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# Insect pests and insect-vectored diseases of palms

# Catherine W Gitau,<sup>1\*</sup> Geoff M Gurr,<sup>1</sup> Charles F Dewhurst,<sup>2</sup> Murray J Fletcher<sup>3</sup> and Andrew Mitchell<sup>4</sup>

<sup>1</sup>EH Graham Centre for Agricultural Innovation, Charles Sturt University, PO Box 883 Orange, NSW 2800, Australia.
<sup>2</sup>PNG Oil Palm Research Association, Kimbe, West New Britain, Papua New Guinea.
<sup>3</sup>NSW Department of Primary Industries, Orange Agricultural Institute, Orange, NSW 2800, Australia.
<sup>4</sup>NSW Department of Primary Industries, Wagga Wagga Agricultural Institute, Wagga Wagga, NSW 2650, Australia.

**Abstract** Palm production faces serious challenges ranging from diseases to damage by insect pests, all of which may reduce productivity by as much as 30%. A number of disorders of unknown aetiology but associated with insects are now recognised. Management practices that ensure the sustainability of palm production systems require a sound understanding of the interactions between biological systems and palms. This paper discusses insect pests that attack palms, pathogens the insects vector as well as other disorders that are associated with these pests. We re-examine the disease aetiologies and procedures that have been used to understand causality. Pest management approaches such as cultural and biological control are discussed.

Key words aetiology, Arecaceae, diagnosis, pathosystems, pest management.

### INTRODUCTION

In many cultures, palms are a symbol of splendour, peace, victory and fertility. Palms constitute one of the best-known and widely cultivated groups of monocotyledons. Their fruits, seeds, leaves (fronds) and stems are put to various uses ranging from food stuffs and versatile oils, including biofuels, to furniture and construction materials. Palm fruits are used in the production of wine and other beverages (e.g. Borassus spp. Carota urens L. and Elaeis guineensis Jacq.) and are sources of mild narcotics (Areca catechu L.). The plants are ornamentals in landscape and interior settings (numerous species). Date palm, Phoenix dactylifera L., is cultivated in the desert oases of northern Africa, the Middle East and Asia and is of significant value because of its edible fruit. The African oil palm E. guineensis produces up to seven tons of oil per hectare per year (Murphy 2007) and this versatile commodity is used in many industries including foods, cosmetics and toiletries. Coconut, Cocos nucifera L., is a source of oil, fibre, food and timber. It is also an important smallholder crop and like oil palm, significantly contributes to food security, improved nutrition, employment and income generation.

Coconut, dates and oil palm production has increased in the last decade resulting in an expansion of cultivated land, leading, in some instances, to the clearing of forests (e.g. in Indonesia). While palms are naturally distributed in different ecological zones, particularly in the tropics, human activity

\*cgitau@csu.edu.au

has transported them from their native habitats to new locations. For example, the date palm is believed to have originated in the Persian Gulf and North Africa but it is now grown worldwide in semi-arid regions (Zaid 1999). Oil palm, which originated in West and Central Africa, is now grown over large areas of South-East Asia, western Melanesia and in central and South America (Azeemoddin 1988).

High-yielding, improved palm varieties have been developed (FAO 1984; Jannot 1998; Wahid et al. 2004), most recently with the aid of DNA markers (Oropeza et al. 1999; Murphy 2007). Breeding programs for coconut, date and oil palms have resulted in the development of cultivars that are resistant to diseases and are adapted to regions in which they are cultivated (Azeemoddin 1988; Jannot 1998; Koczberski et al. 2001; Murphy 2007). However, major breeding efforts have focused on disease resistance, e.g. fusarium wilt of oil and date palm and phytoplasma diseases of coconut. Pest adaptation in changing situations has relatively been less important when selecting a cultivar. Additionally, the sustainability of cultivars in new environments has often not been taken into consideration. For instance, the performance of coconut germplasm developed for management of lethal yellowing (LY) and LY-type diseases is still a major challenge in areas where it has been introduced (Ploetz 2007). The sustainability of new cultivars is compromised by incidences of insect pests and challenges that are encountered in studying insectvectored pathogens. Studies of insect pests and associated diseases have been complicated by difficulty in confirming which insect species are disease vectors (Smith 1980a; Mariau 1994; de Franqueville 2003; Edwin & Mohankumara 2007; Philippe et al. 2007).

Despite these challenges, research into some palm diseases and their insect vectors such as LY and LY-type diseases of coconuts has received much attention (Eden-Green & Tully 1979; Howard et al. 1979; Howard 1980a,b, 1983; Kastelein et al. 1990; Schuiling & Mpunami 1992; Harrison et al. 1994, 2008; Mpunami et al. 1999; Oropeza et al. 1999). These diseases have been studied using a variety of techniques ranging from transmission experiments (Howard 1980a; Smith 1980a) to DNA-based tests (Harrison et al. 2008). However, a number of palm diseases of unknown aetiology have also been documented (Smith 1980a,b; Ploetz et al. 2003). An example is Finschhafen disorder (FD) which was first observed in 1960 on coconut palms near Finschhafen in Papua New Guinea (PNG). FD has now spread to oil palms in mainland and islands of PNG (Smith 1980a,b; Prior et al. 2001). The planthopper Zophiuma lobulata Ghauri (Lophopidae) is the suspected causal agent for FD transmission due to its presence on palms in areas where the disorder is prevalent (Prior et al. 2001).

As monocotyledons, palms have a single meristem (Tomlinson 1990). Damage due to insects, diseases or nutritional deficiencies may result in the death of the whole plant. Some diseases may have long incubation periods during which the disease is not externally visible leading to sudden death of palms. Early detection, therefore, may allow control measures before losing a lot of plants. On the other hand, development of efficient methods for the control of palm diseases is slow due to gaps in knowledge of biology and ecology of insect pests, the associated pathogens and disease epidemiology. In trying to bridge the knowledge gap between insects and the associated diseases on palms, this paper reviews important palm pathosystems, their aetiology, palm disorders and methods that have been used to study causality and management practices in place to date.

# INSECT PESTS ATTACKING PALMS

A number of insect taxa that attack palms have been documented. Some of these insects are associated with diseases and disorders while transmission of pathogens by some is ambiguous or has not been fully investigated. Pests may feed on foliage, fruit, woody tissue or sap. Palm defoliators feed and consume all or parts of the palm foliage by removing the lamina tissues of the lower leaf surfaces, e.g. bagworms (Psychidae), upper epidermis, e.g. Acria sp.nr emarginella (Oecophoridae) and Latoia viridissima Holland (Limacodidae) and leaf edges (e.g. Tettigonidae) leaving the leaf veins intact (Howard et al. 2001a). Most defoliators of palms are in the orders Lepidoptera, Orthoptera and Phasmatodea. Lepidopteran defoliators include coconut black-headed caterpillars Opisina arenosella Walker (Oecophoridae), the coconut leaf caterpillar Artona catoxantha Hampson (Zygaenidae), various bagworms (Psychidae) and nettle caterpillars (Limacodidae). Long-horn grasshoppers (Tettigoniidae) and stick insects (Phasmatidae) are also important

and widely distributed in most palm-growing regions (Howard *et al.* 2001a). Leaf beetles (Chrysomelidae) are also known to consume palm foliage (Mariau *et al.* 1991; Howard 2001; Howard *et al.* 2001a). Defoliators on palms are not known to transmit pathogens but do interfere with photosynthesis due to the reduction of leaf surface area, eventually resulting in low fruit production and reduction in plant vigour.

Sap feeders and trunk borers belonging to diverse hemipteran taxa (Kaloshian & Walling 2005) on the other hand include a wide variety that transmit pathogens in palms (Howard 2001; Howard et al. 2001a). Common families and superfamilies include Tingidae (lace bugs), Fulgoroidea (planthoppers), Aleyrodoidea (whiteflies) and Coccoidea (mealybugs and scale insects) (Mariau et al. 1991; Howard et al. 2001a; Wilson 2005; Wilson & Weintraub 2007). Among the most serious of the scale insects are coconut scales, Aspidiotus destructor Signoret, the Florida red scale, Chrysomphalus aonidum (L.) and the white date scale, Parlatoria blanchardi (Targioni-Tozzetti), a common pest of date palms in the Middle East and North Africa (EI-Sherif et al. 1996; Howard et al. 2001a). Others include the fern scale (Pinnaspis aspidistrae Signoret) found on lady palm (Rhapis sp.), lesser snow scale (Pinnaspis strachani Cooley) and the magnolia white scale (Pseudalacaspis cockerelli Cooley) found on various palms and foliage such as the queen palm (Syagrus romanzoffianum Cham.), C. nucifera, kentia palm (Howea forsteriana) and lady palm (Rhapis sp.) (Gaimari 2005). The larvae of Batrachedra arenosella (Walk.) reportedly damage flowers of coconut palms in Malavsia (Corbet 1932). Seed production on kentia palms in Lord Howe Island is affected by similar flower feeding by Batrachedra eurema (Bradley).

Transmission of most palm pathogens is dependent on insects' feeding behaviour (Liefting et al. 1997; Miles 1999; Mitchell 2004; Kaloshian & Walling 2005; Weintraub & Beanland 2006). Aspects of the feeding process that influence transmission are salivation, saliva composition, size of the stylet bundle and preferred target plant tissue and the sensory ability of the insect (Kabrick & Backus 1990; Miles 1999; Boyd et al. 2002; Backus et al. 2005a). Hemipterans such as Myndus crudus Van Duzee (Cixiidae), a vector of the LY phytoplasma in coconut palm, feed through a proboscis. The proboscis is covered by a modified labium forming a rostrum that pierces plants to extract phloem contents (Kaloshian & Walling 2005). Studies on Hemiptera feeding on plants show that the insects produce a digestive salivary secretion that forms a raised area on the leaf surface, preventing the stylets from slipping and a sheath that insulates the stylet from apoplastic defences of the plant (Boyd et al. 2002; Backus et al. 2005a, 2007; Kaloshian & Walling 2005; Bressan et al. 2006). During probing and feeding from the phloem, phloem-restricted pathogens, such as phytoplasmas and viruses, are transmitted from the alimentary canal to the insects' salivary glands where they reproduce and are subsequently transmitted to healthy plants (Mitchell 2004).

Sap feeders not only transmit pathogens, but they cause various disorders and may kill entire leaves and fronds through 'hopperburn' when present in high densities (Howard et al. 1982, 1984a; Backus et al. 2005b). Hopperburn has been well studied in rice, potatoes and peanuts (Kabrick & Backus 1990; Backus et al. 2005b; Allen & Rieman 2008), but not clearly understood in palms. The condition arises when insect feeding triggers a complex of plant responses (Backus et al. 2005b). The insects may introduce a toxic substance in their saliva which induces vascular blockage (Backus et al. 2005a,b), resulting in leaf chlorosis, wilting of the tips of young plants, stunting and subsequently reduced yield, symptoms that are similar to LY-type diseases of palms (Howard et al. 1984a). In some instances, hopperburn results from wound responses triggered by stylet movement, as is the case with the grapevine leafhoppers Empoasca spp. (Cicadellidae) (Kabrick & Backus 1990; Backus et al. 2005a,b, 2007).

Although most pathogen transmissions are known to be induced by sucking insects (Miles 1969, 1978; Mitchell 2004; Bressan et al. 2008), transmission can occur when non-sucking insects feed at plant wounds or open cuts, especially when wounded palms produce chemicals that attract these vectors. Examples are the black palm weevil, Rhynchophorus palmarum L., and the red palm weevil, Rhynchophorus ferrugineus Olivier (Curculionidae). Rhynchophorus palmarum transmits the red-ring nematode Bursaphelenchus cocophilus (Cobb) (Griffith 1987; Chinchilla et al. 1990) causing red-ring disease in date palms (Griffith 1987; Murphy & Briscoe 1999) while R. ferrugineus is a common pest on coconut palms in South-East Asia (Soroker et al. 2005; Faleiro 2006; Blumberg 2008). Larvae of dynastine beetles (Scarabaeidae) are also common pests which bore into the crown of palms. The most notable of the non-sucking insects are the rhinoceros beetles (Oryctes spp. and Scapanes spp.) and Rhynchophorus spp. (Bedford 1980; Griffith 1987; Chinchilla et al. 1990; Giblin-Davis et al. 1996; Murphy & Briscoe 1999; Howard et al. 2001a). The larvae, the most destructive stage of these beetles, bore through the frond bases and unopened fronds and may reach into the apical meristematic tissues and sometimes the trunk (Howard 2001) to feed. In coconut, oil, ornamental and date palms, Oryctes rhinoceros L. (Scarabaeidae) larvae bore into the base of the spear clusters, which subsequently reduces yields and may kill the palm if the meristem is badly damaged (Bedford 1980). Severe infestations reduce yields of coconut by up to 10% (Zelazny 1979; Zelazny et al. 1992) while defoliation of more than 40% of fronds has long-term effects on the health of coconut (Bailey et al. 1977). Other beetles such as Rhabdoscelus obscurus (Boisduval) (Curculionidae) and Scapanes sp. (Scarabaeidae) may also damage the fruit bunches and female inflorescence (Beaudoin-Ollivier et al. 2000; De Chenon et al. 2001). In Indonesia, attack by the beetles provides entry points for potentially lethal secondary attacks by Rhyncophorus spp. and R. obscurus (Murphy & Briscoe 1999; De Chenon et al. 2001) or pathogens such as nematodes and fungi (Bedford 1980; Agrios 2005).

# PATHOGENS TRANSMITTED BY INSECTS AND DISEASES CAUSED ON PALMS

Table 1 summarises major diseases, of known and unknown aetiology, of palms, the insect vectors and their associated pathogens. Protozoans, nematodes, viroids, viruses, phytoplasmas, fungi and bacteria cause a variety of palm diseases. Many of these pathogens are vectored by insects and the resulting diseases vary according to the type of palm species and the general attributes of the pathogen based on whether its multiplication is localised or systemic. The ecological and climatic conditions where the palms, pathogens and insect vectors occur also influence disease causality. We discuss here the most significant plant pathogens transmitted to palms by insects.

# Protozoa

Uniflagellate protozoans in the genus *Phytomonas* (Kinetoplastida, Trypanosomatidae) are serious pathogens of palms in tropical America. Genetically uniform isolates of *Phytomonas* sp. cause heart rot disease in coconut and oil palm, and 'sudden wilt' ('marchitez sorpresiva') in oil palm (Dollet 1984, 2001). These unicellular eukaryotes are transmitted by the heteropteran families Coreidae, Pentatomidae, Lygaeidae, Miridae and Tingidae. Transmission can also occur through root grafts (Howard *et al.* 2001a). Heart rot disease in coconut palms and sudden wilt in the African oil palm are transmitted by several Pentatomidae of the genera *Lincus* and *Ochlerus* (Agrios 2005). In Peru, Colombia, French Guyana and Brazil, the genus *Lincus* is involved in the transmission of heart rot disease in coconut and 'marchitez sorpresiva' in oil palm (Parthasarathy *et al.* 1976; Dollet 1984; Mariau *et al.* 1992).

# Nematodes

Nematode-palm interactions involve the modification of growing cells and surrounding tissue. Red-ring disease is the most common and serious disease recorded in oil and coconut palm in Central America, South America and many Caribbean islands (Griffith & Koshy 1990). It is caused by the nematode B. cocophilus whose life cycle lasts only 9-10 days. The third larval stage of the nematode is deposited in a palm leaf, stem or roots by the black palm weevil, R. palmarum, as the weevil oviposits. Inside the tree, the nematode feeds, grows and reproduces. Once the weevil eggs hatch, immature nematodes enter the weevil larvae and remain inside them as the latter undergo metamorphosis. The nematodes do not reproduce inside the weevils. However, mature weevils leave the palm carrying with them new batches of third-stage larval nematodes ready to infest a new tree (Chinchilla et al. 1990; Giblin-Davis 2001).

#### Viroids and viruses

Viroids differ from viruses by virtue of their small size (ca. 300 nucleotides of circular, single-stranded RNA) and the absence

Table 1 Diseases of palms vectored by insects, associated pathogens, the diseases aetiology, distribution and diagnostic procedures that have been used to study causality

Disease pathogen/insect vector	Aetiology	Host plants	Distribution	Symptoms	Diagnostic tests/methods/techniques used to understand pathosystem	References
Diseases associated with Bacteria Bacterial bud rot Sudden decline <i>Ervinia</i> spp., including <i>Ervinia</i> <i>chrysanthemi</i> , <i>E. carotovora</i> and <i>E. lathyri</i> A pentatomid bug	Role played by these bacteria has not been clearly demonstrated	Oil palm (Elaeis guineensis) Date palm (Phoenix dactylifera)	Columbia, Cuba, Ecuador, Nigeria, Republic of Congo, South-East Asia, the USA (Florida)	Sudden loss of vigour in infected palms. Symptoms develop rapidly and some blighted inner leaves appear. The whole tree assumes a straw colour in about 2 weeks	Bacterium is subjected to biological and physiological tests. Mostly through artificial inoculation of palm cultivars using the technique of infusion under reduced pressure	de Franqueville (2003)
Diseases associated with phytopla Lethal yellowing Palm lethal yellowing phytoplasma <i>Myndus crudus</i>	smas Significant presence of <i>M. crudus</i> where disease is found. Phytoplasma spread through vectors feeding on palms during incubation period or early disease developmental stage. Vector then spreads fluids from infected to non-infected plants	Coconut (Cocos nucifera) P. dacylifera Phoenix sp.	Florida, Jamaica, Mexico, Caribbean, West Africa, South-East Asia	Premature shedding of fruits. New inflorescences become dark, necrotic, dry and gnarled. Fronds turn yellow, beginning with older towards younger ones. Final stage is death of growing point, causing youngest unfolded leaf ("spear" leaf) to rot and collapse. Entire bud rots within days. Decay of root system and sudden death of plant	Transmission tests in cages with the introduction of <i>M. crudus</i> into cages containing coconut palms. Identification of MLOs by TEM. Analysis using TEM with DAPI tests. Sequencing and phylogenetic analysis of 16S rRNA genes cloned by conventional means or PCR amplification, RFLP analysis of amplified 16S rDNA and supported by sequence variation. DNA probes to detect MLOs	Parthasarathy (1974) Steiner (1978) Howard <i>et al.</i> (1986) Eskafi <i>et al.</i> (1986) Sangare <i>et al.</i> (1992) Harrison <i>et al.</i> (1994) Mpunami <i>et al.</i> (1999)
Cape St Paul wilt Phytoplasma Diostrombus sp. and Myndus adiopodoumeensis	Not well understood. Suspected to have similar actiology to lethal yellow	C. nucifera	Ghana	Premature fruit drop Blackening of new inflorescences followed by yellowing of leaves until crown dies	Molecular techniques using a nested PCR P1/ P7 and Ghana 813f and Awka SR primers followed by sequencing	Tymon <i>et al.</i> (1997) Nipah <i>et al.</i> (2007) Philippe <i>et al.</i> (2007)
Awka disease Phytoplasma Meenoplus proximus and Malenia cocos	Not well understood. Suspected to have similar actiology to lethal yellow	C. nucifera	Ghana, Nigeria	Premature nut fall	Molecular techniques using a nested PCR P1/ P7 and Ghana 813f and Awka SR primers, followed by sequencing	Tymon <i>et al.</i> (1997) Ploetz <i>et al.</i> (2003) Nipah <i>et al.</i> (2007) Philippe <i>et al.</i> (2007)
<b>Kribi disease</b> Phytoplasma Unknown vector	Not well understood. Suspected to have similar aetiology to lethal yellow	C. nucifera	Cameroon	Similar to Awka and Cape St Paul wilt	Similar to Awka and Cape St Paul wilt	Tymon <i>et al.</i> (1997) Ploetz <i>et al.</i> (1999, 2003)
Al-Wijam disease Aster yellows phytoplasma 165rl Candidatus Phytoplasma Cicadulina bipunctata (Melichar)	Not well understood. Suspected to have similar aetiology to lethal yellow	P. dactylifera	Saudi Arabia	Stunting and yellow streaking of the leaves. Reduction of fruits and fruit stalks. Intensity of symptoms varies with variety of date palm	Identification and molecular characterisation by PCR and RFLP analysis	Alhudaib <i>et al.</i> (2007b)
Kalimantan wilt Phytoplasma Sophonia sp. and Idioscopus clypealis (Cicadellidae), Nisia nervosa (Motschulsky) (Meenoplidae)	Phytoplasma spread to neighbouring palms from symptomatic seedlings	C. nucifera	Indonesia	Malformation of mature or middle-aged nuts elongated in shape and poorly developed endosperm. Death of the palm occurs in 6–18 months after symptoms appear	Amplification of phytoplasma DNA by PCR assays. Nested PCR approach using P1/P7 and R16F2n/R16R2 primer combinations	Jones <i>et al.</i> (2000) Warokka <i>et al.</i> (2006)

Table 1 Continued						
Disease pathogen/insect vector	Aetiology	Host plants	Distribution	Symptoms	Diagnostic tests/methods/fechniques used to understand pathosystem	References
Diseases associated with fungi Basal stem rot Ganoderma boninense Insect vectors harbouring Ganoderma such as Oryctes and Rhabdoscelus have been implicated	Landscape practices, such as excessive mulch against the trunk. Irrigation water splashes onto the trunk. Insects carry fungal spores from infected plants	C. mucifera E. guineensis	Florida, Papua New Guinea, South-East Asia, Indonesia, Malaysia	Collapse of xylem and other trunk tissues. Lower fronds turn dark green/grey, droop, and eventually die. Symptoms progress to younger fronds until entire crown collapses	Population structure and genetic studies of <i>G</i> , <i>hominense</i> studied. Inter-fertility between isolates collected from basal stem rots on oil palm investigated	Pilotti (2005) Susanto <i>et al.</i> (2005) D. de Chenon (pers. comm. 2008) Paterson (2007)
Disease caused by a nematode Red-ring disease Red-ring nematode Bursaphelenchus cocophilus Palm weevil Rhynchophorus palmarum and R. ferrugineus Scale insects, ants and weevils are suspected vectors	Palm weevil is internally infested by nematode that lives in the gut with the red-ring nematode. The weevil is attracted to a trunk wound to feed and acts as a vector for $B$ . <i>cocophilus</i> to uninfected trees	C. nucifera E. guineensis P. dactylifera Roystonea sp.	Central America, South America, Caribbean islands	Bright red ring characterised by a circular, coloured band in the trunk of palm. Leaves become short and deformed, turn yellow-bronze, then deep red-brown. Colour change begins at tip of leaf, starting with older leaves. Leaves wilt and die	Transmission assays of nematode by root contact from infected to non-infected palms. Light and scanning electron microscopy analyses on faecal matter of palm weevil	Chinchilla <i>et al.</i> (1990) Warwick and Berzerra (1992) Giblin-Davis (2001) Mani <i>et al.</i> (2005) Magalhães <i>et al.</i> (2008)
Diseases associated with viroids. Cadang-cadang Coconut cadang-cadang Viroid (CCCVd) Beetles and lace bugs. Other insects in the orders Orthoptera, Coleoptera, Hemiptera	and a virus Direct feeding injury. Disease spread through grafting, handling and cutting using contaminated tools	C. nucifera E. guineensis	Malaysia, Philippines, Guam, Solomon islands	Orange spotting on palm fronds. Olive translucent spots on frond veins. Decline in fruit production followed by sterility, brittle leaves, deterioration of root system, stunting and finally death	Two-dimensional polyacrylamide gel electrophoresis. Cloning. Reverse transcription-PCR techniques	Zelazny and Pacumbaba (1982) Hanold and Randles (1989, 1991) Vadamalai <i>et al.</i> (2006)
<b>Tinangaja</b> Coconut Tinangaja Viroid (CTiVd) Scale insects and mealy bugs	Viroid actiology poorly understood	C. nucifera	Island of Guam and Anahatan	In early stage of disease, crown is slightly reduced in size but normal nuts are produced. In the middle stage, crown is reduced, nuts are small and elongated and usually lack kernel. In the late stage, the crown is small in size, nuts are absent, and leaflets are thin. Retention of stipule and leaf stippling are evident	Two-dimensional PAGE. Molecular based hybridisation assay diagnostic oligonucleotide-probe (DOP) to CTIVd or reverse transcription-PCR used for low levels of virus	Hodgson <i>et al.</i> (1998)

332 CW Gitau et al.

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<b>Coconut foliar decay disease</b> Coconut foliar decay virus (CFDV) <i>Myndus</i> sp.	<i>Myndus taffini</i> commonly settles on coconut fronds. The virus is transmitted from neighbouring <i>Hibiscus tiliaceus</i> shrubs harbouring the virus	C. mucifera	Vanuatu	Yellowing in several leaflets of a frond down from the unopened spear leaf followed by general yellowing, necrosis and death of frond. Yellowing and necrosis spread to adjacent fronds. Reduction and death of the crown	Electron microscopy. Molecular techniques using cloned sequences for the identification of CFDV, hybridisation with cDNA or cRNA. Two-dimensional PAGE. Use of PCR for diagnosis of CFDV. Transmission experiments: <i>M. taffini</i> released into cages with coconut palms to investigate development of CFD symptoms	Hanold <i>et al.</i> (1988) Randles (1999) Mariau (2001)
Ringspot Ringspot Fovea-like virus Possibly the aphid <i>Myzus</i> <i>persicae</i> (Sulzer)	Inknown aettology Not well understood	E. guineensis	South America	Ring spots and elongated rings on leaflets and rachis of spear and first leaves. Foliar spots turn yellow eventually reaching lower leaves. The spear leaf develops necrosis and affects the meristem. Roots show necrosis and a cross-section shows a purple coloration. Lower leaves brown and die within 3 months of initial symptom expression. Fruit bunches also get necrotic	Pathogenicity tests using virus isolates obtained from symptomatic and asymptomatic plants. Tests were carried out by mechanically inoculating clean plants with extracts obtained from ring spot-afficeted tissues of palms. Viruses detected using electron microscopy, electrophoresis, nucleic acid hybridisation, molecular cloning and sequencing also used in virus detection	Hanold <i>et al.</i> (1988) Randles (1999) Howard <i>et al.</i> (2001a) Mariau (2001) Morales <i>et al.</i> (2002a,b)
Dry bud rot Pathogen unknown Sogatella kolophon and S. cubarna (Delphacidae) possible insect vectors	Not known	<i>C. nuclfera</i> <i>E. guineensis</i> in the nursery	West Africa	Small yellow and white spots on the spear or first leaf. At an advanced stage, base of the spear is completely brown	Through transmission experiments. 200 individual insects introduced into the cages and symptom development assessed	Howard <i>et al.</i> (2001a) Mariau (2001)
Finschhafen disorder Pathogen unknown Zophiuma lobulata	Attributed to feeding by the vector on palms	C. mucifera E. guineensis Betel nut (Areca catechu)	Papua New Guinea	Bright yellowing/bronzing of tips of fronds, reduced fruits production, stunted growth	FD symptoms have been induced in coconut palms by introducing <i>Z. lobulata</i> into cages with coconut plants. Remission of symptoms was observed 3 years after planthoppers were killed at the sixth-seventh month of continuous feeding on the palms	Ghauri (1966) Smith (1980a,b) Prior <i>et al.</i> (2001)

FD, Finschhafen disorder; PCR, polymerase chain reaction; RFLP, restriction fragment length polymorphism.

#### 334 CW Gitau et al.

of a protein coat. Viroids of importance in palms include Coconut cadang-cadang viroid (CCCVd) and Coconut tinangaja viroid (CTiVd), which have been identified in coconut palms (Wall & Randles 2003; Vadamalai et al. 2006). The epidemiology of viroids is poorly understood and there is no direct proof to date of insect transmission (Randles & Rodriguez 2003). Certain Coleoptera, however, are suspected to transmit viroids through feeding on wounded sections of palms (Singh 2000). Among the viruses, fovea-like virus and potyvirus are most notable, causing chlorotic ring and ring spot respectively on oil palm in South America (Morales et al. 2002a,b). Potyvirus is believed to be transmitted by feeding of the aphid Myzus persicae (Sulzer) which is known to induce cytoplasmic inclusions which are seen as pinwheels, laminated aggregates and scrolls (Narayanasamy 2001) in oil palm.

### Mollicutes

Mollicutes include phytoplasmas and non-culturable plant pathogens in the order Acholeplasmatales (Lee et al. 1998, 2000). Phytoplasmas are obligate parasites of plant phloem tissues and some insects (Gundersen et al. 1994; Lee et al. 2000; Dollet et al. 2001) and are by far the most widely studied group of pathogens transmitted to palms by insects (Table 2). Phytoplasmas belonging to the group 16SrIV are most often associated with palm diseases (Harrison et al. 2002a,b; Narváez 2006; Table 2). The phytoplasmas are transmitted by hemipteran vectors or by vegetative propagation (Oropeza et al. 1999; Howard et al. 2001a) and are associated with LY and other related yellowing diseases. Koch's postulates have not yet been satisfied for phytoplasmas due to the fact that they are unculturable (Howard 1980a, 1983; Chinchilla et al. 1990; Harrison et al. 2008). The involvement of the palm LY phytoplasma in LY diseases is, however, supported by its presence in tissues of affected palms and in the planthopper vector, Myndus crudus Van Duzee (Hemiptera: Cixiidae) (Howard et al. 1982, 1984a; Harrison et al. 1994). The palm LY phytoplasma was absent in healthy plants (Harrison et al. 1994, 2008). Introduction of M. crudus from affected palms onto caged susceptible palms resulted in development of LY symptoms while plants in control cages, which were protected from infestation by M. crudus, remained healthy (Howard 1983).

LY and LY-type diseases of coconuts are widespread in the Caribbean, Americas and Africa (Eden-Green & Tully 1979; Howard *et al.* 1979; Howard 1980b, 1983; Kastelein *et al.* 1990; Schuiling & Mpunami 1992; Harrison *et al.* 1994, 2008; Mpunami *et al.* 1999). In Africa, LY-type diseases are named after the specific region where the disease occurs, e.g. Awka disease (Nigeria), Cape St Paul wilt (Ghana) (Philippe *et al.* 2007), Kaincope disease (Togo) and Kribi disease (Cameroon) (Tymon *et al.* 1997). The phytoplasma group of these diseases in Africa is still unknown (Table 2). Other phytoplasma diseases have been reported on date palm in the Middle East. In Saudi Arabia, 'Al-Wijam' disease is associated with the '*Candidatus* Phytoplasma *asteris*' group, and thought to be trans-

mitted by the cicadellid leafhoppers *Cicadulina bipunctata* (Melichar) and *Asymmetrasca decedens* (Paoli) (Alhudaib *et al.* 2007a). In Sudan, white-tip dieback is spread throughout northern Sudan (Cronjé *et al.* 2000a) whereas slow decline ('El Arkish') occurs commonly along the river Nile between Dongola and Merowe-Karema (Cronjé *et al.* 2000b).

The spread of phytoplasmas relies on phloem-feeding insects such as leafhoppers (Cicadellidae) and planthoppers (Cixiidae, Derbidae, Delphacidae) (Solomon 1997; Howard et al. 2001b). The phytoplasmas are ingested when insects feed on infected plant tissue and then multiply in the insect salivary glands (Weintraub & Beanland 2006; Weintraub 2007). The pathogens are then passed to non-infected plant tissues through the insect salivary secretions during subsequent feeding (Kabrick & Backus 1990; Jeger et al. 2004; Kaloshian & Walling 2005). Symptoms of phytoplasmainfected palms include yellowing of leaves (Howard & Thomas 1980; Smith 1980a; Harrison et al. 2008) which is presumably caused by inhibition of sugar transportation in the phloem vessels by the phytoplasmas (Orenstein et al. 2001, 2003; Hogenhout et al. 2008). Studies have shown that presence of phytoplasmas in plants leads to a decrease in chlorophyll content which can interfere with photosynthetic activity subsequently resulting in yellowing and rapid senescence of leaf tissues (Lepka et al. 1999; Junqueira et al. 2005). Phytoplasmas are also known to down-regulate a gene involved in petal formation and other genes involved in the maintenance of the apical meristem (Maust et al. 2003) and this could explain stunted growth in palms infected by these pathogens.

# METHODS OF STUDYING CAUSALITY

Diagnosis of palm diseases and disorders is important due to the fact that palms live for many years and may harbour pathogens that have an extended incubation period which are however asymptomatic, e.g. LY (Dabek 1974; Howard 1980a). Although some pathogens such as nematodes cause distinct symptoms, there is a distinct overlap in the symptoms that different pathogens invoke. Palms may be attacked simultaneously by a range of lethal diseases. For example, in South America, phytophthora bud rot, red-ring disease, heart rot and LY can all attack coconut palms at the same time (Oropeza et al. 1999). A deep working knowledge of the specific diseases that affect a given palm and the associated symptoms is invaluable when diagnosing these problems. Palm diseases caused by viruses and phytoplasmas have been diagnosed using conventional plant pathology techniques. In cage experiments, disease transmission to healthy palms is conducted by releasing known or suspected insect vectors that carry the pathogens. Symptom development and detection of pathogens may then be assessed using a variety of methods ranging from microscopy to DNA-based techniques.

Microscopic techniques using stains such as safranin have been used for phytoplasma detection in palm tissues (Bashan *et al.* 1980). Symptomatic cells are recognised by a dense red

Disease/disorder	Palm species	Phytoplasma	Phytoplasma group	GenBank accession numbers	Locality	Reference
Awka disease	Cocos nucifera	Unknown	Unknown		Nigeria	Tymon <i>et al.</i> (1997)
Cape St Paul wilt	C. nucifera	Unknown	Unknown		Ghana	Tymon et al. (1997)
Kaincope disease	C. nucifera	Unknown	Unknown		Togo	Tymon et al. (1997)
Kalimantan wilt	C. nucifera	Candidatus Phytoplasma oryzae	16SrXII groups	D12581	Indonesia	Warokka et al. (2006)
Kribi disease	C. nucifera	Unknown	Unknown		Cameroon	Ploetz et al. (2003)
Lethal decline	Phoenix dactylifera	Coconut lethal decline	16SrIV-C	X80117	Tanzania	Tymon et al. (1997)
	C. nucifera				Kenya	
Lethal yellowing	P. dactylifera	Candidatus Phytoplasma palmae	16SrIV group	U18747	Florida	Harrison (2002)
	C. nucifera	Ca. Phytoplasma castaneae			Mexico	Harrison et al. (2002b)
		Ca. Phytoplasma cocosnigeriae				Narváez (2006)
Root wilt disease	P. dactylifera	Date palm phytoplasma	16SrI	DQ913090	Sri-Lanka	Howard et al. (2001a)
	C. nucifera					Ploetz et al. (2003)
Al Wijam	P. dactylifera	Candidatus Phytoplasma asteris	16SrI group	AF322644	Saudi Arabia	Alhudaib et al. (2007c)
Yucatan coconut lethal decline	C. nucifera	Lethal yellowing phytoplasma	16SrIV-B	U18753 32	Yucatan	Tymon et al. (1997)
Date palm white tip dieback	P. dactylifera	Date palm white tip die-back phytoplasma	16SrI-L	SR AF100411	Sudan	Cronjé et al. (2000a)
		Candidatus Phytoplasma cynodontis				
Slow decline	P. dactylifera	Date palm slow decline phytoplasma	16SrXIV-A	SR AF268000	Sudan	Cronjé et al. (2000b)
Kerala wilt	C. nucifera	Kerala wilt disease phytoplasma	16SrIV-C		India	Edwin and Mohankumara (2007)
Texas Phoenix palm decline	P. dactylifera	Carludovica palmata leaf yellowing phytoplasma	16SrIV-D	AF434989	Mexico, Texas	Harrison et al. (2002b)
Coconut yellow decline (in Malaysia)	C. nucifera	Candidatus Phytoplasma cynodontis	16SrXIV	EU328159	Malaysia	Nejat et al. (2008)

Table 2 Palm diseases and associated phytoplasma groupings

colouration. In studies where pathogens such as phytoplasmas and viruses cannot be cultured or observed using light microscope, electron microscopy has been useful (Morales *et al.* 2002a,b). A major constraint on use of specialised microscopy is that it is not often accessible in developing countries. Visualisation of viroids under the electron microscope is also difficult, due to their small size, and the fact that their distribution in the phloem tissues is sometimes localised and unpredictable or particles are in low concentration (Randles & Hanold 1989; Randles 1999).

DAPI test (4, 6-diamidine-2-phenylindole dihydrochloride), which uses a fluorescent counterstain that stains the plant cell nucleus blue, has been successfully used to identify phytoplasmas in coconut palm embryos (Cordova *et al.* 2003). Phytoplasmas have also been detected in the phloem by fluorescence light under the microscope (Chen *et al.* 1994). The DAPI technique has been used effectively in the examination and selection of coconut palm varieties that are resistant to LY-type disease in Tanzania (Schuiling & Mpunami 1992). Serological methods such as enzyme-linked immunosorbent assay (ELISA) or immunofluorescence using specific monoclonal antibodies are more specialised and have been applied in the identification of phytoplasmas in symptomatic palms (Bulletin 2006).

Although disease diagnoses have previously relied on conventional methods, the use of DNA-based molecular diagnostic techniques is gaining popularity among scientists for the study of plant pathogens and insect vectors. Molecular techniques have been used in studying various palm diseases such as those caused by phytoplasmas (Tsai 1975; Howard & Thomas 1980; Smith 1980b; Howard et al. 1982). Techniques such as nucleic acid hybridisation (NAH) and polymerase chain reaction (PCR) allow infection by pathogens to be detected before they induce symptoms on plants (Fránová et al. 2007). Other than pathogen detection and identification, molecular tools can facilitate identification of new pathogen genes. Products and functions of the pathogenic genes can hence be determined and biochemical traits and process by which gene expression is regulated can be elucidated (Hanold & Randles 1991; Harrison et al. 2001, 2002b; Morales et al. 2002a; Alhudaib et al. 2007a,b).

A variety of DNA-based methods has been used to detect and characterise LY phytoplasmas on palms (Oropeza et al. 1999; Harrison et al. 2008) and viroid detection (Hanold & Randles 1991; Morales et al. 2002a). PCR has been particularly valuable in phytoplasma research (Gundersen et al. 1994; Lee et al. 1998; Harrison et al. 2001, 2002b; Weintraub 2007). Total DNA from plants and insect vectors is used as a template for amplification of specific regions of the phytoplasma genome, usually the 16S ribosomal RNA gene since phytoplasma taxonomy is based on this gene, by using sequencespecific PCR primers. Commonly used PCR techniques are the single-step PCR and nested PCR. Single-step PCR amplifies regions of target DNA regions in a single operation while nested PCR involves amplification by one primer pair which is followed by a second amplification with a more specific primer pair that amplifies a smaller region nested within the first product. Nested PCR has recently been used for detecting

phytoplasmas in palms (Cronjé *et al.* 2000a,b; Harrison *et al.* 2002a, 2008; Cordova *et al.* 2003; Mejia *et al.* 2004; Narváez 2006).

Compared with single-step PCR, nested PCR is more sensitive and the chance that a region erroneously amplified in the first round of PCR is re-amplified by the second set of primers is low. The disadvantage of nested PCR, however, is that contaminants, such as bacteria, found in the original tissue samples can present a problem due to the test's sensitivity (Carginalea et al. 2003; Polak et al. 2007; Wally et al. 2008). This problem can, however, be alleviated by using appropriate and independent negative controls at each step (Fránová et al. 2007). Real-time PCR, where the increase in the amount of DNA may be visualised in real time as it is amplified, may be less laborious and is able to detect a wider spectrum of phytoplasmas (e.g. Crosslin et al. 2006). However, while conventional 'end-point' PCR has become a standard tool in many biologists' repertoire, real-time PCR requires highly specialised equipment and extensive assay development and optimisation.

Restriction fragment length polymorphism (RFLP) entails the use of restriction enzymes which cut DNA only at a particular nucleotide sequence to survey DNA for sequence variation. RFLP is often performed on PCR products to reveal patterns specific to a group of phytoplasmas (Heinrich et al. 2001; Bressan et al. 2007). Such PCR-RFLP analyses have been used in investigating pathogens on palms (Oropeza et al. 1999; Alhudaib et al. 2007b; Martinez et al. 2007; Harrison et al. 2008). While PCR-RFLP assays are quicker and cheaper than determining the actual underlying DNA sequence, they are more prone to misinterpretation than DNA sequences because of uncertain homology between fragments of the same size. In addition, RFLP detects novel sequences only if mutations occur at the exact site targeted by the specific restriction enzyme used in the assay, thus the technique is less sensitive than DNA sequencing.

Nucleic acid hybridisation has been used in the detection of the viroid CCCVd (Randles & Palukaitis 1979; Imperial et al. 1985; Hanold & Randles 1991). This method utilises radioactively labelled single RNA strands of known base sequences which are used as a probe to detect the nucleotide sequence of another single-stranded RNA molecule. The method can then be followed by molecular cloning or sequencing in cases where a sufficiently accurate probe has not been developed. Although this method is reliable and sensitive for viroids of known sequence, it is inappropriate for the detection of new viroids where sequence information is unavailable. Recently, a number of reverse transcription-polymerase chain reaction (reverse transcription-PCR) protocols, including real-time reverse transcription-PCR protocols, have been developed for detection of different viroids (Vadamalai et al. 2006). Although they do not always detect all pathogen strains, reverse transcription-PCR techniques are rapid and quite sensitive to single mutations. Ribonuclease protection assay (RPA), a more specific and sensitive liquid-hybridisationbased RNA diagnostic method, has recently been used in the detection of the viroid that causes cadang-cadang disease of oil

palm in Malaysia (Vadamalai *et al.* 2009). RPA uses enzymes that are resistant to inhibitors and is useful for measuring expression of a single-target mRNA species in a complex mixture of total RNA (Aranda *et al.* 1993; Gilman 1993). A limitation of RPA, however, is that not all single-nucleotide mismatches are targeted by the ribonuclease digestion step and the probes used need to be re-synthesised weekly to guard against radiolysis (Aranda *et al.* 1995; Rottman 2002).

Determining the DNA sequence has become useful in diagnostic research and leads to an indication of the kind of pathogens present in tested samples and provides a platform from which aetiology can be examined. Molecular techniques, however, followed by methods that use Koch's postulate approach (Evans 1976; Falkow 1988) and those that are supported by acceptable biological principles would be an excellent system in studying causality of most palm diseases and other unknown conditions. Although molecular techniques are quick and fairly reliable, they have several limitations. The techniques use costly reagents and equipment (López et al. 2008), the development process can be long and highly complex and does not always guarantee a clear-cut outcome. Contamination of reagents and samples during collection and outcomes of false negatives from PCRs are common setbacks (Heinrich et al. 2001; Wally et al. 2008) and hence necessitate optimisations. The choice of a molecular technique hence depends on the organism, plant species, financial resources available and sanitation conditions in the region concerned. It would be prudent to begin with the cheapest conventional method available, such as using disease characteristics and visualisation of the pathogen using a dissecting or compound microscope. Strategies such as those used to study LY-type diseases are useful models for the identification of causal agents and possible vectors and the development of diagnostic tools for palm diseases with unknown aetiologies. Observations using simple tests such as DAPI together with light microscopy can be used as a preliminary test for pathogen detection. This method has been successful in the identification of the red-ring nematode (Flood et al. 2005).

### MANAGEMENT PRACTICES

Management practices of palm diseases related to insects range from chemical, cultural to biological control. Unrestrained use of pesticides has led to severe outbreaks of insect pests. In palm plantations, the use of chemical compounds is normally restricted to heavy pest infestations. Sanitation during palm growth plays a major role in disease management (Mariau 1994). Strict quarantine at international, national and local levels has been the major emphasis in the control of palm diseases and disorders in palm-growing areas in the last decade (Wahid *et al.* 2004; EPPO 2006). Roguing, achieved through exclusion or eradication of infected palm material, has been effective in the control of viroid diseases (Hanold & Randles 1989). Integrated pest management (IPM) approaches that use crop management procedures that include covering fruit bunches with plastic nets and early harvesting have successfully been used for the control of fruit moths in cultivation of date palms in Israel (Kehat 1999; Blumberg 2008). Microbial control, mainly application of *Bacillus thuringiensis* products, has recently been used against the lesser date moth (Blumberg 2008).

Biological control has been used against a number of palm pests (Fediere *et al.* 1990; Howard 1990; Kathirithamby *et al.* 1998). For instance, the entomopathogenic *Oryctes rhinoceros* baculo-virus has been a major success in many areas of the Asia/Pacific region as a classical biocontrol agent of the coconut palm rhinoceros beetle (Bedford 1976; Huger 2005; Jackson *et al.* 2005). On the other hand, the use of nematodes has been successful in the control of the red palm weevil. An entomopathogenic heterorhabditid nematode has been isolated from areas planted with palm trees and has been used for the control of the red palm weevil in Egypt (Salama & Abd-Elgawad 2001). The entomopathogenic nematode *Heterorhabditis* sp. (Rhabditida: Nematoda) has been isolated in the United Arab Emirates and has been shown to have potential for control of the red palm weevil (Elawad *et al.* 2001).

Parasitoids such as the strepsipteran Stichotrema dallatorreanum Hofeneder (Mymecolacidae) (Kathirithamby et al. 1998; Solulu et al. 1998; Kathirithamby 2009) and two egg parasitoids Doirania leefmansi Waterston (Hymenoptera: Trichogrammatidae) and Leefmansia bicolor Waterston (Hymenoptera: Encyrtidae) are used against leaf-feeding Sexava spp. (Orthoptera: Tettigoniidae) in PNG (PNGOPRA, unpubl. data 1985). Parasitoids can be used to complement other control practices that are in place in most palm plantations. For instance, old fronds are routinely pruned off to facilitate access to fruit bunch during harvest. Growers are advised to stack the fronds in specific rows and grow leguminous cover crops to speed up frond breakdown and enhance nitrogen recycling (Howard 1990). Using cover crops not only enhances soil improvement but offers suitable habitat and nectar for beneficial insects, which simultaneously helps to lower pest populations (Turner & Gillbanks 1974; Howard & Oropeza 1998). In addition, some legumes when used as cover crops provide poor breeding sites and/or do not support development of eggs or other immature stages of some insect pests. For example, larval development of *M. crudus* is not supported by Pueraria phaseoloides (Roxb.) Benth (Fabaceae) and Arachis pintoi Krapov and Greg (Fabaceae) (Howard 2006). Maintaining grass that impedes development of M. crudus larvae also has been a successful practice in palm plantations (Howard 1990). On the other hand, the fallen frond material suppresses growth of weeds. By destroying habitats occupied by insects that may be vectoring diseases, cultural and integrated management control practices have been and can be achieved (Howard & Oropeza 1998; Caudwell 2000).

Pheromone trapping (Chinchilla *et al.* 1993; Oehlschlager 1998; Oehlschlager *et al.* 2002) and biological control are options that are generally underutilised in the management of insect pests of palms. The role of semiochemicals has been elucidated and is used in monitoring and control of the black palm weevil *R. palmarum* in oil palm (Oehlschlager *et al.* 

1993, 1995). Male aggregation pheromone has been used to monitor weevil spread and reduce weevil densities when used as trap baits (Giblin-Davis *et al.* 1996; Oehlschlager *et al.* 2002). Semiochemicals have in addition facilitated pest management decisions and helped in detection of non-native pests at ports of entry. Use of these products, whether in natural or synthesised form, may be highly effective but generally require extensive knowledge of the target pest and correct timing and application techniques.

Future management of pests should rely on improved monitoring systems, habitat management and integration of pheromone application (where appropriate). The use of Global Positioning System and Geographical Information Systems (GIS) technologies has recently been applied in locating insect infestations such as corn rootworms and bean leaf beetles (Rami & Ishaaya 2004; Yong-Lak et al. 2007). These spatiotemporal monitoring systems are methods that have not been explored in monitoring the existence and dispersal of insect pests in palm ecosystems. The technology would allow collection and analysis of information on insect pest activities thereby facilitating accurate monitoring of palm pests. Appropriate preventative measures can therefore be taken in a timely manner. Geospatial tools can, in addition, be useful in studying the biological richness and landscape ecology in crop-growing regions (Beheraa et al. 2005) and hence provide information on insect pest dynamics in relation to landscape (Hunter 2002).

#### CONCLUSIONS

There is much to be learned from analysing results of studies that have been conducted on insect pests and associated diseases and disorders in palms. For rational insect pest management practices to be developed, a sound knowledge of palm pathology and ecology of the associated insect pests is a prerequisite. A better understanding of the factors that are a threat to palm husbandry as well as to the development of appropriate diagnostic, monitoring and sustainable management techniques can reduce production costs and help understand and manage new disease incidences. This is possible through a comprehensive understanding of existing circumstances and the nature of biology and ecology of insect pests. For many palm diseases, the possible role of vectors is still unconfirmed or unknown (Table 1). The accurate taxonomic clarification of any suspected pest taxon is essential for the development of pest management programs, especially where biological control is to be used.

While investigating new diseases, there is likely to be an unfortunate bias towards familiar vector taxa. For example, researchers investigating oil palm bud rot tested several families of Hemiptera without positive results but later found that a soil dwelling cydnid bug (Cydnidae, *Scaptocoris* sp.), was the probable vector (de Franqueville 2003). Since the transmission of pathogens such as viruses and phytoplasmas is not limited to one group of insects, a suitable approach to vector research is to test insects in the same taxonomic group as other proven

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#### 342 CW Gitau et al.

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