United States Department of Agriculture

Animal and Plant Health Inspection Service

Plant Protection and Quarantine

New Pest Response Guidelines

Oryctes rhinoceros (L.)
Coleoptera: Scarabaeidae

Coconut Rhinoceros Beetle

First Edition Issued 2014

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O. rhinoceros

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Cover Image

Coconut palm frond damaged by *O. rhinoceros* (photo courtesy of Arnold Hara, University of Hawaii)

Male adult of *O. rhinoceros* (photo courtesy of Pest and Diseases Image Library, Bugwood.org)

Coconut palms killed by *O. rhinoceros* in Fiji (image by Geoffrey Bedford printed with permission from the Annual Review of Entomology, Volume 58 © 2013 by Annual Reviews, http://www.annualreviews.org)

Chapter

1

Introduction

Introduction

Use *New Pest Response Guidelines: Oryctes rhinoceros* (L.) when designing a program to detect, monitor, control, contain or eradicate an outbreak of this pest in the United States and collaborating territories.

The United States Department of Agriculture, Animal and Plant Health Inspection Service, Plant Protection and Quarantine (USDA–APHIS–PPQ) developed the guidelines through discussion, consultation or agreement with staff members at the USDA-Agricultural Research Service and advisors at universities.

Any new detection may require the establishment of an incident command system to facilitate emergency management. This document is meant to provide the necessary information to launch a response to a *O. rhinoceros* detection.

If *O. rhinoceros* is detected, PPQ personnel will produce a site-specific action plan based on the guidelines. As the program develops and new information becomes available, the guidelines will be updated.

Users

The guidelines are intended as a field reference for the following users who have been assigned responsibilities for a plant health emergency involving *O. rhinoceros*:

- PPQ personnel
- Emergency response coordinators
- State agriculture department personnel
- Others concerned with developing local survey or control programs

Contacts

When an emergency program for *O. rhinoceros* has been implemented, the success of the program depends on the cooperation, assistance and understanding of other involved groups. The appropriate liaison and information officers should distribute news of the program's progress and developments to interested groups including the following:

- Academic entities with agricultural interests
- Agricultural interests in other countries
- Commercial interests
- Grower groups such as specific commodity or industry groups
- Land-grant universities and cooperative extension services
- National, state and local news media
- Other federal, state, county and municipal agricultural officials
- Public health agencies
- The public
- State and local law enforcement officials
- Tribal governments

Initiating an Emergency Pest Response Program

An emergency pest response program consists of detection and delimitation and may be followed by programs in regulation, containment, eradication and control. The New Pest Advisory Group (NPAG) will evaluate the pest. After assessing the risk to U.S. plant health and consulting with experts and regulatory personnel, NPAG will recommend a course of action to PPQ management.

Follow this sequence when initiating an emergency pest response program:

1. A new or reintroduced pest is discovered and reported
2. The pest is examined and pre-identified by regional or area identifier
3. The pest's identity is confirmed by a national taxonomic authority recognized by the USDA–APHIS–PPQ National Identification System
4. Published New Pest Response Guidelines are consulted or a new NPAG is assembled to evaluate the pest
5. Depending on the urgency, official notifications are made to the National Plant Board, cooperators and trading partners
6. A delimiting survey is conducted at the site of detection
7. An incident assessment team may be sent to evaluate the site

8. A recommendation is made, based on the assessment of surveys, other data and recommendation of the incident assessment team or the NPAG as follows:
   - **A.** Take no action
   - **B.** Regulate the pest
   - **C.** Contain the pest
   - **D.** Suppress the pest
   - **E.** Eradicate the pest
9. State departments of agriculture are consulted
10. If appropriate, a control strategy is selected
11. A PPQ Deputy Administrator authorizes a response
12. A command post is selected and the incident command system is implemented
13. State departments of agriculture cooperate with parallel actions using a unified command structure
14. Trace-back and trace-forward investigations are conducted
15. Field identification procedures are standardized
16. Data reporting is standardized
17. Regulatory actions are taken
18. Environmental assessments are completed as necessary
19. Treatment is applied for required pest generational time
20. Environmental monitoring surveys are conducted to evaluate program success
21. Pest monitoring surveys are conducted to evaluate program success
22. Programs are designed for eradication, containment or long-term use

Preventing an Infestation

Federal and state regulatory officials must conduct inspections and apply prescribed measures to ensure that pests do not spread within or between properties. Federal and state regulatory officials conducting inspections should follow the sanitation guidelines in the section *Preparation, Sanitization and Clean-Up* on page **Error! Bookmark not defined.** before entering and upon leaving each property to prevent contamination.

Scope

The guidelines are divided into the following chapters:

1. *Introduction* on page **Error! Bookmark not defined.**
2. *Taxonomy* on page **Error! Bookmark not defined.**
3. *Identification* on page **Error! Bookmark not defined.**

4. *Biology* on page **Error! Bookmark not defined.**
5. *Damage* on page **Error! Bookmark not defined.**
6. *Survey Procedures* on page **Error! Bookmark not defined.**
7. *Regulatory Procedures* on page **Error! Bookmark not defined.**
8. *Control Procedures* on page **Error! Bookmark not defined.**
9. *Environmental Compliance* on page **Error! Bookmark not defined.**
10. *Pathways* on page **Error! Bookmark not defined.**

The guidelines also include appendices and a list of literature cited.

Authorities

The regulatory authority for taking the actions listed in the guidelines is contained in the following authorities:

- Plant Protection Act of 2000 (Statute 7 USC 7701-7758)
- Executive Order 13175, Consultation and Coordination with Indian and Tribal Governments
- Fish and Wildlife Coordination Act
- National Historic Preservation Act of 1966
- Endangered Species Act
- Endangered and Threatened Plants (50 CFR 17.12)
- National Environmental Policy Act

Program Safety

The safety of the public and program personnel is a priority in pre-program planning and training and throughout program operations. Safety officers and supervisors must enforce on-the-job safety procedures.

Support for Program Decision Making

The USDA–APHIS–PPQ–Center for Plant Health, Science and Technology (CPHST) provides technical support to emergency pest response program directors concerning risk assessments, survey methods, control strategies, regulatory treatments and other aspects of the pest response programs. PPQ managers consult with state departments of agriculture in developing guidelines and policies for pest response programs.

How to Use the Guidelines

The guidelines are a portable electronic document that is updated periodically. Download the current version from its source and then use Adobe Reader® to view it on your computer screen. You can print the guidelines for convenience; however, links and navigational tools are only functional when the document is viewed in Adobe Reader®. Remember that printed copies of the guidelines are obsolete once a new version has been issued.

Conventions

Conventions are established by custom and are widely recognized and accepted. Conventions used in the guidelines are listed in this section.

Advisories

Advisories are used throughout the guidelines to bring important information to your attention. Please carefully review each advisory. The definitions have been updated to coincide with the America National Standards Institute (ANSI) and are formatted as follows:

Example	Example provides an example of the topic.
Important	Important indicates information that is helpful.

CAUTION

CAUTION indicates that people could possibly be endangered and slightly hurt.

DANGER

DANGEROUS indicates that people could easily be hurt or killed.

NOTICE

NOTICE indicates a possibly dangerous situation where goods might be damaged.

WARNING

WARNING indicates that people could possibly be hurt or killed.

Boldfacing

Boldfaced type is used to highlight negative or important words. These words are: **never**, **not**, **do not**, **other than** and **prohibited**.

Lists

Bulleted lists indicate information listed in no particular order. Numbered lists indicate that information will be used in a particular order.

Disclaimers

All disclaimers are located on the page that follows the cover.

Control Data

Information placed at the top and bottom of each page helps users keep track of where they are in the guidelines. At the top of the page is the chapter. At the bottom of the page is the year, edition, title and page number. PPQ–Pest Detection and Emergency Programs (PDEP) is the unit responsible for the content of the guidelines.

Footnotes

When space allows, figure and table footnotes are located directly below the associated figure or table. However, for multi-page tables or tables that cover the length of a page, footnote numbers and footnote text cannot be listed on the same page. If a table or figure continues beyond 1 page, the associated footnotes will appear on the page following the end of the figure or table.

Heading Levels

Within each chapter and section there can be 4 heading levels; each heading is green and is located within the middle and right side of the page. The first-level heading is indicated by a horizontal line across the page with the heading following directly below. The second-, third- and fourth-level headings each have a font size smaller than the preceding heading level. The fourth-level heading runs in with the text that follows.

Hypertext Links

Figures and tables are cross-referenced in the body of the guidelines and are highlighted in blue hypertext type.

Italics

The following items are italicized throughout the guidelines:

- Cross-references to headings and titles
- Names of publications
- Scientific names

Numbering Scheme

A two-level numbering scheme is used in the guidelines for pages, tables and figures. The first number represents the chapter. The second number represents the page, table or figure. This numbering scheme allows for identification and updating. Dashes are used in the page numbering to differentiate page numbers from decimal points.

Transmittal Number

The transmittal number contains the month, year, and a consecutively issued number (beginning with -01 for the first edition and increasing consecutively for each update to the edition). The transmittal number is only changed when the specific chapter sections, appendices, tables or index is updated. If no changes are made, then the transmittal number remains the unchanged. The transmittal number only changes when a new guidelines edition is issued or changes are made to the entire guidelines.

Acknowledgements

Writers, editors, reviewers, creators of cover images and other contributors to the guidelines are acknowledged in the acknowledgements section. Names, affiliations and Website addresses of the creators of photographic images, illustrations and diagrams, are acknowledged in the caption accompanying the figure.

How to Cite the Guidelines

Cite the guidelines as follows:

U.S. Department of Agriculture, Animal Plant Health Inspection Service, Plant Protection and Quarantine. 2011. New Pest Response Guidelines: *Oryctes rhinoceros* (L.). Washington, D.C.: Government Printing Office.
http://www.aphis.usda.gov/import_export/plants/manuals/online_manuals.shtml

How to Find More Information

Contact USDA–APHIS–PPQ–EDP–Emergency Management for more information regarding the guidelines. Refer to *Resources* on page A-**Error! Bookmark not defined.** for contact information.

Chapter

2

Taxonomy

Linnaeus originally described the species as *Scarabaeus rhinoceros* in 1758; in 1798, Illiger proposed the genus *Oryctes* (Brands, 1989-2005).

Table 2-1 Classification of the coconut rhinoceros beetle (Arnett *et al.*, 2002; Brands, 1989-2005; Schoolmeesters, 2014b; Triplehorn *et al.*, 2005)

Rank	Taxon
Phylum	Arthropoda
Class	Insecta
Order	Coleoptera
Suborder	Polyphaga
Superfamily	Scarabaeoidea
Family	Scarabaeidae
Subfamily	Dynastinae
Tribe	Oryctini
Genus	*Oryctes*
Scientific name (accepted)	*Oryctes rhinoceros* (L.)

In the Catalogue of Life (Schoolmeesters, 2014a), Scarabaeidae and Dynastidae are considered 2 separate families. However, Arnett *et al.* (2002) and Triplehorn *et al.* (2005) included the dynastids in the subfamily Dynastinae and in the family Scarabaeidae. In addition, Systema Naturae provides similar taxonomic classification (Brands, 1989-2005), which is utilized throughout this document.

Synonyms

- *Oryctes stentor* Castelnau
- *Scarabaeus rhinoceros* L.

Common Names

Coconut rhinoceros beetle is the preferred common name of *O. rhinoceros*. Table 2-2 provides a list of names common throughout the world.

Table 2-2 Common names for *O. rhinoceros* (Gressitt, 1953; Mohan *et al.*, 2005; Schoolmeesters, 2014b)

Language	Common names
English	palm rhinoceros beetle, rhinoceros palm beetle, Asiatic rhinoceros beetle, Indian rhinoceros beetle, coconut palm rhinoceros beetle, coconut palm beetle, coconut black beetle, black beetle, coconut beetle, date palm beetle, dung beetle
Palauan	arm-ar-alius
Japanese	yashino kabutomushi, Taiwan- kabutomushi, sai-kabutomushi
Samoan	manu-i-niu, avi-i-vii
Tagalog	uang
Malay	kumbang badak, kumbang kelapa
Dutch	klappertor
German	Indischer nashornkäfer
French	oryctes du cocotier, rhinoceros du cocotier
Spanish	escarabajo rinoceronte Asiático

Chapter

3

Identification

Species Description/Morphology

The developmental stages of the *O. rhinoceros* beetle—egg, larva, pupa, and adult—are presented in Figure 3-1, and all morphometric measurements of the stages are listed in Table 3-1.

Figure 3-1 Developmental stages of *O. rhinoceros*. Top left to right: eggs, first, second and third instars; bottom left to right: pre-pupa, pupa (images by Geoffrey Bedford printed with permission from the Annual Review of Entomology, Volume 58 © 2013 by Annual Reviews, http://www.annualreviews.org)

Egg

A freshly laid egg is minute and cylindrical and gradually absorbs moisture to become a rounded oval. Approaching the hatch date, the color sometimes changes from clear white to yellowish brown. The chorion appears to be tough with minute granulations on its surface (Gressitt, 1953). Figure 3-2 presents *O. rhinoceros* eggs and a first instar larva.

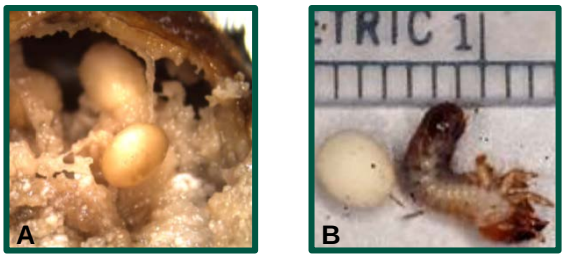

Figure 3-2 (A) Immature eggs inside a dissected female; (B) oviposited egg and first instar (photos courtesy of Aubrey Moore, University of Guam, and Arnold Hara, University of Hawaii)

Larva

The *O. rhinoceros* larva often curls into a crescent shape with the head touching the posterior end of the abdomen (Gressitt, 1953; Monty, 1978). A complete description of third instar larval morphology is available from Bedford (1974).

Garlovsky *et al.* (1971) and Gressitt (1953) have given an overview of the larval morphology. The head capsule is dark reddish brown and sclerotized with strong mouthparts including the mandibles and maxillae (Figure 3-3). The mouthparts project outward from the head in a rounded triangular fashion. The antennae are 4-segmented and located on the dorsum at the base of the mandibles. The head lacks ocelli. Although the thorax of the larva has 3 segments each with a pair of legs, the margins are not typically distinct with only the first thoracic segment possessing a pair of spiracles. The larva has 10 abdominal segments, only the first 8 of which contain spiracles. The exoskeleton is transparent and off-white. The posterior end of an actively feeding larva is darker, possibly indicating undigested food particles (Figure 3-3).

Figure 3-3 Third instar displaying sclerotized mouthparts and a dark abdomen (photo courtesy of Arnold Hara, University of Hawaii)

Instars

There are 3 instars, which can be differentiated by the size of their head capsules (Table 3-1, Figure 3-4). Due to weight loss prior to molting, the larval lengths (8–100 mm) overlap significantly rendering this measurement inappropriate for identification.

Table 3-1 Body measurements of *O. rhinoceros* at different developmental stages (Gressitt, 1953)

Developmental stage	Character	Measurements (mm)
egg	fresh-elongate (L × W)	3.5 × 2.3
	mature-round	4 × 3.7
first instar	head capsule width[1]	2.5–3.1
	body length	7.6–25
	body width	2.5–7
second instar	head capsule width[1]	5–6
	body length	22–65
	body width	6–12
third instar	head capsule width[1]	9.5–11.2
	body length	60–105
	body width	10–20
pupa	body length	39.4–51.5
	body width	19.0–23.6
adult	male length	30–57
	female length	29–51

[1] Diagnostic features used to distinguish the 3 instars

Immediately after hatching, the head capsule of the first instar appears whitish, becoming reddish brown during the first 24 h of inactivity. The width of the head can exceed that of the body. The epicranial suture is visible at the posterior occiput. The acute terminal segment of the antenna is longer than the other segments and has 15 sensory spots (Gressitt, 1953).

The second instar can be distinguished by head capsule width and sometimes body size (Table 3-1). Other identifying morphological features include the terminal segments of the antennae (shorter, less acute), the epicranial suture (more distinct), the node on the middle inner edge of the left mandible (distinctly tooth-like), the thoracic spiracle (broader, oval, non-distinct posterior margins, oval anterior-center tubercle) and rounder posterior abdominal spiracles (Gressitt, 1953).

The third instar can be distinguished from early instars using head capsule width (Table 3-1). In addition, the terminal antennal segment of this instar is short and blunt with 17 sensory spots; the mandibles have complex molar areas; the pronotum is large, reddish brown and heavily sclerotized with approximately 18 dorsal and 50 bilateral pronotal bristles near the spiracles. The fifth abdominal tergite has approximately 250 short bristles on each side. The spiracles are large, rounded and sclerotized. The fecal pellet of the larva is flat, elliptical and approximately 8 mm long. The larval appearance varies throughout development––shiny blue gray for the first 20 d and whitish for most of its stadium (Gressitt, 1953). Refer Bedford (1974) for detailed and most recent information on third instar morphology.

Figure 3-4 Head capsules of (A) first, (B) second, and (C) third instars of *O. rhinoceros* (photo courtesy of Aubrey Moore, University of Guam)

Pre-Pupa and Pupa

Prior to pupation, the larval body shrinks, and the body wall becomes more translucent. During this pre-pupal period, the body changes from whitish to creamy white. The pupa is yellowish brown, exarate, with a rubbery texture and characteristic odor. The sexes may be differentiated at this stage—the cephalic horns in males are approximately 3 times longer than wide, but only 1–2 times longer in females (Gressitt, 1953) (Figure 3-5).

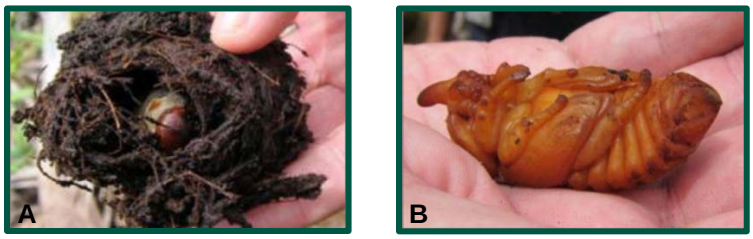

Figure 3-5 (A) Pre-pupa and (B) pupa of *O. rhinoceros* (photo courtesy of Arnold Hara, University of Hawaii)

Adult

Endrödi (1985) described morphological keys to distinguish adults of *O. rhinoceros* from other congeneric species. Illustrations and descriptions of adult morphology were also presented by Gressitt (1953). Adults have a convex reddish dark brown to black elytra and a heavily sclerotized body (Gressitt, 1953). Adults vary in size, largely depending on larval nutrition (Table 3-1).

The compound eye is large and partly masked by a flange extending from the cephalic horn. A cephalic horn is present and in males, the horn is 2.5–3 times longer than its base width, but shorter in females (Doane, 1913; Gressitt, 1953). The antenna is lamellate with 3 thick lobes forming the club (Figure 3-6A). The mouthparts are prominent and include the 4-segmented maxillary and 3-segmented labial palps (Gressitt, 1953).

The prothorax is typically one-third of the total body length and wider than long, tapering toward the concave anterior edge (Figure 3-6A). The thorax contains a single pair of spiracles located toward the dorsal edge of the prothoracic legs. The

scutellum is triangular and blunt at the base; the thoracic sternum has large coxal cavities. The elytra extend from the mesothorax and cover the remaining dorsum (Garlovsky *et al.*, 1971; Gressitt, 1953). Adults are strong fliers with flight muscles that occupy most of the thoracic cavity (Monty, 1978). The hind wings have 9 prominent veins extending close to the posterior wing margin. The elytra protect the hind wings and have a smooth humerus; typically, the elytra have 6 rows of non-distinct punctures apically along the suture (Figure 3-6B). All 3 pairs of legs are stout, with sharp tibial spines that are adapted for burrowing (Gressitt, 1953; Monty, 1978). The fore tibia are larger than the hind tibia (Garlovsky *et al.*, 1971).

There are 8 abdominal segments. Tergum of the first 6 segments are pale and smooth, while the seventh is heavily sclerotized for stridulation with the posterior elytra (Gressitt, 1953). The distribution of pleural abdominal spiracles is also a characteristic feature. The first 4 spiracles are large and elliptical, whereas the last 3 posterior pairs are smaller and broadly oval (Garlovsky *et al.*, 1971; Gressitt, 1953). In males, paramera are medium broad and narrows toward the wide apices (Endrödi, 1985). Mathur *et al.* (1960) described the internal and external genitalia of male and female *O. rhinoceros* specimens, and Jacob *et al.* (2008) further illustrated the structure and development of the male accessory sex glands. A ventral view of the thoracic segments and abdomen is presented in Figure 3-6C.

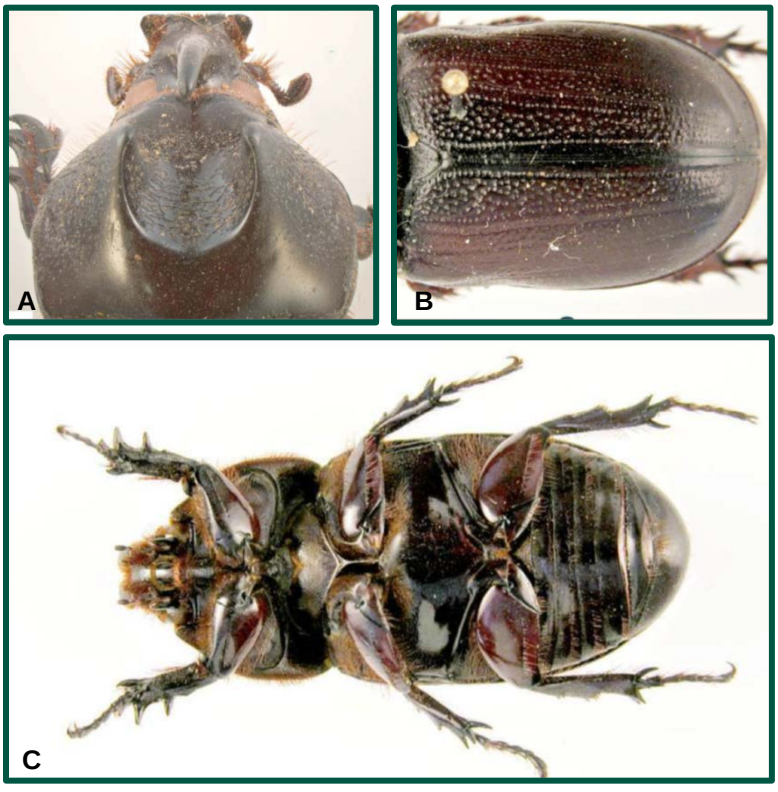

Figure 3-6 (A) Head with horn, antennae and fossorial forelegs, (B) punctures on elytra, (C) ventral view of the adult *O. rhinoceros* (photos courtesy of Pest and Diseases Image Library, Bugwood.org; numbers 5488589, 5488583, 5488582)

Males vs. Females

In adults, females have a blanket of long reddish erect hairs on the pointed pygidium, whereas in males, the pygidium is smooth, rounded and shiny with only a row of stout hairs along the ventral margin (Figure 3-7). Although male cephalic horns are typically longer than female, their size heavily depends on the environment and may not be reliable for differentiating between the sexes (Garlovsky *et al.*, 1971; Gressitt, 1953).

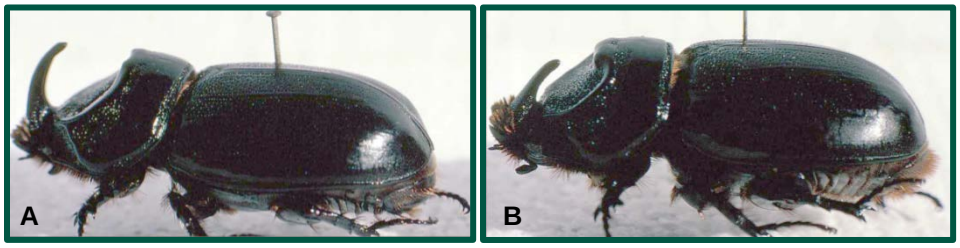

Figure 3-7 (A) Male and (B) female adult *O. rhinoceros* (images by Geoffrey Bedford printed with permission from the Annual Review of Entomology, Volume 58 © 2013 by Annual Reviews, http://www.annualreviews.org)

Young vs. Mature

Newly emerged adults lack abrasions on the elytra and often have pupal exuviae or meconium. The young adults generally co-exist with third instars and pupae and have a strong odor (Gressitt, 1953; Zelazny, 1975). Mature females commonly have abrasions on their elytra and are often associated with eggs, forming galleries in wood and exhibiting a weak musky odor (Gressitt, 1953; Zelazny, 1975). Zelazny and Neville (1972) provided an age–endocuticle layer relationship to determine the ages of young adults, typically within the first 32 d of eclosion.

Diagnostics

Early instars of *O. rhinoceros* could not be distinguished from early instars of other scarab larvae. Therefore, it is ideal to rear the larvae until the third instar before identification. Place young larva in a small container with 50:50 cowdung:rooted wood or sawdust with sufficient aeration until third instar, when it could be transferred to respective authority for identification (Bedford, 2014). Refer Bedford (1974) for the third instar morphology. Adults can be distinguished from congeneric species using the morphological keys by Endrödi (1985).

Similar Species

At present, 7 Oryctini species are found in North America: 5 *Strategus* spp. and 2 *Xyloryctes* spp. (Molet, 2014). Among the 39 species of *Oryctes* found globally, none are present in the contiguous United States; however, *O. rhinoceros* was recently reported in Hawaii (PestLens, 2014). Endrödi (1985) published detailed keys for the adults of 38 *Oryctes* spp. including *O. rhinoceros*.

Beaudoin-Ollivier *et al.* (1998, 2000) published simplified field keys to distinguish *O. rhinoceros* larvae from those of other species that feed on decaying organic matter. The study notes 2 characteristics that can identify the larva of *O. rhinoceros*—the presence of an impressed ring on the last abdominal segment and larvae that move on their side (Beaudoin-Ollivier *et al.*, 2000). However, these tips may not be useful in the case of congeneric species. Bedford (1974) has described a complete set of morphological characteristics for accurate differentiation of third instar *O. rhinoceros* from similar dynastid species.

Chapter

4

Biology

Life Cycle

The life cycle, beginning with breeding sites and extending to the adult stage, of the coconut rhinoceros beetle is summarized in Figure 4-1. The duration of each stage presented in the diagram represents an average from the studies cited in the respective sections.

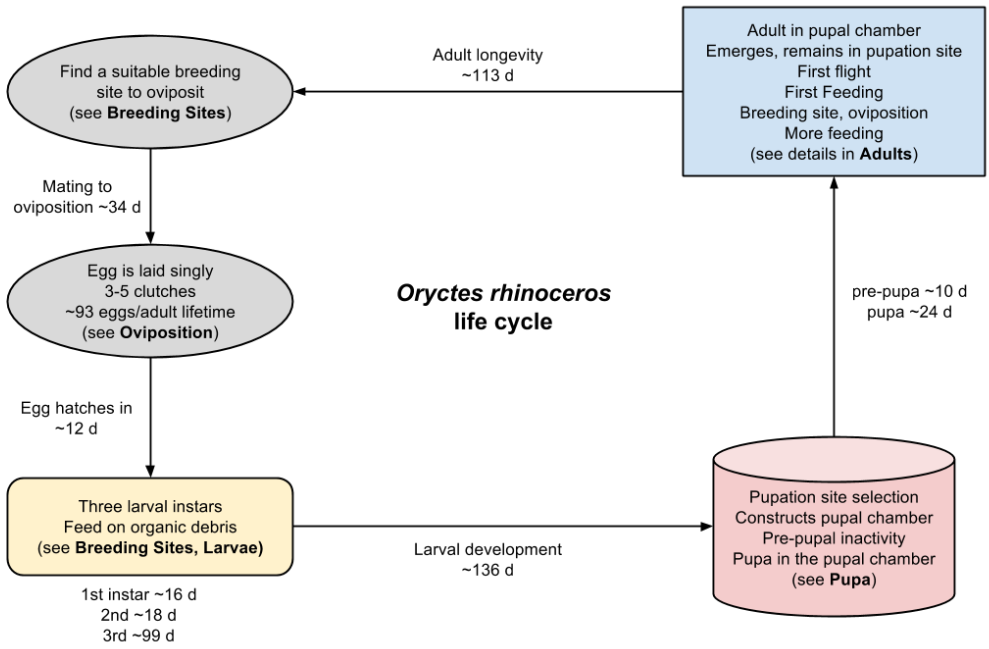

Figure 4-1 Life cycle of *O. rhinoceros*

Breeding Sites

Given the ability of *O. rhinoceros* to adapt to new breeding sites, the risk of impact is high upon introduction to a new location (Bedford, 2013). After finding a favorable breeding site, the female adult burrows into the substrate to lay eggs (Vargo, 1995). Dead standing palms, logs and stumps are the most preferred

breeding sites (Bedford, 1976a; Catley, 1969; Gressitt, 1953). Common breeding sites are presented in Table 4-1 and Figure 4-2.

Table 4-1 Breeding sites of coconut rhinoceros beetle

Breeding substrates	Reference
dead standing palms[1]	(Bedford, 1976a; Gressitt, 1953)
palm logs[1]	(Catley, 1969; Gressitt, 1953)
short coconut stumps[1]	(Gressitt, 1953)
cowdung[1]	(Lever, 1969)
compost[1]	(Gressitt, 1953)
sawdust pits or heaps[1]	(Zelazny, 1975)
oil palm mesocarp[1]	(Ponnamma *et al.*, 2001)
oil palm bunch refuse	(Jacob and Bhumannavar, 1991)
empty oil palm fruit bunches	(Wan Zaki *et al.*, 2009)
dried fronds and shredded palm wood refuse	(Bedford, 2013; Monty, 1978)
wood bark	(Jacob and Bhumannavar, 1991)
miscellaneous wood	(Gressitt, 1953)
soil underneath palm logs	(Gressitt, 1953)
papaya skin and taro refuse	(Gressitt, 1953)
humus from decaying cocoa pods	(Bedford, 1976a)
decaying coir refuse	(Cherian and Anantanarayanan, 1939)
sugarcane bagasse	(Cherian and Anantanarayanan, 1939)
sugarcane leaves preserved for silage	(Monty, 1978)
filter cakes from sugarcane processing plants	(Monty, 1978)
decaying rice straw heaps	(Jacob and Bhumannavar, 1991)
debris lodged near coconut fronds (rare)	(Moore, 2011, 2012a)
heaps of decaying organic matter	(Beaudoin-Ollivier *et al.*, 2000; Monty, 1978)

[1] Preferred breeding sites

The beetles are able to survive in forests with available live and felled hosts (Cumber, 1957; Gressitt, 1953). Other substrates favored by the larvae include the decaying wood of *Pandanus* spp. (screwpine), *Artocarpus* spp. (breadfruit), *Casuarina* spp., *Calophyllum inophyllum* L. (Alexandrian laurel), *Dictyosperma album* (Bory) Scheff. and *Mangifera* spp. (mango) (Gressitt, 1953; Monty, 1978).

Figure 4-2 Breeding substrates: (A) dead standing palm killed by *O. rhinoceros* in Tumon Bay, Guam; (B) cross-section exhibiting feeding activity; (C) larvae extracted from the substrate; (D) larvae inside rotten felled logs; (E, F) palm residue as a potential substrate; (G) potential substrate for arboreal development; (H) sawdust substrate; (I) potential breeding site from hurricane damage in Fiji (photos courtesy of Aubrey Moore, University of Guam [A, B, D, E, G, H]; Arnold Hara, University of Hawaii [F]; images by Geoffrey Bedford printed with permission from the Annual Review of Entomology, Volume 58 © 2013 by Annual Reviews, http://www.annualreviews.org [C, I])

Oviposition

On average, oviposition begins within a month of copulation (Gressitt, 1953; Hurpin and Fresneau, 1973; Sushil and Mukhtar, 2005; Zelazny, 1975). The egg stage lasts approximately 12 d with the typical female laying 4–5 eggs/d singly in the substrate; however, under favorable conditions females may lay 10–14 eggs/d (Bedford, 1976; Gressitt, 1953; Hinckley, 1973). Depending upon the adult size, eggs are laid in 3–5 clutches of 27–38 eggs/clutch with 20 d between clutches (Gressitt, 1953; Hinckley, 1973). The oviposition rate varies based on adult

longevity; adults lay 93 eggs on average in their lifetimes. Moreover, the oviposition location within a substrate can differ, including the fibrous apex of dead standing palms or under the loose bark of decaying coconut logs. In recently killed palms with no decay, the beetle may oviposit singly in small balls of shredded wood constructed during burrowing (Monty, 1978). Hinckley (1973) noted that the adult female chewed through the palm host in a serpentine fashion, ovipositing as it tunneled and subsequently compressing the shredded wood behind it to protect each egg. The number of eggs and reproductive days can vary significantly between individuals and may depend on adult longevity and oviposition substrate (Catley, 1969; Hurpin and Fresneau, 1973). Most adults continue to lay eggs up to 7–14 d prior to death (Hurpin and Fresneau, 1973). Males and females generally co-occur at the breeding sites. The females typically arrive first and lay some eggs; the males then arrive, remain longer at the site to chew and prepare more substrate for the emerging larvae (Zelazny and Alfiler, 1991).

Larvae

Instars

The 3 instars each molt following a brief period of inactivity. The larval development varies with the season and breeding medium (Bedford, 1976a; Catley, 1969; Cherian and Anantanarayanan, 1939; Gressitt, 1953; Indiravathi *et al.*, 2001; Monty, 1978; Sushil and Mukhtar, 2005; Vargo, 1995; Zhong *et al.*, 2013). The average development periods for the first, second and third instars were at 16, 18 and 99 d, respectively. The development period of the instars may overlap (Gressitt, 1953). In Malaysia, Wan Zaki *et al.* (2009) reported a predominant number of second (40 % of all stages) and third instars (36 %) co-existing in empty oil palm fruit bunches. At high insect densities, the instars and newly eclosed adults often co-exist at the breeding site (Gressitt, 1953).

Feeding Behavior

Immediately after hatching, the larva feeds on its egg chorion, frass or host remains left by the adults during tunneling and oviposition (Vargo, 1995). If the oviposition site was near the core of a decaying trunk, the larva feeds outward toward the bark (Gressitt, 1953). At high densities, the larvae can reduce the infested trunk to crumbled fibers, larval frass and decaying tissue (Monty, 1978). Larvae avoid extremely hard regions of the trunk with the population concentrated under the palm bark, at the decaying ends or in the center of the trunk where the tissue is softer (Bedford, 1976a; Gressitt, 1953). Although larvae typically avoid excessively damp wood, they can survive seawater submersion for more than 48 h (Gressitt, 1953; Nirula *et al.*, 1952).

In general, larvae feed on dead wood tissue and although studies have noted larval presence on wood near palm roots, no evidence indicated direct feeding on the root. Occasionally, during periods of high pest density, young larvae may exhibit cannibalistic behavior (Gressitt, 1953). If mortality factors are limited, a single dead standing trunk can hold more than 200 larvae (Monty, 1978). In breeding sites with low nutritional value and in dense logs, the mortality may approach 100 % before adulthood (Hinckley, 1973). Refer to the section on Breeding Sites for information regarding larval feeding.

Movement

Studies under natural conditions suggest that larvae exhibit negative phototaxis, possibly to avoid dessication and/or natural enemies (Bedford, 1980). Larvae are cryptic and hide in the breeding substrate until they develop into adults (Bedford, 2013). In Guam, a survey of the vertical distribution of adults and larvae in dead standing palms suggested that most individuals were found between 3.4 and 4.3 m from the base (Moore, 2011). The larvae can survive temperatures between 16 and 49 °C (Jacob and Bhumannavar, 1991; Nirula *et al.*, 1952). However, the optimal temperature for development is 27–29 °C with a relative humidity up to 85–95 % (Bedford, 1980). In a field study, Moore and Quitugua (2009) demonstrated that larvae and adults survived high temperatures (maximum between 40 and 59 °C) in rotting palm residue. In a preliminary laboratory study using steer manure, Moore (2014b) demonstrated a lethal temperature, LTe_{50}, of approximately 47 °C for third instars, with the compost heap temperatures reaching to 70 °C (Gressitt, 1953; Zimmermann, 1982). The larvae tend to avoid ‘hotspots’ in which temperatures exceed 37 °C, thus surviving unfavorable conditions (El-Shafie, 2014).

Unfavorable environmental conditions reduces larval size and prolongs development up to 420 d (Catley, 1969). Zelazny and Alfiler (1986) reported rainfall as a factor in larval mortality; mortality can also occur through desiccation, unavailability of a suitable pupation site or overcrowding (Cherian and Anantanarayanan, 1939). During active feeding, larval movement within a breeding substrate can be influenced by environmental factors. In a farmyard manure substrate, the larvae are typically found at 5–30 cm below the surface, where early instars often die if the substrate dries out, but third instars can burrow deeper into the moist soil to avoid desiccation (Cherian and Anantanarayanan, 1939). Larval activity and movement may also depend on the breeding substrate. For example, the larva typically moves only a few centimeters in solid wood, but movement is greater in sawdust or compost. The instars exhibit different mechanisms for movement on a flat surface: young larvae may use their thoracic legs, while mature larva typically

moves by contracting and relaxing body segments (Gressitt, 1953). After feeding, the larva finds a favorable site for pupation.

Pre-Pupa and Pupa

After feeding, the third instar searches for a suitable pupation site, preferably a compact location within the same substrate. However, if the current media is not firm, the larva moves to a different substrate. For example, if the breeding site is a decaying coconut log, the third instar might tunnel into a dense portion of the wood and construct a pupation chamber away from the hollow bark, rotten core or tips (Catley, 1969; Gressitt, 1953). Similarly, if the soil below is more compact than the breeding substrate, the larva burrows away from the current substrate and into the soil (Gressitt, 1953). In manure, the pupal chamber is typically found 15–30 cm deep in the soil near the breeding site (Cherian and Anantanarayanan, 1939). In sawdust, the pupal cell may occur deeper (1–1.5 m) below the surface (Catley, 1969; Gressitt, 1953; Vargo, 1995). In soil or other non-firm breeding substrates, *O. rhinoceros* constructs an ovoid pupal cell with thick walls (5.5 × 3.5 × 3.3 cm) using debris and liquid excretions (Gressitt, 1953; Lever, 1969). The pupating larva produces large quantities of liquid from its mouth and semi-solid excrement through its anal opening. After applying the liquid excrements, the larva turns inside the chamber to smooth the inside walls (Cherian and Anantanarayanan, 1939). The entrance of the pupal chamber is covered with loose substrate or frass (Gressitt, 1953). If no soil is available, the larva constructs irregular chambers using fibers and leaves (Monty, 1978). After preparing the chamber, the larva undergoes a brief inactive pre-pupal phase to prepare for pupation. The larvae then empties the alimentary canal, becoming flaccid and changing in appearance from off-white or bluish gray to creamy or pinkish white (Bedford, 1980; Catley, 1969; Gressitt, 1953; Monty, 1978). The pupa is exarate and is yellowish brown (Monty, 1978). The development period of the pre–pupa (~10 d) and pupa (~24 d) vary with location (Bedford, 1976a; Catley, 1969; Cherian and Anantanarayanan, 1939; Gressitt, 1953; Indiravathi *et al.*, 2001; Monty, 1978; Sushil and Mukhtar, 2005; Vargo, 1995; Zhong *et al.*, 2013).

Adults

Before First Flight

At eclosion to adulthood, the insect remains inside the pupal chamber for 10–24 d (Cherian and Anantanarayanan, 1939; Gressitt, 1953; Lever, 1969; Meer, 1987; Sushil and Mukhtar, 2005; Zelazny and Alfiler, 1987). After emergence from the pupal chamber, it remains at the site of pupation for 20–30 d (Jacob and Bhumannavar, 1991; Meer, 1987; Zelazny and Alfiler, 1987). Immediately after emergence, the adult is whitish, but completes pigmentation

within the next 24 h. The cuticle is gradually sclerotized (Gressitt, 1953; Zelazny, 1975).

First Flight

Both males and females fly within 20 d of adult emergence; the youngest adults found near the palm crowns are 20–30 d old (Zelazny, 1975).

First Feeding, Breeding and Later Feeding

Oryctes rhinoceros adults feed only after their first flight, even when food is readily available at their breeding site. A Philippine field study demonstrated that the adult beetles continued to feed for approximately 35 d at their first feeding site, which was sometimes followed by long-distance dispersion. After the first feeding, the adults proceeded to the breeding site where they remained for 32–49 d. After oviposition, the adults continued to visit additional host plants for a late-life feeding, which was shorter and lasted approximately 14 d (Meer, 1987; Zelazny and Alfiler, 1987, 1991).

Survival

Adults prefer a temperature between 28–30 °C and typically occur at elevations below 900 m (Gressitt, 1953; Hurpin and Fresneau, 1973). Females tend to survive longer than males (Bedford, 1976a; Hurpin and Fresneau, 1973). Without other mortality factors, adult longevity can be predicted using the adult weight at the time of eclosion from pupa: an adult dies when its body weight reaches approximately 40 % of its initial value (Meer, 1987). A lab study by Indiravathi (2001) reported that approximately 63 % of eggs and 87 % of larvae successfully developed into adults. Another study examined the biology of the beetle in manure pits and reported 83–91 % adult emergence from the pupal chamber (Sushil and Mukhtar, 2005). On average, the adult longevity observed in the various studies was approximately 113 d (Bedford, 1976a; Catley, 1969; Cumber, 1957; Gressitt, 1953; Hurpin and Fresneau, 1973; Indiravathi *et al.*, 2001; Jacob and Bhumannavar, 1991; Lever, 1969; Meer, 1987; Sushil and Mukhtar, 2005; Zelazny and Alfiler, 1987).

Movement

Adults fly between different sites for mating, oviposition and feeding. They are cryptic and during daylight prefer to stay in shaded regions at the feeding or breeding sites (Catley, 1969). Gressitt (1953) considered adults weak fliers, whereas Young (1986) described *O. rhinoceros* as excellent fliers. Most of the thoracic cavity is occupied by flight muscles, indicating their importance in the biology of the beetles (Monty, 1978). Females often have longer wings than males and may be are more robust fliers (Cumber, 1957). Adult flight

activity is primarily observed around dusk and dawn (Catley, 1969). Adults generally fly in a straight line and are sometimes unable to avoid obstacles in their paths (Monty, 1978). Flight activity decreases on moonlit nights or during heavy rains (Vargo, 1995). Kamarudin and Wahid (2004) noted that the flight activity increased in males during a full moon, which may be associated with a search for mating habitats. Although adults exhibit negative phototaxis—preferring shaded areas and avoiding light during the daytime, they may fly toward light at night (Gressitt, 1953; Monty, 1978; Stride, 1977). Reported flight distances vary. The beetle is thought to prefer short flights, but is capable of long flights if local conditions are unfavorable (Catley, 1969). A lab study demonstrated that palm-fed tethered adult beetles had a flight potential of 2–3 h, covering the equivalent of 2–4 km (Hinckley, 1973). Reports of long distance flight by *O. rhinoceros* include adults flying toward light on a ship anchored 700 m from shore (Catley, 1969), marked adults recaptured at 900 m within 3 d and approximately 1600 m within a month (Cumber, 1957). Kamarudin and Wahid (2004) used mark-release-recapture studies to determine the flight range of *O. rhinoceros* in oil palm replanting regions in Malaysia; their results suggested that the adults moved at the rate of 10–23 m/day and up to 1.3 km/week. However, the flight distance may depend on environmental conditions and the abundance of breeding and feeding sites. Flight may occur at different times depending on location. In Samoa, captures of adults in flight were highest from February–June, whereas in southern India, adult beetles were abundant from March–April (Cherian and Anantanarayanan, 1939; Cumber, 1957). Adult activity is primarily observed near live coconut crowns (Cumber, 1957).

Mating

Mating primarily occurs at night after the first flight and feeding. Dead coconut trunks and other breeding substrates are the reported mating sites for the beetle (Cumber, 1957; Zelazny, 1975). Even though it is possible that copulation may occur near feeding locations including palm crowns and leaf axils, there are no peer-reviewed reports for these events to date (Bedford, 2013, 2014). Multiple matings (~8) may occur in the field, but are not essential because the female can store sperm in a spermatheca after a single mating. The sperm is typically viable for 4–6 months (Catley, 1969; Hurpin and Fresneau, 1973). The male to female ratio is typically 1:1 (Indiravathi *et al.*, 2001), but may sometimes vary from 1:0.65 to 1:3.27 (Al-Habshi *et al.*, 2006; Bedford, 1975; Gressitt, 1953; Hinckley, 1973; Jacob and Bhumannavar, 1991; Sushil and Mukhtar, 2005; Zhong *et al.*, 2013).

Total Lifespan

The lifespan of the beetle appears to depend on substrate availability and seasonal variations at different locations (Cherian and Anantanarayanan, 1939; Gressitt, 1953; Hinckley, 1973; Jacob and Bhumannavar, 1991; Lever, 1969; Sushil and Mukhtar, 2005). The total lifespan generally ranges from 4–10 months, allowing for more than 1 generation/year. In Palau, which is tropical without distinct seasons, the lifespan was approximately 7 months, and under favorable conditions, the insect may have up to 3 generations per year (Gressitt, 1953). In India and China, the lifespan of *O. rhinoceros* varied from 3–9 months (Cherian and Anantanarayanan, 1939; Gressitt, 1953). In Samoa, Hinckley (1973) observed that under favorable conditions, the egg-to-egg generation time was below 5 months.

Hosts

Oryctes rhinoceros adults reportedly feed on approximately 51 plant species from 10 families (Table 4-3). Thirty-seven of the reported host species belong to the palm family, Arecaceae. The coconut palm is the preferred host, followed by oil palms and date palms (Catley, 1969; Gressitt, 1953).

A list of plant hosts that were reported only under laboratory conditions are presented in Table 4-4. The scientific name, synonyms and common names for each plant host were retrieved from the following databases—Plants Database (USDA and NRCS, 2014), Catalogue of Life (Roskov *et al.*, 2014), Crop Protection Compendium (CABI, 2014a), and The Plant List (2013).

Table 4-3 Reported plant hosts of *O. rhinoceros*

Plant host	Common name	References
Agavaceae		
Agave spp.		Cherian and Anantanarayanan (1939)
Agave americana L.	American century plant	Gressitt (1953)
Agave sisalana Perrine	sisal hemp	Gressitt (1953) Chong *et al.* (1991)
Aloaceae		
Aloe spp.	aloe	Sivakumar and Mohan (2013)
Araceae		
Colocasia spp.	colocasia	Gressitt (1953)
Alocasia spp.	taro	Gressitt (1953)
Cyrtosperma spp.		Gressitt (1953)
Xanthosoma spp.	elephant's ear	Gressitt (1953)
Aracaceae		
Acanthophoenix rubra (Bory) H. Wendl.	barbel palm	Gressitt (1953)
Aiphanes horrida (Jacq.) Burret (= *A. caryotifolia*)	ruffle palm, coyure palm	Gressitt (1953)
Areca spp.		Lever (1969)
Areca catechu L. (= *A. cathecu*)[1]	betel palm	Nirula *et al.* (1952) Gressitt (1953)
Arenga spp.		Lever (1969)
Arenga pinnata (Wurmb) Merr.[1]	sugar palm	Gressitt (1953)
Borassus spp.		Lever (1969)
Borassus flabellifer L.	toddy palm, palmyra palm	Nirula *et al.* (1952), Gressitt (1953)
Caryota urens L.	jaggery palm	Gressitt (1953)
Clinostigma samoense H. Wendl. (= *Cyphokentia samoensis*)		Gressitt (1953)
Cocos nucifera L.[1]	coconut	Bedford (1980), Gressitt (1953), Lever (1969), Nirula *et al.* (1952)
Corypha spp.		Lever (1969)
Corypha umbraculifera L.	talipot palm	Cherian and Anantanarayanan (1939), Nirula *et al.* (1952)
Corypha utan Lam. (= *C. elata*)	gebang palm, serdang palm	Gressitt (1953)
Dictyosperma album (Bory) Scheff.	hurricane palm, red palm	Gressitt (1953)
Dypsis pinnatifrons Mart. (= *D. gracilis*)	dypsis palm	Gressitt (1953)
Elaeis spp.[1]	oil palm	Chong *et al.* (1991), Kamarudin and Wahid (1997) Lever (1969)
Elaeis guineensis Jacq.[1]	African oil palm	Gressitt (1953), Hoyt (1963), Bedford (1980), Sullivan *et al.* (2013)
Heterospathe elata var. *palauensis* (Becc.) Becc.		Gressitt (1953)
Hydriastele palauensis (Becc.) W.J. Baker & Loo (= *Gulubiopsis palauensis*)		Gressitt (1953)
Latania spp.		Gressitt (1953)

Livistona spp.		Lever (1969)
Livistona chinensis (Jacq.) R.Br. ex Mart.	fountain palm, latanier palm	Gressitt (1953) Bedford (1980), Monty (1978)
Hyophorbe lagenicaulis (L.H. Bailey) H.E. Moore (= *Mascarena lagenicaulis*)	bottle palm	Gressitt (1953)
Metroxylon spp.		Lever (1969)
Metroxylon amicarum (H. Wendl.) Hook.f. (= *Coelococcus carolinensis*)	caroline ivory nutpalm	Gressitt (1953)
Metroxylon sagu Rottb.	sago palm	Gressitt (1953)
Metroxylon vitiense (H. Wendl.) Hook.f.		Pacific Islands Pest List Database (2009)
Normanbya normanbyi (W. Hill) L.H. Bailey	black palm	Gressitt (1953)
Nypa spp.		Lever (1969)
Nypa fruticans Wurmb	nipa palm	Gressitt (1953) Nirula *et al.* (1952)
Oncosperma spp.		Gressitt (1953)
Oncosperma tigillarium (Jack) Ridl.	niblong palm	Nirula *et al.* (1952)
Phoenix spp.		Lever (1969)
Phoenix dactylifera L.	date palm	Gressitt (1953), El-Shafie (2014)
Phoenix sylvestris (L.) Roxb.	wild date palm	Gressitt (1953), Nirula *et al.* (1952)
Pinanga insignis Becc. (= *Pseudopinanga insignis*)		Gressitt (1953)
Pinanga spp.		Gressitt (1953)
Pritchardia pacifica Seem. & H. Wendl.	Fiji fan palm	Gressitt (1953)
Raphia farinifera (Gaertn.) Hyl. (= *R. ruffia*)[1]	raffia palm	Bedford (1980), Monty (1978) Hoyt (1963)
Raphia vinifera P. Beauv.	West African piassava palm	Gressitt (1953)
Roystonea regia (Kunth) O.F. Cook (= *R. elata, Oreodoxa regia*)	royal palm	Gressitt (1953), Bedford (1980)
Stevensonia spp.		Gressitt (1953)
Syagrus romanzoffiana (Cham.) Glassman (= *Arecastrum plumosa*)	queen palm	Gressitt (1953)
Thrinax spp. (thatch palm)		Gressitt (1953)
Verschaffeltia splendida H. Wendl.	Latanier Latte	Gressitt (1953), Monty (1978)
Wodyetia bifurcata A.K. Irvine	foxtail palm	USDA-APHIS EPICA (2009)
Bromeliaceae		
Ananas comosus (L.) Merr.	pineapple	Nirula *et al.* (1952), Gressitt (1953), Chong *et al.* (1991)
Caricaceae		
Carica papaya L.	papaya	Catley (1969), Chong *et al.* (1991)
Cyatheaceae		
Cyathea spp.	treefern	Gressitt (1953)
Liliaceae		
Musa spp.	banana	Gressitt (1953), Sharma and Gupta (1988), Chong *et al.* (1991), Sivakumar and Mohan (2013)
Pandanaceae		

Pandanus spp.		Gressitt (1953), Lever (1969)
Pandanus tectorius Parkinson ex Du Roi	Tahitian screwpine	Gressitt (1953)
Poaceae		
Saccharum spp.	sugarcane	Gressitt (1953), Chong *et al.* (1991), Sivakumar and Mohan (2013)
Sterculiaceae		
Theobroma cacao L.	cacao	Pacific Islands Pest List Database (2009)

[1] Preferred host

Table 4-4 Plant hosts of *O. rhinoceros* reported only under laboratory conditions (Gressitt, 1953)

Plant host	Common name
Colocasia esculenta (L.) Schott	coco yam
Alocasia macrorrhizos (L.)	giant taro
Cyrtosperma merkusii (Hassk.) Schott (= *C. chamissonis*)	swamp taro
Xanthosoma sagittifolium (L.) Schott	arrowleaf elephant's ear
Tradescantia spathacea Sw. (= *Rhoeo discolor*)	boatlily
Hanguana malayana (Jack) Merr.	
Persea americana Mill.	avocado
Cordyline fruticosa (L.) A. Chev. (= *C. terminalis*)	tiplant
Dracaena angustifolia (Medik.) Roxb.	
Hymenocallis littoralis (Jacq.) Salisb.	beach spiderlily
Tacca leontopetaloides (L.) Kuntze	batflower

Other hosts are not attacked if coconut palms are abundant, depending on the developmental stage of the host (Cherian and Anantanarayanan, 1939). For example, *O. rhinoceros* adults do not damage *Pandanus tectorius* Parkinson ex Du Roi if mature coconut palms are available; however, adults prefer *P. tectorius* to young coconut palms.

Although *Lantana* spp. is listed as a host in the Crop Protection Compendium and in publications citing this reference (CABI, 2014), no supporting information confirms this association; possibly confused with *Latania* spp., a host of *O. rhinoceros* in Palau (Gressitt, 1953). Further, *Casuarina equisetifolia* L. has been erroneously reported as a minor adult host, but the original citation includes the plant as a larval host (Elfers, 1988). For information regarding larval hosts, refer to the section on Breeding Sites on page 4-1.

Dispersal

Active flight

Adults can fly long distances under adverse conditions, but likely will not if breeding and feeding sites are available at the location of origin. The highest flight activity is observed around dusk and dawn (Catley, 1969). The beetle is thought to prefer short flights, but is capable of long flights if local conditions are unfavorable (Catley, 1969). In a field study,

Kamarudin and Wahid (2004) demonstrated that the adults moved at the rate of 10–23 m/day and up to 1.3 km/week. Further, a lab study demonstrated that palm-fed tethered adult beetles had a flight potential of 2–3 h, covering the equivalent of 2–4 km (Hinckley, 1973). Other reports of long distance flight by *O. rhinoceros* include adults flying toward light on a ship anchored 700 m from shore (Catley, 1969), marked adults recaptured at 900 m within 3 d and approximately 1600 m within a month (Cumber, 1957). Flight may occur at different times depending on location. In Samoa, the largest adults were captured in flight from February–June, whereas in southern India, adult beetles were abundant from March–April (Cherian and Anantanarayanan, 1939; Cumber, 1957).

Natural movement of breeding substrates

Coconut palms grow along ocean shores in many locations. After infestation, some decaying palm logs may travel short distances through the sea. Larvae may survive inside the infested substrate and reach other locations aided by the ocean currents (Gressitt, 1953; Lever, 1969).

Human-assisted spread

Cargo such as timber, sawdust and copra are suitable substrates for the larvae; ships carrying infested materials can introduce *O. rhinoceros* to new locations (Gressitt, 1953; Stride, 1977). However, port interceptions and previous reports indicate that the most likely method of introduction occurs through adults that hitchhike aboard ships and flights. Early coconut rhinoceros beetle invasions in the Pacific islands possibly occurred through sea and air traffic during WWII (Catley, 1969; Nishida and Evenhuis, 2000). The beetles are active fliers at night, and containers loaded after sunset are more likely to have hitchhiking adults than those loaded during the daytime. Regulatory personnel have found beetles in empty pallets on shipments from Guam to the mainland (CRB TWG, 2014).

After introduction, movement or availability of the substrates can rapidly spread the beetles to uninfested locations (Gressitt, 1953; Guaminsects.net, 2007b; Sweeney, 2008). In Oman, the percentage of infestation doubled when infested cattle manure was transported to meet the demands of increasing banana cultivation (Kinawy, 2004). In addition to the unintentional movement of infested substrates, the beetles may have been deliberately moved due to their perceived potential for nutrition, collection and cultural amusements (Fakayode and Ugwumba, 2013; New, 2005; Okaraonye and Ikewuchi, 2009; Onyeike *et al.*, 2005; Ratcliffe, 2006). A lack of public awareness may be a key factor in the spread of this insect (Ridgell, 2009).

Geographic Distribution

Ecological Distribution

Oryctes rhinoceros is endemic to south and southeast Asia including Bangladesh, China, India, Sri Lanka, Taiwan, Indonesia, Malaysia, the Philippines and Thailand (Table 4-5). Although the exact origin of the pest is unknown, the earliest reports are in the 1890s' from southern India and Malaysia (Alam, 1975; Cherian and Anantanarayanan, 1939; Gressitt, 1953; Nirula *et al.*, 1952). The pest is suspected to have spread from Malaysia to southern Myanmar, then further north. In Polynesia, the insect was likely introduced to Samoa from Sri Lanka through the import of rubber seedlings. The pest further spread to American Samoa, Niue, Keppel Island, Vava'u and Tonga, Wallis, and Tokelau. The beetle was eradicated from Tonga in the 1930s, but was re-introduced during WWII (Catley, 1969; Pacific Islands Pest List Database, 2009). In Africa, the beetle is present in Mauritius and Réunion (Catley, 1969; Lever, 1969; Monty, 1978; Nirula *et al.*, 1952). Although Nirula *et al.* (1952) and Hoyt (1963) published *O. rhinoceros* in Burundi, Rwanda, Tanganyika, Sierra Leone and Nigeria, it is more likely that these reports are a mistaken identification for the endemic *O. monoceros* (Bedford, 2014; Gressitt, 1953). In Micronesia, the pest is reported in Palau, Guam and Saipan. In the U.S., the only report of *O. rhinoceros* was in Hawaii on 23 December 2013 in Oahu at the Joint Base Pearl Harbor-Hickam (JBPHH). Initially it was contained within a 3 km radius of the first detection, but adults were recently found 0.8 km outside and west to the 3 km radius (Hawaii Department of Agriculture, 2014; Hawaii Invasive Species Council, 2014a, 2014b). The worldwide distribution of *O. rhinoceros* and presented in Figure 4-3.

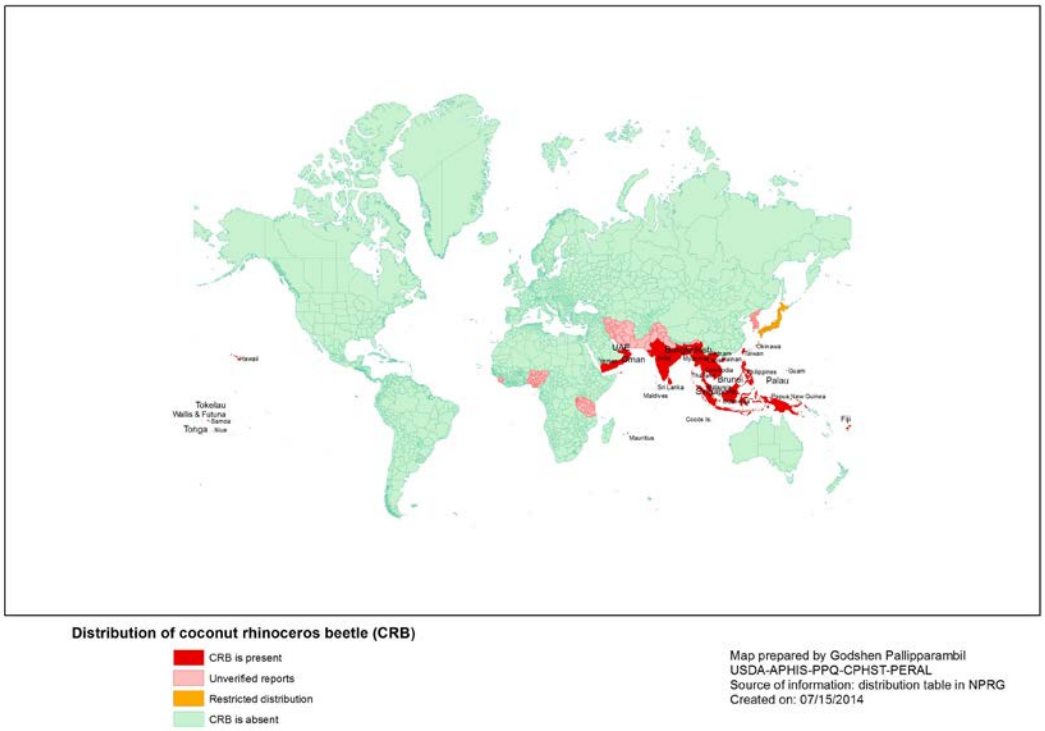

Figure 4-3 Worldwide distribution of *O. rhinoceros*. Labeled locations have confirmed presence of the beetle

Table 4-5 Worldwide distribution of *O. rhinoceros*

Location	Specific locations (if available)	References
Africa		
Burundi, Rwanda, and Tanganyika[3]		Nirula *et al.* (1952)
Sierra Leone[3]		Hoyt (1963)
Mauritius		Catley (1969), Lever (1969)
Nigeria[3]		Hoyt (1963)
Réunion		Monty (1978)
Asia		
the Andaman and Nicobar islands		Catley (1969), Jacob and Bhumannavar (1991) GBIF (2014)
Bangladesh		Alam (1975)
Brunei		Waterhouse (1993)
Cambodia		Waterhouse (1993)
China[1]	Hainan Province	Gressitt (1953), Lin *et al.* (2010)
Cocos (Keeling)		Catley (1969)
Diego Garcia[2]		Catley (1969)
Hong Kong[1]		Gressitt (1953)
India[1]	Kerala, Karnataka, Tamil Nadu, West Bengal, Maharashtra, Orissa, Madhya Pradesh, Assam, Rajasthan, Manipur, Goa, Nagaland, Andhra Pradesh, Gujarat, Bihar, Jharkhand, and Tripura	Cherian and Anantanarayanan (1939), Nirula *et al.* (1952), Bhatnagar (1971), Gope and Prasad (1983), Raju (1983), Sharma and Gupta (1988), Dhileepan (1991), Singh and Gandhi (2010), Coconut Development Board (2013)
Indonesia[1]	Pekalongan, Irian Jaya, Kalimantan, Maluku, Nusa Tenggara, Sulawesi, Sumatra, Mattirobulu, Kabupaten Pinrang, central Java, West Java Regency, Moluccas, Bangka and Mt. Dempo	Muir and Swezey (1916), Gressitt (1953), PQR EPPO (2013), Daud (2007), GBIF (2014)
Iran		Bedford (2013), PQR EPPO (2013)
Japan	Ryukyu[1], Yaeyama, Okinawa	PQR EPPO (2013), Gressitt (1953), Hosoya (2011), GBIF (2014)
Korea		Gressitt (1953), Endrödi (1985)
Lakshadweep		Mohan and Pillai (1993b)
Laos		Waterhouse (1993)
Malaysia[1]		Gressitt (1953), Waterhouse (1993), Darus and Basri (2000)
the Maldives		Zelazny *et al.* (1990)
Myanmar[1]		Nirula *et al.* (1952), Gressitt (1953), Bedford (1980)
Oman		Kinawy (2004)

Pakistan[3]	western Pakistan	Gressitt (1953), Crawford (1981)
the Philippines[1]		Nirula *et al.* (1952), Gressitt (1953), Zelazny and Alfiler (1987), GBIF (2014)
Singapore		Cherian and Anantanarayanan (1939), GBIF (2014)
Sri Lanka[1]		Nirula *et al.* (1952), Gressitt (1953), GBIF (2014)
Taiwan[1]		Gressitt (1953), GBIF (2014)
Thailand[1]		Gressitt (1953) GBIF (2014)
United Arab Emirates[2]		Gassouma (2004)
Vietnam[1]		Bedford (1980) Waterhouse (1993)
Yemen		Al-Habshi *et al.* (2006), El-Shafie (2014)
North America		
Hawaii[2]	First detected on 23 December 2013 on Oahu at the Joint Base Pearl Harbor-Hickam contained within a 3 km radius of first detection adults found 0.8 km outside and west to the 3 km radius	Hawaii Department of Agriculture (2014b) Hawaii Invasive Species Council (2014b) Hawaii Invasive Species Council (2014c)
Oceania		
Fiji[2]		Catley (1969), Gressitt (1953), Bedford (1980), Young (1986)
Guam[2]		USDA-APHIS EPICA (2007), Sweeney (2008), Moore (2012a)
Niue[2]		Dharmaraju (1980)
Palau[2]		Gressitt (1953), Catley (1969), Muniappan (2002)
Papua New Guinea[2]		Gressitt (1953), Bedford (1976a)
Saipan[2]		USDA-APHIS EPICA (2010)
Samoa[2]		Bedford (1980), Cumber (1957), Catley (1969)
American Samoa[2]		Monty (1978), Catley (1969), Pacific Islands Pest List Database (2009)
Tokara[2]		Hosoya (2011)
Tokelau[2]		Catley (1969), Uili (1980)
Tonga[2]		Catley (1969)
Wallis[2]		Cohic (1950)

[1] presumed native; [2] introduced; [3] unverified

Potential Distribution

Among other factors, the distribution of *O. rhinoceros* depends on adult host availability, breeding substrate abundance and favorable abiotic factors. The optimum temperature for an adult is between 28 and 30 °C, with a preliminary study indicating the lethal temperature for the third instar at approximately 47 °C (Gressitt, 1953; Hawaii Invasive Species Council, 2014c; Moore, 2014b). The larvae favor high relative humidity, preferably 85–95 % (Bedford, 1980). Although little information regarding the topography and elevation is available in the literature, Gressitt (1953) indicated that the pest is not typically reported at altitudes above 900 m. A map depicting the potential distribution of the coconut rhinoceros beetle was constructed for important palm hosts in the contiguous U.S. (Figure 4-4). The known distribution of the coconut rhinoceros beetle indicates that its distribution is more host-limited than climate-limited; therefore, climatic parameters were not included in the map preparation. Because no reliable host acreage data was available, data on the presence or absence of coconut and oil palm were collected at a county level using the Biota of North America Program, BONAP database (Christie, 2014; Kartesz, 2013).

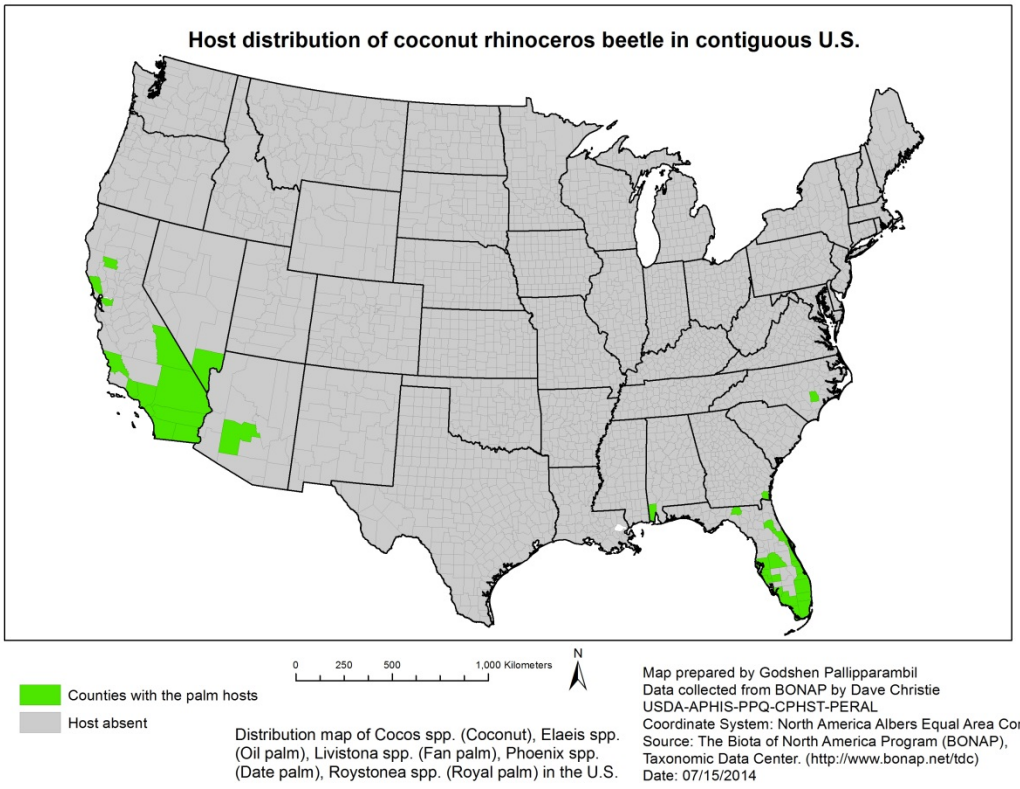

Figure 4-4 Potential distribution of *O. rhinoceros* based on host availability

Chapter

5

Damage

Signs and Symptoms

Direct Damage

Visible Symptoms

As the damaged fronds unfold, a distinct ‘V’ or wedge-shaped cut, characteristic of an *O. rhinoceros* attack—becomes visible. This cut is caused by angular tunneling through the spadix that does not damage the rachis (Figure 5-1D–G). Damage to the rachis tip may cause the terminal portion of the frond to hang or break (Gressitt, 1953; Monty, 1978). During tunneling, fibrous tissue is typically pushed outward from the palm trunk and may be visible at the entrance hole (Figure 5-1A–C) (Giblin-Davis *et al.*, 2001). Entrance holes are approximately 2.5 cm in diameter, and the tunnels are 5–60 cm long; the entrance holes on the petioles and trunk may be visible from the ground depending upon the site and time of attack (Figure 5-1H–I) (Cherian and Anantanarayanan, 1939; Gressitt, 1953; Monty, 1978; Young, 1975).

Figure 5-1 Signs and symptoms of *O. rhinoceros* damage on coconut palms (photos G, H courtesy of Aubrey Moore, University of Guam; Arnold Hara, University of Hawaii [B, C, D, E, I]; images A, F by Geoffrey Bedford posted with permission from the Annual Review of Entomology, Volume 58 © 2013 by Annual Reviews, http://www.annualreviews.org)

Impact on the Host

The damaged rachis may break off during strong winds resulting in frond fall and reducing the area available for photosynthesis. Although a moderate attack may delay or reduce fruit production, a severe attack can cause the shedding of all fronds in a crown, which gradually kills the palm (El-Shafie, 2014; Lever, 1969; Young, 1986). The dead standing palms eventually become a breeding substrate for the beetles (Figure 5-2). An attack on the spathe, inflorescence or immature nuts may cause an early nut fall, whereas a more severe attack directly impacts the yield by destroying the inflorescence (Giblin-Davis *et al.*, 2001; Sullivan *et al.*, 2013). The palm is also killed if the beetle completely destroys the terminal growing point (Garlovsky *et al.*, 1971).

Figure 5-2 Coconut palms killed by *O. rhinoceros* in Fiji (image by Geoffrey Bedford, posted with permission from the Annual Review of Entomology, Volume 58 © 2013 by Annual Reviews, http://www.annualreviews.org)

The extent of palm damage may vary with location, insect density, structure and host maturity. The impact of a beetle attack depends on the structure of the palm crown. For example, the number of cuts may depend on the spacing between fronds, whereas the distance between the frond axils and growing tip may influence either the probability of palm mortality or the damage to the inflorescence. Furthermore, the spear growth, position of the developing inflorescence and the position of beetle attack all contribute to the palm damage (Young, 1975). Adults prefer to attack mature coconut palms, although younger palms are sometimes damaged (Gressitt, 1953). In Papua New Guinea, most damage occurred palms older than 3 years (Bedford, 1976a). An adult attack can be tolerated to an extent if the palm is mature and healthy, but repeated attacks can still lead to palm death (Monty, 1978). Insect attack is more severe for 1–3-year-old palms (Giblin-Davis *et al.*, 2001) in which the growing apices are readily impacted leading to palm death (Young, 1986). The dead standing palms remaining after a beetle attack act as breeding sites and further facilitate the proliferation of *O. rhinoceros*. In many cases, palms that are isolated or situated at the edge of a plantation are more exposed to beetle attack than other palms—this preference may be related to the insects' flight behavior. Adult beetles fly in a straight line and sometimes are unable to avoid obstacles (Monty, 1978). Cumber (1957) notes that, on average, an adult beetle visits 3–4 palms during its lifetime. In a later study, Meer (1987) indicated that adults may have approximately 7 flying events toward old or new hosts, but that the feeding duration on each host could vary. Favorable temperatures and rainfall also promote *O. rhinoceros* outbreaks (Jacob and Bhumannavar, 1991). If insect populations are low, the beetle

damage may not be deadly to the palm but can still affect its aesthetic value. A healthy palm can produce approximately 1 frond/month, which, after unfolding, is resistant to beetle attack. In low insect densities, feeding may occur after long intervals allowing some palm fronds to evade attack (Gressitt, 1953).

Hosts other than the Coconut Palm

Although the *O. rhinoceros* damage reported herein is based on the most studied host, the coconut palm, the signs and symptoms are similar in other palm hosts. For example, in oil palms, the spears and unfolded fronds are attacked, and the crown may appear twisted (Dhileepan, 1992; Gressitt, 1953). Moslim *et al.* (1999) indicated that a severe infestation can kill young oil palms. Damage to the young palms can prolong the immature stage of the tree and therefore cause significant economic loss during the early phase of crop production. If a beetle attack during the first 2 years of planting causes a 15 % reduction in leaf area, up to 25 % of the crop can be lost (Darus and Basri, 2000; Moslim *et al.*, 1999). The attack on sugarcane is different: the beetle enters the stem at ground level and tunnels upward (Gressitt, 1953).

Indirect Damage

An *O. rhinoceros* attack may lead to secondary infestations by other pests. For example, a secondary infestation by palm weevils, *Rhynchophorus* spp., can prove far more dangerous than the direct damage by *O. rhinoceros* (Bedford, 2013; Catley, 1969; Giblin-Davis *et al.*, 2001; Manjeri *et al.*, 2014). Cherian and Anantanarayanan (1939) noted that the reverse is true in most cases, *i.e.*, trees attacked by the palm weevil are more attractive to the rhinoceros beetle, a finding that agrees with an observation by Gressitt (1953) in which multiple rhinoceros beetles infested the same palm when other healthy palms were available in the neighborhood, tending to aggregate on the damaged palm. However, this early study ignored the involvement of host attractants or insect aggregation pheromones. Furthermore, Sivakumar (2001) indicates that *O. rhinoceros* is only attracted to the palms infested with *Rhynchophorus ferrugineus* Herbst once decay begins. Rainwater collecting in the excavated tunnels may also indirectly damage the palm, resulting in rotting (Cherian and Anantanarayanan, 1939; Gressitt, 1953).

Economic Impact

Impact on Yield

In both its endemic and introduced habitats, the coconut rhinoceros beetle primarily threatens coconut and oil palms (Cherian and Anantanarayanan, 1939; Chong *et al.*, 1991; Gressitt, 1953; Kamarudin and Wahid, 1997). A beetle attack reduces the individual palm yield, and severe infestations can significantly damage entire plantations (Catley, 1969; Gressitt, 1953). A reduction in coconut yield is of particular concern where nuts are used for both consumption and the production of copra, oil and other byproducts (Bedford, 1980; Catley, 1969; Smith and Moore, 2008).

Trade

More than 85 countries cultivate coconut with Indonesia, the Philippines, India, Brazil and Sri Lanka as the top five. In 2010, global coconut production was approximately 62.5 million metric tons (Marikkar and Madurapperuma, 2011). The second-most preferred host, the oil palm, is cultivated by approximately 43 countries among which Malaysia and Indonesia are the 2 major producers, together constituting approximately 87 % of global production (Punnuri and Singh, 2013). Information regarding the import and export of coconut and oil palm products in the U.S. is outlined in Table 5-1. Refer to the host information on page 4-10 for comprehensive information regarding other hosts, and refer to Table 5-2 for the impact of *O. rhinoceros* in some locations.

Table 5-1 Coconut and oil palm product imports and exports in United States in 2011 (FAOSTAT, 2014)

Product	Import quantity (tons)	Import value ($1,000)	Export quantity (tons)	Export value ($1,000)
coconut	34,919	28,503	3,084	3,156
desiccated coconut	43,853	124,444	2,929	4,244
copra, oil	498,278	926,591	36,144	42,131
palm kernel oil	321,583	577,605	11,138	16,420
palm oil	1,087,626	1,281,840	94,906	107,491

The trade information in Table 5-1 indicates that the U.S. is not a major exporter of coconut and palm oil, but is an important consumer. Furthermore, the U.S. is the largest importer of desiccated coconut and coconut oil (FAOSTAT, 2014; USDA-FAS, 2014). Therefore, a coconut rhinoceros beetle introduction might decrease domestic production of these commodities requiring additional imports. The European and Mediterranean Plant Protection Organization (EPPO) lists *O. rhinoceros* as an A1 quarantine pest in the Caribbean, Central America, Brazil and the Pacific (PQR EPPO, 2013).

Cost of Control and Domestic Regulations

The cost of removing mature infested palms and replanting is high. Pest management costs are also high due to the phytosanitary measures, pheromone trapping, chemical control and the release of entomopathogens. Furthermore, prophylactic phytosanitary measures and regular monitoring via traps are essential to limit the spread of the beetle even in regions currently devoid of the pest but with the potential for introduction—these measures add to production costs (Bedford, 1980; Catley, 1969; Moslim *et al.*, 2013). An establishment of the pest triggers interstate regulations and international quarantine restrictions, further adding to the cost of an invasion (Campbell, 2011; Smith and Moore, 2008; USDA-APHIS, 2014b).

Socio-Cultural Impact and Aesthetics

In addition to their commercial value as cash crops, palms can be an integral part of a location's identity and culture: Palms may add to the aesthetics of a business, increase property values and promote tourism. The destruction of palms by *O. rhinoceros* could negatively impact these attributes, perceptions and damage otherwise lucrative businesses (Campbell, 2011; Smith and Moore, 2008; USDA-APHIS, 2014b).

Table 5-2 Damage and economic impact of *O. rhinoceros*

Locations	Economic impact	References
Guam	damage to businesses and tourism; expensive removal and replanting (current estimate for replanting mature palm US$ 1,000, replacing lost trees in Tumon may cost US$ 2.5 million; additional expenses for quarantine restrictions; impact on small-scale household businesses that depend on coconut products; potentially high impact on culinary use and copra exports	Campbell (2011) Smith and Moore (2008)
India	10 % yield loss due to spathe damage	Catley (1969)
	major pest of coconut and important but relatively minor pest of oil palm; in southern India, the percentage of damage in coconut palms ranged 0.3–64 %; in oil palms, the incidence below 1.5–20 %	Dhileepan (1992)
	in Kerala leaf damage varied 7.7–15.4 %, based on the coconut cultivar	Muthiah and Bhaskaran (2000)
	crown damage in ~50 % of palms in the Andamans	Jacob and Bhumannavar (1991)
Malaysia	major pest of coconut and oil palm; ~25 % of oil palms attacked	Chong *et al.* (1991), Kamarudin and Wahid (1997)
	most severe damage to oil palms during second and third year of planting; almost no bunches if highly damaged	Oehlschlager (2005)
	~67 % damage in 1–2-year-old tissue-cultured oil palms	Ahmad (2006)
Pacific Islands	major pest of coconut in Palau; 50 % of palms were killed within 10 y of introduction;	Gressitt (1953)

	biological control was not used 1968 annual estimate indicated approximately US$ 1 million impact on South Pacific islands	Catley (1969)

Environmental Impact

The environmental impact of an *O. rhinoceros* introduction could derive from direct damage by the beetle or control measures implemented to manage the invasion.

Direct Impact of the Beetle

The coconut rhinoceros beetle is a destructive pest of mature coconut palms in the agricultural, residential and native forest ecosystems (Cumber, 1957; Gressitt, 1953). In an agroecosystem, the beetle may variably impact the palms and alter the age structures within plantations (Campbell, 2011). In native forests, the self-perpetuating life cycle of *O. rhinoceros* is likely to affect the diversity and distribution of flora by selectively targeting host species (Cumber, 1957; Gressitt, 1953; USDA-APHIS, 2014b). The decayed breeding substrates and hollowed-out palm trunks are favorite habitats for several organisms; therefore, the infestation could influence faunal diversity (Gressitt, 1953; Hinckley, 1967). In some locations, coconut palms are planted along beaches to reduce erosion; because coastlines provide ready targets for beetle attacks, an infestation could increase the rate of soil erosion (Campbell, 2011; Gressitt, 1953). In the U.S., an introduction poses a risk to approximately 400 endangered or threatened species (Pimentel *et al.*, 2001). The introduction of *O. rhinoceros* may damage several protected plants, a couple of which are known insect hosts with a few others that are congeneric to the known hosts and therefore potential hosts (USDA-NRCS, 2014; USFWS, 2014). Refer to Table 5-3 for a list of potential plant hosts with federal protected status.

Table 5-3 Threatened and endangered plant species that are potential adult coconut rhinoceros beetle hosts (USDA-NRCS, 2014; USFWS, 2014)

Potential plant host	Federal protected status
Agave eggersiana Trel. (Eggers' century plant)	proposed endangered
Agave × *arizonica* Gentry & J.Z. Weber	endangered
Pritchardia affinis Becc. (Hawaii pritchardia)	endangered
Pritchardia aylmer-robinsonii H. St. John (Nihoa pritchardia)	endangered
Pritchardia hardyi Rock (Makaleha pritchardia)	endangered
Pritchardia kaalae Rock (Waianae Range pritchardia)	endangered
Pritchardia munroi Rock (Kamalo pritchardia)	endangered
Pritchardia napaliensis H. St. John (Nihoa pritchardia)	endangered
Pritchardia remota Becc. (Nihoa pritchardia)	endangered
Pritchardia schattaueri Hodel (lands-of-papa pritchardia)	endangered
Pritchardia viscosa Rock (stickybud pritchardia)	endangered

Impact of Control Measures

Phytosanitary measures including burning can pollute the environment; therefore, Malaysia has zero-tolerance for any strategy that involves burning (Moslim *et al.*, 2011a; Moslim *et al.*, 2005b). Furthermore, several insecticides used for management and eradication pose environmental and health concerns; for details, refer to the section on Chemical Control on page 8-36.

Chapter

6

Survey Procedures

Survey Types

After the first detection, regulatory officials estimate the core-infestation area and create buffer zones for the delimiting surveys. If the pest is established, regular monitoring surveys are conducted to determine the impact of the pest and evaluate the management strategies.

Preparations, Sanitization and Clean-Up

This section provides information to aid personnel in preparing to conduct a survey, procedures to follow during a survey and instructions for proper cleaning and sanitizing of supplies and equipment after the survey is complete.

1. Prior to beginning a survey, determine whether recent pesticide applications might render the inspection of plants and leaf litter unsafe. Contact the property owner or manager and ask if a re-entry period is in effect due to pesticide application. Look for posted signs indicating recent pesticide applications, particularly in commercial fields or nurseries.

2. Conduct the survey at the proper time. General surveys should focus on months in which host plants are easily accessible and in active growing phases.

3. Obtain permission from the landowner prior to entering a property.

4. Determine whether quarantines for other pests or crops are in effect for the survey area. Comply with any and all quarantine requirements.

5. When visiting the area to conduct surveys or obtain samples, strict measures must be taken to prevent contamination by any pests between properties.

6. Prior to entering a new property, ensure that clothing and footwear are clean and free of pests, soil and litter to avoid transferring soil-borne pests and arthropods between properties.

7. Wash hands with approved antimicrobial soap. If not using an antimicrobial soap, wash hands with regular soap and warm water to remove soil and debris. Then, use an alcohol-based antimicrobial lotion with an equivalent of 60% ethyl alcohol. If hands are free of soil or dirt, the lotion can be applied without washing. Unlike some antimicrobial soaps, antimicrobial lotions are less likely to irritate the hands and thereby improve compliance with hand hygiene recommendations.

8. Gather all supplies. Confirm that equipment and tools are clean. When taking plant samples, disinfect tools with bleach to avoid spreading diseases or other pests. A brief spray or immersion of the cutting portion of the tool in a 5% solution of sodium hypochlorite (bleach) is effective for inactivating bacteria and other diseases, thus preventing their spread.

9. Flag the plant, tree or sampled location whenever possible and draw a map of the immediate area indicating reference points so that the areas can be found again if necessary. Do not rely solely on flags or other markers to re-locate a site as they may be removed. Record the GPS coordinates for each trap or infested tree so that the area or plant may be re-sampled if necessary.

10. Survey task forces should consist of an experienced survey specialist or entomologist familiar with *O. rhinoceros* and the symptoms of its damage.

Detection Survey

The purpose of a detection survey is to determine if a pest is present in a defined area. A detection survey can be broad in scope to assess the presence of a pest or multiple pests over large areas or restricted to determine if a specific pest or pests are present in a focused area.

Statistically, a detection survey is not a valid tool to claim that a pest does not exist in an area, even if results are negative. Negative results can be used to provide clues regarding mode of dispersal, temporal occurrence or industry practices. Negative results are also important when compared with results from similar sites.

Procedure

Use the following procedure when conducting a detection survey for *O. rhinoceros*:

1. Focus the survey on high-risk areas where *O. rhinoceros* is more likely to occur. Refer to the section on potential distribution.
2. Inspect potential breeding sites and hosts as described in the techniques section on page 6-3.
3. If presence of the pest is suspected on a host, collect the larvae or adults to confirm its identity.

4. Submit the specimens to the proper authority. Refer to How to Submit Insect Specimens on page X for further information. Morphological characteristics that may aid in the identification of *O. rhinoceros* adult females are described on pages 3-1 through 3-7.
5. Establish regular sites to inspect *O. rhinoceros* along your normal surveying route.

Techniques for detection

A detection survey investigates the presence of a pest within a broad or focused area of interest using the following techniques:

Inspect potential breeding sites: Refer to Breeding Sites on page 4-1 for substrates suitable for *O. rhinoceros* larval development. In addition to the larvae, these sites also harbor other stages of the beetle. Refer to the Identification chapter on page 3-1 for morphological characteristics.

Visually inspect for host damage: Frond damage and borer holes in the trunk are characteristic of the presence of feeding adults. At some locations, sentinel logs were strategically placed to detect the beetles. For more information, refer to the Damage chapter on page 5-1 and the Palm Damage section on page 6-8. If the damage is minor and the palms are tall, the attack signature on the host may not be easily detected (Gressitt, 1953). In addition to trained researchers, visual inspection is also crowd sourced with the aid of multimedia, pamphlets and other online resources: Trifold pamphlets were issued in Hawaii with information regarding the identification, biology and damage of the coconut rhinoceros beetle. (Hawaii Department of Agriculture, 2014d; Moore, 2012b, 2014d, 2014g; Smith and Moore, 2008).

Install pheromone traps: Ethyl (S)-4-methyloctanoate (E4-MO) is an aggregation pheromone produced by the male *O. rhinoceros* (Hallett *et al.*, 1995) and is widely used to trap adults of both sexes. For further information regarding the traps and their placement, refer to the Traps section on page 6-8 and the Pheromone Traps section on page 8-25.

Other attractants: Artificial traps using food substrates, light traps and a non-pheromone attractant ethyl chrysanthemumate (now superseded by E4-MO) are also utilized to detect the presence of adults. Refer to Non-Pheromone Traps on page 8-24 for further information.

Acoustic detection: The chewing, scraping, movement and tunneling activities of the larvae and adults in wood substrates produce distinct temporal and spectral acoustic patterns that can be detected using vibration sensors attached to the substrate, which allows the surveyor to detect the cryptic *O. rhinoceros* without obliterating or dissecting the suspected host (Mankin *et al.*, 2011; Mankin and Moore, 2010; Moore-Linn, 2009).

Furthermore, adult beetles can stridulate by rubbing the elytra and abdominal tergite and these characteristic stridulations vary with age, sex, courtship, aggression and distress (Laartech, 2004; Mankin *et al.*, 2009; Mini and Prabhu, 1990). Accoustic detection is not feasible after *O. rhinoceros* establishment (Bedford, 2014).

Detector dogs: During beetle eradication efforts in Guam, dogs were trained to detect the *O. rhinoceros* larvae. The dogs were equipped with GPS and tracked to monitor new survey sites. More than 350 new breeding sites were discovered using this method (Moore, 2012a; Quitugua, 2010).

The two most prominent detection methods are the CAPS-approved survey methods: visual inspection of host damage (by scanning fronds) and trapping using the aggregation pheromone (Bedford, 2014; Mankin *et al.*, 2009); specific information is available in the CPHST pest datasheet for the coconut rhinoceros beetle (Molet, 2014). To report the pest on the mainland, inform the local USDA-APHIS office using the 'www.hungrypests.com/what-you-can-do/' website and the 'Report a Pest' dropdown menu. For Hawaii, a pest hotline number and email is available to report any signs or symptoms—(808) 679-5244, or e-mail stoprhino@gmail.com (USDA-APHIS, 2014).

Delimiting Survey after Initial United States Detection

After detection, the source of the infestation is determined and an area is delimited for additional surveys to investigate the extent of infestation and to take necessary measures to prevent the spread to nearby uninfested areas. Using the sampling results, buffer zones for the delimited area are extended if necessary.

Procedure

Follow the same procedures used in the Detection Survey on pages **Error! Bookmark not defined.**2 to 6-4. Additional surveys should continue in nearby areas to determine the full extent of the infestation. Other palm species should also be inspected; for example, survey royal palms in the vicinity for damage (Bedford, 2014). Inspections should encompass larger areas particularly where hosts are known to occur. Once *O. rhinoceros* has been confirmed, surveys should be most intensive around the known positive detections and any discovered through trace-back and trace-forward investigations.

Two cases of delimited surveys:

In Guam

The beetle was first detected in Tumon Bay in September 2007 (USDA-APHIS EPICA, 2007). After detection, a delimiting survey by the Guam Department of Agriculture and the University of Guam suggested that the infestation extended only to Tumon Bay and Fai Fai Beach. Therefore, an eradication zone of 1,360

acres was delimited at this location; however, a larger quarantined area including 5,830 acres outside the eradication zone was designated to consider potential spreading. Delimiting traps were spread along Guam roadsides at the rate of 1 trap/1,340 acres, a rate below the mass-trapping control rates of 1 trap/acre. (Guaminsects.net, 2007a; Smith and Moore, 2008). The quarantine area was later expanded to 28,362 acres (Campbell, 2011). In Guam, new trap designs for pheromone lures are being investigated to increase the number of captures. For further information regarding the traps and their designs, refer to the Pheromone Traps section on page 8-25.

In Hawaii

An adult was first detected on Oahu on 23 December 2013 near the Joint Base Pearl Harbor-Hickam; 2 weeks later, a severely infested mulch pile was identified nearby Hickam's Mamala Bay Golf Course. A few adults were subsequently discovered in adjacent traps, which led to the formation of an Incident Command System (ICS) involving the USDA-PPQ, the Hawaii Department of Agriculture, the University of Hawaii at Manoa and the Hawaii Department of Land and Natural Resources; the objective was to prevent the spread of the rhinoceros beetle and to coordinate eradication procedures. After discovery of the core-infestation area, a 3.2-km delimiting buffer zone was established around the reported spots for intensive monitoring. Although the method is similar to mass trapping, delimitation trapping is intended to survey for the beetles allowing for a reduced trap density. The desired rate is 64 traps/1.6 km, and at present, approximately 280 traps are deployed in and around the infested area; visual inspections were also conducted within the delimited zone (Hawaii Department of Agriculture, 2014c; Hawaii Invasive Species Council, 2014b; USDA-APHIS, 2014b). Although most reports were within the buffer zone, on 21 May 2014, a male adult was detected in a panel pheromone trap in Barbers Point outside the zone, extending the buffer zone to the west and increasing the delimited area by 14.5 km^2 (Hawaii Department of Agriculture, 2014a). As part of the eradication effort, approximately 66,000 palms and 150 breeding sites had been surveyed as of 28 May 2014, and approximately 1,200 panel traps set throughout Oahu to monitor the incidence. The newly developed pheromone traps coupled with solar-powered UV LEDs are more effective than the traditional pheromone traps and were deployed on poles or suspended from non-host branches (USDA-APHIS, 2014b). Refer to the section on Pheromone Traps on page 8-25 for additional details. As of 28 May 2014, approximately 520 larvae, 16 pupae and 360 adults were discovered at the breeding sites and in traps (Hawaii Department of Agriculture, 2014a). A current map of the reported pest locations and buffer zones is available from the Hawaii Department of Agriculture website (USDA, 2014). The most recent map is included in Figure 6-1.

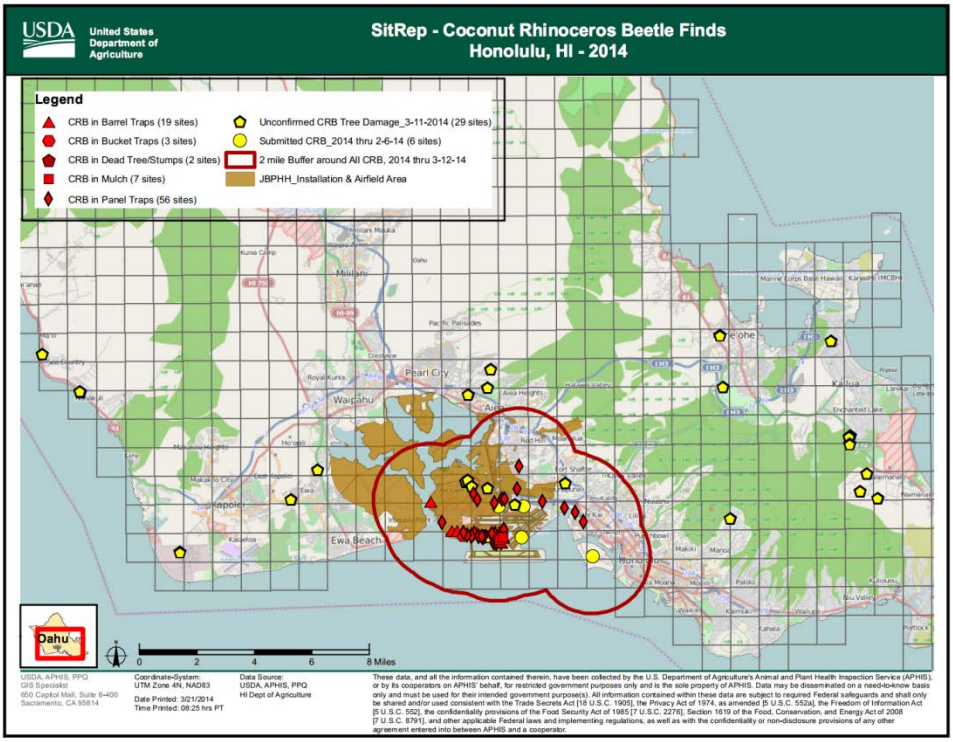

Figure 6-1 Hawaiian detection sites of *O. rhinoceros* adults and larvae. The red border separates the quarantined buffer zone for delimited surveys.

Trace-Back and Trace-Forward Investigations

Trace-back and trace-forward investigations aid in prioritizing delimiting survey activities after an initial detection. Trace-back investigations attempt to determine the source of the infestation. Once a positive detection is confirmed, efforts should be made to determine the extent of the infestation or find potentially infested areas in which to conduct further investigations. Trace-forward investigations attempt to define further potential dissemination through different routes as described in pages 4-10 to 4-11.

Homeowner Properties

For positive detections on homeowner properties, ask the owner of the infested material about the time of first detection, potential origin of infestation and any possible sites of further distribution.

Nursery Properties

As noted in the pathway section on page 10-1, the main risk of *O. rhinoceros* introduction is through hitchhicking adults. Nursery stocks are not known to be potential sources of introduction; however, the procedure in this section may be

useful if larvae are detected in breeding sources associated with nursery stocks or if adults hitchhike or infest on nursery plants.

For nursery hosts, a list of facilities associated with potentially infested nursery stock will be compiled. These lists will be distributed by the state to the field offices and are not to be shared with individuals outside the USDA-APHIS-PPQ regulatory cooperators. Grower names and field locations on these lists are strictly confidential, and any distribution of the lists beyond the appropriate regulatory agencies is prohibited.

Each state is only authorized to see locations within their state, and the sharing of confidential business information may be restricted between state and federal entities. Check the privacy laws with the State Plant Health Director for that state.

When notifying growers on the list, be sure to identify yourself as a USDA or state regulatory official conducting an investigation of facilities that may have received *O. rhinoceros* -infested material. Speak to the growers or farm managers and obtain proper permission prior to entering private property.

Several actions should occur immediately upon confirmation that a nursery host was infested with *O. rhinoceros*:

- Check nursery records to obtain names and addresses for all sales or distribution sites (if any sales or distribution has occurred from infested nursery during the previous 6 months).
- Evaluate the pest situation, including identification and inspection of the infested plant and the location within the nursery.
- Check nursery records to identify potential sources of the infestation inside or outside the nursery.
- Note the time of first detection and subsequent reports.

Analyzing Information

Use trace-back information gathered from the surveys and interactions to determine the origin of infestation. With timely submitted records from landowners and growers prioritized lists for further surveys can be prepared.

Monitoring Survey

Conduct a monitoring survey if you have applied a control procedure and need to measure its effectiveness. If *O. rhinoceros* is detected in the U.S., a technical working

group will be assembled to provide guidance on using a monitoring survey to measure the effectiveness of applied treatments on the pest. Refer to Control Procedures on page **Error! Bookmark not defined.** for further information regarding control option.

Procedure

All methods used in the detection and delimitation surveys can be used for monitoring. However, a monitoring survey focusses on monitoring the movement and seasonal dynamics. Because monitoring survey investigates the density and spread of the pest, the sample size would be much larger than that is required for a detection survey. Two important survey techniques—visual inspection of plant damage and the placement of traps—are briefly discussed and examples provided in this section.

Traps

The aggregation pheromone produced by males can attract adults of both sexes and may be helpful to study population dynamics of *O. rhinoceros*. In endemic locations, the typical density of pheromone traps is trap/2 ha (Bedford, 2014). Kamarudin and Wahid (2004) monitored the dynamics and movement of *O. rhinoceros* in and near a target oil palm plantation in Selangor, Malaysia. The survey used 49 pheromone traps spaced at 27 × 45 m to study the movement of *O. rhinoceros* in and out of the oil palm replanting areas; the study area was bordered by mature oil palm plantations. The traps were placed in 3 tiers: the first outer tier was at the fringes, the second tier was just inside the replanting area, and the third tier—noted as the ‘core’—was further inside. The distances between the tiers were approximately equal. Trapping was initiated approximately 5 months after the ageing oil palms were felled, and all traps were placed at a height of 1.5 m. Lures were replaced every 6 weeks with monitoring continuing for 2 years. The study noted that female adults immigrated into the replanting blocks much earlier and more frequently than males, indicating an active search for breeding sites. The core region was infested within 4–7 months of the logging of old palms (Norman and Mohd Basri, 2004). Although pheromone traps provide an index of the *O. rhinoceros* population in the vicinity, no studies have related the proportion of catches to the actual insect densities in the field. Therefore, the results from monitoring pheromone traps should be interpreted with caution (Bedford, 2013, 2014).

Palm Damage

Zelazny and Alfiler (1987) noted that the number of catches did not truly represent the *O. rhinoceros* population when ethylchrysanthemumate baits were used. However, the position of damage on the palm fronds may correlate with the number of adults feeding at a location. Based on the age of the palm, the number of fronds produced per year can be determined. For example, in 1-year-old palms

as few as 8.5 fronds may be produced per year, whereas mature palms produce approximately 16 fronds/year. For a majority of their lifetime, palms have 25–40 fronds in their crown (Young, 1975). Because the new fronds emerge every 3–4 weeks, the position of a frond in the crown of a coconut palm can aid in determining the age of each frond. The damage to the frond may vary depending on the type and time of attack; the damage severity and frequency can be used to estimate the population dynamics of the feeding adults on a plantation (Young, 1975; Zelazny and Alfiler, 1987). For more detailed sampling procedures, see the review by Bedford (2013).

Table 6-1 provides a general outline of the survey techniques used at some locations. The techniques vary somewhat with the location, the availability of resources and the time of publication.

Table 6-1 Survey techniques for *O. rhinoceros*

Location	Summary	References
Guam	traps with pheromone and food substrates; detection dogs; preliminary studies using acoustics	Mankin and Moore (2010), Mankin *et al.* (2009), Moore (2012a)
Hawaii	solar-powered UV LED pheromone traps, serviced every 1–2 weeks; crowd sourcing information using social networks and citizen scientists; visual inspection of breeding sites and the palm hosts	Hawaii Department of Agriculture (2014a, 2014b, 2014c, 2014d), Hawaii Invasive Species Council (2014a, 2014b, 2014c, 2014d), USDA-APHIS (2014a)
India	aggregation pheromone, weekly trap counts May–February on coconut plantations	Bhanu *et al.* (2012)
Thailand	November–May in aromatic coconut; coconut fronds and breeding sites examined	Thai Agricultural Standard (2008)
Yemen	year-round monitoring using light traps at 1-km distance; weekly monitoring; significant numbers noted in March, gradually increasing and peaking in June with drastic decrease after September	Al-Habshi *et al.* (2006)

Sentinel Sites

Sentinel sites are regularly inspected locations along a surveyor's normal route. The sites can be established using a known host plant. The plant used as a sentinel site should be visually inspected for signs or symptoms of damage; if possible, the host plant should be evaluated. A Global Positioning System (GPS) device should be utilized to record the location of the host plant, and draw a map of the immediate area drawn including reference points to allow others to easily find the area if necessary. Once established, the surveyor should re-inspect the sentinel site on a regular basis (bimonthly or monthly) as permitted by the individual's regular survey schedule. A geographic information system (GIS) can be used to map the sentinel sites to ensure even coverage, particularly in high-risk areas.

Procedure

At the time of publication, a defined method for *O. rhinoceros* was unavailable.

Other Pests

Other pests can cause similar symptoms on their host plants and may have similar morphological features. Refer to the identification section on page 3-1 for more information.

Targeted Surveys

Regular targeted surveys must be conducted in areas with regular traffic from countries with known infestations.

Procedure

At the time of publication, a defined method for *O. rhinoceros* was unavailable.

Survey Records

Records should be maintained for each survey site. Survey records and data recording formats should be consistent to standardize the collection of information. Repeat surveys are recommended at the same location or palm to record the chronological progress or reduction of *O. rhinoceros* damage at a specific site (Bedford, 2014).

If automated field collection devices are used, such as the Integrated Survey Information System (ISIS), ensure that all surveyors are trained in the technology before beginning the survey. Use the appropriate ISIS templates for this pest. To reduce the burden on field data collectors, enter any known contact or address information into the database and hand-held data recorders prior to field operations. At the survey's conclusion, all survey data should be entered into a designated state or national pest database.

Data Collection

Surveyors visiting sites to place holds or take samples should collect the following information:

- Date of survey
- Collector's name and affiliation
- Grower's or landowners field identification numbers if available
- Full name of business, institution, or agency
- Full mailing address including country
- Type of property (commercial nursery, hotel, natural field, residence)
- GPS coordinates of the host plant and property
- Host plant species and cultivar
- Presence or absence of the pest or symptoms
- Evidence of existing or previous infestations
- General conditions or any other relevant information

In the absence of inspection officials, take the following actions immediately if symptoms are noted:

1. Mark the location
2. Collect evidence of infestation and flag the specific sample locations within the field
3. Notify the state or PPQ inspector
4. Place the larvae or adults of *O. rhinoceros* inside secure plastic containers
5. Label the sealed containers with the following information:
    a. Date
    b. Name of person responsible
    c. Location of sample collection
6. Keep samples cool or refrigerated until the inspector arrives
7. Do not freeze the sample

Cooperation with Other Surveys

Other surveyors regularly sent to the field should be trained to recognize outbreaks that could be associated with *O rhinoceros*.

Chapter

7

Regulatory Procedures

Chapter

8

Control Procedures

Overview of Emergency Programs

Plant Protection and Quarantine (PPQ) develops control measures and makes them available to involved states. Environmental Protection Agency (EPA)-approved treatments will be recommended when available. If selected treatments are not labeled for use against the organism or in a particular environment, PPQ's FIFRA (Federal Insecticide, Fungicide and Rodenticide Act) coordinator is available to explore the appropriateness of an emergency exemption under section 18, or a State Special Local Need under section 24(c) of FIFRA as amended. The PPQ FIFRA coordinator and pesticide-use coordinators are also available upon request to work with the EPA to expedite approval of a product that may not be registered in the United States, or to obtain labeling for a new use. Refer to the Resources on page A-1 for information on contacting the coordinator.

Treatment Options

Treatments may include the following:

- Biological Control on page 8-1
- Cultural Control and Sanitary Measures on page 8-24
- Chemical Control on page 8-36
- Host Resistance on page 8-42
- Integrated Pest Management on page 8-42

Biological Control

Biological control and cultural control are useful for managing the coconut rhinoceros beetle. Using new biological control agents may not be plausible for an eradication program given the expected delays in approval, introduction and establishment of a biological control agent. Some applications of these agents are similar to synthetic insecticides with formulations that immediately impact the pest.

Predators and Parasitoids

A number of predators and parasitoids reportedly attack *O. rhinoceros*; however, only a few species are viable biological control agents (Table 8-1).

Table 8-1 Natural arthropod enemies introduced as biological control agents against *O. rhinoceros*

Predator	Host stage attacked	Country of origin	Notes	Reference
Alaus speciosus L. (Coleoptera: Elateridae)	all larval instars	Sri Lanka	established in Samoa	Catley (1969); Cumber (1957)
Catascopus facialis (Wiedemann) (Coleoptera: Carabidae)	larva		did not establish in Samoa	Gressitt (1953)
Hypoaspis rhinocerotis Oudemans (Acari)	egg			Khanjani *et al.* (2013)
Lanelater fuscipes L. (Coleoptera: Elateridae)	egg, all larval instars		established in Samoa; no impact in Mauritius	Catley (1969) Monty (1978)
Neochryopus savagei (Hope) (Coleoptera: Carabidae)	adults	Nigeria	did not establish in Fiji or New Guinea	Catley (1969) Bedford (1980)
Pachylister chinensis Quensel (Coleoptera: Histeridae)	first instar	Samoa	established in Palau	Gressitt (1953)
Pheropsophus sobrinus (Dejean) (Coleoptera: Carabidae)	larva	Kerala, India		Catley (1969)
Pheropsophus spp. (Coleoptera: Carabidae)		India	established, but no impact in Mauritius	Monty (1978), Bedford (1980)
Platymeris laevicollis Distant (Hemiptera: Reduviidae)	adults	Zanzibar Malaysia India Sri Lanka	established in Samoa and Solomon Islands did not establish in Mauritius,New Guinea, Tonga	Catley (1969) Lever (1969), Monty (1978), Bedford (1980)
Scarites madagascariensis Dejean (Coleoptera: Carabidae)	larva	Madagascar	did not establish in Mauritius	Surany (1960) Monty (1978)
Parasitoids				
Elis romandi de Saussure (Hymenoptera: Scoliidae)	larva	Madagascar		Lever (1969)
Scolia oryctophaga Coquillett (Hymenoptera: Scoliidae)	larva	Madagascar	did not establish in the Pacific	Gressitt (1953), Monty (1978), Bedford (1980)
Scolia patricialis	larva	Singapore	did not establish in	Gressitt (1953)

Burmeister (Hymenoptera: Scoliidae)			Palau	
Scolia procer Illiger (Hymenoptera: Scoliidae)	larva	Malaysia	did not establish in Palau or Mauritius	Gressitt (1953), Monty (1978)
Scolia ruficornis F. (Hymenoptera: Scoliidae)	larva	Zanzibar	established in Samoa did not establish in Palau or Diego Garcia	Gressitt (1953), Catley (1969), Lever (1969), Bedford (1980)

The aforementioned scoliid wasps are larval ectoparasitoids of *O. rhinoceros* and have been studied extensively as biological control agents (Surany, 1960). For successful development, the scoliids require a cool period during the pupal stage and heavy rain during adult emergence. Dry weather is unfavorable (Gressitt, 1953). *Scolia ruficornis* is unable to enter hard wood, and its activity is restricted to decomposing and easily accessible friable breeding substrates (Catley, 1969). Furthermore, the parasitoids are extremely susceptible to insecticides; therefore, chemical control is not recommended at the release sites (Lever, 1969).

Several species of ants, carabids, click beetles and histerid beetles feed on various life stages of *O. rhinoceros* (Cherian and Anantanarayanan, 1939; Gressitt, 1953). Some species of mites (*Hypoaspis* spp., *Macrocheles* sp., *Uropoda* sp.) are also recorded on *Oryctes* spp. (Cherian and Anantanarayanan, 1939; Jacob, 2000; Khanjani *et al.*, 2013). The impact of several of these predators remains unknown rendering them poor choices for biological control (Bedford, 1976a). In addition to the arthropods, several animals and birds feed on the coconut rhinoceros beetle, but these predators are not significant sources of mortality (Cherian and Anantanarayanan, 1939; Gressitt, 1953). Surany (1960) found that the control status of the coconut rhinoceros beetle in Southeast Asia cannot be attributed to its parasitoids and predators. Most of the predators and parasitoids introduced into the Pacific likely perished by the early 1970s (Young, 1986). Stiling (1993) notes that biological control campaigns in Samoa and Mauritius failed due to predation, climate and the habitat preferences of the released agents.

Nematodes

A few nematode species are associated with the various stages of *O. rhinoceros*, but their host–parasite relationships are not well studied (Catley, 1969). *Oryctonema genitalis* n. gen., n. sp., is associated with the male aedeagus and female bursa copulatrix of *Oryctes* spp. (Bedford, 1980; Catley, 1969). Another nematode, *Rhabditis adenobia* sp. n., was reported from the colleterial glands of female *Oryctes* spp. (Bedford, 1980). *Thelastoma pterygoton* sp. n. was described from the larvae of *O. monoceros*; this nematode is also found in *O. boas* (Poinar, 1973).

Green Muscardine Fungus

The green muscardine fungus, *Metarhizium majus* (J.R. Johnston) J.F. Bischoff, Rehner & Humber (=*Metarhizium anisopliae* var. *major*), occurs in the soil and attacks various *O. rhinoceros* stages (Bischoff *et al.*, 2009; Kepler and Rehner, 2013; Roskov *et al.*, 2014; Sathiamma *et al.*, 2001). Although the rates of natural infection are low, the fungus is an effective biological control agent against the rhinoceros beetle (Fernando *et al.*, 1995; Nirula, 1957; Tey and Ho, 1995). The fungus is more useful as a biopesticide component of integrated pest management at established locations than as a strategy for quarantine or eradication efforts (Bedford, 2014).

Identification of Infection

The fungus infects the *O. rhinoceros* larval, pupal and adult stages; the eggs are not infected, and symptoms typically do not appear in early instars (Cherian and Anantanarayanan, 1939; Nirula, 1957). In a larva, the initial symptoms of infection include sluggish movement, visible discoloration and dark brown lesions appearing throughout the body 3–7 d after inoculation (Figure 8-2A). The larva subsequently loses its appetite, sometimes moving out or onto the surface of the breeding site a few hours prior to its death (George and Kurian, 1970; Nirula *et al.*, 1955). Post-mortem, the host body becomes opaque, white and soft, gradually shrinking and becoming mummified. The fungus fills the *O. rhinoceros* body cavity, and 2–3 d after death, breaks through the host integument as a white mycelial growth (George and Kurian, 1970; Nirula *et al.*, 1955). In another 3–5 d, the characteristic dense mass of green conidiospores appears and covers the cuticular surface of the rhinoceros beetle (Figure 8-2B), promoting spore dispersal (Nirula *et al.*, 1955; Philippine Coconut Authority, 1998a, 2005). Larvae rarely survive the infection and molt into adults. If the pre-pupal larva is infected, they typically die in the cocoon before the final molt. Pupae are susceptible to infection; but generally do not exhibit symptoms prior to death; in some cases, a minor discoloration of the integument is noted. If the emerging adults are infected, they move out of the breeding substrate and exhibit a symptomatology similar to that of the larvae (Nirula *et al.*, 1955). Overall larval mortality occurs 1–3 weeks after the first sign of infection. The first instar dies in 1–2 weeks, and the second and third instars are typically dead in 2 weeks (Gopal and Gupta, 2001; Nirula *et al.*, 1955).

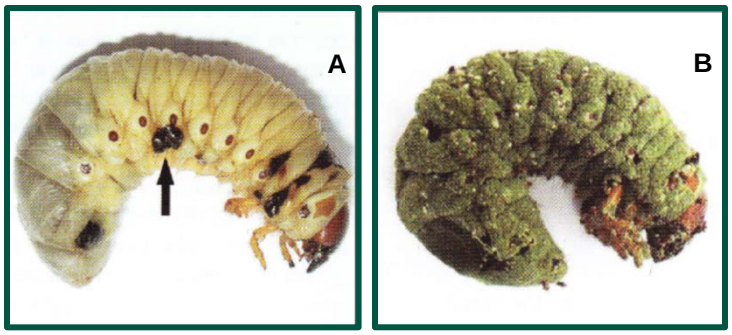

Figure 8-2 (A) Dark lesions characteristic of the initial stage of infection; (B) green conidiospores of *M. majus* covering the larval cuticle of the *O. rhinoceros* (photo courtesy of Ramle Moslim of MPOB)

Inoculum Concentration

Concentrations of 10^5–10^6 spores/mL cause mycosis in 50 % of the adults (the lethal time, LT_{50}) in 75–80 d for fungal isolates from *Oryctes* spp. (Ferron *et al.*, 1975). Although high inoculum concentrations improved the control of *O. rhinoceros* larvae, Darwis (1990) demonstrated that a minimum of 10^6 conidia/kg of sawdust provided efficient control, killing 90 % of the beetle larvae. However, this result may vary with the substrate, environment, larval distribution and other biotic and abiotic factors. In India, Gopal and Gupta (2001) recommended a concentration of 5×10^{11} spores/kL for the initial establishment of the fungus and successful control of *O. rhinoceros*. In a laboratory study, Bhide and Patil (2005) recorded maximum larval mortality at multiple concentrations: 4×10^8 spores/kg cow dung led to 70 % mortality in first instars after 10 d, 5×10^8 spores/kg led to 43 % mortality in second instars after 18 d, and 5×10^8 spores/kg led to 53 % mortality in third instars after 22 d.

Isolate Selection

The fungal isolates from *Oryctes* spp. are highly pathogenic to *O. rhinoceros*. In isolates from scarab species other than *Oryctes*, much higher (10–100-times) concentrations are required for a similar rate of larval mycosis (Ferron *et al.*, 1975). The pathogenicity of *M. majus* isolates from 5 different *Oryctes* species and 2 other scarab species is presented in Figure 8-4. There was no difference in the pathogenicity of isolates from *O. rhinoceros* larvae collected from multiple countries, all of which exhibited equally high virulence (Latch, 1976). The virulence reduce if the isolates are cultivated in artificial media for a prolonged duration, but it can be recovered after infecting an *O. rhinoceros* host (Fargues and Robert, 1983). For example, Gallego and Aterrado (2003) demonstrated that an *in vivo* cultured inoculum of the green muscardine fungus caused mycosis in 100 % of the beetle larvae in 10 d, whereas an *in*

vitro culture killed 96 % of the larvae in 13 d. The reduced time to mycosis and higher mortality confirmed the increased virulence of fungal isolates cultured on the target host.

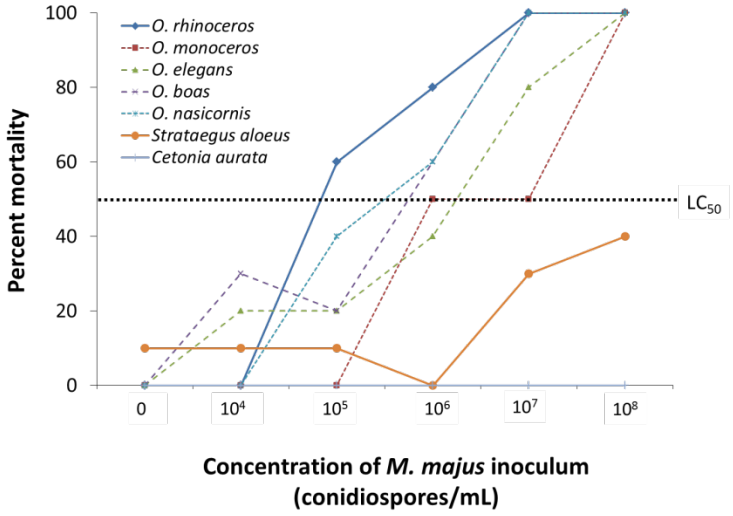

Figure 8-4 Percentage of *O. rhinoceros* adult mortality at increasing concentrations of 5 *M. majus* isolates. Prepared using results from Ferron *et al.* (1975).

Persistence

Metarhizium majus is present at the breeding site throughout the year, but survival and the effectiveness of the inoculum may vary over time (Nirula *et al.*, 1955). In Malaysia, application of a pathogenic strain at breeding sites increased the rate of infection from 5 to 83 % 4 months after inoculation; however, no long-term residual information was available (Tey and Ho, 1995). In Tonga, Latch and Falloon (1976) demonstrated that some *M. majus* isolates survived up to 2 y at the breeding sites; however, infection reduced over time, dropping to 50–70 % after a year. Persistence may vary; the fungus can remain from 1–3 y at a breeding site (Marschall, 1980). Cultural practices may also increase or conserve pathogenic fungal inoculums. Moslim *et al.* (2007) suggested that the cover crops often growing at breeding sites may interfere with the application of spores and delay the impact of fungal treatment on *O. rhinoceros* larvae; however, these crops may also provide favorable conditions for disease development. The establishment of the fungus at a location depends heavily on high humidity conditions (Subaharan, 2004).

Favorable conditions for infection

Favorable temperature, relative humidity and an overcast sky are important predisposing factors for the green muscardine disease. The optimum

temperature for *M. majus* sporulation is approximately 28 °C (Moslim *et al.*, 2005a; Moslim *et al.*, 2006). In southern India, fungal infections were highest during the monsoon seasons under high rainfall and humid conditions (Nirula *et al.*, 1955). Independent of temperature, increased humidity appears to favor the spread of infection and insect mortality. In Samoa, the fungus, which occurs naturally in the soil, killed 25–30 % of the larvae during wet years and 1–5 % during dry years (Marschall, 1980). However, the performance of the fungus on *O. rhinoceros* depends on the interaction between temperature and humidity. The LTe_{50} of the fungal conidia is negatively correlated with the relative humidity (RH) (Figure 8-3). This relationship implies that a humid environment is detrimental to the viability of the spores if the temperature is extremely high. In contrast, at average temperatures, a high humidity is favored for infection. For example, Nirula (1957) demonstrated that an RH above 70 % at 23–31 °C was preferable for disease development. Other studies indicated that spore germination was highest at 27–28 °C and relative humidities exceeding 95 % (El Damir, 2006; Gopal and Gupta, 2001). Direct sunlight may also negatively affect the fructification of the fungus; cloudy weather facilitates the disease development. In southern India, the disease developed most efficiently during overcast days with intermittent rainfall. A favorable period consisted of cloudy conditions on more than 50 % of the days with rainfall varying from 13–61 cm/month (Nirula, 1957).

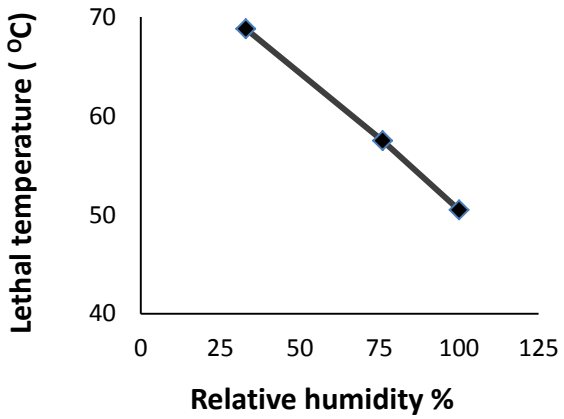

Figure 8-3 Relationship between the relative humidity and lethal temperatures of *M. anisopliae* conidia. Graph prepared using results from a study by Zimmermann (1982).

Formulations and Impact

Solid Substrate with Fresh Spores Sporulating solid substrates can be broadcast directly onto the breeding sites. The spores can be mass cultured on rice (Zimmermann, 1993), oat grain (Latch and Falloon, 1976), or cooked corn (Tey and Ho, 1995) in autoclavable polypropylene bags. Moslim *et al.* (1999) demonstrated that a wet solid substrate is more effective and economical than a dry solid substrate. In India, an inexpensive supplementary nitrogen source such as fishmeal or urea is added to the solid substrate to balance the C:N ratio and prevent variations in the pH that may deteriorate the substrate (Subaharan, 2004). The larval population at the treatment sites may be impacted as as early as 3 months (Latch and Falloon, 1976). However, the direct use of solid substrates carries limitations—the viability of the spores on the substrates may decline over time, and the substrate quantity required to treat large areas may pose handling restrictions (Moslim *et al.*, 2013; Moslim *et al.*, 2006).

Spore Suspensions A spore suspension can be prepared by harvesting spores from the liquid or solid substrate during the mass production procedure. A field study suggested that wet inocula, 10^8–10^{10} conidia in 10 L of water, were more effective and economical than dry inocula, 3–6 kg broadcast over a corn substrate. Three months after the application of dry or wet inocula, the treatment sites exhibited 37 or 51 % mycosis, respectively, for all *O. rhinoceros* stages (Moslim *et al.*, 1999). In Malaysia, application of a fresh suspension, 1×10^{10} spores/mL, at the breeding site significantly reduced the number of larvae. The spore suspension can be delivered using hydraulic sprayers or trunk injection (Moslim *et al.*, 2013). In some cases, live larvae are dipped in the suspension and introduced into decomposing felled logs at a rate of 2 larvae/m of the log (Philippine Coconut Authority, 1998a, 2005).

Granules A granular formulation is easy to apply and provides long-term control. Among the combinations tested, highest insect mortality was recorded with a mixture of 925 g of kaolin and 400 g of rice bran added to a 2-L solution of *M. majus* mycelia (Moslim *et al.*, 2009). Granules prepared from mycelia or spores were equally effective, both killing 100% of the larvae in 18 d (Moslim *et al.*, 2008). Mycelia production was higher at pH 8 and lower at pH values of 5–7 (Moslim *et al.*, 2009).

Spore Dust The Philippine Coconut Authority (1998a, 2005) recommends the use of spore dust on coconut stumps (1–2 tbsp/stump), piles of sawdust (1 tbsp/m^2) and traps filled with suitable breeding media (50 g/box). In some cases, powdered infected larvae are applied directly to the breeding sites (Gallego and Aterrado, 2003).

Other Formulations In addition to the conidiospores, other forms such as dry mycelia and blastospores are used against *O. rhinoceros*. Pellets prepared from these fungal structures can be stored at low temperatures in vacuum-sealed plastic bags (Gopal and Gupta, 2001).

Delivery Techniques

Ideally, field application of the fungal inoculum would occur under high humidity that typically coincides with rainfall (Subaharan, 2004). Optimum control is achieved when most of the larvae at the breeding site are molting. The *M. majus* formulations can be delivered to the target breeding or feeding sites using several techniques as depicted in Figure 8-7.

Moslim *et al.* (2013) describes fungus formulations on dry substrates or fresh spore solutions as effective for smaller breeding sites, but unsuitable for large-scale applications.

Figure 8-7 Delivery of *M. majus*: (A) treatment of oil palm residues using a high-volume sprayer; (B) application of a spore solution at a breeding site (photo courtesy of Ramle Moslim of MPOB)

Mist blowers, power sprayers and high-volume sprayers are effective for field applications (Moslim *et al.*, 2013). For the first two, application rates as low as 0.5 g of spores in 3 L of water per m^2 of rotting palm heaps caused 100 % larval mortality 3–5 weeks after treatment (Hamid *et al.*, 2005). The solution for high-volume spray was prepared by mixing 200–400 g of spores in 30–40 L of water for an application rate of 0.75 L/m^2 on oil palm debris heaps. The first impact of the application was observed after 8 months when the insect population decreased 70 % in the treated plots (Moslim *et al.*, 2013). Moslim *et al.* (2013) recommends the use of spore solutions in large flat areas infested with *O. rhinoceros*; the equipment and water resources necessary for high-volume sprays are typically available in such locations.

Adults may play an important role in the spread of the disease as they are strong fliers and may transfer the fungal spores to other breeding or feeding sites. This ability of the adult can be exploited as a strategy for population control. For example, in Samoa infected live larvae and adult beetles smeared with a mixture of butter and spores were released at the target sites (Marschall, 1980). In Malaysia, adult beetles were collected using pheromone lures, dusted with *M. majus* spores and manually released in a large target area to spread the spores (Moslim *et al.*, 2013). Spores can also be spread through auto dissemination traps. The adults are lured to traps using an aggregation pheromone mixed with a spore solution (2–4 g/L). The trap design allows most of the adults to escape and spread the fungus (**Figure 8-8**).

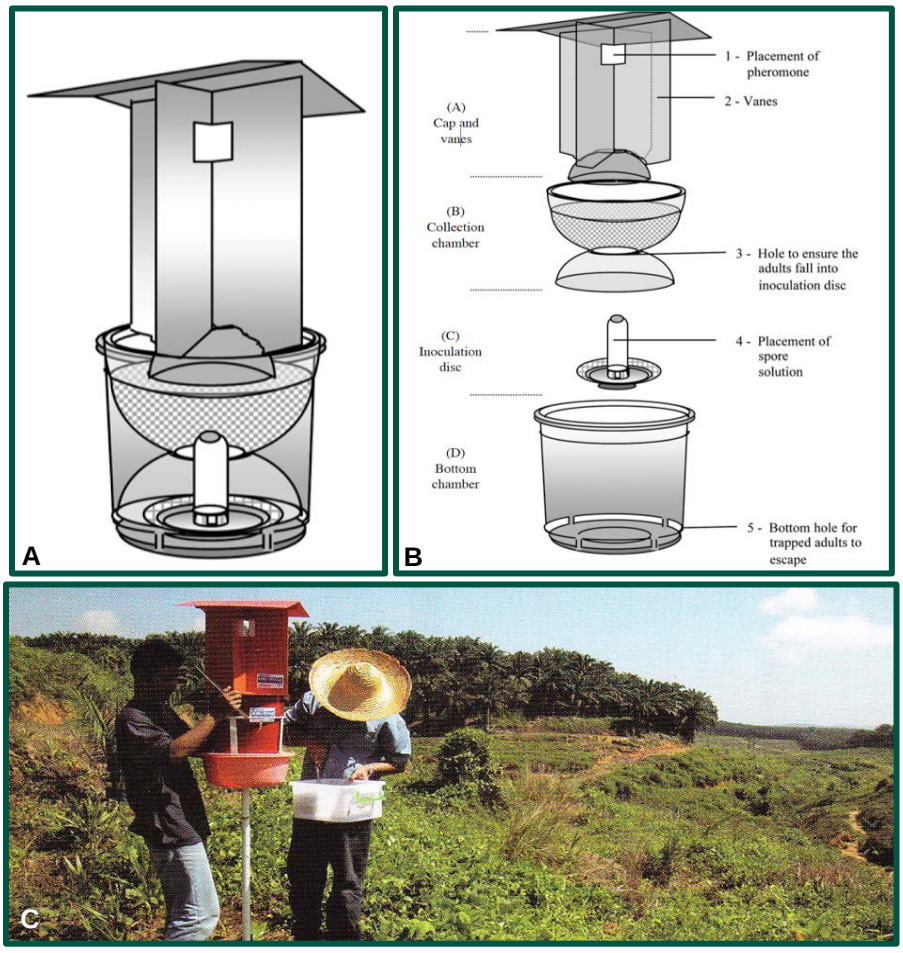

Figure 8-8 Trap for the auto dissemination of *M. majus*; (A–B) trap design; (C) field use (photo courtesy of Ramle Moslim of MPOB)

A field study demonstrated that among the trapped adults, 85 % were able to escape and 67 % were infected. The escaped adults further infected and killed 92 % of the larvae at the breeding sites. However, the viability of the spores spread through this method was low and did not significantly reduce the beetle population. Most of the escaped infected adults died within 15–30 d (Moslim

et al., 2011b). Given challenges in application, the use of auto dissemination traps remains preferable to sprays in undulating topographies. Moslim *et al.* (2013) recommends high trap densities to enhance spore concentration and distribution.

Moslim *et al.* (2013) reviewed the use of artificial breeding sites (1 trap/5 Ha) to spread the spores. For the breeding trap, a trough was prepared with an approximate length of 4 m, a width of 2 m and a depth of 1 m. The trough was filled with palm trunk chips, which were allowed to decompose. To attract adults, a pheromone lure was placed adjacent to the trough. Different doses of *M. majus* inocula were sprayed every 3 months for 1 year to infect the visiting adults and breeding larvae. Most adults were infected after contact and spread the spores to other breeding sites. This method produced 43 % infection at the trap breeding sites, an improvement over the 32 % achieved with the blanket spray of a spore solution (Moslim *et al.*, 2013). However, dispersion of the inoculum by the infected beetles is inefficient if the breeding sites are widely distributed (Young, 1986). In a similar study in Samoa and Tonga, 'trap' breeding sites were sprayed with fungal spores to increase larval mortality and serve as an inoculum source to be spread by emerging or visiting adults (Marschall, 1980; Prior and Arura, 1985). A review by Moslim *et al.* (2013)indicated that the breeding traps were most effective on plantations with replanting programs.

Few studies have investigated the use of the green muscardine fungus against the feeding stage of rhinoceros beetles and other secondary pests. An *O. rhinoceros* attack can be followed by secondary and more serious infestations of *Rhynchophorus ferrugineus* Herbst and/or *Scapanes australis* (Boisduval). In Papua New Guinea, the green muscardine fungus was applied to the palm leaf axils using a pre-cooked rice medium (100 g/palm), which reduced the *S. australis* damage by 32 %, but had no significant impact on *R. ferrugineus* (Prior and Arura, 1985). Therefore, a broader use of the biological control agent requires further investigation.

Safety

The fungus does not harm mammals and is considered to be of minimum risk to non-targets including humans (Gopal and Gupta, 2001; Zimmermann, 2007). *Metarhizium majus* can also be used at the vermicomposting sites without impact to the non-target earthworms. The fungus selectively killed *O. rhinoceros* larvae at the concentrations (10^2–10^4 spores/g of substrate) tested (Gopal *et al.*, 2006). Moslim *et al.* (2007) demonstrated that the fungus had no significant impact on the oil palm pollinating weevil *Elaeidobius kamerunicus*

Faust or the stag beetle *Aegus chelifer* MacLeay after the spores were applied in rotting palm residues.

Advantages and Challenges in Implementation

The use of entomopathogenic fungi is increasingly common in integrated pest management (IPM) and insecticide resistance management (IRM) programs. The fungi aid the IRM programs because a fungal infection suppresses enzyme activities in a pest, possibly increasing the insect's susceptibility to pesticides. Therefore, some IRM programs use the entomopathogenic fungi in conjunction with sub-lethal insecticide doses (Ambethgar, 2009). However, some insecticides, like chlorpyrifos, have an inhibitory effect against the green muscardine fungus and some carbamates are highly toxic to the fungus (Kao *et al.*, 2012). Therefore, future research should examine the compatibility of *M. majus* with IRM and IPM programs.

Oryctes rhinoceros Nudivirus

Oryctes rhinoceros nudivirus (OrNV) [=*Rhabdionvirus oryctes* ; *Oryctes baculovirus* ; *Oryctes* virus (OrV)] is endemic to Malaysia, Philippines, Indonesia, and India (Mohan *et al.*, 1983; Sujatha and Rao, 2004; Zelazny, 1977a) and has been effectively used worldwide in integrated management programs against the coconut rhinoceros beetle (Huger, 2005).

Isolates, Virulence and Host Resistance

The isolate from Malaysia was used globally in several virus release programs, but multiple isolates with distinct variations in virulence have since been identified from different locations. Zelazny (1979) carried out the initial studies, examining ten virus isolates obtained from Samoa and the Philippines, and determined that the isolate, PB, from Leyte Island in the Philippines caused higher mortality than the isolates from other locations. This study indicated differences in virulence among the isolates; but, it is to be noted that the number of virions in a 'standard dose' to test the virulence was not accurately determined and therefore, the differences reported in virulence may not be conclusive. Further, the genomic information for the virus was unavailable at the time. In 1985, an OrNV genomic map was prepared using restriction enzymes on the virus isolate PV505 from Southern Luzon, the Philippines (Crawford *et al.*, 1985). This genomic information allowed subsequent studies to determine significant genetic variations among the OrNV isolates. In the Maldives, a field-study evaluated 12 geographical isolates of the virus and determined that the X2B isolate from Palawan, the Philippines, best reduced the *O. rhinoceros* populations in the Maldives (Zelazny *et al.*, 1990). A survey conducted in Malaysia identified 4 distinct

isolates—types A, B, C and D—of varying virulence. Although type A, similar to PV505 from the Philippines, was naturally widespread at the location, type B (= Ma07) was much more virulent and effective against *O. rhinoceros* in Malaysia (Ramle *et al.*, 2005). The virus types A, B and C, caused 27, 87 and 13 % mortality, respectively, in third instars. The LT_{50} was also significantly shorter for type B (= 34 d) than for other types (~100 d). Likewise, in younger adults the mortality and infection rates were significantly higher for type B, though type A had higher infection and mortality rates in mature adults (Ramle *et al.*, 2011). Although the number of virions in a dose could not be determined, the study by Ramle *et al.* (2005) attempted to quantify virus DNA content in doses administered by PCR; this provides an index that may be helpful in determining virulence (Bedford, 2013, 2014). Previous studies have examined the probability of the development of resistance in *O. rhinoceros* against different isolates of OrNV; however, no conclusive evidence has been obtained to suggest counter-resistance in the host insect (Crawford, 1988; Zelazny, 1979; Zelazny *et al.*, 1989). In Guam, Moore (2012) tested the efficacy of 8 OrNV strains—obtained from Dr. Trevor Jackson's lab in New Zealand—on locally obtained *O. rhinoceros*. Preliminary assays determined that the strains did not affect the beetles, indicating that the local population may be resistant to OrNV. Alternatively, the lab-cultured OrNV in the cell lines may have lost its virulence over time. Therefore, further studies are underway to examine the impact of OrNV on an imported susceptible population of the rhinoceros beetle (Moore, 2014).

Detection of infection

Pre- and post-release monitoring of OrNV levels in the *O. rhinoceros* population relies on accurate detection.

Visual Infection is lethal, visible and distinct for *O. rhinoceros* larvae (Zelazny, 1972). This infection, initially referred to as the 'Malaya disease' (Huger, 1969), causes the thoracic tergum to appear pearly, waxy and translucent in infected larvae; this obvious external symptom may be due to the cessation of feeding and absence of food in the midgut (Huger, 1966; Mohan *et al.*, 1985). The translucency, which directly contrasts with the clear, dark midgut of healthy larvae, typically appears 5–8 d after the entry of virus into the larva (Mohan *et al.*, 1985). As the infection progresses, the fat in the abdomen appears to disintegrate as observed through the integument. The body becomes more turgid with an apparent increase in hemolymph that may lead to a prolapse of the rectum (Huger, 2005). During the final stages of infection, the layer beneath the abdominal integument develops chalky-white mottled accumulations. Post-mortem, the larva becomes flaccid, then shrinks

and mummifies. Initially, the cadaver appears brownish, later turning bluish-black (Huger, 1966; Mohan *et al.*, 1985). The adults do not typically exhibit external symptoms of infection; but, a few cases of wing malformations have been reported after virus release in Mauritius (Monty, 1974; Zelazny, 1973a), although these malformations may be associated with damage to the pupal chambers (Zelazny, 1976). Adults also exhibit reduced feeding, flight activity and longevity (Zelazny, 1973a; Zelazny, 1977b).

Laboratory Various techniques have been used to identify OrNV infection. For example, the structure of the virus can be examined through electron microscopy (Huger, 1966; Jackson *et al.*, 2010; Mohan *et al.*, 1983; Payne, 1974). Gut symptoms could be examined using light microscopy (Gorick, 1980; Ramlah Ali *et al.*, 2001), inspection of adult excreta (Monsarrat and Veyrunes, 1976), dot-blot assay (Crawford, 1988), enzyme-linked immunosorbent assay, ELISA (Longworth and Carey, 1980; Mohan and Gopinathan, 1989), immuno-osmophoresis (Mohan *et al.*, 1983; Mohan and Pillai, 1983), immunofluorescence (Croizer and Monsarrat, 1974), host mortality bioassays (Jackson *et al.*, 2010), DNA restriction endonuclease activity (Eberle *et al.*, 2012; Ramle *et al.*, 2011), solid-phase radioimmunoassay (Crawford *et al.*, 1978), direct antigen coating-indirect ELISA and dot-immunobinding assay (Rajamannar and Indiravathi, 2000), and polymerase chain reaction (PCR) (Eberle *et al.*, 2012; Jackson *et al.*, 2010; Ramlah Ali *et al.*, 2001; Ramle *et al.*, 2010; Ramle *et al.*, 2001). Ramle *et al.* (2010) optimized the PCR reaction to yield the single 945 bp band using a primer pair developed by Richards *et al.* (1999):

OrV515a = 5’-ATTACGTCGTAGAGGCAATC
OrV515b = 5’-CATGATCGATTCGTCTATGG

During pre and post-release monitoring of OrNV at a location, cross-contamination can occur in beetles collected together in traps; this data collection technique may inflate the infection results (Ramle *et al.*, 2005). To reduce cross-contamination, adults collected from traps could be processed immediately, kept separate or stored under conditions that do not allow disease spread. Early stages of viral infection are detectable via PCR and ELISA (Bedford, 2013; Ramle *et al.*, 2011).

Impact on the Host

The virus readily infects the larva and adult stages of *O. rhinoceros* without affecting the pre-pupa and pupa (Huger, 2005; Moslim *et al.*, 2011a). Further, percentage of infection is greater in adults than larvae (Moslim *et al.*, 2011a).

Larva Infected larvae develop diarrhea, become lethargic, discontinue feeding and eventually move to the surface of the breeding substrate (Huger, 1966;

Lacey, 2012). A significant reduction in food consumption, growth rate, digestibility and food conversion efficiency were observed after infection (Paulose and Abraham, 1997). The virus typically kills the larvae in 1–4 weeks (Huger, 2005; Zelazny, 1972). Zelazny (1972) reported that 94 % of all larvae died within 5 weeks of ingesting the virus with total mortality occurring in 8 weeks; however, this may vary based on the dose and as there is no accurate methods to determine the number of virions, the information about the longevity of infected larvae may not be useful (Bedford, 2013, 2014).

Adult Zelazny (1973b) suggests that the OrNV infection in adults impacts the *O. rhinoceros* population more significantly than that in larvae. Although symptoms are not explicitly visible in an adult, an OrNV infection modifies their behavior and biology. In adults, a viral infection leads to a cessation of feeding within 1 week, decreases flight activity and reduces longevity (Zelazny, 1973a; Zelazny, 1977b). The infected adult dies within 4–5 weeks (Zelazny and Alfiler, 1991). In males, decreased mating activity was also reported following infection (Zelazny, 1977b). In females, the infection rate is typically higher and leads to reduced fecundity; thus, females in advanced stages of infection made significantly fewer visits to the breeding sites for oviposition than healthy females (Zelazny, 1973a, 1973b; Zelazny and Alfiler, 1991).

Dissemination of the virus

The autodissemination of the virus by adult beetles is the key to its success as a biological control agent. After infection, the gut lumen of the infected adults fills with sloughed-off midgut epithelial cells that contain the proliferating virus particles in their nuclei. The infection creates diarrhea symptoms in the adults causing them to actively defecate, possibly spreading the virus at the mating, feeding and breeding sites (Huger, 1966). An infected adult can typically produce 0.3 mg of virus in their excrement each day (Monsarrat and Veyrunes, 1976). Huger (2005) noted that the cytopathic process in the adult midgut, the chronic infection and the autodissemination process rendered the adults “flying virus reservoirs,” providing a suitable strategy for regulating the beetle populations.

Virus transmission is prevalent where male and female adults co-exist; these locations include dead standing palms, other breeding sites and possibly at the feeding sites (Bedford, 2013, 2014; Zelazny and Alfiler, 1991). The horizontal transmission of the virus to adults probably occurs through 3 methods: (1) during mating, copulation itself may not transmit the virus, but may expose an uninfected adult to the fresh excrement of its infected partner, facilitating rapid per os entry. In the field, the number of infected mated females

significantly exceeded that of unmated females (Zelazny, 1976). (2) During feeding, the virus may be transmitted through successive or simultaneous feeding by adults at the palm axils or similar feeding sites. Adults defecate at the feeding sites, exposing the uninfected insects to the inoculum. Nevertheless, transmission of the virus at the feeding sites may occur infrequently because the virus is rapidly inactivated under dry conditions. Furthermore, adults may not come into contact during feeding (Zelazny, 1975, 1976). Hochberg and Waage (1991) constructed a model to investigate the efficiency of various OrNV transmission pathways and indicated that the dominant route may be from infected to feeding adults. Zelazny and Alfiler (1991) indicated that virus transmission among young adults at a feeding site may be attributed to mating events at the site (3) At breeding sites, the survival of the virus is reduced, but presence of the virus at breeding sites may be critical for its persistence in the environment and its utility for long-term control (Hochberg and Waage, 1991). Although there were several assumptions about the spread of the virus, Bedford (2013, 2014) noted that there are no peer-reviewed studies to prove the presence of virus in feeding holes, crowns or frond axils and further, there is no evidence of copulation at these locations.

In the field, the virus may occur naturally at only a few breeding sites, but artificial OrNV inoculations at these sites can facilitate dissemination. The virus can be transmitted to adults from the inoculated breeding sites; however, this mode of transmission has limitations. Although the older females frequently visit the breeding sites for oviposition, they rarely become infected, whereas younger females with higher probabilities of infection seldom visit the breeding sites (Zelazny, 1976). Although an increase in the availability of decaying logs or other substrates can slow the spread of the virus, a limited substrate ensures a location for transmission. A large number of breeding sites reduces the chance for contact between infected and healthy adults. Therefore, a strict balance in the substrate availability could facilitate the spread of the disease and regulate the *O. rhinoceros* populations (Zelazny and Alfiler, 1991).

The type of the substrate available may also play a role in spreading the virus. For example, in the Philippines, Zelazny and Alfiler (1986) demonstrated that increasing the number of dead standing palms can increase the spread of the virus; however, additional stumps and felled logs did not appear to enhance disease incidence. These observations may be location specific and may not display a similar pattern in populations with different substrate preferences. Adults visiting the breeding sites may also become infected if the sites contain fresh virus inoculum from newly dead infected larvae; likewise, healthy larvae

may become infected if they are exposed to the virus inoculum from the excrement of visiting beetles—though this scenario is rare (Jackson *et al.*, 2005; Zelazny, 1973b; Zelazny, 1976; Zelazny and Alfiler, 1991).

Previous studies have also investigated other modes of OrNV transmission: Virus particles on the culticular surfaces of the adult beetles are rapidly inactivated under warm and dry conditions; therefore, cuticular contact is not a common mode of transmission (Zelazny, 1976). Although Huger (1969) noted that vertical transmission of the pathogen was possible, a study by Zelazny (1976) indicated that the pathogen cannot be vertically transmitted from larva to adult. Moreover, although virus particles occur in oviducts and oocytes of infected females, most of the older infected females do not lay eggs, and if they oviposit, the larvae emerging from the surface-contaminated eggs are rarely infected (Zelazny, 1973a; Zelazny, 1976). Furthermore, newly emerging adults are not vectors until they acquire the virus from other individuals, the environment or artificial inoculation (Zelazny and Alfiler, 1991).

Production, Maintenance and Storage of the Inoculum

The artificial cell line, DSIR-HA-1179, derived from the scarab, *Heteronychus arator* (F.) can be used for multiplying OrNV (Crawford, 1982). Jackson *et al.* (2010) indicated that pure culture vials can only be stored in a dark refrigerator for 1–2 weeks without significant degradation, and cultures should be used on the same day that the vial is opened. Deep-freezing may reduce viral activity (Jackson *et al.*, 2010).

In the later iterations of release, Zelazny *et al.* (1987) described a new procedure to produce OrNV inoculum in a laboratory with limited facilities. The Philippine Coconut Authority had adopted a purified suspension technique using a virus-infected DSIR-HA-1179 cell culture, the guts of infected insects, glass permeation chromatography, sucrose and a final filtration method to seal the purified inoculum in sterile serum vials. The sealed inoculum could then be stored at room temperature for 2 weeks or refrigerated for 20 weeks. Furthermore, Zelazny (1972) demonstrated that high temperature can significantly reduce the viral activity in a suspension.

During a field release study in Fiji, Bedford (1976) suggested that the virus-packed cadavers of infected larvae could be stored in deep freeze indefinitely for later use as a source of inoculum; the virus particles may be gradually inactivate after thawing (Bedford, 2013, 2014). Zelazny (1972) investigated the storage and inactivation of the virus in a formulation of powdered infected larvae mixed with sawdust. After storage for 1 week, the viral activity dropped to 0.091 % of its initial value, and after storage for 1 month, the

inoculum was no longer infectious. The inactivity rate increased at high temperature and low humidity (Zelazny, 1972). However, this was a lab study and in natural conditions the inactivation rates may vary with the specific condition and location (Bedford, 2014). Bedford (2013) noted that specific enzymes from microbes in the breeding substrate may also influence inactivation rate. The OrNV or the virus-containing substrates are fed to the coconut rhinoceros beetle adults to produce and maintain the pathogen (Bedford, 1976).

Release of the Virus at Various Locations

Among the 37 species of entomopathogens used in classical biological control programs for any pest, OrNV, with the highest number of global releases (= 18), is considered the most successful microbe that has established at all locations of introduction (Hajek *et al.*, 2007). This virus was released against *O. rhinoceros* in the South Pacific (Hammes and Monsarrat, 1974; Marschall, 1980), against *O. monoceros* in Seychelles, Tanzania and the Ivory Coast (Julia and Mariau, 1976; Lomer, 1986; Paul, 1985; Purrini, 1989) and was re-released in OrNV-native locations (Mohan and Gopinathan, 1991; Mohan *et al.*, 1983; Zelazny, 1977a). The safety and environmental concerns with the pathogen were addressed prior to widespread OrNV introduction (Gourreau *et al.*, 1981).

Samoa In 1967, OrNV was released in Manono and Savai'i in decaying sawdust under split coconut logs simulating a breeding site to attract adults. After 18 months, the beetle population nearly disappeared from Manono. In addition, infected larvae were recovered from untreated locations in Savai'i and another island, Upolu (Marschall, 1970). Although the virus strain was originally from Malaysia and was cultured in Darmstadt, it was multiplied on native larvae in Samoa prior to release. The virus continued to spread without further assistance, allowing the recovery of coconut plantations in Samoa (Huger, 2005; Zelazny, 1973b). In 1975, a re-release program was established and the study confirmed that the virus levels can be increased through a periodic re-release of OrNV (Marschall and Ioane, 1982).

Tokelau In 1967, OrNV was released at this location (Uili, 1980; Zelazny, 1977). A few years after the initial release, the virus levels in the population plummeted necessitating re-releases in 1973 and 1974. Zelazny (1977) reported that there was a significant decline in adult population 10 months after OrNV introduction.

Wallis Island In 1970, OrNV was introduced and as a result, adult beetle populations declined 60–70 % in 1 year.

Mauritius In 1970, OrNV was introduced from Samoa and in late 1974, a survey of the breeding grounds confirmed a decline in *O. rhinoceros* larvae (Monty, 1978). Overall, the surveys from 1973 to 1977 reported a 60–95 % reduction in damage caused by *O. rhinoceros* (Hammes, 1978).

Tonga In 1970, OrNV was introduced from Samoa. Epizootic levels developed in less than 5 months and the virus spread across the island in 15 months (Young, 1974). In 1978, approximately 7 y after the first release, surveys indicated high percentage of infected breeding sites and adults, low levels of palm damage and reduced beetle populations (Young and Longworth, 1981). Bedford (1986) reviewed that due to the lack of damage and persistence of OrNV, the virus was not re-released at this location.

Fiji Multiple OrNV releases were made in Fiji from 1970 to 1974 (Bedford, 1976a, 1976b). Approximately 12–18 months after field release, a significant reduction in palm damage was reported in Fiji. Bedford (1986) noted the same timeline for population decline in most release locations in the South Pacific. Approximately 57–68 % of the beetles were infected at this location.

Papua New Guinea In 1977, OrNV was imported from Samoa for field release in 1978 and 1979 (Gorick, 1980). Post-release monitoring, which revealed a minimum interval of 8 months between the OrNV release and the capture of a newly infected adult at this location. Trap surveys following the release suggested that the infection spread at a rate of 1 km per month (Gorick, 1980).

Maldives Between 1984 and 1985, the first OrNV releases occurred in Meemu, Lawiyani, North Ari, the Baa atolls and a few islands close to Male. Within a year of the release, the quantity of beetle damage decreased by 25 %, and the virus-infected adults increased to 50 % yielding a 10 % increase in the coconut yield (FAO, 1986).

Caroline and Marshal Islands OrNV was imported from Samoa. After release, the virus became established in Babeldoab. In 1983, the virus was re-imported from Samoa and released in Peleliu and a few other locations because of an increase in *O. rhinoceros* populations (Schreiner, 1989).

Sultanate of Oman In 1989, virus-infected beetles were released (Kinawy, 2004). Two months after release, OrNV was detected in approximately 41 % of the local beetle population. Prior to release, approximately 85 % of all palms were damaged; however, after OrNV release, the number of damaged palms decreased to 48, 31, 17, 10, 6 and 4.2% from the first through sixth year, respectively (Kinawy, 2004; Kinawy *et al.*, 2008).

Lakshadweep In 1983, an OrNV isolate, OBV-KI, obtained from Kerala, India was released on Minicoy Island (Mohan *et al.*, 1989). After 19 months, the spathe damage in the coconut palms decreased by 93 %, and after 3 y, a survey indicated that approximately 7.5 % of the breeding sites in Minicoy were infected with the virus—a result comparable to that in Samoa after 4 y. After 2.5 y, nearly 50 % of the beetles in trap catches were infected with OrNV (Mohan and Pillai, 1993b).

India In 1985, OrNV-infected adults were released in oil palm plantations in Palode, Kerala, India, where the virus is indigenous (Dhileepan, 1994; Mohan *et al.*, 1983). Prior to the release, the indigenous virus had already infected approximately 60 % of the beetles surveyed. After the supplementary OrNV release, the beetle damage in palms declined significantly during the first 3 y—70 % damage was reduced to 20 % by the third year. The quantity of damage appeared to reach equilibrium at the fourth year, and increased gradually until the final surveys in 1991. The decline in beetle attacks was not due to increased palm maturity, but to the reduced virus inoculum levels at the sampled sites—as evidenced by a decrease in infected larvae at the breeding sites (Dhileepan, 1994). In 1989, Biju *et al.* (1995) studied 3–4 year old coconut palms in OrNV-endemic Thrissur, Kerala, India and obtained similar results indicating that re-release programs increase virus inoculum levels in the field. These studies confirmed that even in endemic areas a re-release of OrNV can reduce palm damage and that post-release surveys should regularly monitor inoculum loads in the field.

Andaman Islands In 1987, the Kerala isolate OBV-KI was introduced. Approximately 1.5 y after release, the palm damage decreased an average of 60 %; and after 3.5 y, the damage decreased by 90 % (Jacob, 1996). However, from 1999–2000, outbreaks of *O. rhinoceros* were associated with an increase in area-wide coconut replanting programs in South Andaman. The outbreaks led to the re-release of infected beetles and approximately 23 months after release, a 90 % reduction in palm damage was observed at these locations with a three-fold reduction in the adult *O. rhinoceros* population (Prasad *et al.*, 2008a).

Malaysia OrNV was first discovered in Malaysia, and although the virus is endemic, it resurfaced in the 1990s as an important pest of oil palm because of the the ‘zero-burn’ policy in oil palm plantations. Moslim *et al.* (2011a; 2005b) demonstrated that the type B strain of the virus was effective at the release sites.

OrNV Inoculation Methods for Field Release

The exact mechanism of transmission was unknown during the initial virus release programs in the South Pacific. The inoculation of the virus in the field occurred primarily through the application of OrNV at the breeding sites, for which the imported virus strain was first multiplied in local host larvae in the laboratory to confirm and maintain virulence in the local populations. After death, the infected larval cadavers were stored in deep freeze for an extended period. Immediately prior to application, the cadavers were triturated to prepare a fresh virus suspension, which was then used to inoculate many breeding sites. The treatment sites included natural breeding habitats, artificial compost heaps and split coconut log heaps. OrNV inoculation at breeding sites was successful in Samoa, Wallis, Mauritius and Tonga. The primary disadvantage of the method was the possible inactivation of the virus under ambient conditions outside the host; nevertheless, the breeding site inoculation programs were successful, likely due to large adult populations at the target site that may have acquired the pathogen before inactivation (Bedford, 1976b; Burand, 2008; Hammes and Monsarrat, 1974; Huger, 1973; Marschall, 1970; Monty, 1974; Young, 1974). However, after the insects acquire the virus, an increase in beetle population may not always result in an increase in the virus inoculum at the release site (Marschall and Ioane, 1982; Zelazny, 1977b).

Once the role of adult beetles in the dissemination of the virus was discovered, all OrNV release programs shifted focus to the direct release of infected adults. *Oryctes rhinoceros* adults can be collected in pheromone traps or from breeding heaps for infection and release (Bedford, 2013; Jackson *et al.*, 2010; Moslim *et al.*, 2010). Previous studies investigated various techniques to maximize adult infection prior to release. For example, Bedford (1976b) dipped healthy adults in a virus suspension for 2–3 minutes and fed them an infected sawdust substrate for a day prior to their release at target sites in Fiji. Zelazny (1978) indicated that injecting the adults with virus-infected hemolymph was more effective for infection than submerging the adults in a virus suspension, but this method may not be suitable for large-scale releases. Marschall (1980) suggested another inoculation technique—the adult was fed infected substrates, and after 5–8 d, the infected midguts were dissected, macerated and prepared into a solution. After adding sugar to increase the palatability, the solution was applied, at a dosage of 10^{-4} to 10^{-5} g to individual beetle mouthparts. This technique was adapted with slight modifications at several release locations (Jackson *et al.*, 2010; Jacob, 1996; Mohan *et al.*, 1989; Moslim *et al.*, 2005b; Zelazny, 1978). A pause in pheromone trap operations for ~ 2 weeks at the time of OrNV release is recommended for a uniform distribution of the infected adults (Moslim *et al.*, 2005b). The virus

dosage used to infect the adults for release may also impact the efficiency of the OrNV release program (Mohan and Pillai, 1993; Prasad *et al.*, 2007; Prasad *et al.*, 2008); however, to date, there is no conclusive method for determining the virus dose (Bedford, 2013, 2014).

Challenges to and Prospects for Implementation

The release of OrNV was successful at all introduced locations and reduced the impact of *O. rhinoceros* (Figure 8-10) and, in some cases, *O. monoceros* damage to palms. However, the reduction in *O. rhinoceros* population and the duration of impact varied with each release. The impact depends on intrinsic factors of *O. rhinoceros* and OrNV and their interactions with the environment, which include the virulence of the isolate against the beetle population at the release location, the inoculation methods and dosage and activities that may impact the pre- and post-release *O. rhinoceros* density (Mohan *et al.*, 1989; Mohan and Pillai, 1993b; Zelazny and Alfiler, 1986; Zelazny *et al.*, 1990). For example, natural disasters, palm replanting and changes in plantation management such as the introduction of zero-burn policies can increase the availability of breeding sites. An abundant larval habitat may diminish opportunities for infected adults to contact healthy beetles. An increase in breeding sites will also increase the proportion of healthy adults in the population leading to a reduced equilibrium of viral incidence (Mohan and Pillai, 1993b; Moslim *et al.*, 2011a; Prasad *et al.*, 2008a; Zelazny and Alfiler, 1991). This limitation encourages the development of strategies to occasionally boost the number of infected adults. Jackson *et al.* (2005) reviewed the potential of a ‘lure and infect’ autodissemination system for OrNV release programs. Ideally, the aggregation pheromone, E4-MO, can lure the adults, facilitate infection with a virulent OrNV isolate and allow their escape for dissemination. However, execution requires a persistent and reliable virus inoculum, and the benefit of the ‘lure and infect’ strategy to a ‘lure and kill’ method has not been established (Jackson *et al.*, 2005).

Abiotic factors can also inhibit the activity of OrNV, which typically does not persist outside its host for a prolonged duration; dry, warm conditions can increase its inactivation rate (Zelazny, 1972, 1977b). According to Zalazny (1972) and Mohan *et al.* (1985), complete inactivation of the virus particles could occur after exposure to 56–70 °C for 10 minutes, though the inactivation time may be influenced by several intrinsic factors. In a field study, Gopal *et al.* (2002) demonstrated that OrNV infection in adults and larvae was negatively correlated with the minimum temperature and positively correlated with the relative humidity.

Figure 8-10 A comparison of *O. rhinoceros* damage in Fiji (A) before and (B) after the release of OrNV (images by Geoffrey Bedford posted with permission from the Annual Review of Entomology, Volume 58 © 2013 by Annual Reviews, http://www.annualreviews.org)

The 2 entomopathogens—green muscardine fungus and OrNV, were used simultaneously as biological control agents on several occasions to reduce the damage from coconut rhinoceros beetles. In 1969–1970, both pathogens were released in Tonga to combat *O. rhinoceros*, and the fungus exhibited no significant impact on the pest population, whereas the OrNV led to epizootic levels of infection and substantially reduced the number of beetles (Young, 1974). However, Bedford (1986) notes that the fungus may have established at the breeding sites and contributed to the impact on the pest. Hochberg and Waage (1991) constructed a mathematical population model to understand the influence of various factors on the efficient dissemination of OrNV throughout the beetle population. In the model, the treatment of breeding sites with *M. majus* was considered a persistent, density-independent factor that reduces the pest population. The model suggested that applying the fungus significantly impacted the larvae and emerging adults, whereas feeding adults were unaffected, which suggests that the application of *M. majus* might cause population instability in *O. rhinoceros* and lead to the gradual elimination of

OrNV at the release site. In a later review, Bedford (2013) argued that although this negative interaction could occur on small islands, it should not be a problem on large landmasses because the diminishing OrNV inoculum will be replenished by immigrating infected adults. Furthermore, because *M. majus* primarily affects the larvae and OrNV the adults, their simultaneous use may lead to a rapid decline in the pest population. Interactions between OrNV and other microbes are not well studied; a preliminary study has examined the impact of the enterobacterium, *Pseudomonas alcaligenes* Monias, on *O. rhinoceros* larvae in Kerala, India. An OrNV infection of the host can act as a stressor to promote infection and septicemia by *P. alcaligenes*, which could lead to a gradual reduction in the total OrNV inoculum in the environment. However, this stressor could also reduce the pest population (Murali and Alka, 2002). A survey conducted in Kerala, India from 1996–1999 demonstrated that 5 and 3 % of the larvae died from viral and fungal infections, respectively, whereas 20 % of the sampled larvae had evidence of bacterial septicemia that interfered with the OrNV efficiency. The adults were not infected with the fungal and bacterial pathogens, but OrNV infection occurred in 22 % of the sampled population (Gopal *et al.*, 2002). The interactions between the 3 microbes merit further investigation.

Cultural Control and Sanitary Measures

Trapping

In pest management programs for *O. rhinoceros*, traps are used to monitor the incidence and population dynamics of the adults, auto-disseminate green muscardine fungus, capture adults for OrNV inoculation and release and mass-trap and kill the adults for a direct immediate impact on the population (Jackson *et al.*, 2010; Moslim *et al.*, 2011b; Moslim *et al.*, 2005b; Young, 1986). To control *O. rhinoceros*, the efficiency of the traps may depend on the lure/attractant, the trap design, spacing or distribution and the timing of their use.

Non-Pheromone Traps

Traps using food substrates In Vailele, Samoa, log traps were used during the 1950s to control the adult population (Cumber, 1957). The traps were constructed using 1-m-long decaying palm logs that were split longitudinally at the center. Approximately 12 split logs were arranged adjacent and parallel at an approximate distance of 25 m from the affected palm plantations. Adult beetles were attracted to these log traps after sunset and were collected and removed every 2 d. Occasionally during rainy days, the adults remained at the palm crown. For this method to prove successful, other breeding sites should

be eliminated from the target location, and trap activity should be closely monitored (Stride, 1977). Although the split-log trap design was not effective in Nigeria, a modified design attracted more adults (Hoyt, 1963). In Palau, Gressitt (1953) also noted that the split-log traps were not effective if compost pits or other attractive breeding sites were abundant at the trap location. In southern India, castor cake in combination with starch water is used as an indigenous technique adopted by growers to attract the adults to the bait. The fermented starch serves as the attractant, and the toxic alkaloids in the castor cake kill the beetles (Swapna and Ahamed, 2005). Previous studies have reviewed the use of carbon bisulfide, rotten vegetables, compost pits, sawdust, fermented garbage water, green petiole leaves, coconut water, coconut debris, ragi water, yeast and acetic acid and bait traps using other host plants (Gressitt, 1953; Rajamanickam *et al.*, 1992; Stride, 1977).

Light Traps Adults are only moderately attracted to light traps even at high population densities (Gressitt, 1953). In Yemen, Al-Habshi *et al.* (2006) used light traps to monitor the population dynamics of *O. rhinoceros* adults in the field. However, further verification is required to confirm that the species in Yemen was indeed *O. rhinoceros* and not *O. elegans* or *O. agamemnon* that are readily attracted to light and widely reported in the Arabian Peninsula (Bedford, 2014). In laboratory experiments in Hawaii, *O. rhinoceros* adults were attracted to ultraviolet light-emitting diodes, UV LEDs (Moore, 2013).

Non-Pheromone Attractants Prior to the discovery of the aggregation pheromone in *O. rhinoceros*, several synthetic attractants were evaluated for trapping adults, the most promising of which was ethyl chrysanthemumate (Barber *et al.*, 1971; Maddison *et al.*, 1973; Meer *et al.*, 1979). This attractant was compatible with food substrates for trapping breeding *O. rhinoceros* adults (Young, 1986; Zelazny and Alfiler, 1987). However, the popularity of this product lasted only until the commercial synthesis of a more effective aggregation pheromone, E4-MO.

Pheromone Traps

After its commercial production, the male-secreted aggregation pheromone, ethyl 4-methyloctanoate, became the most important attractant used for trapping *O. rhinoceros* adults on coconut and oil palm plantations (Bedford, 2013; Muñoz *et al.*, 2009; Ragoussis *et al.*, 2007). In 1994, the compound was identified from *O. monoceros* and later described as a major pheromone in adult *O. rhinoceros* males (Allou *et al.*, 2006; Gries *et al.*, 1994; Hallett *et al.*, 1995; Morin *et al.*, 1996). Hallett *et al.* (1995) determined that ethyl 4-methyloctanoate was preferred 10 times more often by *O. rhinoceros* adults than the non-pheromone attractant ethyl chrysanthemumate. The racemic

ethyl 4-methyloctanoate and the (S)-stereoisomer of the aggregation pheromone attracted the adult beetles equally; therefore, the chemical was characterized as ethyl (S)-4-methyloctanoate (Hallett *et al.*, 1995). Commonly used abbreviations include ethyl 4-me-octanoate and E4-MO (Bedford, 2013; El-Sayed, 2007).

Efficiency of the Pheromone Lure The efficiency of the lure may depend (among other parameters) on the dosage, longevity, trap design, trap placement, trap density, additives and abiotic factors. After identification of the aggregation pheromone, initial studies examined various rates of pheromone release in the lures. Although the capture rate increased with the release rate of the pheromone in a vane trap, the ratio of the impact to the release rate gradually plateaued above 6 mg/day (Hallett *et al.*, 1995). At present, commercial formulations are available with different dosages and release rates (BCRL, n.d.; ISCA, 2006; Sime Darby Plantation, n.d.). In many southeast Asian countries, a pheromone lure sachet that contain 800 mg active ingredient is used to trap the beetles (Loring, 2007). In a Thai study, Loring (2007) compared 2 commercial pheromone dispensers and noted that the lures were equally attractive. In Guam, initial studies indicated that the traps were not successful in mass capturing adults, but were a useful survey tool for detecting and monitoring the spread of the beetles (Moore, 2011). However, subsequent studies have focused on increasing the trap efficiency. In a preliminary laboratory study, Moore (2013b) noted that the release rate of a lure changed over time and that the new lures exhibited the highest release rates during the first few hours, after which rates plummeted. This result may indicate that slow and steady release may provide lures with similar or higher efficiencies than standard lures. In a pilot assay, Moore (2013c) found no significant difference in the trap catch for the lures with standard (14.32 mg/day) and slow (1.41 mg/day) rates of pheromone release.

The efficiency of a trap also depends on the lure longevity, which appeared relatively consistent across several studies. A survey in Malaysia noted that adults were caught in the traps as early as the second day and that the traps continued to attract for 6–9 weeks (Fee, 1997). The guidelines from a commonly used slow-release formulation recommends replacement of the lures every 8–10 weeks (Sime Darby Plantation, n.d.). In Kerala, India, Sujatha *et al.* (2002) confirmed that the pheromone lures were effective for approximately 8 weeks and that the trap catches were highest in the second and fourth weeks. A study in Karnataka, India, tested the efficiency of 2 commercial pheromone lures and reported lure longevities reaching 10 and 14 weeks; however, a weekly breakdown of the captures was not available to examine the efficiency of the traps over time (Swamy and Puttaswamy, 2004).

Some commercial formulations reportedly release the pheromones more efficiently and last slightly longer, approximately 11 weeks (Loring, 2007). Thus, the longevity of the traps could be extended.

The efficiency of the traps also depends upon environmental conditions. Based on preliminary studies conducted as part of the *O. rhinoceros* eradication program in Guam, Moore (2013b) noted that the lure longevity may depend on wind, sunlight exposure and rainfall. A study at the oil palm plantations in southern India indicated that even a slight increase in the maximum daytime temperature above 33.5 °C impacted the efficiency of the pheromone lures and reduced the number of beetles caught in the traps, which may be due to the volatilization of active pheromone ingredient. Variations in nighttime temperature had no impact on the lure captures. The lures lasted longer during the winter than the summer (Kalidas, 2004). Small landowners may be able to reduce costs by placing the lures in the trap only during the evening, thus eliminating excessive dissipation of the pheromones during the day (Desmier *et al.*, 2001). Furthermore, some pheromone formulations may be relatively more efficient in regulating their release at high temperatures (Loring, 2007). In contrast, relative humidity exhibited a positive correlation with the number of beetles trapped (Kalidas, 2004), an observation that may not be due to the attractiveness of the pheromone but to the behavior of the beetles. In general, flight activity increased during wet weather (Kamarudin and Wahid, 2004). Moore (2013b) indicated that rainwater may enter the lures via capillary action and interfere with the trap efficiency.

Increasing the trap density may also enhance the level of control; however, at low densities traps may be ineffective, and at high densities they may attract additional pests to the plantation increasing the damage (Fee, 1997; Sujatha *et al.*, 2002). A study on the oil palm plantations in Malaysia indicated 1 trap/2 ha as the optimum density for pheromone traps; at this density, the traps captured 5.72 adults per week (Fee, 1997). In Tamil Nadu, India, the pheromone traps are typically placed at a density of 1–2 traps/2 ha (TNAU, n.d.). A Malaysian study compared the impact of normal (1 trap/2 ha) and high (11 traps/ha) densities on *O. rhinoceros*: At normal trapping density, the *O. rhinoceros* larval population declined and stabilized below 10 larvae/m^2. At a high trapping density, the population declined below 10 larvae/m^2 for 16 months, after which no new individuals were detected (Kamarudin *et al.*, 2007). A popular slow-release commercial formulation of the pheromone recommends 1 trap/2 ha (Sime Darby Plantation, n.d.). Loring (2007) indicated that these trap densities may be ineffective at high pest population densities; thus, based on the extent of infestation, the number of traps should be increased. Although the trap density influences the number of insects

trapped, other factors such as lure formulation and seasonal fluctuations also contribute. A study in 2000–2002 tested the efficiency of a pheromone lure in Kerala, India, and revealed that the average trap counts were low at 3.3 adults per month, but the traps caught more beetles, 18 per month, from March to June (Sujatha *et al.*, 2002). Swamy and Puttaswamy (2004) tested the efficiency of 2 commercial pheromone lures in southern India. Twelve traps at a density of 1 trap/2 ha captured 196 (lure 1) and 508 adults (lure 2) in 10–14 weeks. The study found differences in the sex ratios of the adults captured using the aggregation pheromone traps. Some traps caught more females than males (Jayanth *et al.*, 2009; Sujatha *et al.*, 2002); however, the sex ratio was approximately 1:1 in most studies and male biased in some (Sakthivel *et al.*, 2008; Swamy and Puttaswamy, 2004). A 10-month multi-state study in southern India captured approximately 13,000 adults in pheromone traps, approximately 8,500 of which were females; many of these were gravid (Bhanu *et al.*, 2012; Jayanth *et al.*, 2009). Female flight activity increases when they actively search for breeding sites; therefore, the sex ratio bias at the time could have been behavioral and was not due to differences in pheromone attractiveness (Kamarudin and Wahid, 2004).

Trap design After the adult senses the semiochemical, it flies toward the pheromone source, hits the barrier in the trap and drops into a 13–20-L bucket where it is killed using compatible insecticides or drowned in water (Loring, 2007; Swamy and Puttaswamy, 2004). Hallett *et al.* (1995) compared 3 trap designs and determined that vane traps were more effective than pitfall or simple barrier traps; the cross vanes provided efficient barriers (Hallett, 1996; Hallett *et al.*, 1995). In a more recent review, Bedford (2013) indicated the use of 4 trap types to manage *O. rhinoceros* using pheromone lures—plastic bucket traps, parabolic traps, single- or double-vane barrier traps and a PVC tube trap. Several other trap designs are available from the guidelines for commercial pheromone formulations and previous studies (BCRL, n.d.; ISCA, 2006; Jackson *et al.*, 2010; Sime Darby Plantation, n.d.).

For optimum capture rates, Fee (1997) suggested the use of pheromone lure sachets in black-painted vane traps suspended from wooden stands at a height of 3 m. The study indicated that the black-painted vane traps captured 1.5 times more adults than the non-painted vane traps. Kalidas (2004) later suggested that the vane bucket traps needed improvement as they allow some beetles to escape, eventually leading to high infestations in the field. Although Oehlschlager (2005) indicated that the beetles may not be able to escape if the cross vanes extended inside to within 5 cm of the bucket base, this was not confirmed in subsequent studies. Moore (2014c) observed that some of the beetles were able to escape by vertically ‘helicoptering’ from the traps, which

may occur even if the vanes are extended to the base of the bucket. As part of the *O. rhinoceros* eradication program in Guam and Hawaii, researchers are testing modified and improved traps using 'minibuckets' and barrels covered with plastic tops, bird netting or chicken wire (Moore, 2013a, 2014a, 2014e, 2014f, 2014h; Moore and Quitugua, 2014a).

No consensus has been reached on trap height and placement. In Kerala, India, the pheromone lures were installed in vane bucket traps at 8 m to match the palm crown height (Sujatha *et al.*, 2002). In Karnataka, India, pheromone traps were attached to the palm trunk at a height of 2.5 m during a study to evaluate multiple commercial pheromone formulations (Swamy and Puttaswamy, 2004), whereas in Tamil Nadu, India, Sakthivel *et al.* (2008) secured the pheromone trap to a tree trunk using wire at a height of 1 m. In an area-wide study in southern India, the traps efficiently caught adult beetles at a height of 5 m; the sex ratio of trapped adults was female biased (Jayanth *et al.*, 2009). Bhanu and Chandrasekharaiah (2013) described effective mass trapping at a rate of 2/ha, when the traps are placed at chest height on the coconut palms. In Indonesia, Hallett *et al.* (1999) determined that the traps were efficient at either ground level or 2 m. In Malaysia, a height of 1.5–2 m was typical (Kamarudin and Wahid, 2000), but Oehlschlager (2005) reviewed the use of vane traps at canopy level. Although previous studies have used coconut palms to secure the traps, non-host stands are preferred for trap placement to avoid attraction to the host when a beetle approaches the pheromone lure.

Previous studies are not comparable due to variations in experimental design, location and associated parameters; however, reasonable assumptions could be drawn based on *O. rhinoceros* flight activities at a location. In his recent review, Bedford (2013) poses several questions for future research such as whether the males release more pheromone at the breeding sites or the mating sites. The answers to these questions may aid in streamlining trap placement and target specific behavioral phases of the pest. *Oryctes rhinoceros* flight activity and movement patterns are now tracked through visual observations, infrared trail cameras and radio using miniature radio tags.

Implementation, impact and integration Areas replanted with young oil palms may be more susceptible to pest infestations than those with older mature palms. In Malaysia, the Palm Oil Board recommends the use of pheromone traps 6–12 months prior to replanting, *i.e.*, approximately 6 months after the old palms are cut down inside the block. The removal of aging oil palms and the zero-burning policy increase the availability of breeding substrates, thus the *O. rhinoceros* population. Therefore, trapping focuses on reducing the number of *O. rhinoceros* already existing inside the block and simultaneously

trapping at the borders of the replanting block to reduce the number of beetles that may immigrate from adjacent mature palms. The pheromone traps are placed in 2–3 tiers encircling the replanting block—one at the immediate boundary of the replanting block and the others approximately 15 palms inside the block. The distance between the tiers and traps is equivalent to the required plant-to-plant distance in a row of 15 oil palms. The trap density is increased if the pest damage and populations are higher in the surrounding areas. Field studies in Malaysia indicated that the number of adults captured using pheromone traps correlated with a reduction in palm damage and that additional gravid females were trapped at the borders; these females may have been searching for breeding sites (Kamarudin and Wahid, 2000, 2004). Placement of traps at the border is only effective in the absence of infested breeding sites within the block (Desmier *et al.*, 2001; Loring, 2007).

Pheromone traps can effectively capture adults in both oil palm and coconut plantations (Jayanth *et al.*, 2009; Muthiah and Mohan, 2002; Sakthivel *et al.*, 2008). Oehlschlager (2005) noted that trapping at a rate of 1 pheromone lure/2 ha reduced oil palm damage by more than 90 % and further suggested that trapping was less expensive and more effective than chemical control. In the Middle East, more than 50,000 ha are mass trapped annually (Wraight and Hajek, 2009). The traps are also critical components of integrated pest management programs. In West Malaysia, Fee (1997) noted that mass trapping using the pheromone was only effective at low *O. rhinoceros* densities. However, pheromone traps may prove beneficial even at high pest densities if used in conjunction with biological control agents such as OrNV or *M. majus*. Just as multiple strategies are adopted to efficiently manage a pest, a strategy can target multiple pests. In many coconut plantations worldwide, *O. rhinoceros* and *R. ferrugineus* co-exist, necessitating a set of management strategies with a broader impact. In a field study, Hallett *et al.* (1999) demonstrated that the effectiveness of 2 lures did not diminish when placed in the same trap to attract both *O. rhinoceros* and *R. ferrugineus*. However, the flight activity of the 2 beetles may not coincide (Chakravarthy *et al.*, 2014). For example, in southern India, *R. ferrugineus* traps are recommended from October to December, whereas the preferred trapping period for *O. rhinoceros* is from September to February. The population dynamics and flight activity of the rhinoceros beetle can vary; ideally, the time and duration of trapping should be based on the pest–host dynamics at a given location, which is likely to be unknown.

Moore (2014c) recently reported that the use of pheromone traps alone may not reduce insect population; on the contrary, palms in the trapped areas continue to become damaged suggesting that the hosts may be more attractive

than the pheromone lures. A release and re-capture study supports this assumption as none of the marked beetles were re-captured in pheromone traps at the release location. At present, the University of Guam in collaboration with the USDA-ARS is investigating new semiochemical candidates using gas chromatography–mass spectrometry (GC-MS) and electroantennograms. Previous studies have also analyzed the cost of pheromone trapping (Loring, 2007).

Combination Traps

Combination traps were tested as part of the coconut rhinoceros beetle eradication program in Guam and Hawaii (Moore, 2014c). Some commercial pheromone formulation guidelines have also mentioned the use of pheromones in combination with other attractants (BCRL, n.d.).

Food and Light As part of the eradication program in Guam, Moore (2013a, 2013e) investigated the use of barrel traps. Large 208-L barrels filled with a breeding substrate and coupled with solar-powered UV LEDs were used to attract *O. rhinoceros* adults. A chicken wire covering on the barrel allowed the adults to land and enter the barrel, but discouraged the insects from leaving. A preliminary study suggested that this food-based trap may be more effective than the pheromone lures. Furthermore, the addition of UV LEDs increased adult capture by 50 %, and a comparison indicated that UV LEDs were more effective than white LEDs (Moore, 2014c).

Pheromone and Food In Indonesia, Hallett *et al.* (1995) and Sudharto *et al.* (2001) demonstrated that the aggregation pheromone is more effective when combined with the empty or rotting oil palm fruit bunches, indicating potentially synergistic interactions between the pheromone and the early products of fruit-bunch fermentation. Bhanu and Chandrasekharaiah (2013) suggested using a combination of the outer layers of tender coconut, detergent and water to attract and drown *O. rhinoceros* adults. High pheromone doses can be expensive, but the use of pheromones in combination with readily available attractants may allow reduced dosages (Gunawardena, 2014).

Pheromone and Light In a field trial, Moore (2013c, 2013e) evaluated the use of solar UV LEDs with slow-release pheromone lures (Figure 8-11). The addition of the light source enhanced the trap catches of *O. rhinoceros* 3-fold (Moore, 2013d). In 2014, these panel traps were deployed in Oahu, HI, to survey and trap the coconut rhinoceros beetle (Hawaii Department of Agriculture, 2014a). A ‘minibucket’ trap with a pheromone lure and UV LED is also being tested for efficacy (Moore, 2014c).

Figure 8-11 Traps and components used to monitor and control *O. rhinoceros* in Guam: (A) minibucket trap; (B) standard pheromone trap with UV LED; (C) trap components—slow release pheromone and UV LED; (D) solar powered and UV LED-fitted vane pheromone trap (photo courtesy of Aubrey Moore, University of Guam)

Food, pheromone, and light In Guam, a combination of food, pheromone and light is used to lure and trap beetles in large barrels (Hara, 2014; Moore, 2014c). The traps are prepared by filling the barrels with palm residue up to ~15 cm below the opening, which is covered with chicken wire. The 15-cm distance prevents the beetle from climbing out of the barrel, and the chicken wire prevents escape of the flying adults (Moore and Quitugua, 2014b). The traps and some modifications are presented in Figure 8-12.

Figure 8-12 Barrel trap and components used in Guam: (A–C) barrel filled with palm residue; (D) UV LED placed above the trap; (E) a pan is sometimes used to collect the beetles; (F) pheromone lure (photos courtesy of Aubrey Moore, University of Guam [A–C]; Arnold Hara, University of Hawaii [D–F])

Phytosanitary Measures

Dead standing palms, stumps, sawdust, compost and rotting organic matter all serve as substrates for *O. rhinoceros* larval development. For further information regarding suitable breeding sites, refer to the Biology section on page 4-1. Refer to Table 8-4 for substrate management strategies adopted at various locations. Considering the larval development period, infested locations should be surveyed at least every 2 months to detect and eliminate breeding substrates (Gressitt, 1953).

Table 8-4 Management of substrates to prevent *O. rhinoceros* adults from breeding at specific sites

Substrate	Management	References
dead standing palms, other logs	use for construction; cut, split, stack, dry and burn; soak in water	Gressitt (1953), Philippine Coconut Authority (1998b), Stride (1977)
palm stumps	grow cover crops over stumps; treat with insecticides; treat with *M. majus* (200-g suspension in 16 L water)	Philippine Coconut Authority (1998b, 2005), Stride (1977)
rotting organic matter, palm residue	treat with *M. majus* or thinly scatter and mix with soil; construct incinerators for disposing infested material if needed; chip for use as feedstock for composting; fumigate with methyl bromide and bury immediately	Gressitt (1953), Moore (2012a), Muthiah and Mohan (2002), Philippine Coconut Authority (1998b), Stride (1977)
compost, farmyard manure	treat with *M. majus*; should not be placed inside palm plantations; prepare deep pits with small surface area, cover or screen to prevent oviposition; turn at regular intervals and manually remove larvae	Catley (1969), Gressitt (1953), Muthiah and Mohan (2002), Philippine Coconut Authority (1998b), Stride (1977)
sugarcane bagasse	thinly scatter and mix with *M. majus;* feed for livestock; burn	Philippine Coconut Authority (1998b, 2005)
corn cobs	thinly scatter and mix with soil	Philippine Coconut Authority (1998b)
rice straw heaps	thinly scatter and inspect regularly	Philippine Coconut Authority (1998b)
garbage	burn or bury weekly	Stride (1977)
sawdust	thinly scatter and mix with *M. majus*; burn	Gressitt (1953), Philippine Coconut Authority (1998b), Stride (1977)

Burning is among the preferred methods for disposing of potential substrates. However, this method can prove unsuitable depending upon environmental conditions, government policies or the substrate condition. For example, dry spells are brief in Samoa, which prevents the processing of fresh palm residue prior to burning; thus, logs were soaked in water and sometimes cast off into the open sea; however, the effectiveness of this method is unknown as the larvae can survive submerged in seawater for more than 48 h (Catley, 1969; Gressitt, 1953; Nirula *et al.*, 1952). In Malaysia, the zero-burning policy established in the 1990s for the sustainable management of oil palm plantations interferes with phytosanitary measures. Although environmentally desirable, palm residue may not be removed from the oil palm plantations even if infested (Ahmad, 2006; Darus and Basri, 2000). Burning is also unsuitable for heaps of partially composted materials. In Guam, alternative treatments such as the on-site

application of insecticides or entomopathogens, burial and modified compost management are being examined. For the latter, the infested materials should be transported to a large-scale composting facility that produces high temperatures to kill the different *O. rhinoceros* stages, which requires the construction of large-scale infrastructure creating a limitation at some locations (Moore, 2012a). Grinding is considered an alternative disposal method for heavily infested breeding substrates, but this too depends on the availability of equipment capable of processing large quantities of organic matter (Hawaii Invasive Species Council, 2014b). In Hawaii, the infested debris from the breeding sites was excavated and ground using 'tub grinders,' then returned to the excavation sites, placed on plastic sheeting and covered with a net to allow the beetle to enter, but prevent their escape. This method kills the different life stages during grinding and further provides a 'substrate trap' to lure and trap the visiting beetles (Navy Region Hawaii, 2014). Ideally, the ground debris should be incinerated (USDA-APHIS, 2014b).

Although previous studies have recommended deep burial, this method is often ineffective as the emerging beetles can tunnel through the soil (Gressitt, 1953). As an alternative, the palm residue could be fumigated prior to burial, but this method is expensive and environmentally unfriendly (Moore, 2012a). Other methods of sterilization such as solarization were examined, but *O. rhinoceros* easily survived the daily maximum temperatures; therefore, these methods were not pursued further (Moore, 2009, 2014b). The impact of steam sterilization requires further examination (Hawaii Invasive Species Council, 2014b). The coconut rhinoceros beetle larvae can withstand high temperatures, and preliminary studies suggested that the LTe_{50} for the third instars was approximately 47 °C (Moore, 2014b). Therefore, the lowest optimum temperature for killing the larvae via composting operations is being investigated in Hawaii; substrates are being treated, composted and re-treated at high temperatures of 55–77 °C (Hawaii Invasive Species Council, 2014c).

Cultivation practices, natural disasters, and an abundance of hosts can also interfere with phytosanitary measures. For example, underplanting is a replanting method utilized in oil palms in which young palms are planted under aging palms that are due to be felled in the following years; however, destruction of the infested mature palms prior to replanting is preferred. In some situations, natural disasters may interfere with the sanitation of plantations; these may leave an abundance of uprooted palm trees (Figure 4-2-I) that can potentially serve as oviposition sites (Bedford, 2013; Monty, 1978). At times, the elimination of alternate beetle hosts may prove impossible. *Pandanus* spp. is an important beetle host in Palau, but it is also economically important and abundant. In such cases,

scouting for infestation and selective elimination may provide a reasonable approach (Gressitt, 1953).

When removal is impossible, the substrates should be treated with insecticides or the entomopathogen, *M. majus* (Chong *et al.*, 1991; Murphy, 2007; Stride, 1977). Currently, no available insecticides are effective against all stages of the insect; therefore, this method is unreliable for eradication (Hawaii Invasive Species Council, 2014b). Alternative methods were explored in some locations: leguminous cover crops are grown over palm residues as barriers to obscure the breeding sites from the adults (Vargo, 2000; Young, 1986). In the Philippines, intercropping utilizes the space between the palms, thus reducing the accumulation of breeding substrates on unused land (Philippine Coconut Authority, 1998b). Replacement of isolated older unproductive palms with younger trees in a coconut plantation can prevent selective infestations by *O. rhinoceros* and potentially improve the overall health of the plantation (Vargo, 2000). Another 'green' technique involves chipping the entire mass of palm residue to use as feedstock for later composting operations. However, this method requires the infrastructure for large-scale composting (Moore, 2012a). Educating and involving the community in phytosanitary measures proves necessary in many cases (Moore, 2012a; Nair *et al.*, 1998; Peter and Kenmore, 2005; Secretariat of the Pacific Community, 2004; Young, 1986).

Mechanical Control

The adult beetles can be manually removed from the palm crowns, axils and short borer holes using a rod or wire-hook assembly—a 50-cm-long iron rod or wire with a hook at its end (Cherian and Anantanarayanan, 1939; Muthiah and Mohan, 2002). This method is also known as 'beetle winkling,' and is recommended at weekly intervals (Chong *et al.*, 1991). In southern India, the use of beetle hooks is recommended from June–September when the rhinoceros beetle adult population peaks (Nair *et al.*, 1998). By sorting materials at the breeding sites, all *O. rhinoceros* life stages can also be manually collected and removed from breeding sites; however, this method is laborious and may have only limited impact on the overall population (Muthiah and Mohan, 2002).

Chemical Control

Oryctes rhinoceros populations can be regulated by limiting the availability of larval breeding substrates. The substrates could be eliminated through phytosanitary measures, but not in all locations due to 'zero-burn' policies aimed at reducing air pollution. In some cases, treating the breeding sites with insecticides may be preferred (Dhondt *et al.*,

1976; Howard, 2001). Bedford (2014) noted that chemicals may not be useful for eradication efforts and may complement IPM strategies more.

Juveline Hormone (JH) analogs

The JH analogs used as insect growth regulators (IGRs) can interfere with the action of naturally occurring JH, with the highest impact occurring at metamorphosis. The JH analogs may reduce egg hatch, increase adult sterility or increase pupal mortality (Wilson, 2004).

Methoprene

Dhondt *et al.* (1976) tested 53 JH mimics and demonstrated that pupal mortality was highest for methoprene (isopropyl (2E, 4E)-11methoxy-3,7,11-trimethyl-2-4 dodecadienoate). In contrast, a recent lab study by Moore (2012a) indicated that methoprene was ineffective.

Pyriproxyfen

Moore (2012a) demonstrated that pyriproxyfen (2-[1-methyl-2-(4-phen-oxyphenoxy) ethoxy] pyridine) prolongs the third instar stadium and prevents pupation. To reduce adult emergence in *O. rhinoceros*, Moore (2012a) recommends treatment of the palm stumps or other breeding sites with pyriproxyfen (10% AI) at a maximum rate of 56 mL per 190 L water using backpack sprayers.

Although JH mimics have low non-target toxicity and high target specificity, they lack an ability to rapidly impact an insect population. (Wilson, 2004).

Organophosphates

Phorate

Phorate (O,O-diethyl S-[(ethylsulfanyl)methyl] phosphorodithioate) utilizes a cholinesterase inhibitor mode of action (Stenersen, 2004) and is commonly used against *O. rhinoceros* in Southeast Asia. Phorate granules (10G) can be placed in perforated sachets (5 g each, twice per 6 months) in the inner leaf whorls to reduce leaf damage by adult beetles (Rajamanickam *et al.*, 2002). Sometimes, phorate is used sequentially with naphthalene balls, neem seed kernel powder and carbofuran to maximize control and slow the development of insect resistance (Kumar and Ahmad, 2008; Muthiah and Bhaskaran, 2000; Rajamanickam *et al.*, 2002).

Trichlorfon

In Mauritius, trichlorfon (RS-dimethyl (2,2,2-trichloro-1-hydroxyethyl) phosphonate) was extensively applied (2% granules) to the youngest frond axils of coconut palms. Treatment of breeding sites was not recommended due to possible non-target impacts (Monty, 1978).

Chlorpyrifos

As part of the *O. rhinoceros* eradication program in Guam, chlorpyrifos (0,0 diethyl 0-(3,5,6 trichloro-2-pyridinyl) phosphorothioate) was sprayed into the bored holes and frond axils (23% AI, 0.5% solution) and applied to felled palm stumps (21.4% AI, 0.23% solution). The insecticide residues in the soil and plants were expected to remain active for 2 y after application (Berringer, 2007). Subsequent studies suggested that this insecticide was ineffective and therefore was not pursued further for the eradication program (Moore, 2012a).

N-Methyl Carbamates

Carbaryl

Carbaryl (1-naphthyl N-methyl carbamate) is used to control *O. rhinoceros* in some locations (Pardede and Utomo, 1992). Rajamanickam *et al.* (2002) reviewed a 1:1 (or 1:2) mixture of carbaryl (5% D) and sand placed in the palm leaf axils (50 and 100 g per palm) as an effective prophylactic measure against beetle infestation. However, a further evaluation suggested less impact with the following ranking of effectiveness: phorate > naphthalene > carbofuran > carbaryl. As part of the coconut rhinoceros beetle eradication program in Guam, carbaryl (43% AI solution; 10G) was originally evaluated as an alternative to the no-action plan (Berringer, 2007). Preliminary studies suggested that this insecticide was not as effective and was therefore not pursued further (Moore, 2012a).

Carbofuran

Carbofuran is widely used in some south Asian countries against the coconut rhinoceros beetle (Darus and Basri, 2000; Kamarudin and Wahid, 1997; Kumar and Ahmad, 2008; Muthiah and Bhaskaran, 2000; Padmasheela and Krishnan, 1996; Rajamanickam *et al.*, 2002; Stenersen, 2004). Although 12 products (US EPA PC code=090601) are currently registered in the U.S., the EPA has now concluded that the risks with its usage are unacceptable at any level and has published a notice of intent to cancel the registration (EPA, 2011; Kegley *et al.*, 2010).

Synthetic Pyrethroids

Tefluthrin, bifenthrin, permethrin, fenpropathrin, cyhalothrin, cypermethrin, cyfluthrin, flucythrinate, fenvalerate, deltamethrin, fluvalinate, allethrin, lamda cyhalothrin and tetramethrin were previously evaluated for use against *O. rhinoceros* (Darus and Basri, 2000; Faridah *et al.*, 2003). Darus and Basri (2000) reviewed 2 studies, one of which indicated that lamda-cyhalothrin was most effective against the pest on young oil palm plantations, and the other reported that both lamda-cyhalothrin and cypermethrin impacted the beetle even at low concentrations. Most trials used cypermethrin as the pyrethroid of choice. The eradication program in Guam evaluated bifenthrin, but was later discarded because this pyrethroid was relatively less efficient (Berringer, 2007; Moore, 2012a).

Cypermethrin

Cypermethrin ($C_{22}H_{19}Cl_2$ NO_3) is widely used to control the coconut rhinoceros beetle. In nursery and field trials, cypermethrin treatment significantly reduced the beetle population after 11 weeks and was effective even at low doses (Darus and Basri, 2000). In Malaysia, Oehlschlager (2005) reviewed the application of cypermethrin on each palm at biweekly intervals. Faridah *et al.* (2003) notes that severely impacted areas can be selectively and regularly sprayed with cypermethrin at 0.05% at biweekly intervals. The cypermethrin residues accumulate in the digestive systems of *O. rhinoceros* larvae and concentrate in their body walls. Various cypermethrin 10 EC concentrations (0.125, 0.25 and 0.5%) were evaluated by Venkatarajappa (2001), with the most residue detected in the body wall from the 0.25% solution, and the least from the 0.5% solution. In this study, toxicity was highest during the first 12 h and declined to a minimum in 24 h.

Cypermethrin (beta, 25.3% AI, EC) is being extensively used in Guam as part of the *O. rhinoceros* eradication project (Figure 8-13) (Moore, 2012a), and may be the ideal candidate for chemical treatment (Moore, 2014i). Borer holes, frond axils, palm stumps and breeding sites including large compost piles were treated with cypermethrin; a maximum 0.1 % EC was used for all treatments. The primary limitation of this pyrethroid was its rapid degradation in the environment, which necessitated frequent re-treatment of the breeding sites (Moore, 2012a).

Currently, 143 active products containing cypermethrin (US EPA PC code 109702 (beta); code 129064 (zeta)) are registered in the U.S. with formulations including an emulsifiable concentrate (EC), water-soluble powder (WSP), wettable powder (WP), suspension concentrate (SC), soluble concentrate (SL), granular (G), water-dispersible granule (WDG), dust (D), technical-grade,

pressurized liquid, impregnated material, formulation intermediate and ready-to-use solution (Kegley *et al.*, 2010). Cypermethrin is registered for foliar application in food and feed crops, as a soil residual insecticide against structural pests and for direct application in animal husbandry (EPA, 2008; Kegley *et al.*, 2010). Like other synthetic pyrethroids, cypermethrin has low mammalian toxicity and a short environmental persistence (Faridah *et al.*, 2003).

Figure 8-13 Application of cypermethrin to young palm crowns (photo courtesy of Aubrey Moore, University of Guam)

Neonicotinoids

Although imidacloprid was initially recommended for the integrated eradication program in Guam, the neonicotinoid—along with carbaryl, chlorpyrifos and bifenthrin—was subsequently determined to be ineffective (Berringer, 2007; Moore, 2008a, 2008b, 2012a).

Insect Repellants

Along with phorate and carbofuran, naphthalene balls were widely used in southern Asia to repel *O. rhinoceros* adults from coconut palms. In southern India, placing naphthalene balls (3 g each) at the base of the leaf sheath at the rate of 3 per palm over 45 d intervals proved effective (Sadakathulla and Ramachandran, 1990). In Indonesia, naphthalene balls placed at the axils of palm fronds (5 balls per palm, 14 d interval) provided up to 97% control (Pardede and Utomo, 1992). Similarly in Malaysia, naphthalene balls in the frond axils yielded over 95% control, but at high beetle densities was ineffective (Darus and Basri, 2000). In Guam, the use of naphthalene balls did not have any impact on the insect population or infestation levels (Moore, 2014i). Naphthalene has a moderate level of acute toxicity and is possibly carcinogenic; therefore, some health risks are associated with its usage (EPA, 2003; Kegley *et al.*, 2010).

Fumigants

In Guam, methyl bromide was used as a fumigant to sterilize large volumes of *O. rhinoceros*-infested breeding substrates (Moore, 2012a). Although 164 products (US EPA PC codes 053201, 853201) are currently registered in the U.S., the EPA completed a phaseout of this fumigant in 2005 and at present, its use is restricted to critical-use exemptions as defined by the Montreal Protocol (EPA, 2014; Kegley *et al.*, 2010).

Botanicals

Some indigenous techniques adopted by the farmers in India were reported to be highly effective against larvae and adults. These treatments were predominantly used in combination or as a follow-up treatment. Neem oil, neem seed kernel powder (Padmasheela and Delvi, 2002; Rajamanickam *et al.*, 2002), dried cakes of *Hydnocarpus wightiana* Blume plants (Swapna and Ahamed, 2005) and powdered *Tephrosia purpurea* (L.) can be applied at leaf axils against the adults (Unnikrishnan Nair, 2012). Powdered leaves of *Clerodendron infortunatum* L. (Unnikrishnan Nair, 2012), *Chromolaena odorata* (L.) (Leena *et al.*, 2008), *Eupatorium odoratum* L. (Sreelatha and Geetha, 2008, 2010), *Adhatoda vasica* Nees, *Gliricidia maculate* (Humb., Bonpl. & Kunth) Steud. (Sreelatha *et al.*, 2011), *Ailanthus malabarica* DC. (Swapna and Ahamed, 2005), *Mikania micrantha* Kunth (Zhong *et al.*, 2012), and methanol extract of *Annona squamosa* L. leaves can be applied at the breeding sites against the larvae of *O. rhinoceros* (Sreelatha and Geetha, 2008, 2010).

Labeling

Although a proposed formulation may be approved for an effective eradication or control program, it may not be labeled, at the time of pest detection, for the specific use required. If a formulation is not labeled for the necessary use, one can request a Federal Crisis or Quarantine Exemption from the EPA under section 18 of FIFRA. For further information, refer to Regulatory Procedures on page 7-1. The prescribed formulation must be labeled for use on the site at which it is to be applied and must be registered for use in the state in which the eradication program is occurring. All applicable label directions must be followed, including requirements for personal protection equipment, maximum treatment rates, storage and disposal.

Host Resistance

Host plant resistance is not currently used as a strategy against *O. rhinoceros*; however, evidence suggests a host preference by the beetle. In Kerala, India, out of 5 different banana cultivars, only the pseudostems of 'Nendran' and 'Njalipoovan' were infested (Sivakumar and Mohan, 2013). A study of the insect damage on banana fruits indicated that the beetle preferred smooth-skinned high sugar content varieties (Sharma and Gupta, 1988). In coconut, mature palms are preferred to younger ones. The insect also reportedly prefers coconut cultivars from specific locations (Nirula *et al.*, 1952). In a different study, Muthiah and Bhaskaran (2000) screened different cultivars of coconut and determined that West Coast Tall has the lowest, 7.7%, leaf damage, whereas; the Malaysian Yellow Dwarf had the highest, 15.4%, leaf damage. For further information about preferred hosts refer to the 'Hosts' section in Chapter 3, Biology.

Integrated Pest Management (IPM)

The following strategies were the most commonly adopted at locations infested with *O. rhinoceros* (Table 8-5). Further information regarding these categories is available from their respective sections.

1. Phytosanitary Measures—to eliminate breeding substrates; the most important strategy for eradication
2. Trapping—mostly useful for survey; some reduction of adult population
3. *Oryctes rhinoceros* Nudivirus—highest impact on adult population
4. Green Muscardine Fungus—to control the larval population especially during wet and cold conditions
5. Chemical—cypermethrin sprays on treetops and drenching at breeding sites

Table 8-5 Integrated pest management strategies against *O. rhinoceros*

Location	Summary	Reference
Guam	traps, phytosanitary measures, cypermethrin, surveys, outreach, preliminary studies using green muscardine fungus and OrNV	Moore (2012a)
India	repellents and abrasives at leaf axils, granular insecticides at leaf axils, chemicals at breeding sites, green muscardine fungus or botanicals at breeding sites, OrNV, mass trapping, mechanical control	Bhanu *et al.* (2012), Kumar and Ahmad (2008), Nair *et al.* (1998), Unnikrishnan Nair (2012), Varma (2013), Vidyasagar and Bhat (1991)
Pacific Islands	OrNV, green muscardine fungus, phytosanitary measures, chemicals	Catley (1969), Gressitt (1953), Huger (2005), Nirula *et al.* (1955), Zelazny (1975)
the Philippines	green muscardine fungus, phytosanitary measures, OrNV, chemical, mechanical,	Philippine Coconut Authority (1998b, 2005), Zelazny and Alfiler

	cultural, mass trapping	(1987)
Malaysia	mass trapping, cover crops, OrNV, green muscardine fungus, synthetic pyrethroids, coal tar on frond rachis, mechanical control, trapping and removing breeding sites considered most important	Ahmad (2006),Chong *et al.* (1991), Darus and Basri (2000), Murphy (2007), Oehlschlager (2005)
the Middle East	mass trapping using pheromones and light traps, phytosanitary measures	El-Shafie (2014), Wraight and Hajek (2009)

Bedford (2013, 2014) noted that pheromone traps are widely used for control in oil palm plantations, but that is not the case in coconut growing South Pacific countries where the pheromone traps are only economical for monitoring of the endemic *O. rhinoceros*. Trap maintenance can be expensive overtime.

Educational outreach activities and active progress reports are also important to the integrated pest management and eradication process. In Guam and Hawaii, the recent outbreak of *O. rhinoceros* and the management strategies adopted were reported in online news channels and blogs (Kelman, 2007; Orth, 2007; Paco, 2013; Smith, 2014; Sweeney, 2008). This information also attracts comments from readers who may communicate the extent of adoption, the success of pest management methods and the socio-cultural concerns at these locations (Rumsey, 2012).

Chapter

9

Environmental Compliance

Chapter

10

Pathways

Natural Movement

Coconut palms grow along ocean shores in many locations. After infestation, some decaying palm logs may travel short distances through the sea (Gressitt, 1953; Lever, 1969). Adults can fly long distances under adverse conditions, but likely will not if breeding and feeding sites are available at the location of origin. Refer to the Dispersal section for additional information.

Human-Assisted Spread

Cargo such as timber, sawdust and copra are suitable substrates for the larvae; ships carrying infested materials can introduce *O. rhinoceros* to new locations (Gressitt, 1953; Stride, 1977). However, port interceptions and previous reports indicate that the most likely method of introduction occurs through adults that hitchhike aboard ships and flights. Early coconut rhinoceros beetle invasions in the Pacific islands possibly occurred through sea and air traffic during WWII (Catley, 1969; Nishida and Evenhuis, 2000). The beetles are active fliers at night, and containers loaded after sunset are more likely to have hitchhiking adults than those loaded during the daytime. Regulatory personnel have found beetles in empty pallets on shipments from Guam to the mainland (CRB TWG, 2014). Moore (2007) indicated that the introduction into Guam possibly occurred through gravid female stowaways aboard cargo ships originating from Asia. Gressitt (1953) and Stride (1977) reviewed a previously used method to reduce introduction through ships—the vessels were anchored at least 4 km from the shore, and shipboard infestation were detected using light traps after sunset and monitoring adult flight activity until dawn. The port interception database revealed that *Oryctes* spp. was intercepted 14 times, of which 10 reports came from airports and 2 from seaports indicating that hitchhiking aboard flights was more likely (PestID, 2014).

In Oahu, Hawaii, an adult *O. rhinoceros* was detected at the international baggage claim area of the Honolulu airport in November 2013; however, no additional adults were reported nearby at the time, and the incident was considered an isolated event. In December 2013 the first adult was reported in Hawaii outside the ports, and breeding

sites and multiple adult detections were subsequently made near the Joint Base Pearl Harbor-Hickam and the Honolulu International Airport, suggesting that the introduction may have occurred through air transport from either location (USDA-APHIS, 2014b). Using restriction fragment length polymorphism (RFLP), Moore and Marshall (2014) compared DNA samples from beetle populations in Hawaii to those in Diego Garcia, Fiji, Guam, Samoa and Papua New Guinea; results suggested that that the infestation in Hawaii may have originated from Guam. Table 10-1 lists suspected methods of *O. rhinoceros* introduction at various locations.

Table 10-1 Suspected methods of *O. rhinoceros* introduction in some locations

Location	Introduction pathway	Reference
Diego Garcia	suspected introduction on WWI troop ships	Catley (1969)
Guam	2006, 200 miles from Guam, an adult beetle captured in seaport warehouse; unrelated to the 2007 introduction in Guam; suspected hitchhiking on ships from Asia bearing construction materials	Moore (2007), Smith and Moore (2008)
Hawaii	suspected introduction from Guam via air transports at Joint Base Pearl Harbor-Hickam or Honolulu International Airport	Moore and Marshall (2014), USDA-APHIS (2014b)
Mauritius	suspected introduction via ships	Catley (1969)
Palau	suspected introduction into Koror from Asia by Japanese ships; possible transported during WWII	Catley (1969), Gressitt (1953)
Samoa	import of *Hevea* seedlings to Upolu from Sri Lanka; movement of nursery stock potential source of risk	Catley (1969)
Tokara (part of Ryukyu Archipelago)	possible introduction from Amami Ōshima Island via ferry	Hosoya (2011)
Tokelau	suspected introduction from Samoa via shipments of infested soil for vegetable production	Dale and Maddison (1984)
United Arab Emirates	introduced through imported planting material	Gassouma (2004)

After introduction, movement or availability of the substrates can rapidly spread the beetles to uninfested locations (Gressitt, 1953; Guaminsects.net, 2007b; Sweeney, 2008). In Oman, the percentage of infestation doubled when infested cattle manure was transported to meet the demands of increasing banana cultivation (Kinawy, 2004). In addition to the unintentional movement of infested substrates, the rhinoceros beetles (includes other *Oryctes* species) may have been deliberately moved due to their perceived potential for nutrition, collection and cultural amusements (Fakayode and Ugwumba, 2013; New, 2005; Okaraonye and Ikewuchi, 2009; Onyeike *et al.*, 2005; Ratcliffe, 2006). Furthermore, the beetles can fly long distances, but typically will not if breeding sites are nearby. A lack of public awareness may be a key factor in the spread of this insect (Ridgell, 2009).

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