type or because of the presence of dead trees.

The line transect method is recommended. A large-scale map of the stand should be created from an overview map of the area (1:5000 or better). Before conducting the ground survey, transects are laid out within the stand with the assistance of aerial photographs and walk-through reconnaissance data (when available).

A continuous line transect consisting of a strip between 2 and 5 m wide should be placed 50 m from the edge of the stand and not come closer than 10 m to any stand boundary. Parallel transects between 50 and 100 m apart are surveyed over the entire plantation. The length depends on the size of the blocks. Transect lines are flagged and marked for easy relocation and inspection.

### Step 13. Data collected

The following information was collected along each transect line:

- a. location of any dead or infected tree
- b. status of tree (i.e. healthy, standing-infected, standing-dead, windthrown)
- c. presence and extent along transect of infection centres.

Trees fallen due to windthrow were noted only if root disease could be identified as contributing to the cause of windthrow.

Root disease incidence can be calculated as follows:

$$\text{root disease incidence (\%)} = \frac{\text{total number of infected trees} \times 100}{\text{total number of trees examined}}$$

## Step 14. Samples collected

Samples are collected when appropriate.

## Step 12. Timing of survey

It is best to avoid conducting the survey during the dry season or if trees have shed their leaves (for species that defoliate seasonally) so as to prevent confusion of seasonal defoliation with defoliation due to root disease.

## Comments

Various methods for the survey and assessment of root diseases can be found in the Root disease management guidebook, July 1995, authorised by the Forest Practices Code of British Columbia Act, Government of Canada. This can be found on the Internet at <<http://www.gov.bc.ca/tasb/legsregs/fpc/fpcguide/root/chap3a.htm>>.

## References

Lee, S.S. and Noraini Sikin Yahya 1999. Fungi associated with heart rot of *Acacia mangium* trees in Peninsular Malaysia and East Kalimantan. *Journal of Tropical Forest Science*, 11, 240–254.

Núñez, M. and Ryvardeen, L. 2000. East Asian Polypores. Vol. 1. Ganodermataceae and Hymenochaetaceae. Oslo, Norway, Fungiflora. Synopsis Fungorum 13.

Old, K.M., Lee, S.S. and Sharma, J.K., ed. 1997. Diseases of tropical acacias. Proceedings of an international workshop held at Subanjeriji (South Sumatra), 28 April–3 May 1996. CIFOR Special Publication, 53–61.

Pegler, D.N. and Waterston, J.M. 1968. *Phellinus noxius*. Commonwealth Mycological Institute Descriptions of Pathogenic Fungi and Bacteria No. 195.

# 8.16. Case study O. Monitoring survey of defoliation caused by a leaf disease in a plantation

## Step 1. Purpose of the survey

This survey was used to measure the severity of damage (loss of functional leaf area) to a plantation after a leaf disease epidemic. The survey is appropriate for any type of crown damage caused by leaf pathogens or defoliating insects.



## Step 2. Targeted pest names and diagnostic characteristics

*Mycosphaerella* leaf blight is caused by the pathogen *Mycosphaerella nubilosa*. This fungal pathogen infects the young, juvenile foliage of *Eucalyptus globulus* causing large blotchy lesions (Figure O1). The disease causes the soft, expanding leaves near the end of shoots to rapidly shrivel and detach producing a top-down pattern of defoliation (Figure O2).

While *M. nubilosa* is the main pathogen involved in the disease, a range of other *Mycosphaerella* species can be associated with the leaf lesions. The different species can be confidently identified and differentiated only by using DNA analysis.



Figure O1. Large blotchy leaf lesion on a juvenile leaf of *Eucalyptus globulus* due to infection by *Mycosphaerella nubilosa*



Figure O2. Top-down defoliation caused from the blighting infection by *Mycosphaerella nubilosa* of soft, young expanding leaves near the end of shoots

### Step 3. Target host

*Eucalyptus globulus*

### Step 4. Alternative hosts

No alternative hosts were surveyed.

### Step 7. The area

The area was a *Eucalyptus globulus* plantation in northwestern Tasmania, Australia. The plantation was 2 years old and covered an area of 62 ha.

### Steps 10 and 11. Site choice and sample size

We followed the method outlined in Stone et al. (2003) and divided the plantation into eight sub-areas to survey (Figure O3).

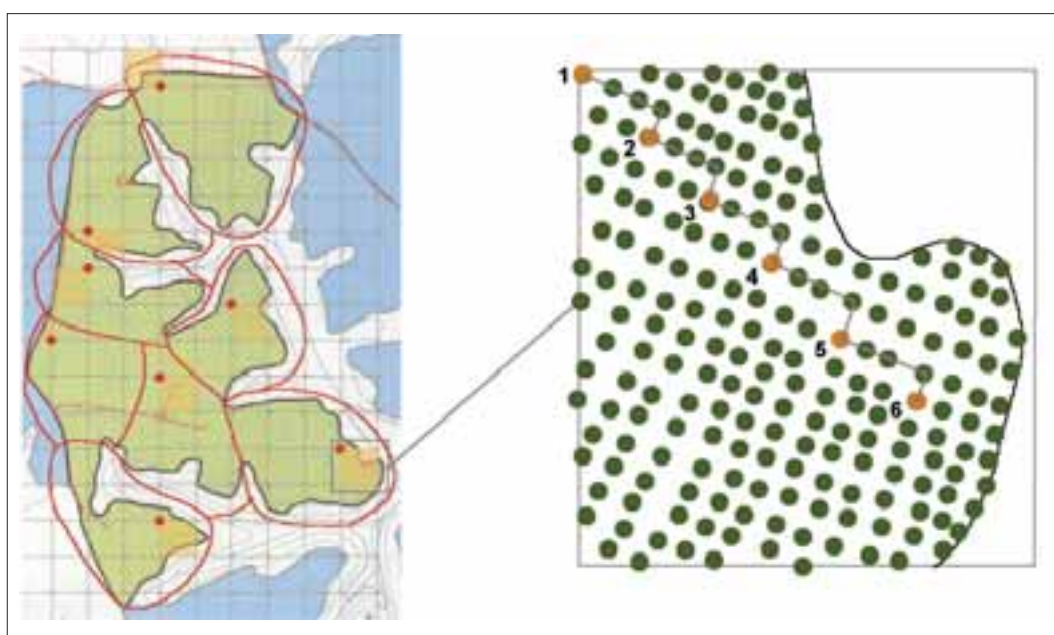


Figure O3. Map of the plantation to be surveyed showing how the plantation was subdivided into eight sub-areas and the use of a 100 m x 100 m grid to randomly select a 1 ha cell within each of the eight sub-areas. The enlarged diagram on the right shows how six trees are selected for measurement from within the 1 ha cell using a stepped transect.

We drew a 1 x 1 cm grid on a map of the plantation (map scale equivalent to 100 m x 100 m on a 1:10,000 map). Each grid cell within each of the eight sub-areas was numbered (starting from the top left-hand grid cell in the sub-area) and a correspondingly numbered piece of paper was put into a jar. We then drew one number out of the jar to randomly select one of the grid cells within each of the eight sub-areas. (Figure O3). The top-left corner of the selected grid cell located the starting point of a stepped transect to select six trees to measure. Where the top-left corner fell outside the plantation, we progressed

in a clockwise direction to the first corner of the grid that fell within the plantation. We walked to each of the selected grid corners, using the map for navigation. Once there, we found the tree closest to the corner and chose it as our first measurement tree. We then moved in a stepped transect (zigzag pattern) towards the diagonally opposite corner of the grid to select the remaining five measurement trees for that grid. The stepped transect involved moving three trees down the row from the tree just selected and then stepping across into the next row to select the closest tree (Figure O3).

## Step 12. Timing of survey

The survey was carried out in late spring following a disease epidemic in late winter and early spring.

## Step 13. Data collected

For each of the trees selected for measurement we estimated (i) the percentage of the tree crown that had been defoliated and (ii) the amount of leaf spotting in the remaining crown. These data are called crown damage index data. We estimated defoliation to the nearest 10% using a visual standard (Figure O4) to assist in making that estimate.

To estimate the amount of leaf spotting we estimated (i) the proportion of the remaining crown leaves with spots, and (ii) the average area of spots per leaf (Stone et al. 2003). The product of those two factors was converted to a percentage (by multiplying by 100) as shown below.

The total leaf area lost as a result of *Mycosphaerella* leaf blight was the sum of the defoliation estimate and the leaf-spotting estimate, e.g.

30% defoliation (i.e.  $100 - 30\% = 70\%$  of leaves remained) [1]

50% of the leaves remaining had leaf spots [2]

the leaf spots affected an average of 30% of the leaf area [3]

Total leaf area lost = % defoliated + (leaves remaining %  $\times 0.5 \times 0.3$ )

Total leaf area lost =  $30\% + (0.5 \times (100\% - 30\%) \times 0.3) = 40.5\%$

We entered the field data into an Excel spreadsheet for calculating the crown damage index of a plantation. A reformatted version of this worksheet showing the data we recorded and the calculated value for average loss of leaf area ('CDI mean') is shown in Figure O5. A copy of the spreadsheet can be downloaded from the National Forest Inventory web site at <http://www.affa.gov.au/nfi>.

A selection of leaves with blotchy necrotic lesions was collected, bagged and returned to the laboratory for pressing and drying. The dried, pressed leaves were put into an appropriately labelled envelope (with accession number, collector, collection date, host species, location) and added to the disease herbarium if needed for future reference.



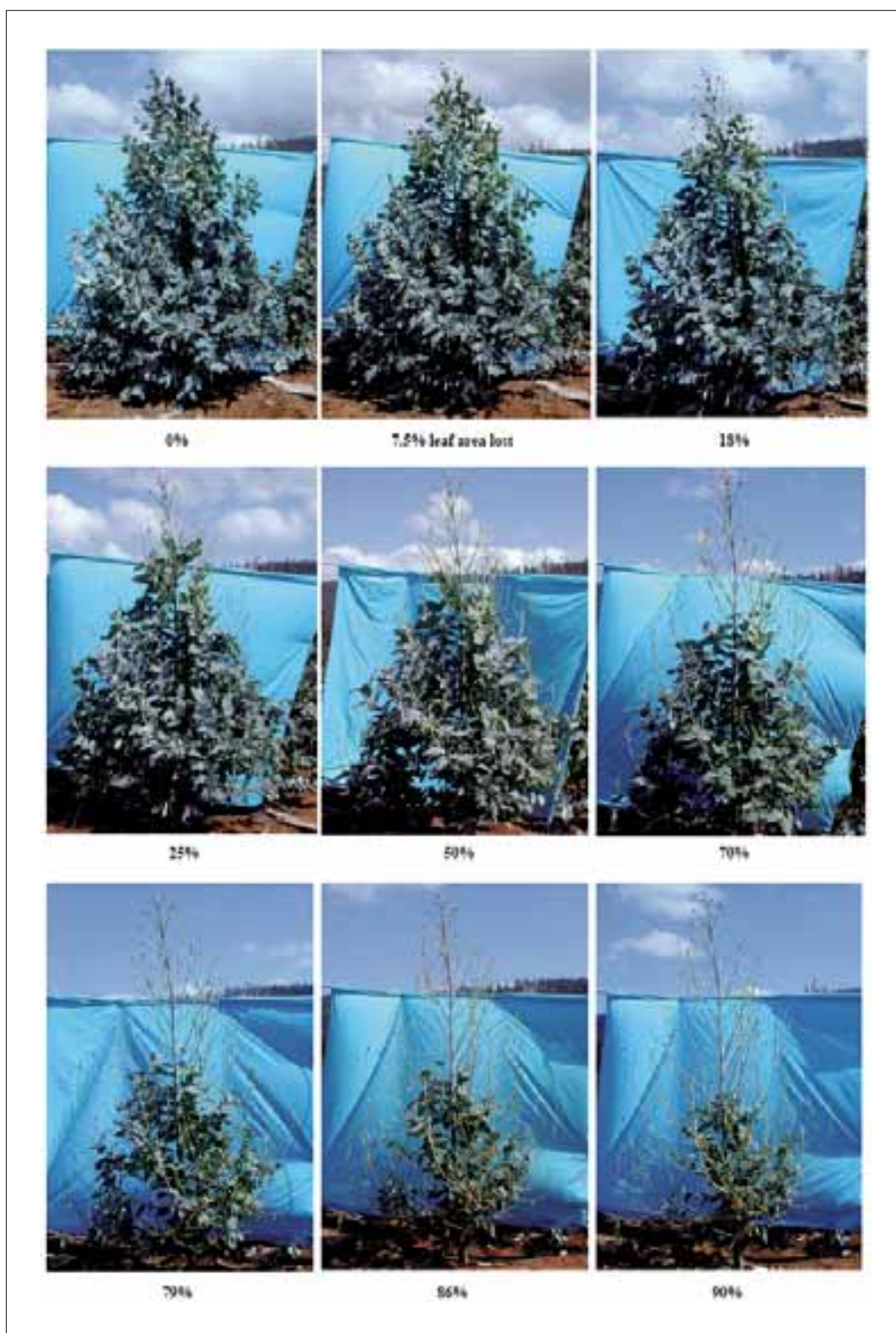


Figure O4. Visual standard for top-down defoliation of 2-year-old *Eucalyptus globulus* trees

date		10/23/01			CDI		
assessor name		Tim Wardlaw			mean		41
plantation name		NW Tas			95%CI		6
year planted		1999			target 95%CI		10
species		globulus			upper bound of 95%CI		47
planted area for that species (ha)		62			lower bound of 95%CI		35
size of grid cell (ha)		1			number of trees assessed		48
stocking rate (stems/grid cell)		1100					
total number of grid cells		62			Defoliation		
target 95%CI width (% of mean)		25			mean		29.4
					95%CI		6.2
					upper bound of 95%CI		34.6
					lower bound of 95%CI		24.2
					Spots		
					mean		11.8
					95%CI		1.5
					upper bound of 95%CI		13.3
					lower bound of 95%CI		10.4
cell	tree	Defoliation	Spots	CDI	comments		
1	1	40	15	55			
1	2	50	15	65			
1	3	20	20	40			
1	4	40	7	47			
1	5	30	12	42			
1	6	20	15	35			
2	1	10	7	17			
2	2	20	6	26			
2	3	20	12	32			
2	4	10	5	15			
2	5	30	12	42			
2	6	10	8	18			
3	1	30	12	42			
3	2	20	8	28			
3	3	30	10	40			
3	4	40	12	52			
3	5	20	12	32			
3	6	20	8	28			
4	1	60	8	68			
4	2	40	12	52			
4	3	20	12	32			
4	4	50	8	58			
4	5	40	10	50			
4	6	20	14	34			
5	1	20	16	36			
5	2	30	12	42			
5	3	30	15	45			
5	4	40	18	58			
5	5	20	12	32			
5	6	20	14	34			
6	1	30	12	42			
6	2	30	18	48			
6	3	10	12	22			
6	4	30	10	40			
6	5	60	12	72			
6	6	40	14	54			
7	1	30	12	42			
7	2	20	8	28			
7	3	20	6	26			
7	4	30	14	44			
7	5	10	12	22			
7	6	20	10	30			
7	10						
8	1	40	15	55			
8	2	50	16	66			
8	3	30	14	44			
8	4	20	12	32			
8	5	40	12	52			
8	6	50	12	62			

Figure O5. Completed spreadsheet showing the defoliation and leaf spotting measurements of 48 trees (six trees in each of eight sub-areas) and calculated values of crown damage index both for individual trees and overall for the whole plantation

## Reference

Stone, C., Matsuki, M. and Carnegie, A. 2003. Pest and disease assessment in young eucalypt plantations: field manual for using the crown damage index. In: Parsons, M., ed., National Forest Inventory. Canberra, Australia, Bureau of Rural Sciences.

## Comments

Equipment needed

One person is able to do this defoliation survey using:

- a map of the plantation drawn at an appropriate scale (e.g. 1: 10,000)
- visual standards appropriate for the type of damage being assessed (defoliation and leaf spots)
- a form for recording the crown damage index data.

For more information

Pest and disease assessment in young eucalypt plantations: field manual for using the crown damage index. September 2003. Australian Government Department of Agriculture, Fisheries and Forestry.

This handbook can be downloaded free from the Internet at:

<<http://www.daff.gov.au/nfi>>

and selecting 'Pest and disease assessment in plantations'. This page will also allow you to download a copy of the Excel spreadsheet that can be used.

## 8.17. Case study P. Survey to measure the incidence of trees with stem wounds

### Step 1. Purpose of the survey

This survey was used to measure the prevalence of trees with stem damage. The survey is appropriate for any type of stem damage whether the cause is biotic (e.g. canker pathogens, stem boring insects), physical (e.g. fire) or mechanical (e.g. thinning wounds).

### Step 2. Targeted pest name and diagnostic characteristics

*Endothia gyrosa* stem cankers were targeted. This fungal pathogen infects and kills the bark of a wide range of woody plants. *Endothia* can be recognised in the field by its small, black, fruiting bodies immersed within a pad of orange fungal tissue that erupts through the bark (Figure P1). The degree of damage caused by the pathogen can vary from negligible in the case of superficial cankers confined to the outer layers of bark (Figure P2) to extreme in the case of deep cankers (Figure P3) that can girdle the stem and kill the tree. In an outbreak, both superficial and deep cankers can occur.



Stem wounds are not easily seen, particularly if the forest has a dense understorey. Because of this, the survey needed to be ground-based to allow each tree in the sample to be closely inspected all the way around its stem.



Figure P1. Black fruiting bodies of *Endothia* immersed within a pad of orange fungal tissue that erupts through the bark



Figure P2. Superficial canker caused by *Endothia*: the infection has not penetrated the entire thickness of the bark so the cambium remains intact. This type of canker has little impact on stem quality



Figure P3. Deep canker caused by *Endothia*: the infection has penetrated the entire thickness of the bark and killed the underlying cambium. The bark within the canker splits and eventually sloughs off to reveal a stem wound.

### Step 3. Targeted host

Shining gum (*Eucalyptus nitens*)

### Step 4. Alternative hosts

No alternative hosts were surveyed.

### Step 7. The area

The area surveyed was a *Eucalyptus nitens* plantation in northern Tasmania, Australia. The 25-ha plantation was 11 years old and had been recently thinned to leave behind a final crop of about 300 pruned trees for future harvest.



## Steps 10 and 11. Site choice and sample size

One rectangular plot of  $100 \times 10$  m was surveyed per 2 ha of forest. Once the number of plots required per coupe was calculated, plot locations were marked on a map of the coupe (1:10,000 scale was ideal). The plots were arranged at right angles from the previous plot without overlapping, to provide the best coverage of the coupe in a zigzag pattern (Figure P4). Whenever possible, the plots ran diagonally along the long axis of the coupe. We avoided locating the plots on log landings or other atypical areas.

A line 100 m long was marked out and trees with the middle of their trunk within 5 m of either side of the line were surveyed.

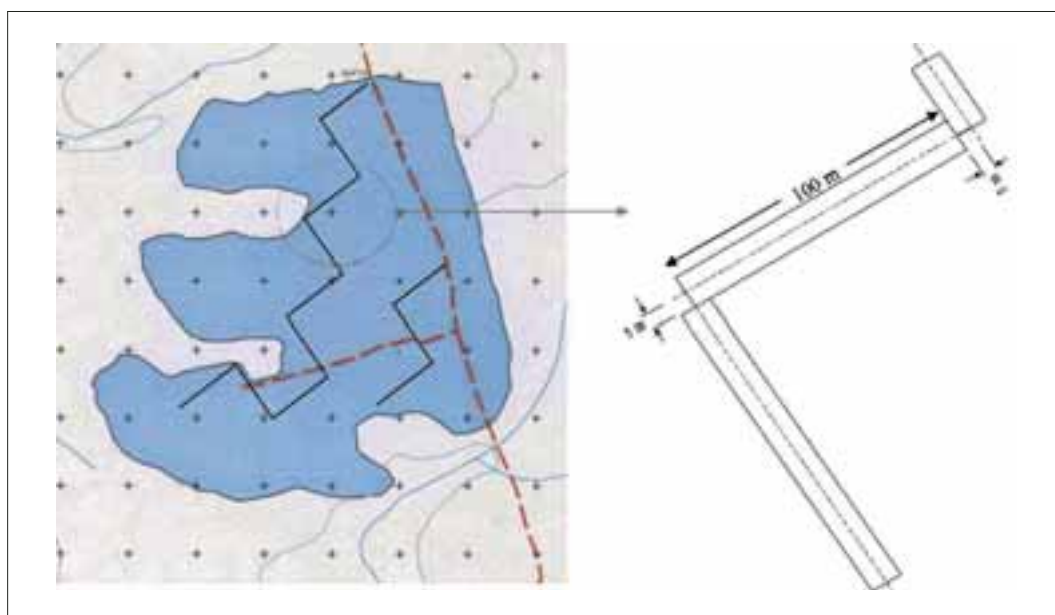


Figure P4. Zig-zag arrangement of  $100 \times 10$  metre plots used for sampling stands to measure stem damage

## Step 12. Timing of survey

The survey was performed in autumn soon after the damage was detected during routine health surveillance, but such a survey can be performed at any time of the year.

## Step 13. Data collected

The survey was used to assess both superficial and deep cankers at the same time. A data sheet was used to record if each tree had superficial, deep or no cankers.

Data were collected for each plot separately (Figure P5). This permitted the total number of trees to be counted and the area covered was known. A percentage of trees per plot with each kind of canker was calculated. The data were used to calculate the average, standard deviation and 95% confidence interval for the data.

## Step 14. Samples collected

The disease can be identified in the field with reasonable confidence solely on the basis of macro symptoms of the dark perithecia immersed in the orange fungal stroma (Figure P1). Nevertheless, several pieces of bark containing the fruiting bodies of the canker pathogen were collected using a hammer and chisel. These were returned to the laboratory, dried and stored with appropriate information (collector, collection date, host, location) as a record of the disease if required for future reference.

## Comments

All plots were marked at each end and at the middle point, i.e. at 0, 50 and 100 m, using a coloured wooden peg or by marking a tree if at the correct distance.

Ensure there is sufficient room for the final 100 × 10 m plot. Only complete 100 m plots should be sampled. Where there is a chance that there will be insufficient room to complete a plot, do not survey it. Many errors have been perpetrated using half or parts of plots.

STEM DAMAGE ASSESSMENT FORM										
Block: Northern Tasmania		Date: 28/04/2005		Damage definitions						
Census point: 1032		Take off point: 10302		Type 1: Superficial canker						
Assessor: T. Wardlaw		Bearing (degrees): 133		Type 2: Deep canker						
		Distance to plot (m): 5								
Unassessed		Assessed								
		Type 1			Type 2			Grand		
Plot No.	Tally	tree	%	Tally	tree	%	Tally	tree	%	Total
1	00 00 00	24	75	00 01	6	10	00	4	12	34
2	00 00 00	10	70	00 00	6	10	00	3	11	27
3	00 00 00	10	60	00 01	6	20	00 01	8	20	30
4	00 00 00	21	60	00	4	13	00 01	6	16	31
5	00 00 00	10	60	00	4	10	00	3	14	22
6	00 00 00	10	50	00 00	6	30	00	4	16	27
7	00 00 00	12	60	0	2	7	00 01	3	30	27
8	00 00 00	10	60	00 01	6	21	0	3	11	26
9	00 00 00	21	60	00 00	7	22	00	4	13	32
10	00 00 00	10	60	00 01	6	20	0	2	9	23
11	00 00 00	13	60	00	6	20	00 00	7	26	26
12	00 00 00	10	70	4	2	8	14	4	17	24
13										
14										
15										
16										
17										
18										
19										
20										
21										
22										
23										
24										
25										
Summary statistics										
		Average	Std. Dev.	95% Conf. Int.		95% C.I. up to % of av.				
Meaning (coloured in 100)		20	3.7	2.1		8				
Incidence: Damage type 1 (%)		10	6.6	3.7		20				
Incidence: Damage type 2 (%)		16	6.7	3.8		23				

Figure P5. An example showing how tree tallies are recorded and calculations made in the damage assessment form

### Equipment needed

A team of at least two people is required to properly do a damage survey. They will need the following equipment:

- a compass to ensure successive plots run at 90° to one another
- a 50 m measuring tape to measure two 50 m lengths
- a 10 m measuring tape to check the plot width
- a damage assessment form
- a calculator
- a hammer, chisel and paper bag to collect and store samples of the canker.

### Reference

Wardlaw, T.J. 1999. *Endothia gyrosa* associated with severe stem cankers on plantation grown *Eucalyptus nitens* in Tasmania, Australia. European Journal of Forestry Pathology, 29, 199–208.

## 8.18. Case study Q. Monitoring survey in pine plantations

### Step 1. Purpose of the survey

This survey was used to monitor the presence of pests or diseases in plantations at a level where the resultant damage is probably sufficient to require remedial treatments. The survey is appropriate for detecting pests and diseases that produce obvious symptoms, such as mortality, dead tops and significant defoliation. Pests and diseases that produce symptoms confined to the stem, such as canker pathogens or stem-boring insects, cannot be reliably detected using this method unless damage is severe enough to result in tree death. Ground surveys are needed to detect pests and diseases that cause such cryptic symptoms.

### Step 2. Targeted pest names and diagnostic characteristics

Usually the target pest will not be known and is not normally encountered. However, early detection surveys will often be an important part of the management of pests that are uncommon but can build to damaging populations very quickly. *Sirex noctilio* (Sirex) wood wasp is one such pest. This stem-boring insect lays its eggs in the sapwood of *Pinus* spp. When the wasp lays its eggs it also secretes a toxic mucus and introduces the fungus *Amylostereum aureolatum*. The combination of the mucus and the fungus causes heavily attacked trees to wilt and die. Trees killed by Sirex have beads of resin flowing down the trunk from the egg-laying sites (Figure Q1). If the adult wasps have emerged, the trunk will also have circular exit holes about 5 mm in diameter (Figure Q2). The wasp can very quickly (over 2–3 years) build up to large populations causing widespread tree losses.



Figure Q1. Pine tree showing beads of resin flowing from egg-laying sites of the wood wasp, *Sirex noctilio*



Figure Q2. Pine tree showing circular exit holes where adult *Sirex* wasps have emerged

### Step 3. Target host

Pine tree (*Pinus radiata*)

### Step 4. Alternative hosts

No alternative hosts were surveyed.

### Step 7. The area

The area was a *Pinus radiata* plantation estate in northern Tasmania, Australia.

### Steps 10 and 11. Site choice and sample size

Within the estate, the focus for Sirex detection was on densely stocked plantations approaching mid-rotation (10–15 years old) on drier sites. These plantations are most susceptible to attack by Sirex

Because of their high density, plantations that are most susceptible to Sirex attack are very difficult to inspect from the roadside or the ground. An overview inspection from the air (Figure Q3) or a high vantage-point (Figure Q4) is the best way to look for Sirex-killed trees. Aerial inspection uses either a helicopter or fixed-wing plane flying at an altitude of 150–200 m above the ground and at speeds of no more than 180 km/h. Vantage-point inspection involves driving or walking to high vantage-points such as a hill top or fire tower and systematically viewing the plantation either with the naked eye or using binoculars. The entire target area needs to be inspected regardless of the method used (air or vantage-point). Where an overview inspection is not possible (this includes areas that cannot be seen from vantage-points) an intensive ground survey needs to be done. This involves walking up or down every third row and inspecting the crowns of individual trees. If recently dead or dying trees are detected during the overview inspection, their location needs to be mapped for follow-up inspection from the ground to confirm the cause of death and, in particular, whether or not Sirex is present.

Static traps using  $\alpha$ -pinene as an attractant (Figure Q5) can be used in susceptible plantations as an alternative to overview inspection or ground surveys for detection of Sirex. Static traps are able to detect Sirex at very low populations. However, they do require regular (fortnightly) servicing during the trapping period.

### Step 12. Timing of survey

This survey was performed in spring, but the work can be done at any time of the year. Response actions to a detection of Sirex need to commence by late autumn to early winter. Because of this, detection surveys targeting Sirex are usually carried out between spring and early autumn.

### Step 13. Data collected

The locations of confirmed Sirex-killed trees were recorded. The locations were recorded either as an annotation on a map or as grid co-ordinates obtained using a GPS instrument.





Figure Q3. Tree death caused by *Sirex noctilio* wood wasp can be visible from small plane or high vantage-point



Figure Q4. Example of hilly terrain where trees could be surveyed for damage from a high vantage-point



Figure Q5. Static traps using  $\alpha$ -pinene as an attractant for *Sirex noctilio* wood wasp

## Equipment needed

A team of two people is needed to properly do a detection survey.

Accurate maps of the plantations being inspected are vital. Maps with a scale of between 1:10,000 and 1:25,000 are most useful for detection surveys. Larger scale maps with scales of between 1:100,000 and 1:250,000 are useful for navigating if you are inspecting the plantations from an aircraft or helicopter. Maps showing contours, water-courses, roads and forest compartments are most useful for mapping the location of trees with symptoms detected from the air or vantage-points.

A compass, protractor and ruler are useful to locate affected trees detected from vantage-points (obtain a compass bearing from the vantage-point to the tree of interest and draw a line of that bearing on the map using a protractor and ruler).

A GPS instrument is useful for obtaining an accurate location of affected trees.

Binoculars are useful for vantage-point inspections of plantations. They allow you to inspect the crowns of individual trees.

## 8.19. Case study R. Monitoring survey of aphids on crucifers

### Step 1. Purpose of the survey

This survey in Vietnam was performed to establish which aphids were present on cruciferous crops in different provinces and which crops they preferred as hosts.

### Step 2. Targeted pest names and diagnostic characteristics

The four aphid species that have cruciferous hosts in Vietnam are:

- *Aphis craccivora* (Koch)—apterae small, adults shiny black. This species is sometimes found between April and July in small colonies on crucifers.
- *Aphis gossypii* (Glover)—apterae very variable in colour on many crops, but on crucifers the adults are dark green and found early in the season (October–November), in small colonies.
- *Brevicoryne brassicae* (L.)—apterae of medium size, 1.5–2.5 mm, greyish green with dark head and dark dorsal thoracic and abdominal markings. The body is covered with greyish white mealy wax which is also secreted onto the surface of host plants. Alatae are 1.3–2.4 mm, with a dark head and thorax and black transverse bars on the dorsal side of the abdomen.
- *Myzus persicae* (Sulzer)—apterae small to medium size, 1.2–2.1 mm, whitish green, pale yellow-green, grey-green, mid-green, pink or red. Alatae have a central black patch on the dorsal side of the abdomen; immatures of the alate morph, especially in autumn populations, are often pink or red.

*Brevicoryne brassicae* and *M. persicae* are typically the dominant aphids. The life-span of *B. brassicae* is 5–10 days, and one apterous female can produce 19–33 nymphs. The life span of *M. persicae* is 6–13 days and females can produce 25–60 nymphs.

In most cases, aphids affect the tender, growing parts of plants. Large colonies can form under the leaves and also on the flower heads of seed crops. When plants are heavily affected, host symptoms are curling of tender leaves, twisting of the tender shoots, yellowing of foliage and stunted growth.

Identification of aphids was confirmed by a taxonomist.

### Step 3. Target host

*Brevicoryne brassicae* (L.) lives on most cruciferous plants, which are planted every month of the year in many provinces. The targeted plants included cabbage, brussels sprouts, radish and cauliflower.

*Myzus persicae* (Sulz.) also lives on most cruciferous plants and tobacco, from October to June in the Red River Delta region. In summer (June–September) we surveyed Cruciferae in only northern mountain areas.



## Step 4. Alternative hosts

*Brevicoryne brassicae* was also surveyed on grape and *M. persicae* on tobacco, peach, pawpaw, water chickweed (*Myosoton aquaticum*), citrus and water spinach (*Ipomoea aquatica* Fosc-Laporte).

## Step 7. The area

The areas surveyed were the largest crucifer production areas in Vietnam. These were suburbs of Hanoi, Haiphong and Sapa in the north of the country and Dalat in the south. These areas have diverse topography, soil conditions, seasonal patterns and host varieties.

## Steps 10 and 11. Site choice and sample size

The places were production areas. Field sites were defined as crop fields.

Due to the limited time, we surveyed 3–5 representative fields of the main seasonal crops and alternative hosts (about 27 fields). Fields were visited five times at 5-day intervals.

We surveyed five sampling sites in each field. Within each sampling site, we selected 10–12 plant parts (shoots, flowers, young plants) that belong to one of five arbitrary classes of infestation (10–12 very light leaves, 10–12 light leaves, 10 medium, 10 heavy). See below.

## Step 12. Timing of survey

Survey was carried out at 5-day intervals because the shortest life span of the aphids is approximately 6 days.

## Step 13. Data collected

At each site, aphids were counted on 5–10 plants or a 20 cm<sup>2</sup> area for seedlings.

Aphids on the plant parts (leaves, stems, shoots, flowers or whole seedlings) were ranked into one of five arbitrary classes of infestation:

zero:

no aphids seen

very light:

from one aphid to a small colony on the leaves

light:

several aphid colonies on the leaves

medium:

aphids present in large numbers, not recognisable colonies but diffusing and infesting a large proportion of the leaves and stem

heavy:

aphids present in large numbers, very dense, infesting all the leaves and stems.

The main data recorded were:

- number of aphids per leaf, shoot, flower, stem or seedling
- number of plant parts showing aphid symptoms per field
- number of natural enemies of aphids observed
- phenology of crop plants
- daily weather conditions.

Entire plants were pressed and photographs were taken.

Data sheets were used to record the data, which were then transferred to Microsoft Excel data sheets.

## Step 14. Samples collected

Twelve sample leaves in each class were collected and aphids from each leaf were collected in 90% alcohol in sealed containers to be later counted in a laboratory. Counts per leaf were recorded.

# 8.20. Case study S. Monitoring survey for phosphine resistant stored grain insects

## Step 1. Purpose of the survey

To monitor any grain insects resistant to the fumigant, phosphine.

## Step 2. Targeted pest names and diagnostic characteristics

Targeted were any phosphine resistant grain insects including beetles, weevils and moth larvae. All grain beetles are small, 2–5 mm long and brown to black. True weevils (a type of beetle) are identified by their elongated snout. The larvae of the grain moths are generally pinkish or cream and may produce webs. Those species that are able to attack whole grains and cause primary damage are the more economically important and include:

- lesser grain borer (*Rhyzopertha dominica* (F.))—Figure S1
- rice weevil (*Sitophilus oryzae* (L.))—Figure S2
- granary weevil (*Sitophilus granarius* (L.)).

The other species cannot attack whole grain but cause secondary damage by eating cracked and broken grains which are found with whole grain. These include:

- rust-red flour beetle (*Tribolium castaneum* (Herbst))—Figure S3
- confused flour beetle (*Tribolium confusum* Jacquelin du Val)
- sawtoothed grain beetle (*Oryzaephilus surinamensis* (L.))—Figure S4
- flat grain beetles (*Cryptolestes* spp.)—Figure S5
- some species of booklice (order Psocoptera).



Figure S1. Lesser grain borer (*Rhyzopertha dominica*)



Figure S2. Rice weevil (*Sitophilus oryzae*)



Figure S3. Rust-red flour beetle (*Tribolium castaneum*)



Figure S4. Sawtoothed grain beetle (*Oryzaephilus surinamensis*)



Figure S5. Flat grain beetles (*Cryptolestes* spp.)

The ‘rapid test’ (Reichmuth 1991), was used to give a quick resistant/not resistant (+/–) result with field collected insects, allowing immediate action (control, eradication or quarantine) to be taken where appropriate.

To more accurately assess insects found to be resistant, one or other of two assays was used.

The first bioassay used the standard FAO technique of placing insects into gastight desiccators and adding phosphine (FAO 1975). Two discriminating doses were used. A lower one discriminated between susceptible and resistant insects and a higher one is designed to detect resistances higher than the common ‘weak’ resistance (Daglish and Collins 1999). The discriminating doses listed in the original method have been adapted according to responses of Australian laboratory reference strains. Insects believed to be homozygous for phosphine susceptibility have been used to determine the lower discriminating dose, while strains homozygous for weak resistance were used to determine the upper discriminating dose. Susceptibility was determined by the dose at which the insects died.

The other assay method used was the flow-through technique that exposes mixed-age cultures of insects to a continuous flow of phosphine at a constant concentration (Winks and Hyne 1997; Daglish et al. 2002). This method is very laborious and lengthy but it gives an accurate prediction of the time required for complete extinction of an insect population at a given phosphine concentration (Daglish and Collins 1999). It is used to characterise the resistance and predict concentrations and exposure periods needed to control insects in the field.

### Step 3. Target host

Grain, cereals and products including wheat, barley, oats, rye, maize, rice, flour, malt, and noodles.

### Step 4. Alternative hosts

None were surveyed.

## Steps 7, 8 and 9. The area, place, district and field sites

The survey targeted grain-export terminals, freight terminals, farm storages, bulk-handling companies and grain processors across Australia, where the grain insects are known to be present or at risk of infesting. Other sources included household samples and quarantine interceptions which could be a source of resistant strains from outside the country.

## Steps 10 and 11. Site choice and sample size

Sampling sites were targeted during routine inspection of all the field sites and locations listed at Step 7. Staff targeted field sites where the practices were not hygienic or where it was suspected that insects might be resistant to phosphine.

## Step 12. Timing of survey

Inspections were performed during the warmer summer months, when beetle activity is at it highest (October–April). In warmer climates, the beetles can survive all year round and so continuous surveillance is required. At terminals, the trapping was continuous.

## Step 13. Data collected

Collector, date, location (including latitude/longitude), property name, property type, owner, survey type, commodity infestation level, and comments including sample position within the storage site were recorded. Following resistance assays, the test date, dose, actual dose, exposure period, number of insects tested and the number surviving the discriminating dose.

## Step 14. Samples collected

Insects were collected using a grain sieve. The recommendation was that at least 100 live insects of each species be collected from each site for the assays.

Pheromone and probe traps were of limited use as the insects were often dead by the time they were returned to the laboratory.

## References:

Daglish, G.J. and Collins, P.J. 1999. Improving the relevance of assays for phosphine resistance. In: Jin, Z., Liang, Q., Liang, Y., Tan, X. and Guan, L., ed., Proceedings of the 7th International Working Conference on Stored-Product Protection, Beijing, 14–19 October 1998. Chengdu, China, Sichuan Publishing House of Science and Technology, 584–593.

Daglish, G.J., Collins, P.J., Pavic, H. and Kopittke, R.A. 2002. Effects of time and concentration on mortality of phosphine-resistant *Sitophilus oryzae* (L.) fumigated with phosphine. Pest Management Science, 58, 1015–1021.

FAO (Food and Agriculture Organization) 1975. Recommended methods for detection and measurement of resistance of agricultural pests to pesticides. Tentative method for adults of some major pests of stored cereals, with methyl bromide and phosphine. FAO Method No. 16. Rome, FAO Plant Protection Bulletin, 23, 12–26.

Reichmuth, C. 1991. A quick test to determine phosphine resistance in stored product pests. GASGA Newsletter, 15, 14–15.

Winks, R.G. and Hyne, E.A. 1997. The use of mixed-age cultures in the measurement of response to phosphine. In: Donahaye, E., Navarro, S. and Varnava, A., ed., International Conference on Controlled Atmospheres and Fumigation in Stored Products, Nicosia, Cyprus, 1996. Printco Limited, 3–16.

## 8.21. Case study T. Papaya infecting strain of papaya ringspot virus (PRSV-P): a delimiting survey

### Step 1. Purpose of the survey

To determine whether an outbreak of papaya ringspot virus in a single papaya tree on the island of Rarotonga was an isolated occurrence or indicated more widespread infestation. This followed confirmation of PRSV infection in leaf samples sent to Fiji and Australia.

Papaya growers and staff of the Ministry of Agriculture, Cook Islands, were on the alert for symptoms of this exotic disease. It had recently been confirmed in neighbouring French Polynesia and SPC had distributed a PestAlert (a one-page information flyer with colour photographs).

### Step 2. Targeted pest name/s and diagnostic characteristics

The principal symptom of papaya ringspot virus is a strong yellow, mosaic and mottling pattern on leaves that is visible, when advanced, from a long distance. Other leaf symptoms are blistering, distortion and sometimes a 'shoestring' symptom (leaf lamina reduction). Symptoms on fruits are characteristic to only this disease. These are dark-green on light-green target-like rings, spots and C-shaped markings which become dark orange-brown as the fruit ripens.

Diagnostic testing in Fiji was performed by double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) and the back-up confirmation in Australia was performed using reverse transcription polymerase chain reaction (RT-PCR).

### Step 3. Target host

*Carica papaya* (papaya, pawpaw)

### Step 4. Alternative hosts

None surveyed.

## Step 7. The area

Rarotonga is an island 32 km in circumference, with a rugged interior of mountains (highest point is 658 m) covered in native bush, surrounded by a narrow band of agricultural land. This includes many large and small commercial plantings of papaya (cv. Waimanalo), grown for export to New Zealand and also for the local market (the value of annual production exceeded NZ\$1 million in 2004). Throughout the cultivable area, papaya trees grow in many domestic and tourist industry properties.

The temperature averages 18–28°C in winter and 21–29°C in summer.

## Steps 10 and 11. Site choice and sample size

The sites were chosen based on where the pest was found and by the anticipated rate of spread. Aphid-borne movement of virus would be limited in two directions as flights in one would meet the forested interior (where aphids would lose a non-persistent virus when feeding on non-hosts) and in another would meet the sea. Spread by humans (via infected seedlings) is possible to anywhere.

The sites surveyed were:

1. the closest 55 trees to the single diseased tree
2. the remainder of the 300 trees in the plot that contained the original diseased tree plus four adjacent plantings
3. all commercial and domestic plantings within 2 km of the diseased tree
4. every other known commercial plot.

Over 5000 trees were individually surveyed and thousands more were observed from various distances. Observations were made by walking every second to fifth row, depending on size of the plantations.

## Step 12. Timing of survey

The survey took place 5–6 weeks after the outbreak was discovered. This was to ensure detection of any natural spread that may have occurred from the original infected tree to other trees before its death. Additionally, symptoms usually take 3–4 weeks to appear in the field following aphid transmission.

## Step 14. Samples collected

Time available for laboratory testing limited the number of leaf samples that could be collected. Apart from sub-area 1, leaf samples were collected because they were showing various leaf abnormalities slightly similar in some ways to those caused by the viral disease.

The 281 leaf samples collected comprised:

1. one from each of the 55 nearest trees, regardless of the appearance of their leaves
2. 16 samples from the affected planting (tree plot) and 15 from the four nearest plantings
3. 83 from plantings or domestic compounds within a 2 km radius of the first detection
4. 112 from commercial plots elsewhere.

Fresh leaf samples were collected, symptoms noted, then stored at 4°C before testing (up to 8 days later) by DAS-ELISA at Totokoitu Research Station. The plus or minus test result threshold used was absorbance readings of greater than three times the mean of four negative control leaves included on each test plate.

## Comments

The eradication was successful because of a rapid response by the government and laboratories involved and by the early detection of the pest which was spotted by a grower who was aware of what to look for thanks to the SPC publicity.

## 8.22. Case study U. Delimiting survey for Huanglongbing disease of citrus and its vector the Asian citrus psyllid in Papua New Guinea

### Step 1. Purpose of the survey

The purpose was to perform a delimiting survey following the detection of Huanglongbing in Vanimo, Papua New Guinea. In the initial detection, 1 of 20 trees examined during a plant health survey had tested positive.

### Step 2. Targeted pest name and diagnostic characteristics

Huanglongbing ('*Candidatus Liberibacter asiaticus*') is an unculturable, phloem-limited bacterium that is vectored by the Asian citrus psyllid, *Diaphorina citri*. The disease caused by Huanglongbing (HLB) is also known as citrus greening disease.

Diagnosis of HLB is difficult because the symptoms are the same as for deficiencies of nutrients such as zinc and manganese, and similar to other disorders. Confirmation of HLB is performed by DNA based PCR tests of leaf tissues from trees that are showing possible HLB symptoms. A yellowing of one section of the tree is the clearest indication of early stages of infection. Leaves display interveinal chlorosis, leaf size is reduced and leaf growth tends to be more upright. Chlorotic blotching, together with one or more other leaf or branch symptoms, especially if accompanied by noticeably swollen veins, may also be a sign of infection. Chronically infected trees are sparsely foliated and stunted, with leaves almost completely devoid of chlorophyll. Fruit may have a lopsided shape with a curved columella.

The Asian citrus psyllid has a high fecundity and a short life cycle (around 14 days) in the absence of natural controls. Eggs are approximately 0.3 mm long, almond shaped, thicker at the base and laid on new shoots. Freshly laid eggs are light yellow but turn bright orange with two distinct red eye spots at maturity. There are five nymphal stages, ranging from 0.25 mm to 1.7 mm. Nymphs have a light pink body and a pair of red compound eyes. In some mature nymphs, the abdomen turns bluish-green instead of pale pink. Adult psyllids can live for 6 months and are 3–4 mm long with a yellow-brown body and grey-



brown legs. Wings are transparent with a broad light-brown marginal band distally on the fore wings. Adults are often found resting on the terminal portion of plant, especially on the lower side of the leaves with their heads pointing downward to the leaf surface at an angle of 30°. When disturbed they readily take flight for a short distance.

*Diaphorina citri* stunts and twists young shoots, so that the growing tips present a rosetted appearance. Leaves are badly curled, and may be covered with honeydew and sooty mould. Leaves may drop prematurely.

### Step 3. Target hosts

Targeted hosts for HLB and the psyllid were all citrus plants. There are different susceptibilities amongst citrus species. HLB symptoms are most severe on mandarins, sweet oranges and their hybrids, moderate on grapefruit, lemon and sour orange, with weak symptoms evident on lime and pomelo.

### Step 4. Alternative hosts

None were surveyed.

### Step 7. The area

The initial infection was detected in Vanimo, in the Sandaun Province of PNG. The town of Vanimo is in a remote area and has a population of about 10,000 (PNG National Census data, 2000). All villages with road access or frequent boat contact with the site of the first detection, Vanimo, were also surveyed.

Survey 1 was conducted in and near Wewak, East Sepik Province and in and near Vanimo, Sandaun Province, totalling 12 villages and including 2 towns. In Survey 2, the same places were surveyed plus coastal villages that have regular contact with Vanimo (both east and west of Wewak as far as Aitape and nearby villages). Survey 2 also included villages inland from Vanimo up to the Bewani region, totalling 23 villages and including 3 towns.

### Steps 10 and 11. Site choice and sample size

In Survey 1, within Vanimo, approximately one tree in every third private backyard in the surrounding streets was examined. Around Wewak the survey was less intensive.

In Survey 2, the streets around the initial infection site were heavily targeted so that all accessible trees were surveyed. At the remaining villages, the most suspicious looking tree was surveyed.

Seventy-two trees were surveyed in the first survey and 48 in the second survey.

### Step 12. Timing of survey

The initial detection of the disease arose from an identification of the psyllid, which prompted an extensive collection of citrus leaves for testing. The first follow-up survey was conducted as soon as it could be arranged (2 months after detection in November 2002), to determine the extent of the disease infestation. The second follow-up survey was conducted 12 months later in November 2003.

Psyllid numbers can fluctuate throughout the year, depending on rainfall and when new growth appears on citrus hosts. The surveys were conducted in November for two reasons; November was still relatively dry, which was important because periods of rain (as often follows from December to April) depress psyllid populations, and the new active leaf growth had begun.

### Step 13. Data collected

The following data were recorded for all samples: sample identification number, date of collection, country, a description of the location—e.g. person's house, street number or nearest major town, GPS coordinates, plant type and name, and name of collector.

### Step 14. Samples collected

Citrus trees showing possible HLB-like symptoms were sampled. Samples consisted of 10–20 leaves per tree, prepared according to the method outlined below.

To sample for psyllids, new growth flushes of host plants were examined for the presence of adults or nymphs. Where psyllids were observed, trees were sampled by sweep net and psyllids collected from the net using an aspirator or pooter. Nymphs were collected using either fine forceps, a scalpel blade or paint brush. Psyllids were then stored in glass vials containing 70% alcohol.

### Comments

Collection and desiccation technique of leaves for Huanglongbing identification

- Collect 10 to 20 leaves displaying symptoms (aim for 1–2 g fresh weight of excised petioles and midribs). Number is dependent upon leaf size. Smaller leaves will require a larger number to be collected.
- If possible, surface sterilise leaves with 70% alcohol or 1% pool chlorine.
- Using a sharp knife cut midribs and/or petioles from leaves and chop midribs and petioles into approximately 2–3 mm lengths (Figure U1). The disease organism is limited to the midribs and petioles of the plant. It is important to collect only this material as any extra leaf material may reduce the sensitivity of the test
- Wrap in paper (facial) tissue or medical gauze bandage and place over calcium chloride in 25 mL plastic vials, wrap Parafilm or insulation tape around the join of the lid and vial and place it immediately in the refrigerator. Calcium chloride will dry the leaf material so that it can be sent for testing.
- The following day replace the paper tissue or gauze with fresh, dry paper tissue or gauze and reseal vials. If possible store in the refrigerator or in a cool box with ice. For long-term storage, material must be held in a refrigerator.
- If sending material to a country free of HLB for testing, ensure quarantine import permits are in place before consigning the samples. Sample vials must be consigned in a robust screw-cap container.



Figure U1. Preparation of leaves for later plant pathogen identification

## 8.23. Case study V. Delimiting survey for red-banded mango caterpillar in northern Queensland

### Step 1. Purpose of the survey

Delimiting surveys for red-banded mango caterpillar (RBMCM, *Deanolis sublimbalis*) help the Queensland Department of Primary Industries and Fisheries (QDPI&F) formulate risk-management options that reduce the potential impact upon commercial mango production areas to the south. The survey also involves a general surveillance component of public awareness and reporting campaigns.

### Step 2. Targeted pest name/s and diagnostic characteristics

RBMCM has slowly spread from Papua New Guinea through the Torres Strait Islands and was first detected on mainland Australia in 2001. The pest is under active control, with regulations prohibiting movement of mango fruit or plants on northern Cape York Peninsula.

Caterpillars burrow into the mango fruit causing severe damage and fruit drop. Holes in fruit with weeping sap stains are a good indicator of pest presence. The caterpillar is distinctively coloured and easy to identify when the fruit and seed are cut. Samples

are taken to compare to existing reference collection material and to substantiate pest records. Identification can be confirmed by DNA sequencing at the Australian National Insect Collection.

### Step 3. Target host

RBMC host range is restricted to mangoes (*Mangifera* spp. and *Bouea* spp.).

### Step 4. Alternative hosts

None.

### Step 7. The area

The survey area covers Cape York Peninsula in northern Queensland, commercial production areas around the Atherton Tablelands near Cairns and the regional cities of Cairns, Townsville and Mackay (Figure V1).

The current infested area of feral mango trees is in rainforest in the northernmost 30 km of Cape York Peninsula known as the Northern Peninsula Area. Scattered mango trees near old sites of human activity make a bridge to the Northern Peninsula Area Indigenous communities where hundreds of mango trees are present. RBMC is currently separated from commercial mango production areas by 700 km of sparsely populated, inhospitable habitat so the main threat comes from caterpillars being carried in fruit.

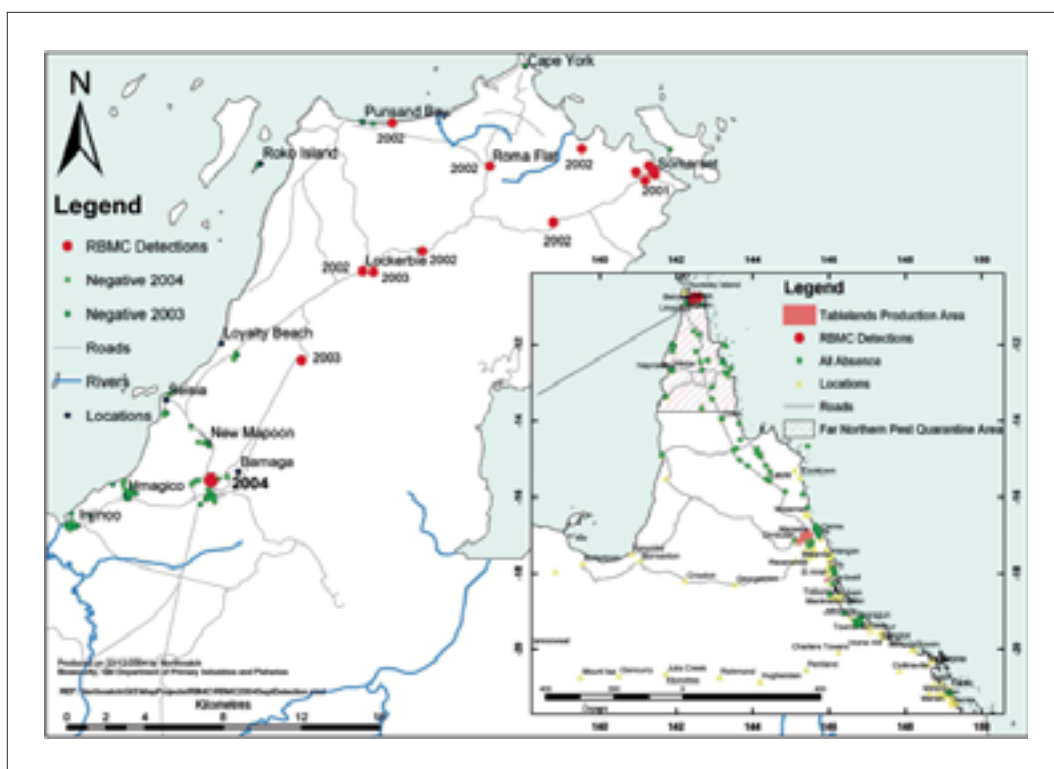


Figure V1. QDPI&F delimiting surveillance for RBMC

## Steps 10 and 11. Site choice and sample size

Surveys targeted the pathways that the pest might use to reach production areas. Early delimiting surveillance was used to support an eradication attempt. After the eradication area was breached, the control options concentrated on regulatory restrictions on fruit movement and public awareness campaigns.

The most immediate pathway is natural spread through the Northern Peninsula Area communities, followed by movement in fruit to tourist sites, communities on Cape York Peninsula, major regional cities and production area towns.

Range expansion from natural spread is delimited by examining all known trees around the perimeter of the infested area annually. The Northern Peninsula Area communities are exposed to both the threat of natural spread and the threat of illegal movement of infested fruit. Approximately one quarter of all trees in the communities are examined and at least 10 of the most suspicious fruit per tree are cut. This surveillance intensity will detect RBMC before populations reach a size that imposes a significant additional risk of transport from the communities towards production areas.

As far as possible, a uniform coverage for each community is surveyed to increase the chance of detecting an incursion. Random sampling is too time-consuming to implement and can result in larger areas not being inspected and adjacent areas that are likely to have similar fauna being over-examined.

NAQS surveillance and regulatory activity target air and sea pathways for RBMC which reduces the delimiting surveillance activity required of the QDPI&F. A single road pathway connects the infested area to the production area. All mango trees at tourist stops on this road are examined annually with additional trees examined around the few towns. Surveillance of fruit is conducted at a quarantine control point at which all mangoes are confiscated from travellers.

Around production areas, roadside and backyard trees are examined as they are not treated with insecticides and are more likely to be in areas exposed to discarded, infested fruit. Public awareness material distributed to growers is the most effective way of monitoring for the pest on farms. Annual surveys in these areas give confidence to growers that the pest is not common in the immediate vicinity and is unlikely to affect them in the season.

Year	Sites	Trees at sites	Trees examined	Fruit cut
2001	240	1,050	898	657
2002	98	999	746	770
2003	129	1,128	647	293
2004	48	357	351	2,701
Total	515	3,534	2,642	4,421

Surveillance of a wide range of pests in urban areas (see also Case study D) targets the spectrum of potential exotic pests. As most of the work involved in surveillance is in getting to sites, gardens that have a broad host range are targeted as they could support high-priority pests. RBMC is one of these pests and information on its absence is collected if fruit is available to be cut.

Public awareness campaigns encourage the public to report the pest, which augments confidence in pest absence records. Material asking people to report the pests is specifically designed for Indigenous communities, tourists, growers and urban residents. Extensive signage informing people of the regulatory restrictions is deployed along paths where the pest is likely to move.

### Step 12. Timing of survey

Surveys around the infested area are timed to coincide with good fruit development, but early in the season before the roads become impassable. Late season surveys are conducted around production areas, while urban area surveys are staggered throughout the year for different pests.

### Step 13. Data collected

Data recorded for each site are the observers' names, date, site description, GPS location, number of trees present, number of trees examined and number of fruit cut. Absence data are recorded explicitly on the form and suspect samples are collected into ethanol.

### Comments

Knowing where the pest is helps authorities to distribute public awareness material to the right place, modify internal quarantine inspection so as to mitigate the risk of fruit movement and keeps commercial growers informed of the level of threat to their industry.

## 8.24. Case Study W. Delimiting survey of the Queensland fruit fly in Rarotonga, Cook Islands

### Step 1. Purpose of the survey

Queensland fruit fly, *Bactrocera tryoni* (Froggatt), was detected at the Punanga Nui market in Avarua, Rarotonga, Cook Islands on 21 November 2001, 500 m from the wharf. This survey was part of an emergency response and eradication program.

### Step 2. Targeted pest name and diagnostic characteristics

*Bactrocera tryoni* is considered to be a fruit fly exotic to the Cook Islands. It is about 7 mm long, almost the same size as a housefly. It is distinguished from two other species present in the Cook Islands by the predominantly red–brown colour of the dorsal part of its thorax and abdomen, and the bright yellow scutellum (Figure W1). It has a single pair of transparent wings with a large black dot on the wing tip and black cross-streak on each wing.

*Bactrocera tryoni* is considered the most damaging fruit fly species in Australia, and is common in the eastern half of Queensland, eastern New South Wales, and the extreme east of Victoria. It is widespread in New Caledonia, French Polynesia and Pitcairn Islands. It was introduced into, but subsequently eradicated from Perth (Western Australia) and Easter Island in the mid-Pacific.



Figure W1. Adult Queensland fruit fly in dorsal (left) and lateral (right) views

### Steps 3 and 4. Target and alternative hosts

*Bactrocera tryoni* is a polyphagous species recorded from more than 113 host plant species in Australia and the Pacific. High risk hosts in the Pacific include breadfruit (*Artocarpus altilis*), guava (*Psidium guajava*), mango (*Mangifera indica*), Tahitian chestnut (*Inocarpus fagifer*), syzygium apples (*Syzygium* spp.) and tropical almond (*Terminalia catappa*). As the survey used a trapping grid, the hosts targeted were those at the grid cross-points.

### Step 5. The area

Rarotonga is a volcanic island 32 km in circumference, with a rugged interior of mountains (the highest point is 658 m) covered in native bush, surrounded by a narrow band of agricultural land, which in turn is encircled by a ring of swamps used largely for growing taro. Along the coasts, coconut plantations, beaches, villages and small hotels fringe the island.

The temperature averages 18–28°C in winter and 21–29°C in summer.

### Steps 10 and 11. Site choice and sample size

The Ministry of Agriculture of the Cook Islands had in place a Fruit Fly Emergency Response Plan before the incursion, designed to allow a rapid, organised response.

To identify the trapping site locations, a grid of the recommended distances for the traps outlined in the Emergency Response Plan was laid over a map of the quarantined area using a geographic information system. The magnified maps were then used as guidelines to allocate the trapping sites.

Traps were placed in host trees at the sites wherever possible (most of the time), or in non-host trees.



### Pre-incursion

There were 15 trapping sites, each with a modified Lynfield trap (Figure W2) baited with *Cue-lure* and methyl ethyl eugenol attractant. All traps were installed at high-risk locations such as the ports of entry, major tourist accommodation sites, diplomatic missions and the rubbish dump.



Figure W2.  
Lynfield trap

### Post-incursion

#### *Cue-lure* pheromone traps

Twenty-four hours after the incursion, the Ministry of Agriculture increased the intensity of the trapping network by setting out five additional *Cue-lure* pheromone traps. This resulted in the detection of the second male Queensland fruit fly 280 m from the first site.

The second detection prompted the increase of the trapping radius to 1 km and the whole area was labelled as Zone A. In Zone A, 25 traps were set up in a grid of 300 m<sup>2</sup>.

A third Queensland fruit fly picked up in Zone A initiated an increase in the density of traps within the Zone covering an area of 800 m<sup>2</sup> (called the 'intensive zone'). In the intensive zone, 30 traps were set up in a grid of 150 m<sup>2</sup>. The Ministry of Agriculture extended the quarantine area to a further 2.5 km radius (Zone B) where 38 *Cue-lure* traps were set in a grid of 500 m<sup>2</sup>.

The decisions pertaining to the identification and establishment of Zones A and B were as guided under the Service Outline section of the General and Cook Islands Fruit Fly Emergency Response Plan. The different zoning technically signifies the different response levels, which are initiated depending on the circumstance of the find, and the whole response system is made up of different components that all contribute to trying to contain, control and eradicate the find.



### Capi-lure traps

Capi-lure traps were set up at seven selected sites (Figure W3).



Figure W3. Map of the Quarantine Fruit Fly Surveillance Trapping Network

### Protein bait spraying

A protein bait spraying program targeting female fruit flies was initiated. This involved spot spraying of trees at about 30 m apart all throughout the infested area covering a distance of about 2.6 km<sup>2</sup>.

### Destruction of breeding sites

Fallen fruits, which amounted to approximately 50,000 kg, were also collected from the surrounding area and buried in the quarantined region.

### BactroMAT C-L bait stations distribution (male annihilation technique)

BactroMAT C-L bait stations were distributed at 800/km<sup>2</sup> covering an area of 8 km<sup>2</sup>. The process involved the tying of pheromone baits onto trees at a spacing of 30 m.

## Step 12. Timing of survey

The traps were checked every fortnight and the flies caught examined and identified by an entomologist. The lures were re-dipped every 2 months.

## Steps 13 and 14. Data and samples collected

### Trapping

Any Queensland fruit flies collected in the traps were recorded.

### Fruits

A fruit-collecting program was initiated to assess fruit-fly damage and data collected were used in the delimiting survey.

Fallen fruits and fruit picked from trees which could have been infested by the Queensland fruit fly were collected from Zones A and B. Generally, the fruit samples collected were the ones examined and seen to contain symptoms of fruit-fly damage. An average of 25 samples was collected weekly, totalling 940 samples over 14 months.

The fruit were counted, and their weight and site of collection recorded. No Queensland fruit fly were reared from the fruit sampled.

## Comments

The success of the Ministry of Agriculture in eradicating *B. tryoni* from Rarotonga rested on the following:

1. There was an existing trapping network that was regularly serviced. This enabled early warning while the fruit fly was still in low numbers.
2. Ministry of Agriculture staff were well-trained in recognising indigenous fruit fly species as well as the high-risk exotic species in the Pacific.
3. Staff responded quickly and in an organised way to the incursion because Cook Islands had an Emergency Response Plan in place. The plan clearly outlined the activities that had to be undertaken upon the detection of an exotic fruit fly species.

## Further reading:

Kassim, A., Allwood, A.J., Wigmore, W., Leblanc, L. and Tora Vueti, E. 2001. Fruit flies in Cook Islands. Suva, Fiji Islands, Secretariat of the Pacific Community, Plant Protection Service, Pest Advisory Leaflet No. 35.

Maddison, P.A. 1983. Queensland fruit fly. Noumea Cedex, New Caledonia, Secretariat of the Pacific Community, Plant Protection Service, Pest Advisory Leaflet No. 18.

SPC (Secretariat of the Pacific Community) 2001. Queensland fruit fly (*Bactrocera tryoni*) found in Rarotonga, Cook Islands. Suva, Fiji Islands, SPC, Plant Protection Service, PestAlert No. 25.