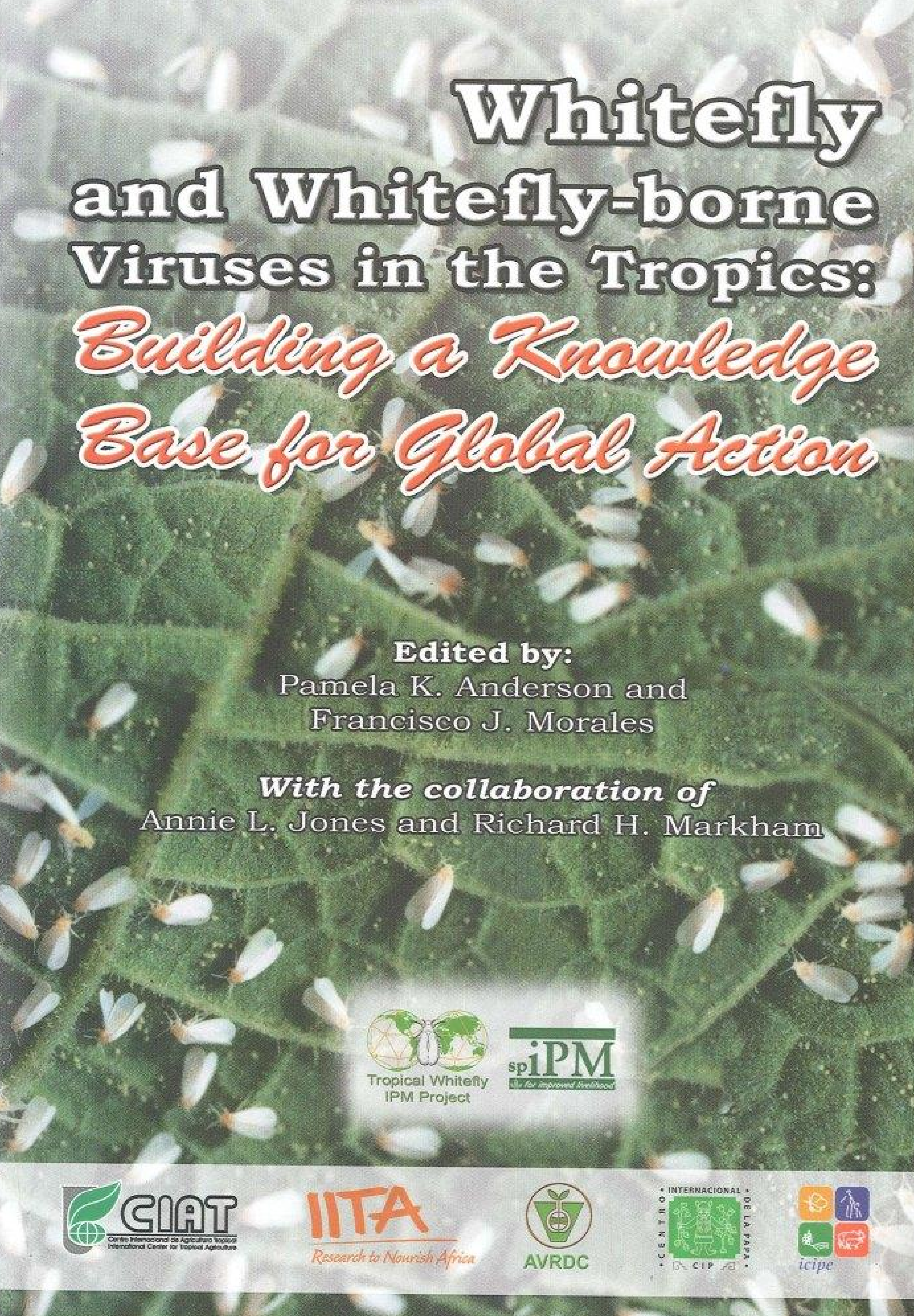

Whitefly and Whitefly-borne Viruses in the Tropics: *Building a Knowledge Base for Global Action*

Edited by:
Pamela K. Anderson and
Francisco J. Morales

With the collaboration of
Annie L. Jones and Richard H. Markham

Tropical Whitefly IPM Project
spIPM
CIAT Centro Internacional de Agricultura Tropical International Center for Tropical Agriculture
IITA Research to Nourish Africa
AVRDC
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Special Topics on Pest and Disease Management

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Foreword

*Pamela Anderson**

This book is the product of the collective knowledge accumulated over 3 decades by hundreds of agricultural professionals and scientists involved in the fight against two of the worst pests of all times: whiteflies and whitefly-transmitted viruses. Millions of farmers affected by these pests have also contributed a great deal of valuable information regarding the nature of the problem and the extent of the damage caused by these pests in important food and industrial crops. Examples of such are: cassava (*Manihot esculenta* Crantz), common bean (*Phaseolus vulgaris* L.), sweetpotato (*Ipomoea batatas* [L.] Lam.), lima bean (*Phaseolus lunatus* L. var. *lunatus*), mung bean (*Vigna radiata* [L.] R. Wilczek), tomato (*Lycopersicon esculentum* Mill.), peppers (*Capsicum* spp. L.), squash (*Cucurbita moschata* Duchesne), melon (*Cucumis melo* L.), other cucurbits, cotton (*Gossypium hirsutum* L.), tobacco (*Nicotiana tabacum* L.), soybean (*Glycine max* [L.] Merr.), lettuce (*Lactuca sativa* L. var. *capitata* L.) and eggplant (*Solanum melongena* L.).

The devastating capacity of these pests, and their global distribution in the main tropical and sub-tropical regions of the world, has been a major challenge to agricultural scientists in both developing and industrialized nations. Obviously, their impact has been far greater in the rural areas of developing countries, where resource-poor farmers have lost both traditional food crops (e.g., common bean and cassava) and cash crops (e.g., tomato and peppers) to these pests. Most of the national agricultural research institutes and ministries of agriculture in the developing countries affected by these pests have implemented different whitefly control measures, including legal measures, which constitute a unique event in the history of most developing countries where agriculture in general is not regulated by the state.

The research activities described in this book provide a general view of the problem for agricultural scientists and laypeople alike, and contributes a great deal of pertinent information on the nature and management of the problem in the different crops affected by whiteflies and whitefly-transmitted viruses. In doing so, the Tropical Whitefly Integrated Pest Management Project (TWF-IPM) has paid considerable attention to agricultural sustainability, ecosystem health and human health issues, related to the appalling misuse of highly toxic pesticides associated with whitefly control practices in developing countries.

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* Former coordinator for Phase 1 of the Tropical Whitefly IPM Project; currently Research Director at the Centro Internacional de la Papa (CIP), Lima, Peru.

The information contained in the following chapters will form the basis of a worldwide effort aimed at disseminating the knowledge accumulated on the nature of the whitefly/virus problems and the most efficient integrated pest and disease management measures available to develop sustainable mixed cropping systems. Moreover, this knowledge should ultimately contribute to improve the livelihoods of resource-poor farmers affected by these pests throughout the tropics, by safeguarding their food staples and high-value crops, reducing production costs and pesticide contamination, and increasing their family income.

Introduction

*Pamela K. Anderson**

The Problem of Whiteflies and Whitefly-Transmitted Viruses in the Tropics

Although Mound and Halsey (1978) catalogued 1156 species of whiteflies (Homoptera: Aleyrodidae), only a limited number of whitefly species are considered pests of economic importance. Two whitefly species are key pests throughout the tropics: *Bemisia tabaci* (Gennadius) and *Trialeurodes vaporariorum* (Westwood). Five additional whitefly species are considered as important pests in specific regions: *Aleurotrachelus socialis* (Bondar), *Trialeurodes variabilis* (Quaintance), *Bemisia tuberculata* (Bondar), *Trialeurodes abutiloneus* (Haldman) and *Aleurocanthus woglumi* (Ashby).

Whiteflies are phloem (sap) feeders. They cause direct damage in some hosts by extracting large quantities of sap. The honeydew that they excrete, as a result of the copious sap intake, serves as substrate for sooty mould fungi, which can also damage hosts by blocking photosynthesis. Sooty mould can discolour harvestable fruits and fibre, affecting the quality of produce, and can cause plant death in crops such as potato (*Solanum tuberosum* L.), tomato (*Lycopersicon esculentum* Mill.) and common bean (*Phaseolus vulgaris* L.). In addition, *B. tabaci* is a vector of plant begomoviruses (Geminiviridae: Begomovirus). These whitefly-transmitted viruses (WTVs) are among the most destructive plant viruses; early virus infection often results in total crop loss.

Although the problems caused by *B. tabaci*, both as pest and vector, have been recognized for more than 100 years, serious damage had been limited to a handful of crops in particular geographic areas. This scenario has changed over the past 2 decades. The known WTVs have extended their geographic range and other WTVs are emerging in new crops and geographic zones, globally. Whitefly infestations now have become severe in both traditional and non-traditional food and industrial crops throughout the tropics.

In Africa, cassava mosaic disease (CMD) was first described in 1894 from Tanzania (Warburg, 1894). In East Africa, the disease was not reported to cause serious losses until the 1920s. In West Africa, it was first recorded in the coastal areas of Nigeria, Sierra Leone and Ghana in 1929 and had spread northward by 1945. By 1987, CMD had been reported from all countries in Africa producing cassava (*Manihot*

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esculenta Crantz) (Fauquet and Fargette, 1990). The disease had been caused by African cassava mosaic virus (ACMV) in South Africa and Africa west of the Rift Valley, and by East African cassava mosaic virus (EACMV) in Madagascar and Africa east of the Rift Valley (Hong et al., 1993; Swanson and Harrison, 1994). ACMV and EACMV are both *B. tabaci*-transmitted begomoviruses.

In 1988, an extremely severe form of CMD was reported from north-eastern Uganda. Infected cassava plants showed severe disease symptoms and produced little or no yield. The epidemic caused total crop failure in north-eastern Uganda (Otim-Nape et al., 1997). Each year since 1988, the epidemic moved progressively southwards along a broad front at a rate of approximately 20 km per year (Otim-Nape et al., 2000). By 1995, the epidemic had reached Kenya (Otim-Nape et al., 2000), with losses estimated at 140,000 tons of cassava per year, conservatively worth US$14 million (Legg, 1999). The severe outbreak in Uganda and neighbouring countries has been associated with a novel recombinant (EACMV+ACMV) begomovirus, currently recognized as EACMV-Ug (Pita et al., 2001).

Soon after, Latin America was struggling with *Bemisia*-transmitted bean and tomato viruses. *Bean golden mosaic virus* (BGMV) was first described in Brazil in the early 1960s as a minor disease (5%-10% incidence) of common bean in the state of São Paulo (Costa, 1965). Since then, BGMV has become a widespread problem in Brazil and now is known to cause epidemics in north-western Argentina and eastern Bolivia. Simultaneously, another begomovirus, *Bean golden yellow mosaic virus* (BGYMV), which causes similar symptoms, was ravaging common bean production in over 12 countries of Central America and the Caribbean (Morales, 1994). Yield loss from either BGMV or BGYMV is often 100% because of the high incidence of flower abortion and the malformation of pods in infected plants (Morales and Niessen, 1988). BGMV and BGYMV are considered to be the limiting biotic constraint to bean production in Latin America (Gálvez and Morales, 1989).

Then, in the late 1980s, tomato-producing areas in Latin America and across the tropics began to suffer from high incidences of whitefly-borne begomoviruses with devastating economic consequences. While no formal assessment studies of crop loss had been undertaken for the tomato diseases caused by begomoviruses, the empirical data were impressive. One of the more complete data sets came from the Dominican Republic (Polston and Anderson, 1997). In 1988, multiple begomoviruses began to affect tomato production in the Dominican Republic. Crop damage from 1988 to 1995 ranged from 5% to 95%. Economic losses in 1988 were estimated at US$10 million dollars, with losses from 1989 to 1995 totalling an estimated US$50 million dollars.

These scenarios were being repeated across the tropics, with significant consequences to food security and poverty alleviation efforts as well as human and ecosystem health.

Food security

In Africa, cassava is one of the most widely grown staple crops and second only to maize (*Zea mays* L.) in terms of calorie intake in sub-Saharan Africa. About 200 million people, or 40% of the sub-Saharan Africa population, rely on cassava for their well-being. In some countries, people derive approximately 1000 calories a day, or 50% of daily food intake, from cassava (IITA, 1988).

In addition to the yield losses caused by CMD, an estimated 150,000 ha of cassava-growing land was abandoned in Uganda during the height of the epidemic; that is, equivalent to over 2.2 million metric tons (US$440 million). This caused food shortages and famine in a number of districts, particularly in the eastern and northern regions where the crop was the principle staple food (Otim-Nape, 1997).

In Latin America, common bean is one of the main staple foods, particularly among the rural and urban poor. In Central America, beans are the most important source of protein, usually being consumed three times a day. Central America, despite its small area (498,368 km^2), devotes twice as much of its geographical area to the cultivation of beans (735,00 ha), when compared to major bean producers such as Brazil (>5,000,000 ha). Beans also are produced in some Caribbean islands, such as Cuba (26,000 t), the Dominican Republic (55,000 t) and Haiti (56,000 t) where they play an important nutritional role in the diet of the lower socio-economic classes. Smallholdings cultivated by farmers with limited resources are characteristic of bean production in Central America and the Caribbean. In El Salvador, for instance, 85% of the bean producers cultivate less than 14 ha, and 50% of these producers have holdings of less than 3.5 ha.

Despite the large area planted to beans in Central America, average productivity is low (495 kg/ha) compared to the average yield expected (over 1500 kg/ha) in most bean-producing regions of the USA and other temperate countries in the world. The main factor identified as responsible for the low bean productivity in Latin America has been the incidence of biotic constraints, particularly diseases. Morales (1992) estimated that by the early 1990s, approximately 2,500,000 ha were under attack by BGMV/BGYMV and that at least 1 million additional hectares could not be planted to beans each year because of the possibility of total yield losses, mainly during the dry seasons, when whitefly populations peak. Throughout Central America and the Caribbean Basin, figures for crop damage indicated that the BGYMV infection was devastating. And there was continuing consensus that BGYMV was the major biotic factor limiting bean production in Latin America (Morales, 1994).

Poverty alleviation

Many resource-poor producers in the tropics modified their traditional cropping systems in the 1980s, to incorporate non-traditional cash and export crops. The shift from subsistence to commercial agriculture is an opportunity to alleviate poverty in rural areas. Smallholder producers across the tropics have begun to cultivate cash crops such as tomato (*Lycopersicon esculentum*), pepper (*Capsicum* spp. L.), melon (*Cucumis melo* L.), watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai), squash (*Cucurbita moschata* Duchesne), grape (*Vitis vinifera* L.), okra (*Abelmoschus esculentus* [L.] Moench), cucumber (*Cucumis sativus* L. var. *sativus*), cabbage (*Brassica oleracea* L.), eggplant (*Solanum melongena* L.), broccoli (*Brassica oleracea* L. var. *italica* Plenck) and ornamentals. However, most of these horticultural crops are particularly susceptible to pests previously unknown to most small-scale farmers.

In the absence of adequate technical assistance for small-scale farmers in the tropics, the new pest problems were primarily dealt with by a myriad of pesticides applied indiscriminately. To further complicate this situation, a new biotype (B) of *B. tabaci* was introduced in the Americas in the early 1990s. This biotype proved to be far more polyphagous and fecund that the original A biotype and has been an important factor in most of the major outbreaks of whiteflies and whitefly-borne viruses. During the past decade, the outbreak of whitefly pests and the epidemics caused by whitefly-transmitted begomoviruses in cash crops has been often devastating. Pesticide abuse is the norm in all whitefly-stricken regions of the tropics, which increases production costs and disqualifies contaminated (pesticide residues) produce for export.

At present, many of the small-scale farmers that attempted to diversify their cropping systems have failed to do so because of whitefly-associated problems. Prime agricultural land remains unexploited in many developing countries during the dry seasons, when whitefly populations peak, despite the availability of water (irrigation districts) in these areas. The welfare of developing countries and their low-income citizens is tightly linked to the existence of cash-earning commodities, principally agricultural products.

The main force driving whitefly outbreaks and crop losses due to whitefly attack and WTVs is the lack of qualified technical assistance to small- and medium-scale farmers in the tropics. The Tropical Whitefly Integrated Pest Management (TWF-IPM) Project is currently entering a final phase when most of the information presented in this book and the lessons learned during the validation of IPM practices in Phase II will be translated into practical recommendations for resource-poor farmers throughout the tropics.

Human and ecosystem health

The reliance of farmers on agrochemicals to protect their cash crops against whitefly pests and whitefly-borne viruses has resulted in the systematic destruction of natural enemies that were once effective in providing natural control, whitefly populations with high levels of insecticide resistance and the creation of new secondary pest problems. The rejection of contaminated produce in international markets has led to the sale of highly contaminated food products in developing countries, with obvious detrimental health consequences for rural and urban consumers alike. Whiteflies provide a classic example of the pesticide treadmill. Insecticide abuse has become a serious threat to the environment as well as a health hazard to producers and consumers.

About 10% of the world’s population lives in the main highlands and mountainous areas of the developing world (the Andes of South America, the Africa ranges and the Himalayas). The management of resources in those ecosystems affects an additional 40% of the world’s population, which inhabits adjacent areas such as Inter-Andean valleys in the Andean Zone. Traditionally, people in the highlands have been marginalized from major development efforts, with significant repercussions on poverty, migration and social unrest as well as environmental deterioration *in situ* and downstream (IFPRI, 1995).

One major issue regarding the welfare of those living in highland areas of Latin America is environmental degradation caused by excessive pesticide use. It is well known that pesticide consumption in the developing world is increasing rapidly. It was thought initially that increases in pesticide use in Latin America were mainly due to the growth of plantation crops, an important source of export revenue (Bellotti et al., 1990). However, as pointed out by Whitaker (1993), the developing country share of the world agrochemical usage, currently valued at US$10.6 billion, was forecast to rise from 19% in 1988 to an estimated 35% by the year 2000. Much of this projected growth was attributed to a wider and more intensive use of chemical protection by smallholder farmers. Unfortunately, highly toxic insecticides represent up to 45% of pesticide use in the developing world.

In the early 1980s, the greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood), became a very serious pest of several hillside-grown crops in the Andes. Major outbreaks occurred in 1987, 1991 and 1994 in selected areas of Colombia, northern Ecuador and the Constanza Valley in Dominican Republic. For example, insecticide use by small-scale bean farmers, which was negligible until 1975-77 (Schoonhoven and Cardona, 1980), has increased steadily and became excessive in Colombia and Ecuador. Surveys conducted in two regions of Colombia and the northern part of Ecuador (CIAT, 1994) revealed that 100% of 893 farmers surveyed sprayed their crops in an attempt to control the greenhouse whitefly. Highest insecticide use occurred in the Sumapaz region of Colombia where farmers make an average of 11.1 applications per season.

Most alarming was the fact that some growers sprayed their crops up to 24 times in a crop cycle of 90-100 days, that is, every 3 to 4 days. Insecticide abuse on beans in Colombia (5.6 kg active ingredient per ha per season) could be compared to insecticide consumption for cotton (*Gossypium hirsutum* L.) (6.2 kg active ingredient per hectare per season), a crop that has been known traditionally as the worst offender in terms of insecticide use. And, of the insecticides utilized for whiteflies, 78% were classified in toxicological category I (highly toxic) and most often applied in mixture with other insecticides, broad-spectrum fungicides and foliar fertilizers. Farmers usually did not take precautions when handling pesticides; up to 24% of those surveyed admit that they have been intoxicated at least once in the prior 10 years (CIAT, 1994).

In the east African highlands, a contributing factor to environmental degradation was the indiscriminate and/or excessive use of broad-spectrum insecticides as part of the intensive crop protection system in industrial crops, especially cotton, and to some extent on plantation crops such as coffee (*Coffea* spp. L.) and tea (*Camellia sinensis* [L.] Kuntze). The problems of pesticide resistance and pest resurgence although initially created on these target crops had extended to the other seasonal crops grown in rotation and/or combination with the plantation crops, as in the case of whiteflies.

The increasing importance of horticultural production also brought with it the attendant strategy of pesticide-based protection in an effort to harvest damage-free produce. Intensive use of pesticides in vegetable crops had become quite common in large areas of Sudan, Uganda, Kenya, Tanzania and Zimbabwe. In a survey of vegetable farmers in Kenya it was found that the majority perceived that they would lose up to 90% of their harvest if they did not use pesticides. By the early 1990s, these vegetable farmers were applying up to 19 sprays a season, with a significant proportion of farmers spraying 9-12 times per season (KARI-GTZ, 1994). It was critical to begin work in eastern Africa in hopes of intervening with alternative whitefly crop protection measures, before the whitefly pests and WTVs reach the severity witnessed in the Neotropics.

There was an urgent need to develop IPM systems that would reduce pesticide use and help re-establish the ecological equilibrium by means of non-chemical approaches to whitefly management. For whitefly pests this implies identifying the principal crop hosts, establishing economic injury levels (EILs) and developing new approaches to maintain whiteflies below the EIL. For whitefly vectors, however, the traditional IPM approach will not suffice. Vectors must be studied and managed within an epidemiological framework, that is, study and analysis of the WTV system with IPM intervention strategies resulting from the epidemiological analysis.

Origin and Organization of the CGIAR TWF-IPM Project

The Systemwide Programme on Integrated Pest Management (SP-IPM)

The Consultative Group for International Agricultural Research (CGIAR) consists of 15 International Agricultural Research Centres (IARCs) located around the world. Historically, each Centre has worked in a relatively independent and autonomous fashion. During the 1990s, the CGIAR Secretariat recognized that significant benefits could be derived by pooling human capital, infrastructure and economic resources to jointly tackle research problems that numerous centres were working on independently. New inter-centre initiatives included the creation of the SP-IPM. Whitefly and WTV research had been ongoing at several of the IARCs since the 1970s. Due to the growing importance of the whitefly and WTV problem, the TWF-IPM Project was the first inter-centre project approved within the new programme. The International Center for Tropical Agriculture (CIAT, the Spanish acronym) was designated as the convening centre to organize the Inter-Centre Whitefly IPM Task Force and to formulate the Inter-Centre proposal on Sustainable Integrated Management of Whiteflies as Pests and Vectors of Plant Viruses in the Tropics (the CGIAR TWF-IPM Project).

A Task Force Meeting was held at CIAT in Cali, Colombia, from 13-15 February 1996. The objectives of the meeting were to discuss: (a) the goal, purpose, outputs and activities that should be proposed for the project; and (b) a structure for the global Whitefly IPM project, as well as how to link and coordinate the institutions that would be involved in the project.

The Task Force Meeting included 24 participants representing IARCs, National Agricultural Research Systems (NARS) and Advanced Research Institutions (ARIs). After considerable discussion on the nature of the whitefly problem, the Task Force agreed that it was possible to define three whitefly problems that should be prioritized:

(1) Whiteflies as vectors in mixed cropping systems in low to mid altitudes of the tropics;
(2) Whiteflies as pests in mixed cropping systems in tropical highlands; and
(3) Whiteflies as vectors and pests of the semi-perennial cassava.

The first problem focused on whiteflies as vectors of plant viruses in annual crops, especially legumes and vegetables, in the tropical lowlands. The second problem focused on whiteflies as direct pests in annual crops in the highlands. And the third problem focused on whitefly pests and vectors in a semi-perennial crop, cassava.

Based on the initial Whitefly IPM Task Force Meeting, and within the CG framework for an eco-regional problem approach (Bouma et al., 1995), the project was structured into six sub-projects:

(1) *Bemisia tabaci* as a virus vector in cassava and sweet potato in sub-Saharan Africa – led by IITA;
(2) *Bemisia tabaci* as a virus vector in mixed cropping systems of the Caribbean, Mexico and Central America – led by CIAT;
(3) *Bemisia tabaci* as a virus vector in mixed cropping systems of Eastern and Southern Africa – led by the International Centre of Insect Physiology and Ecology (ICIPE);
(4) *Bemisia tabaci* as a virus vector in mixed cropping systems of S.E. Asia – led by the Asian Vegetable Research and Development Center (AVRDC);
(5) *Trialeurodes vaporariorum* as a direct pest in the tropical highlands of Latin America – led by CIAT; and
(6) Whiteflies as direct pests on cassava in South America – led by CIAT.

There was consensus by the Task Force on the project goal, project purpose and project outputs for the TWF-IPM project. The project goal is to improve living conditions of rural families through the effective management of whiteflies, resulting in increased crop production and a safer environment. The project purpose is to reduce crop losses due to whitefly feeding damage and WTVs. The Task Force agreed to organize the project around six outputs:

(1) Formation of an international network for research on whiteflies and WTVs in the tropics;
(2) Characterization of whitefly problems in order to prioritize critical target areas;
(3) Improvement of the understanding of whitefly pest and disease dynamics in critical target areas;
(4) Development and testing of IPM strategies and tactics;
(5) Strengthening of NARS research capacity, policy formulation and IPM implementation; and
(6) Assessment of project impact.

Phase 1 of the CGIAR TWF-IPM Project

The project was conceptualized in three phases, to be carried out over 10 years. Phase 1 of the project would focus primarily on Output 1 (network formation) and Output 2 (problem characterization) as the basis for Phase 2 work, with limited activities in Output 3 (pest and disease dynamics) and Output 4 (development and testing of IPM strategies and tactics). The Project was officially launched in 1997 and evolved into a pan-tropical partnership. Five IARCs were included:

CIAT	Centro Internacional de Agricultura Tropical
IITA	International Institute of Tropical Agriculture
AVRDC	Asian Vegetable Research and Development Center
ICIPE	International Centre of Insect Physiology and Ecology
CIP	Centro Internacional de la Papa

Twelve ARIs were also included:

NRI	Natural Resources Institute (UK)
JIC	John Innes Centre (UK)
BBA	Biologische Bundesanstalt für Land-und Fortwirtschaft (Germany)
CATIE	Centro Agronómico Tropical de Investigación y Enseñanza (Costa Rica)
UA	University of Arizona-Tucson (USA)
UFL	University of Florida-Gainesville (USA)
UW	University of Wisconsin-Madison (USA)
MSU	Montana State University (USA)
FSAC	Florida State Arthropod Collection
USDA-(ARS)	U.S. Department of Agriculture-Agricultural Research Service
CSIRO	Commonwealth Scientific and Industrial Research Organisation (Australia)
C&F	New Zealand Crop and Food Research

And 31 National Agricultural Research and Extension Systems (NARES) in Latin America, the Caribbean, Africa and Asia were included from:

Mexico, Guatemala, Honduras, El Salvador, Nicaragua, Costa Rica, Panama, El Salvador, Cuba, Haiti, Dominican Republic, Colombia, Ecuador, Sudan, Benin, Ghana, Nigeria, Cameroon, Kenya, Uganda, Tanzania, Malawi, Madagascar, Nepal, Sri Lanka, Bangladesh, Thailand, Malaysia, Vietnam, Indonesia and the Philippines.

This collaborative partnership was possible due to the support of six donor partners:

DANIDA	Danish International Development Agency
ACIAR	Australian Centre for International Agricultural Research
USAID	United States Agency for International Development
MFAT	Ministry of Foreign Affairs and Trade (New Zealand, now New Zealand Aid)
USDA-(ARS)	U.S. Department of Agriculture-Agricultural Research Service
DFID	Department for International Development (UK)

The CGIAR TWF-IPM Project was launched in 1997 with the DANIDA-funded Phase 1 project on Sustainable Integrated Management of Whiteflies as Pests and Vectors of Plant Viruses in the tropics. This project covered activities in Output 1 (network formation) and Output 2 (characterization) activities in 23 African and Latin American partner countries in Sub-projects 1, 2, 3 and 5. ACIAR then approved funding for the project Sustainable Integrated Management of Whiteflies as Pests and Vectors of Plant Viruses in S.E. Asia, which covered Output 2 (characterization) activities in the eight Asian countries in Sub-project 4. The USAID-funded project on Biological Control of Whiteflies by Indigenous Natural Enemies for Major Food Crops in the Neotropics and the MFAT-funded project on Sustainable Integrated Management of Whiteflies through Host

Plant Resistance contributed to Output 2 (characterization) and Output 4 (IPM) activities in Sub-project 6. USDA-ARS signed a Scientific Cooperative Agreement to link the ARS national research on whitefly IPM research with the international effort. This agreement on Integrated Pest Management Strategies for Whitefly-transmitted Viruses supported Output 1 (network formation) and preliminary Output 3 (disease dynamics) research activities on epidemiology of whitefly-transmitted begomoviruses.

The CMD epidemic in eastern Africa was of particular concern to the TWF-IPM Project for humanitarian reasons. The USAID Office of Foreign Disaster Assistance (OFDA) granted special funding for the Emergency Programme to Combat the Cassava Mosaic Disease Pandemic in East Africa. The objective of this disaster assistance is to boost production of cassava in Uganda, Kenya and Tanzania and enhance both short- and longer-term food security through the implementation of an emergency program to multiply and disseminate CMD-resistant cassava (Output 4 activities). And DFID funded the project on Promotion of and Technical Support for Methods of Controlling Whitefly-borne Viruses in Sweet Potato in East Africa; sweetpotato (*Ipomoea batatas* [L.] Lam.) served as the principal food security crop in the midst of the CMD epidemic.

Whiteflies and Whitefly-Borne Viruses in the Tropics: Building a Knowledge Base for Global Action reports the results from Phase 1 of the CGIAR TWF-IPM Project. The purpose of Output 2 (problem characterization) was to gather, review, generate and analyse baseline data relevant to the diagnosis and characterization of whitefly and WTV problems in the tropics, in order to propose a sound research agenda for improved understanding of pest and disease dynamics, IPM development and IPM implementation. Several of the project coordinators have published review articles (Polston and Anderson, 1997; Legg, 1999; Morales, 2000; Bellotti and Arias, 2001; Morales, 2001; Morales and Anderson, 2001; Morales and Jones, 2004). Baseline data were generated through extensive survey work in each of the participating tropical countries. Surveys included the collection and identification of biological specimens (whitefly species, whitefly biotypes, geminiviruses, and *B. tabaci* natural enemies) and collection and analysis of socio-economic data and IPM practices, based on producer interviews. All partners have presented preliminary results of the extensive survey work and IPM measures in international, regional and national meetings. This book is the first presentation of detailed results generated by the Phase 1 projects contributing to the CGIAR TWF-IPM Project. For further information on specific methodologies used to produce the results published in this book, please contact the information and communication assistant of the project (www.tropicalwhiteflyipmproject.cgiar.org).

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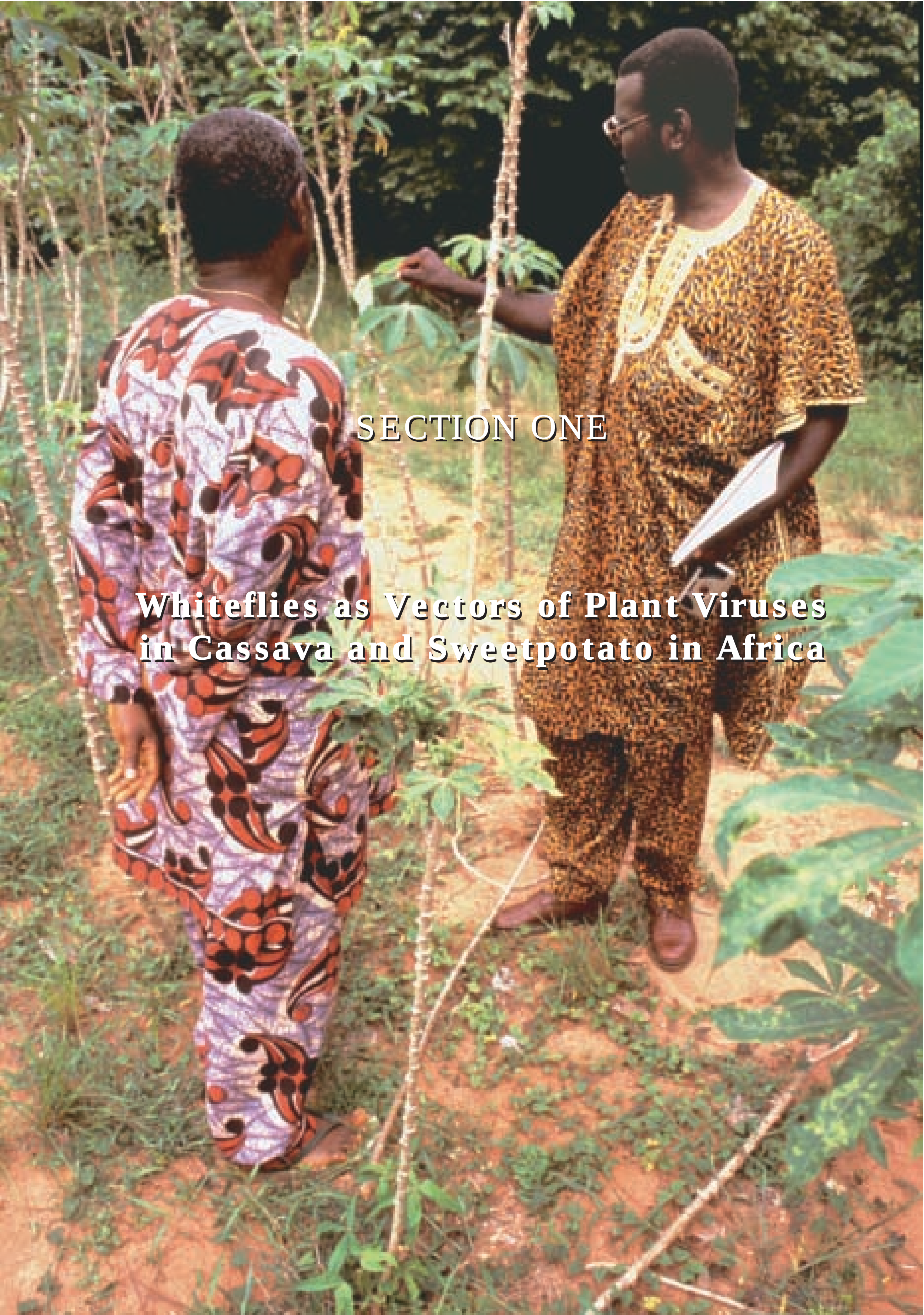

SECTION ONE

Whiteflies as Vectors of Plant Viruses in Cassava and Sweetpotato in Africa

BLANCA 14

CHAPTER 1.1

Introduction

*James Legg**

More than 1200 species of whitefly (Homoptera: Aleyrodidae) have been described worldwide, of which roughly one quarter occurs in Africa (Mound and Halsey, 1978). Of these diverse whitefly species, only a few are of significant economic importance. These include: *Aleurocanthus spiniferus* (Quaintance), *Aleurocanthus woglumi* Ashby, *Aleurodicus dispersus* Russell, *Aleurothrixus floccosus* (Maskell), *Aleyrodes proletella* (Linnaeus), *Bemisia tabaci* (Gennadius), *Dialeurodes citri* (Ashmead), *Parabemisia myricae* (Kuwana), *Siphoninus phillyreae* (Haliday), *Trialeurodes vaporariorum* (Westwood) and *Vasdavidius indicus* (David and Subramaniam) Russell. Principal among these, however, is *B. tabaci*, which is a pest on a wide range of field crops throughout the continent.

B. tabaci was first described from tobacco (*Nicotiana tabacum* L.) in Greece and is thought to have originated from the Old World (Frohlich et al., 1996). It has been recognized as an important agricultural pest in Africa since at least the 1930s, with several of the earliest reports relating to damage in cotton (*Gossypium hirsutum* L.) (Kirkpatrick, 1931) and cassava (*Manihot esculenta* Crantz) (Kufferath and Ghesquière, 1932; Storey and Nichols, 1938). By the 1950s, *B. tabaci* was reported causing major losses in cotton as a result of population resurgence of this species when it developed resistance to pesticides applied against other pests (Joyce, 1955). And, by the 1980s, *B. tabaci* had been associated with a major pandemic of cassava mosaic disease (CMD) in East Africa (Otim-Nape et al., 1997; Legg, 1999).

Cassava and Sweetpotato in Sub-Saharan Africa

According to international production statistics (FAO, 2004), Africa produces almost half of the world's crop of cassava and East Africa is one of the world's most important areas for growing sweetpotato (*Ipomoea batatas* [L.] Lam). Nigeria is the world's leading producer of cassava, while Uganda ranks third, worldwide, in total sweetpotato production. These two crops are of major importance to sub-Saharan Africa and play vital roles in sustaining food security, particularly in view of their ability to provide reasonable yields even under conditions of poor soil fertility or drought (Jennings, 1970).

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Cassava was brought to Africa in the sixteenth century by Portuguese seafarers and, perhaps because of this relatively recent introduction, has few major pests and diseases. This is in contrast to the wide diversity of pests and diseases that affect this crop in its native South America (Bellotti et al., 1994). The two most important arthropod pests of cassava in Africa, the cassava mealybug (*Phenacoccus manihoti* Matile-Ferrero) and the cassava green mite (*Mononychellus tanajoa* [Bondar]) were both introduced from South America in the early 1970s (Yaninek and Herren, 1988; Neuenschwander, 1994). They spread rapidly to all cassava-producing zones of mainland Africa during the 1980s and 1990s but both have been controlled successfully through the implementation of continent-wide biological control programs (Herren and Neuenschwander, 1991; Yaninek et al., 1993).

The principal diseases of cassava in Africa are CMD and cassava bacterial blight. The latter, like the two arthropod pests described above, was introduced into Africa in the 1970s and spread rapidly to all cassava-growing areas (Boher and Verdier, 1994). The disease is caused by the bacterium *Xanthomonas axonopodis* pv. *manihotis* (Xam). Although, under certain circumstances, damage may be very severe and result in total crop loss, outbreaks with serious effects are usually localized. Cassava bacterial blight is therefore less economically important than CMD.

CMD was first described more than a century ago (Warburg, 1894) from what is modern-day Tanzania. It was assumed from the early years of the twentieth century that the disease was caused by a virus, since no pathogens were visible on affected plants and the condition was shown to be graft transmissible (Zimmerman, 1906). It was not until the 1970s, however, that the viral etiology was confirmed (Bock and Woods, 1983). CMD is caused by begomoviruses (Bock and Woods, 1983; Hong et al., 1993), which are transmitted by *B. tabaci* (Storey and Nichols, 1938; Chant, 1958; Dubern, 1979). Cassava mosaic begomoviruses occurring in Africa are thought to be indigenous to that continent; it is assumed that they moved into cassava from wild host plants following the introduction of cassava from South America (Swanson and Harrison, 1994).

Cassava was subsidiary in importance to other staple crops until the early part of the twentieth century but promotion of the crop as a famine reserve by colonial authorities in the 1920s and 1930s rapidly led to it being more widely grown and consumed (Hillocks, 2002). It appears that this increase in cultivation area and intensity, coupled possibly with the unrestricted movement of germplasm between countries, catalyzed the continent-wide spread of CMD. New occurrences as well as the increasing importance of CMD were reported from numerous countries in East and West Africa, including Sierra Leone (Deighton, 1926), Uganda (Hall, 1928), Ghana (Dade, 1930), Nigeria (Golding, 1936) and Madagascar (François, 1937). Losses due to CMD in Africa have been estimated at between 12 and 23 million tons annually, equivalent to between US$1200 and US$2300 million (Thresh et al., 1997). Following the success of biological control in reducing losses caused by cassava mealy bug and cassava green mite, CMD generally is regarded as the most important biotic constraint on cassava production in Africa.

Sweetpotato in Africa is attacked by a greater diversity of pests and diseases than is cassava, although, as with cassava, few are of major economic importance. The most important pests include the sweetpotato weevil (*Cylas* spp.), the striped sweetpotato weevil (*Alcidodes dentipes* [Oliver]), the sweetpotato butterfly (*Acraea acerata* Hewitson), leaf blight (*Alternaria* spp.) and sweetpotato virus disease (SPVD). The latter has been ranked as the single most important constraint to sweetpotato production in the East African region and is the most important disease throughout Africa (Geddes, 1990). It results from co-infection of sweetpotato plants with the aphid-borne *Sweetpotato feathery mottle virus* (SPFMV) and the *B. tabaci*-borne *Sweetpotato chlorotic stunt virus* (SPCSV) (Schaefers and Terry, 1976; Gibson et al., 1998).

Status of CMD and SPVD Research

Since its description more than a century ago, CMD has received significant research attention. Fauquet and Fargette (1990) and Thresh et al. (1994; 1998) have reviewed a wide range of studies on the biological and molecular characterization, etiology, epidemiology, yield loss and control of these viruses. Fishpool and Burban (1994) and Legg (1994) have reviewed studies on *B. tabaci* as the vector of cassava mosaic geminiviruses, including work on biotype characterization, population dynamics, virus transmission and movement within cassava fields. The emergence of a severe form of CMD in the 1990s (Otim-Nape et al., 1997; Legg and Ogwal, 1998; Legg, 1999) has had a devastating effect on cassava cultivation across a large area of East and Central Africa. Rapid expansion of the pandemic has been associated with a new, more virulent, cassava mosaic begomovirus (Deng et al., 1997; Zhou et al., 1997). Super-abundant *B. tabaci* populations associated with the pandemic also have been shown to result from a synergistic interaction with severely CMD-diseased cassava plants, in which the diseased plants appear to promote *B. tabaci* population increase, possibly through the disease improving the food quality of the plant for *B. tabaci* (Colvin et al., 1999). However, evidence has been presented for a link between a distinct *B. tabaci* genotype cluster and the pandemic in Uganda (Legg et al., 2002).

In contrast to CMD, knowledge of SPVD and the role of *B. tabaci* in the ecology of the disease is extremely limited. Studies have been restricted largely to virus characterization, etiology and transmission, virus-virus interactions, and yield loss assessments (Schaefers and Terry, 1976; Hahn, 1979; Gibson et al., 1997; 1998). Although the disease is known to occur widely in sub-Saharan Africa, virtually nothing is known about its prevalence in the major sweetpotato cultivation areas and at the inception of this study published incidence data were available only for Uganda (Aritua et al., 1998).

Although significant progress had been made previously in characterizing whitefly-transmitted viruses and whiteflies associated with CMD and SPVD, at the time of launching the present study, significant gaps in understanding remained (Table 1). This was most apparent for *B. tabaci*, where very little research other than one-time assessments of abundance and limited population dynamics studies had been done outside Uganda and the Ivory Coast.

Table 1. Scope of published work in sub-Saharan Africa for key fields of cassava mosaic disease (CMD) and sweetpotato virus disease (SPVD) research at time of starting present study in Project target countries of sub-Saharan Africa.

Disease	Country	Research topic[a]							
		Virus characterisation	*Bemisia tabaci* characterisation	CMD / SPVD prevalence	*B. tabaci* abundance	CMD / SPVD epidemiology	CMD / SPVD yield losses	Farmer perceptions	Natural enemies of *B. tabaci*
CMD	Uganda	(+)	(+)	+	+	+	+	(+)	(+)
	Kenya	(+)	-	(+)	(+)	+	+	-	-
	Tanzania	(+)	-	+	+	(+)	-	-	-
	Malawi	(+)	-	-	-	-	-	-	-
	Madagascar	(+)	-	-	-	-	-	-	-
	Ghana	(+)	-	+	+	-	-	(+)	-
	Benin	(+)	-	+	+	-	-	(+)	-
	Nigeria	(+)	-	+	+	+	+	(+)	-
	Cameroon	(+)	-	+	+	+	+	(+)	-
SPVD	Uganda	+	-	+	-	(+)	+	-	-
	Kenya	+	-	+	-	-	-	-	-
	Tanzania	(+)	-	-	-	-	-	-	-
	Malawi	-	-	-	-	-	-	-	-
	Madagascar	-	-	-	-	-	-	-	-

a. + Published literature available; (+) Limited study published; - No published literature available.

Sub-project on Whiteflies as Vectors of Viruses of Cassava and Sweetpotato in Sub-Saharan Africa

Within the framework of the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project, the purpose of the diagnostic phase of the sub-project was to gather, generate and analyse baseline data relevant to the diagnosis and characterization of whitefly problems and whitefly-transmitted virus problems in cassava and sweetpotato. The rationale behind the diagnostic phase was therefore to provide essential baseline information on CMD, SPVD and whiteflies on cassava and sweetpotato, to serve as the basis for the development and implementation of targeted and appropriate IPM strategies in subsequent phases of the project.

The sub-project involved collaboration among three international agricultural research centres, one national agricultural research organization in each of nine African countries, and three other research institutions outside Africa (Table 2). The inclusion of nine countries at this stage of the project and the use of a standardized protocol provided a unique opportunity to obtain data that could be compared from country to country and region to region. The scope of the study also facilitated the first systematic collection and characterization of whitefly-transmitted viruses, whitefly and whitefly natural enemy specimens from cassava and sweetpotato in sub-Saharan Africa.

Collaboration and selection of target areas

Countries were identified for participation in the sub-project primarily on the basis of the importance of cassava and/or sweetpotato cultivation to their farmers, although security concerns precluded the participation of some of the major producers of each crop. The nine African countries that were extensively surveyed—Ghana, Benin, Nigeria, Cameroon, Uganda, Tanzania, Kenya, Malawi and Madagascar—together represented 62% of cassava and 56% of sweetpotato production in Africa (FAO, 1999). Zambia was involved in the project only to a limited extent, implementing a sweetpotato virus diagnostic survey in collaboration with the Natural Resources Institute, UK. Sweetpotato work was carried out only in eastern and southern Africa, because the crop is not grown widely in West Africa. Table 2 summarizes the roles of each of the partners involved in the diagnostic phase.

Implementation of the work plan, results and analysis

Diagnostic surveys were conducted according to protocols set out in the standardized methodology agreed among the project partners. In each country, three or four target areas were identified for the survey. The criteria for selection of these areas were that they should be major root crop-producing zones and preferably should have contrasting agro-ecological characteristics. In each ecozone, the sites were selected at about 20-km intervals along roads. Where no cassava farms were encountered at the 20-km point, the next suitable site thereafter was selected. Field data and biological specimens were collected from 3-6 month-old cassava plantings on surveyed farms and producers were interviewed using the standardized project questionnaires.

Between 10 and 42 locations were used for sampling in each target area, and at each location a single set of samples and data was collected. The data set comprised: (1) a producer questionnaire relating to perceptions

Table 2. Partners involved in the diagnostic phase of the sub-project on Whiteflies as Vectors of Viruses of Cassava and Sweetpotato in sub-Saharan Africa and their roles.

Institution	Country	Role[a]
International agricultural research centres:		
International Institute of Tropical Agriculture (IITA)	Uganda/Benin	Co-ordination
Centro Internacional de la Papa (CIP)	Uganda	SPVD epidemiology
International Centre of Insect Physiology and Ecology (ICIPE)	Kenya	Whitefly and natural enemy species identification
National agricultural research organizations/institutes:		
Plant Protection and Regulatory Services Directorate (PPRSD)	Ghana	Diagnostic survey
Institut National de Recherche Agronomique du Bénin (INRAB)	Benin	Diagnostic survey
National Root Crops Research Institute (NRCRI)	Nigeria	Diagnostic survey
Institut de Recherche Agronomique (IRA)	Cameroon	Diagnostic survey
National Agricultural Research Organisation (NARO)	Uganda	Diagnostic survey
Tanzania Root and Tuber Crops Programme (TRTCP)	Tanzania	Diagnostic survey
Kenya Agricultural Research Institute (KARI)	Kenya	Diagnostic survey
Chitedze Research Station (CRS)	Malawi	Diagnostic survey
Centre National de Recherche Appliquée au Développement Rural (FOFIFA)	Madagascar	Diagnostic survey
Mount Makulu Research Station	Zambia	Diagnostic survey (SPVD only)
Specialized research organizations:		
Natural Resources Institute (NRI)	United Kingdom	Epidemiology of CMD and SPVD
Biologische Bundesanstalt für Land und Fortwirtschaft (BBA)	Germany	Sweetpotato virus diagnostics
John Innes Centre (JIC)	United Kingdom	Molecular diagnostics of cassava viruses and whiteflies

a. CMD, cassava mosaic disease; SPVD, sweetpotato virus disease.

and management of whiteflies and whitefly-transmitted virus problems; (2) biological samples of whiteflies, virus-diseased leaf and stem tissues, and whitefly natural enemies; and (3) field assessments of disease incidence, virus symptoms and whitefly abundance. Before carrying out the survey, lead scientists in each country were trained in survey methods. Surveys were conducted between November 1997 and September 1998. The results are reported in the subsequent country chapters in this section. Data from the producer questionnaires for all countries were entered into a single database from which they could be summarized and queried. Field data from all countries were summarized and integrated to develop maps (Chapter 1.14, this volume). Studies conducted in partnership with research institutions outside Africa were initiated in early 1998 and completed by mid-1999; results are reported later in this section (Chapters 1.11, 1.12 and 1.13) or in Section 5 (Chapter 5.3).

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CHAPTER 1.2

Ghana

*Anthony Cudjoe**, *Joseph Gyamenah** and
*Braima James***

Introduction

Cassava mosaic disease (CMD) is a major food production constraint in Africa (Thresh et al., 1994). It is caused by begomoviruses, which are vectored by the whitefly *Bemisia tabaci* (Gennadius) and can be spread through infected planting material. Yield losses caused by the disease are higher if vegetative planting materials sprout with symptoms (cutting infection) than if initially disease-free material becomes infected by *B. tabaci* following sprouting (current season whitefly-borne infection) (Fauquet and Fargette, 1990). In Ghana, recent work on *Bemisia* whiteflies and CMD has been carried out under the auspices of a regional project, Ecologically Sustainable Cassava Plant Protection (ESCaPP) (Yaninek et al., 1994). Unpublished results of ESCaPP diagnostic surveys showed cassava (*Manihot esculenta* Crantz) to be the most widely planted arable crop in the country (45% site incidence), followed by maize (*Zea mays* L.) (15%) and yam (*Dioscorea* spp.) (5%).

Whitefly survey samples consisted largely of *B. tabaci*. *B. afer* (Priesner and Hosny) was unevenly distributed and less abundant (Sotomey et al., 1995). Across ecozones and seasons, CMD was the most common cassava pest constraint both in terms of field data and farmer perceptions. In the field, total plant incidence of CMD was 76% in the dry season and 68% in the wet and, in 56% of the villages, farmers ranked CMD as the main disease. On an ascending 1-5 scale of damage classes, 22% of plants were in class 2 (mild), 51% in class 3 (moderate), 26% in class 4 (severe) and none in class 5 (very severe). Previous attempts to develop CMD control measures in Ghana have included preliminary evaluation of local cassava varieties in relation to their susceptibility to CMD and laboratory studies to produce virus-free tissue culture materials of local germplasm (Bieler et al., 1996; Kissiedu et al., 1996). In subsequent collaborative epidemiology trials, whitefly infection was consistently lowest in the variety TMS 30001 when compared to other improved or local varieties (Legg et al., 1997; IITA, 1998; James et al., 1998). The role of parasitoids, other natural enemies, genetic heterogeneity in *Bemisia* populations and the identity of cassava viruses/viral strains are yet to be investigated in the country.

This chapter gives results of a countrywide survey in Ghana, conducted in November and December

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1997, on 80 farms. These included 25 sites in each of the rainforest and transition forest, 10 sites in the "coastal" savannah and 20 sites in the wet savannah (Figure 1).

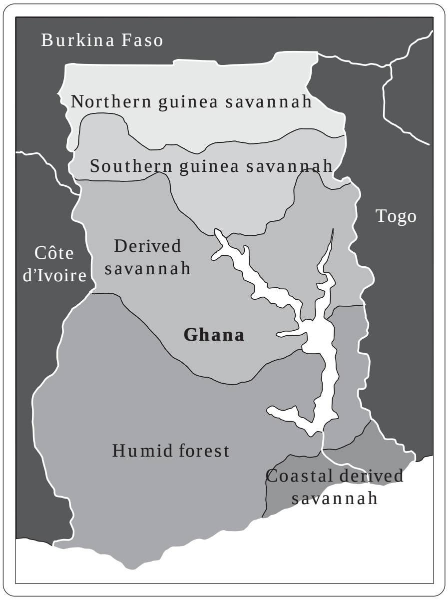

Figure 1. Areas surveyed for whitefly incidence and cassava mosaic disease in Ghana.

Increased Biological Understanding

B. tabaci, *B. afer* and *Trialeurodes vaporariorum* (Westwood) were identified from adult whitefly samples collected from cassava in Ghana. Whilst *B. tabaci* occurred at all survey sites, *B. afer* was recorded from only 3.8% of the sites and comprised 1.9% of the 151 specimens determined. *T. vaporariorum* was collected from only one location. Generally, farmers were unaware of *Bemisia* whiteflies and did not recognize them by specific local names; only 2% of them knew that the insects caused serious problems to cassava. By contrast, 54% of farmers knew that CMD caused serious problems and described the disease symptoms in various local names. Some of these, for example, *bankye kwata* and *kwata*, described the appearance of CMD symptoms. Such indigenous knowledge will facilitate farmer participatory learning and research activities. *Bemisia* whitefly abundance averaged 1.3 adults per cassava shoot tip in the rainforest, 1.0 in transition forest, 11.8 in coastal savannah and 0.3 in wet savannah (see also Figure 2 in Chapter 1.14, this volume). Whitefly mean abundance (logarithm transformed counts) was significantly higher in the coastal savannah than in each of the other ecozones ($t > 12.1$; $P < 0.05$). The natural enemy fauna associated with the whiteflies included the hymenopteran parasitoid *Encarsia sophia* (Girault and Dodd), which was reared from *Bemisia* mummies at four survey sites.

Increased Socio-Economic Understanding

Cassava production was characterized by small landholdings, typically less than 1 ha in size, and a preponderance of local landraces. The intensity of cassava farming decreased along the rainforest-savannah axis. The five most profitable crops listed by farmers were cassava (45%), yam (18%), maize (11%), cowpea (*Vigna unguiculata* [L.] Walp.) (6%) and tomato (*Lycopersicon esculentum* Mill.) (5%). These other crops were planted in association with, or near to, cassava fields but cassava was the most common crop (40% site incidence) planted in nearby fields. Among the associated crops, both cowpea and tomato are known host plants of *B. tabaci*. However, experimental data from other countries in Africa show that the *B. tabaci* biotype that occurs on cassava is largely restricted to that crop (Burban et al., 1992; Legg et al., 1994).

Priority cassava pests listed by farmers were vertebrates (16%), weeds (13%), termites and weevils (10% each) and CMD (6%). Overall, CMD incidence was mainly attributable to cutting infection (Figure 2). The incidence of cutting infection averaged 60% in the rainforest, 55% in the transition forest, 63% in the coastal savannah and 51% in the wet savannah. The incidence of whitefly infection, transformed to allow for the effect of multiple infection (Gregory, 1948), was significantly higher in the coastal savannah and rainforest than in the transition forest or wet savannah ($t > 2.9$; $P < 0.05$). Whitefly abundance showed no correlation with the incidence of whitefly infection. In terms of reducing CMD incidence, the transition forest and wet savannah would seem to provide better sites for multiplication of clean planting material.

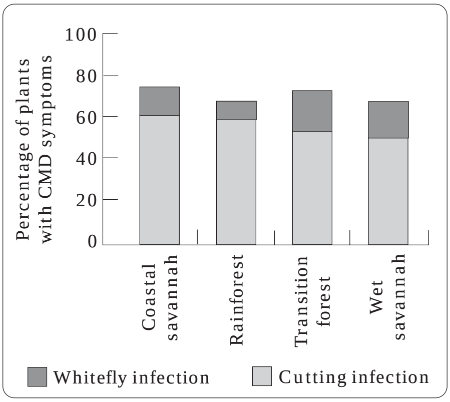

Figure 2. Cassava mosaic disease (CMD) incidence and source of infection in the ecozones of Ghana.

Across ecozones, 24%-32% of plants showed no CMD symptoms. About 10% of the plants showed serious (score 4) to severe (score 5) damage symptoms (Figure 3). The plants were 3-6 months old, a growth stage during which storage root formation and development is initiated and root yield is particularly vulnerable to pest-induced losses. In view of the higher proportion of plants in the moderate and severe damage categories in the transition forest, significant root yield losses could be expected in this ecozone. However, yield loss estimates provided by farmers appeared to be unrealistically high. For example, about 26% of farmers estimated losses at 50% and 20% of farmers at 75%. Such loss estimates would need to be validated, especially since 91% of the farmers reported that they sell their cassava harvest.

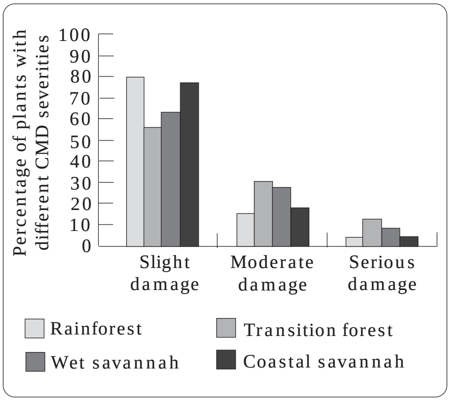

Figure 3. Cassava mosaic disease (CMD) damage severity in the ecozones of Ghana.

Farmers' opinion was almost equally divided on whether or not CMD was becoming more severe, and a sizeable proportion (34%) considered that the disease was not a yearly occurrence. Extension and training support for whitefly/CMD problems appeared low, since 89% of farmers reported receiving no technical information or assistance with these problems. In view of this, it is not surprising that only 1% of farmers practiced any deliberate whitefly/CMD control. Local varieties predominated (54% site incidence) and 18% of the farmers ranked Busuminsia as their single most preferred variety, followed by three other local varieties, Santom

(6.2%), Bakentenma (5%) and Kokoo (5%). Adoption of improved varieties was low: only 11% of farmers planted TMS 30572 (originating from the International Institute of Tropical Agriculture [IITA]) and “Agric” varieties (a locally adapted material of IITA origin) recommended by the national extension service on the basis of their CMD resistance. Agronomic features were more important criteria for selecting planting material than were disease-specific characteristics.

Where disease-specific selection criteria were used, only 10% of the farmers reported that the method was effective, while 9% noticed that the “clean” material became diseased after planting. Since the single most important primary source of planting material was farmers’ own fields (Figure 4), adoption of appropriate procedures for selecting planting material would significantly reduce recycling of cassava mosaic viruses. About 60% of farmers reported that they encountered shortage of planting material in some years. Shortages like these, coupled with the observed high incidence of cutting infection, imply that rapid multiplication schemes would be required to ensure timely availability of clean and healthy planting materials at farm level. No farmer reported pesticide use as a control method against the whiteflies or the disease but about 61% of farmers rogue diseased plants, mostly (55% of farmers) within the first month of planting.

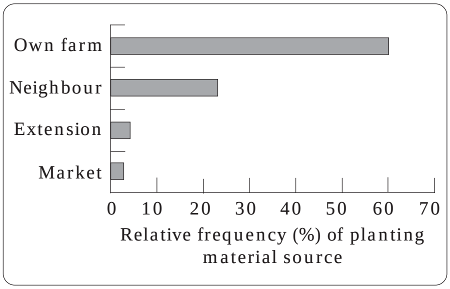

Figure 4. Farmers’ sources of cassava planting material in Ghana.

Conclusions

This study has increased field-level understanding of whiteflies and CMD incidence, and provides a strong basis for the development of ecologically sound management practices for whitefly/CMD problems in Ghana. The incidence of CMD infection by *B. tabaci* was low and the disease appears to be spread mainly through infected cuttings. The preponderance of local cassava varieties over proven CMD-resistant improved varieties contributes to the incidence of the disease in the country. Two key issues that need to be addressed are the quantification of yield losses in the preferred local cultivars and, associated with this, an assessment of why CMD-resistant varieties have not been adopted widely.

Assuming that yield losses warrant the use of CMD control measures, farmer participatory learning activities will promote farm-level selection of parent planting material that is CMD symptom free for making cuttings. Related to this, farmer-led rapid multiplication schemes would be required to increase the availability and adoption of CMD-resistant varieties, provided that evaluations linked to such schemes indicate that some of these varieties are acceptable to farmers. Research should also evaluate farmers’ preferred local varieties as possible sources of CMD resistance in breeding programs since this approach has provided important new impetus to resistance breeding programs for cassava elsewhere (Dixon et al., 1992).

Little is known about temporal and spatial changes in the abundance of *B. tabaci* in Ghana, particularly in relation to its ability to transmit cassava mosaic viruses over the growing period of the crop. Field studies therefore might be necessary to quantify such changes, assess whether parasitism by *E. sophia* has any significant impact on the insect's abundance and, in epidemiology trials, to ascertain the ability of *B. tabaci* to spread cassava mosaic viruses to a range of cassava varieties.

National research capacity needs to be enhanced in order to allow project partners to undertake further research on the whitefly/CMD issues raised here. In this regard, training of national program partners in appropriate areas would strengthen the collaborative linkages initiated in the pilot phase of the project and provide a strong base for providing sustainable technical support to cassava farmers. Key areas for strengthened national research capacity include post-graduate degree training in *B. tabaci* bionomics and cassava mosaic virology, and field-based training of technicians and extension workers in participatory processes and facilitation. Technician and extension training will increase scientific literacy in farming communities and foster action research and learning with farmers. This will improve farmers' currently poor access to information, will help them understand the causes and nature of the disease problem and will highlight effective cultural control options such as planting material selection and roguing.

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CHAPTER 1.3

Benin

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*Symphorien Saizonou***

Introduction

Preliminary research on cassava pests and diseases in Benin was carried out as part of a regional project for Ecologically Sustainable Cassava Plant Protection (ESCaPP) (Yaninek et al., 1994). Unpublished results of ESCaPP diagnostic surveys show cassava (*Manihot esculenta* Crantz) to be the predominant arable crop (87% site incidence) in the country and the next most common crops to be maize (*Zea mays* L.) with 30% site incidence and cowpea (*Vigna unguiculata* [L.] Walp.) with 26%. Summaries of plant protection aspects of the survey show cassava mosaic disease (CMD) caused by cassava mosaic begomoviruses (CMBs) and vectored by the whitefly *Bemisia tabaci* (Gennadius) to be the most common biotic constraint on cassava in the country, with total plant incidence of 49% in the dry season and 57% in the wet. Survey samples of red-eyed whitefly nymphs comprised *B. tabaci* and *B. afer* (Priesner and Hosny) although the latter was less abundant, occurring in only 4% of the samples (Sotomey et al., 1995). The CMD damage score was low, the average falling within class 2 (indicating “slight damage” on a 1-5 scale) across ecozones and seasons. During the surveys, in focus group interviews at village level, farmers at 68% of survey sites rated CMD as the main disease problem. In subsequent preliminary CMD epidemiology trials (Legg et al., 1997; IITA, 1998; James et al., 1998), current season whitefly infection was consistently lowest in the variety TMS 30001 when compared with other improved or local varieties. In a related population dynamics study, the parasitoid *Encarsia sophia* (Girault and Dodd) was identified from *Bemisia* mummies at each of six trial sites in the transition forest, and wet and dry savannahs (James and Gbaguidi, unpublished data). Genetic heterogeneity in *B. tabaci* populations, which is known to influence CMD epidemiology (Burban et al., 1992; Legg et al., 1994), and the identities of cassava viruses and virus strains are yet to be investigated in the country.

This chapter reports the results of a countrywide diagnostic survey of whiteflies and CMD in Benin, conducted in November and December 1997, on 60 farms distributed in the transition forest (28 sites), dry savannah (19 sites) and wet savannah (13 sites) (Figure 1). Farmer respondents in the survey were mostly men (93%) and landowners with over 5 years of cassava farming experience.

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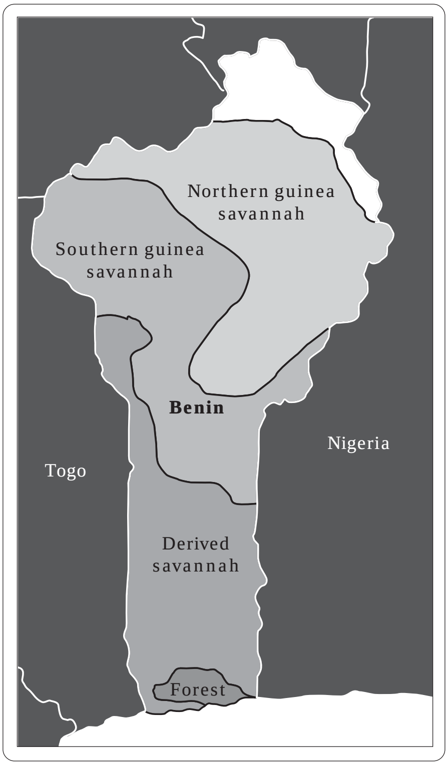

Figure 1. Areas surveyed for whitefly incidence and cassava mosaic disease in Benin.

Based on the survey results, the chapter identifies areas requiring more research attention to further our understanding of CMD in the country and to serve as a basis for developing and implementing an integrated approach to managing this disease.

Increased Biological Understanding

The co-existence of *B. tabaci* and *B. afer* on cassava in Benin was confirmed by the determination of these species from adult specimens. In various local languages, farmers recognized whiteflies simply as "insects". On the other hand, among the various local names used to describe CMD, the names *edjekpo*, *ekpo*, *goudou* and *kpanro* refer specifically to the resemblance of CMD symptoms to those of leprosy. This indigenous knowledge will greatly facilitate farmer participatory research and learning activities—activities that seem especially important given that a high proportion of farmers did not know that whiteflies (60% farmers) and CMD (50% farmers) caused serious problems to cassava. Whilst *B. tabaci* occurred at all 60 survey sites, *B. afer* was recorded in 50% of the sites and comprised 15% of the 212 specimens determined. Whitefly abundance was low and averaged 3.2 adults per cassava shoot tip in the transition forest, 0.5 in the wet savannah and 0.9 in the dry savannah. Whitefly mean abundance (logarithm transformed counts) was significantly higher in the transition forest than in either wet or dry savannah ($t > 5.04$; $P < 0.05$). The parasitoids *E. sophia* and *Eretmocerus* sp. were identified in natural enemy collections. The presence of *E. sophia* was confirmed at 15 sites and *Eretmocerus* sp. at only one site. Diseased whiteflies were not observed in the field and hence no entomopathogens were collected. Biotypes of cassava viruses identified are reported elsewhere (Chapter 1.11, this volume).

Increased Socio-Economic Understanding

Across ecozones, cassava was typically produced on farms of less than 0.25 ha. The improved variety "Agric", originating from the International Institute of Tropical Agriculture (IITA), was grown by 13% of farmers. The local variety Odounbo, grown by 7% of farmers, was the next most popular variety, while the remainder comprised a wide diversity of local cultivars. Fifty-eight percent of farmers ranked cassava as their most profitable crop, compared to 33% for maize, 10% each for cowpea and groundnut (*Arachis hypogaea* L.) and 5% for cotton (*Gossypium hirsutum* L.). These crops were planted also in association with cassava or near to cassava fields. Cowpea, groundnut and cotton are known crop host plants of *B. tabaci*, although experimental data from other countries in Africa show that the *B. tabaci* biotype occurring on cassava is more or less restricted to that crop (Burban et al., 1992; Legg et al., 1994). Cassava was also the most common crop planted in nearby fields (38% of all instances) and the intensity of cassava farming gradually decreased from the transition forest (2.4 nearby fields) to wet savannah (1.9 nearby fields) and dry savannah (0.2 nearby fields).

Average CMD incidence ranged from 25% to 49% across ecozones (Figure 2), with infected cuttings providing the more important source of infection. The incidence of cutting infection did not differ significantly between ecozones. The incidence of whitefly infection, transformed to allow for the effect of multiple infection (Gregory, 1948), differed significantly across ecozones and was higher in both the transition forest and wet savannah than in the dry savannah ($t > 2.06$; $P < 0.05$). Across ecozones, whitefly abundance showed no correlation with the incidence of whitefly infection. The pattern of damage was similar across ecozones, with most plants showing no CMD symptoms and, of those visibly infected with the disease, only a very small proportion showing moderate or severe damage (Figure 3). This is important in the context of the age of cassava farms sampled. The plants were 3-6 months old at the time of the survey, an age at which storage roots form and begin to develop and root yield is particularly vulnerable to pest-induced losses. None of the farmers reported total loss of their cassava to CMD, probably a reflection of the mild disease symptoms observed. However, 25% of farmers attributed losses of at least 25% of the yield to the disease. Whilst this high loss estimate seems to conflict with the low damage severity observed, 58% of farmers indicated that CMD is becoming more severe.

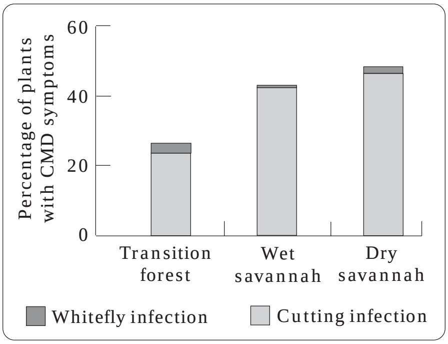

Figure 2. Cassava mosaic disease (CMD) incidence and source of infection in the ecozones of Benin.

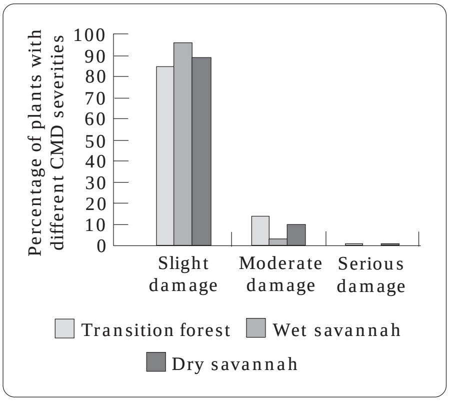

Figure 3. Cassava mosaic disease (CMD) damage severity in the ecozones of Benin.

Most farmers (78%) undertook no specific control measures against CMD or whiteflies, and less than 10% had received technical information or assistance on them. Among farmers who attempted to control the whitefly or disease problem, the main methods used were weeding and application of wood ash. Even though no farmer reported the use of resistant varieties as a whitefly/disease control method, further questioning revealed that 8% of farmers listed “Agric” and 5% listed Odounbo as varieties they had planted on the basis of recommendations that they were tolerant to CMD. Twenty percent of these farmers indicated that the better performance of these varieties was mainly because of better sprouting and establishment of the cutting. Agronomic features were more important than pest/disease resistance as criteria for the selection of planting material. About 12% of the selection criteria related to disease-free stems, health of stems and disease resistance, whilst 23% related to size, maturity, better yield, colour and size of the parent stem.

Where disease-specific criteria were used for selecting planting material, only 10% of the farmers reported that the method was effective and 7% noticed that the “clean” material became diseased after planting. Because the single most frequently cited sources of planting material were farmers’ own fields (30%) and neighbours’ fields (22%), adoption of appropriate planting material selection would be expected to reduce recycling of CMBs considerably. Whilst no farmer indicated roguing as a specific CMD control method, 7% of them conducted roguing to prevent and reduce disease and 2% because of poor plant growth; 7% of the farmers reported some impact from this control measure. However, the current high incidence of cutting infection would make roguing very labor intensive and unattractive for adoption, unless carried out in combination with the selection of CMD-free planting material. Pesticide use was rare.

Conclusions

In Africa, cassava root yield losses due to CMD have been estimated at 28%-40% annually (Thresh et al., 1994). In Benin, there appears to be a contradiction between field data and farmers’ perception of the CMD problem. The research data indicate only a very small proportion of cassava plants with moderate or severe disease damage but 25% of the farmers attribute high losses to the disease. In such cases, farmer participatory trials would help farmers and researchers jointly to better understand the relationship between disease incidence and yield, and to validate loss estimates. The studies would help establish whether losses to CMD really are significant in Benin and, if so, convince farmers of the need to adopt integrated management practices against the disease. Such an approach will provide for action learning by farmers to improve their access to information and understanding of the causes and nature of the disease problem. For example, cutting infection was found to be much more important than *Bemisia* whiteflies in the spread of CMD, suggesting that a participatory learning and extension effort focusing on planting material selection and sanitation and roguing could have a positive impact.

Generally, training of national program partners would strengthen the collaborative links established in the pilot phase of the project and further enhance national capacity to undertake priority CMD and whitefly research activities. For example, field studies on the population dynamics of *B. tabaci* would be needed to understand temporal and spatial changes in the insect’s abundance in relation to its ability to spread CMD. There is also the need to quantify the effectiveness of parasitism by *E. sophia*, particularly as a possible factor influencing the importance of the vector in spreading CMD. In view of the low abundance of *B. afer* in the country, it would not be appropriate to commit further resources to investigating its role in CMD epidemiology.

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CHAPTER 1.4

Nigeria

*Nnamdi C. Echendu**, *Joseph B. Ojo***,
*Braima James**** and *Brice Gbaguidi****

Introduction

Cassava (*Manihot esculenta* Crantz) is an important staple food crop in sub-Saharan Africa. In Nigeria, unpublished results of diagnostic surveys by a project for Ecologically Sustainable Cassava Plant Protection (ESCaPP) (Yaninek et al., 1994) show cassava to be the most important crop in the country, followed by yam (*Dioscorea* spp.) and maize (*Zea mays* L.). Cassava has the potential to yield 60 t/ha of storage roots (Nweke and Lynam, 1997). However, average yields in farmers' fields are usually less than 10 t/ha (FAO, 1989). The shortfall is associated largely with pest and disease problems, of which cassava mosaic disease (CMD) is one of the most important. In sub-Saharan Africa as a whole, losses to CMD have been estimated at 15-27 million tons, representing 15%-24% of total production (Thresh et al., 1997). The disease is caused by begomoviruses transmitted by the whitefly *Bemisia tabaci* (Gennadius) and is spread also by man through planting of infected planting materials (Hahn et al., 1981). The ESCaPP survey results showed CMD to be the single most common pest constraint on cassava in Nigeria with, on average, 85% of plants showing symptoms in the dry season and 79% in the wet (IITA, 1997). The surveys indicated the co-existence of *B. tabaci* and *B. afer* (Priesner & Hosny) on cassava in Nigeria, although *B. afer* was less abundant and widespread than *B. tabaci* (Sotomey et al., 1995).

In Nigeria, studies are continuing to examine the begomoviruses infecting cassava and their variability (Ogbe et al., 1998). These have recorded the presence of both *African cassava mosaic virus* (ACMV) and *East African cassava mosaic virus* (EACMV), although earlier less comprehensive studies had suggested the occurrence of only ACMV in Nigeria (Swanson and Harrison, 1994). Genetic heterogeneity in *B. tabaci* populations, which is known to influence CMD epidemiology (Burban et al., 1992; Legg et al., 1994), has yet to be investigated in the country. In preliminary CMD epidemiology trials, current season whitefly infection was consistently lowest in the variety TMS 30001 when compared to other improved or local varieties (Legg et al., 1997; IITA, 1998; James et al., 1998). The development and deployment of resistant varieties has been the main approach to CMD management pursued in Nigeria (Hahn et al., 1980; 1981). Little attention has been given to the potential for using natural enemies to manage the whitefly vector, although Jerath (1967) suggested that the

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parasitoid *Encarsia strenua* (Silvestri) and a predatory mite, *Typhlodromus* sp., were possible biocontrol agents.

This report presents results of a countrywide survey of *Bemisia* whiteflies and CMD conducted in November and December 1997, on 80 farms distributed across the main cassava-producing agro-ecological zones of Nigeria (Figure 1). Sites were sampled in rainforest (25), transition forest (25), wet savannah (20) and dry savannah (10) zones, and cassava plantings selected were 3 to 4 months old. This chapter summarizes the results of the survey and identifies areas requiring subsequent attention in order to further our understanding of the disease and to serve as a basis for developing and implementing the integrated management of CMD in Nigeria.

Figure 1. Areas surveyed for whitefly incidence and cassava mosaic disease in Nigeria.

Increased Biological Understanding

The insect vector of CMD, *B. tabaci*, occurred at all survey sites, while a second whitefly species, *Trialeurodes vaporariorum* (Westwood), hitherto unreported on cassava, was identified from nymphs collected from one location in the country. No examples of *B. afer* were found in samples collected. Some farmers recognized whiteflies and 17.5% of farmers knew that the insects caused a serious problem to cassava. In various local languages, farmers recognized *Bemisia* whiteflies simply as “insects”. About 24% of farmers knew that CMD caused serious problems and farmers in different localities described the disease in various local languages as *akpa*, *ekikang*, *ikwukwo*, *kpuojuju* and *nkpuk*—mostly terms that refer to the appearance of the disease symptoms. *Bemisia* abundance was less than three adults per cassava shoot tip (defined as the terminal shoot with the first five open leaves) in each of the ecozones.

Five species of hymenopterous parasitoids of *Bemisia* were recorded: *Encarsia sophia* (Girault and Dodd) comprising 93.7% of samples, *Eretmocerus* sp., 3.5%, *Encarsia* sp. (*luteola* group), 0.7%, *Encarsia lutea* (Masi), 1.2% and *Encarsia mineoi* Viggiani, 0.8%. Potential whitefly predators recorded were *Cheilomenes sulphurea* (Olivier) and phytoseiid mites. Diseased whiteflies were not observed in the field and entomopathogens therefore were not collected. The diversity in cassava viruses identified is reported in Chapter 1.11 (this volume). Other cassava pests listed by farmers were grasshoppers, reported by 15% of farmers, termites by 6.3%, and vertebrates by 2.5%.

Increased Socio-Economic Understanding

Cassava farm sizes averaged 0.38 hectares in the rain and transition forests, 0.56 hectares in the wet savannahs and 0.10 hectares in the dry savannahs. Both local and improved varieties were recorded in farmers' fields, although local varieties predominated. The most widely occurring improved varieties were

TMS 30572 (18.8%) and TMS 30555 (2.5%). There was a marked difference in the incidence of improved varieties between ecozones: they were cultivated in 80% of sampled fields in the transition forest but occurred in only about 30% of fields in the rainforest and wet savannah and were not recorded at all in the dry savannah. There was a direct negative correlation between the frequency of cultivation of improved varieties and CMD incidence. Farmers ranked cassava as their most profitable crop followed by maize, yam, sorghum (*Sorghum bicolor* [L.] Moench) and cowpea (*Vigna unguiculata* [L.] Walp.), in descending order. Of the crops planted in association with cassava or in adjacent fields, cowpea is a known host plant of *B. tabaci*. Experimental data from other countries in Africa, however, show that the *B. tabaci* biotype occurring on cassava is more or less restricted to that crop (Burban et al., 1992; Legg et al., 1994).

Average CMD incidence ranged from 45% to 83% across ecozones, with cuttings providing by far the more important source of infection (43% to 83%) (Figure 2). The incidence of whitefly infection, transformed to allow for the effect of multiple infection (Gregory, 1948), was low in each of the ecozones. Among plants with disease, slight damage symptoms (score 2 on a five-point scale from no damage to severe damage) predominated (Figure 3). Plant damage severity varied little by ecozone. Moderate damage symptoms (score 3) were more pronounced in the rainforest (30% of plants) and wet savannah (20% of plants) than in the other ecozones, whilst serious (score 4) to severe (score 5) damage symptoms were negligible (Figure 3). Plants were less than 4 months old at the time of the surveys, a growth stage during which storage root formation and development is initiated and root yield is particularly vulnerable to pest-induced losses. Very few farmers (1.2%) reported total yield loss in the first planting season but 18.8% of farmers attributed losses of at least one quarter of their yield to the disease. However, about 70% of the farmers provided no yield loss estimates for the first or second crop.

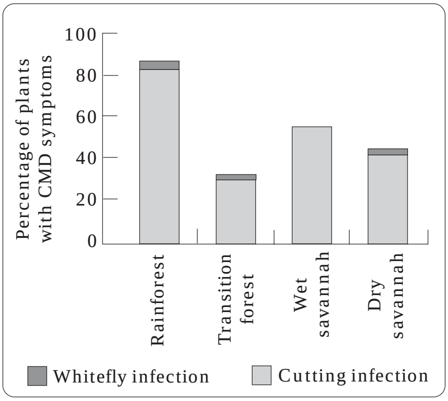

Figure 2. Cassava mosaic disease (CMD) incidence and source of infection in the ecozones of Nigeria.

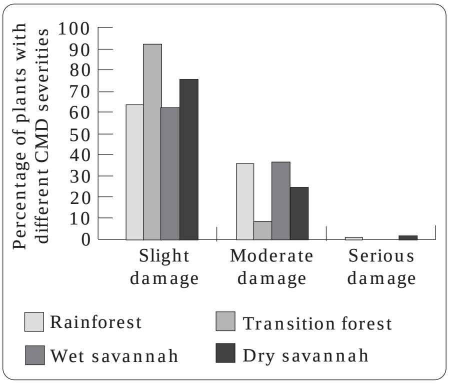

Figure 3. Cassava mosaic disease (CMD) damage severity in the ecozones of Nigeria.

Nearly all farmers reported that CMD occurred every year, while slightly more than half of them confirmed that CMD was most severe during the early growth stages of cassava. More than three-quarters of farmers (77.5%) reported that they had received no technical information or assistance on

CMD or whiteflies. Among farmers who attempted to manage the whitefly/disease problem, the main practices used were selection of planting materials, application of wood ash, roguing and proper weeding. Agronomic features were more important than pest/disease resistance as criteria for the selection of planting material. Since the most frequently cited sources of planting material were neighbours' and farmers' own fields (Figure 4), adoption of appropriate strategies for selecting planting material would be expected to reduce the incidence of CMD appreciably. About 53% of farmers rogued CMD affected plants, usually within 3 months of planting. Most farmers (61%), however, rated roguing as partially effective, with only 4% considering it to be highly effective. In the choice of varieties, 25% of farmers cited “Agric” as the single most commonly planted variety and indicated that the choice was based on recommendations that it was resistant to CMD. Other CMD-resistant varieties commonly recorded were TMS 30572, TMS 30001 and TMS 30555. No farmer used pesticides against CMD and fewer than 2% of farmers used pesticides against whiteflies.

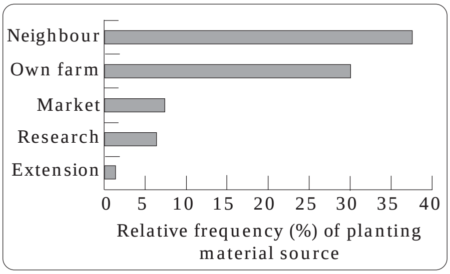

Figure 4. Farmers' sources of cassava planting material in Nigeria.

Conclusions

In Nigeria, whilst incidence of CMD is high, the economic impact of the disease is believed to be only moderate, based on the relatively mild symptoms that were observed in the survey. There is an obvious need, however, to obtain yield loss data for the commonly grown varieties in order to make an accurate assessment of economic impact. In view of the overwhelming importance of cutting infection, as compared with vector-borne infection, in the spread of CMD, research and extension efforts on assuring the health of planting material should be prioritized. The farmers' generally high level of awareness of CMD and their willingness to rogue diseased plants to prevent or slow its spread could provide a good basis for establishing participatory experiments to help farmers multiply clean planting material of their preferred varieties. Action learning activities, involving farmers and extension agents, could help improve their access to information and their understanding of the causes and nature of the disease problem. This will provide opportunities to promote cultural control options such as appropriate methods of selecting planting material, sanitation and roguing. Additionally, training of national program partners would strengthen national capacity to investigate and promote the sustainable management of whiteflies and CMD.

Acknowledgements

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CHAPTER 1.5

Cameroon

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Introduction

Cassava mosaic begomoviruses transmitted by *Bemisia tabaci* and causing cassava mosaic disease (CMD) are among the most important vector-borne pathogens of crop plants in sub-Saharan Africa (Geddes, 1990). Yield losses attributable to CMD are estimated at 28%-40% (Thresh et al., 1994). Fauquet and Fargette (1990) reported yield losses ranging from 20% to 95% for particular varieties of cassava (*Manihot esculenta* Crantz) under specific conditions. The incidence and effects of CMD in different ecozones are influenced by environmental factors, the intensity of cassava cultivation, the relative susceptibility and sensitivity to infection of cassava genotypes grown, and the virulence and abundance of virus and vector species (Fargette and Thresh, 1994). Disease epidemiology may be influenced also by the incidence of *Bemisia tabaci* (Gennadius), the principal vector of CMD, and of *B. afer* (Priesner and Hosny), a non-vector species that also occurs on cassava (Robertson, 1987; Fishpool and Burban, 1994; Legg, 1995), as well as by man through the transport and replanting of infected cassava cuttings. In Cameroon, Fondong et al. (1997), using infection of initially CMD-free cuttings as an indicator, showed that the spread of the disease was more rapid in the lowland forest than in the mid-altitude forest and savannah areas. It was also observed that CMD spreads faster in monocultures of cassava or when cassava is intercropped with cowpea (*Vigna unguiculata* [L.] Walp.) than in cassava intercropped with maize (*Zea mays* L.) (Fondong et al., 1997). In a regional CMD epidemiology trial covering Benin, Cameroon, Ghana and Nigeria, varietal differences in the rate of CMD infection were observed among improved and local varieties (James et al., 1998).

Given that many factors influence CMD epidemiology, a detailed understanding of the various cassava agro-ecologies, the associated whitefly species composition and the cultivation practices of the farmers may be helpful in designing the most appropriate CMD control strategy. Furthermore, since the disease is prevalent in almost all the major cassava production areas of Africa, potential advantages are to be gained in collecting such baseline data from different areas using a common

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protocol, to enable valid comparisons to be made between countries and thus facilitate the search for effective solutions. This chapter reports the status of the CMD problem, the species composition and relative abundance of *Bemisia* whitefly populations, and the cassava production practices in three major cassava agro-ecologies of Cameroon.

Surveys carried out in November and December 1997, and later in March 1998, followed the methods previously agreed among project partners. Seventy fields were surveyed in three cassava growing regions chosen to represent the major agro-ecological regimes under which cassava is grown in Cameroon: South-West Province, representing rainforest (26 fields); Central/South Province, representing transition forest (28 fields); and North-West Province, representing wet savannah (16 fields) (Figure 1).

Figure 1. Areas surveyed for whitefly incidence and cassava mosaic disease in Cameroon.

Increased Biological Understanding

Two whitefly species, *B. tabaci* and *B. afer*, were identified on cassava in Cameroon. *B. tabaci* was the prevalent species, comprising at least 92% of whitefly adult collections in each of the zones surveyed. Whitefly abundance averaged 4.1 adults per cassava shoot tip in the rainforest, 3.1 in the transition forest and 2.4 in the wet savannah. Logarithm-transformed whitefly counts were significantly lower in the wet savannah than in each of the other ecozones ($t > 3.11$; $P < 0.05$). Two parasitoid species, *Encarsia sophia* (Girault and Dodd) and *Eretmocerus* sp. were hatched from *Bemisia* mummies; *E. sophia* predominated in the collections.

Total CMD incidence averaged 44% in the rainforest, 72% in the transition forest and 45% in the wet savannah. Incidence of stem cutting infection contributed far more to total CMD incidence than did whitefly infection (Figure 2) and was significantly higher in the transition forest than in either of the other ecozones ($t = > 3.09$; $P < 0.05$). A possible reason for the high incidence of cutting infection observed in the transition forest is that at least 90% of farmers in the region grow CMD-susceptible local cassava varieties whose tender leaves are consumed as leafy vegetables. The incidence of whitefly infection, transformed to allow for the effect of multiple infection (Gregory, 1948), was significantly lower in the transition forest than in either of the other ecozones ($t > 6.27$; $P < 0.05$). In all ecozones, most of the diseased plants had a mean damage severity score of 2 (“slight damage”, on an ascending damage severity scale of 1 to 5) and the proportion of plants with damage severity score 4 was negligible; there were no plants with damage score 5 (Figure 3).

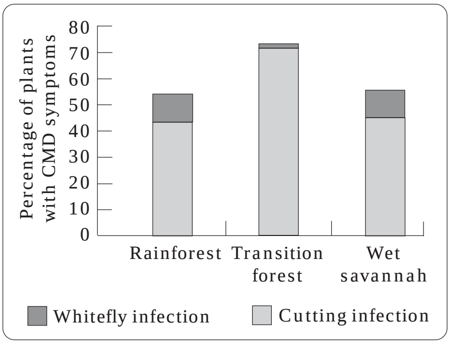

Figure 2. Cassava mosaic disease (CMD) incidence and source of infection in the ecozones of Cameroon.

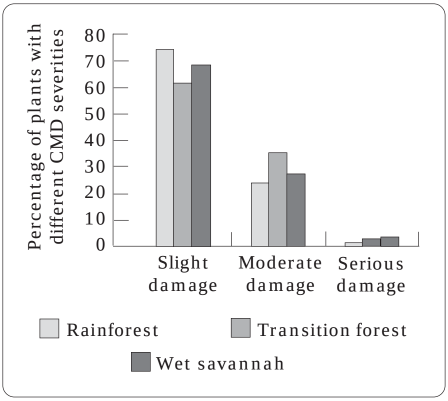

Figure 3. Cassava mosaic disease (CMD) damage severity in the ecozones of Cameroon.

Increased Socio-Economic Understanding

Cassava farmers were mostly women (87%). Sixty-six percent of farmers owned land whilst 20% rented land for farming; the remaining 14% offered no response on the issue of land tenure. Farm size averaged 0.29 hectares (2.5 nearby fields) in South West Province, 0.6 hectares (2.4 nearby fields) in Central/South Province and 0.32 hectares (4.2 nearby fields) in North-West Province. Farmers grew cassava mostly on land that had been worked for more than 5 years (60% of sites). Fourteen percent of the farms were on land that had been worked for between 2 and 5 years, and 26% on land that had been cultivated for less than 2 years. Only 21% of cassava farmers practiced crop rotation. Nineteen percent of farmers ranked cassava as the most profitable crop and 90% of farmers sold all or part of their cassava crop.

Farmers generally attached no significance to whiteflies and in their local languages they simply referred to them as “flies” or “white insects”. In contrast, 51% of farmers recognized CMD as a problem and in local languages referred to the disease symptoms variously as “crazy disease”, “curl”, “jelly cassava”, “leaf curl”, “leprosy”, *kalle*, *khumbo*, *nome* or *pama*. Use of these local names, which describe the symptoms, will be helpful in training farmers to improve their understanding of the causes and nature of the disease.

No farmer reported total yield losses attributable to CMD; however, 16% of farmers reported losses of 25% in the first season, 20% reported 50% losses and 10% reported 75% losses. These figures are surprising in view of the mild damage observed in the survey, which, based on experience elsewhere, would not be expected to lead to significant yield loss. Most farmers indicated that climate affected whitefly/CMD problems and stated that the problem was most severe during the early growth stages of cassava. This period corresponds to March-April during the first season and to August during the second growing season in Cameroon. Even though most farmers recognized CMD as a problem that occurs every year, 61% of them had received no technical information or assistance on the problem.

Farmers undertook various measures to combat the whitefly/CMD problem. The common practices were roguing, choice of resistant varieties, selection of planting material and the application of wood ash or of fertilizers (Figure 4). Less than 2% of farmers used pesticides against whiteflies and/or CMD. Even though it might be supposed that the high incidence of cutting infection observed in the survey would make roguing an unattractive strategy, it was nevertheless the predominant practice.

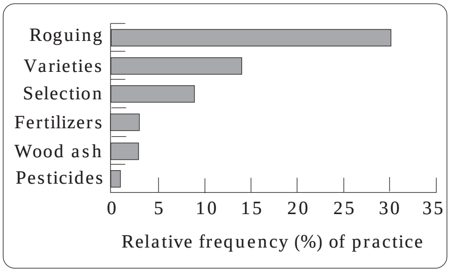

Figure 4. Cassava mosaic disease management practices used by farmers in Cameroon.

In their choice of varieties, 86% of the farmers were aware of the differences in susceptibility to the disease among cassava varieties. Among improved varieties recommended for their disease resistance, “Agric” (of International Institute of Tropical Agriculture, Nigeria [IITA] origin), chosen by 47% of farmers, was the most commonly selected. The variety Pawpaw leaf was the single most frequently planted CMD-resistant local variety (chosen by 9% of farmers). Other CMD-resistant local varieties cited were Metta agric, Mfont and Ndongo.

Most farmers (96%) indicated that in selecting planting materials they intentionally chose parent plants with minimal or without CMD symptoms. This practice would inherently favour CMD-resistant varieties. Farmer-led rapid multiplication schemes to produce CMD-resistant varieties may therefore be relatively easy to promote under such circumstances by building upon previous technology transfer experiences and farmer training programmes in the country (Dahniya et al., 1994; Akoroda, 1997; Bakia et al., 1999). In selecting planting material, the five most important criteria were yield (34.3% of farmers), followed by stem health (25.7% of farmers), absence of diseases (17.1% of farmers), stem size (5.7%) and stem maturity (4.3%). Most farmers expressed willingness to change planting dates for CMD control but 30% of them indicated unwillingness to alter planting date.

Conclusions

There was only limited evidence of CMD being spread by its whitefly vector in farmers’ fields in Cameroon. Infected planting material was the major source of CMD and a major extension effort will be required to encourage farmers to select disease-free and healthy planting materials. If effective measures to improve crop health are to be developed, the importance of latent infection will have to be investigated. The expressed willingness of farmers to alter planting dates and their conviction that CMD incidence varies with climatic conditions paves the way for participatory action research to optimize times of planting in the various ecozones so as to reduce CMD incidence and damage. In such studies, it may be useful to monitor the population dynamics of *B. tabaci* to see whether its abundance varies at the different planting times. Evaluation of parasitism by existing natural enemies (*E. sophia*, for example) may help show whether biological control contributes significantly to regulating populations of the vector. Such information will contribute to understanding the role of *B. tabaci* in the epidemiology of CMD.

Even though the CMD damage observed in the survey was slight, farmers provided high yield loss estimates. This suggests the need for farmer participatory yield loss trials to quantify losses both of storage roots and leaves, especially since the latter are widely consumed as leafy vegetables, but losses have been little studied. Farmer participatory varietal selection trials also need to be conducted on the local cassava varieties (such as Pawpaw leaf), which farmers report to be CMD-resistant, and comparisons made with cultivars whose response to CMD is well documented. Such studies will provide valuable input into breeding programmes that aim to achieve sustainable high yields in varieties preferred by farmers and to increase adoption of such varieties.

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CHAPTER 1.6

Uganda

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Introduction

Cassava mosaic disease (CMD) is the most important viral disease of cassava (*Manihot esculenta* Crantz) (Otim-Nape, 1993) in Uganda. It is caused by begomoviruses transmitted by the whitefly vector, *Bemisia tabaci* (Gennadius) and spread through virus-infected planting material (Harrison, 1987).

The first recording of CMD in Uganda was in 1928 (Martin, 1928). Severe epidemics from 1933 to 1944 were successfully controlled by planting resistant cassava varieties and through phytosanitation measures enforced by local government statute (Jameson, 1964). The disease then was considered a relatively minor problem until the late 1980s, when severe epidemics were again reported in northern Luwero District (Otim-Nape, 1988). By 1990, CMD had severely affected other cassava-growing districts of northern and eastern Uganda (Otim-Nape et al., 1997). In 1993 and 1994, the widespread crop losses associated with the CMD epidemic, coupled with drought, led to food shortages and localized famine (Otim-Nape et al., 1997). Between 1994 and 1998, the CMD epidemic continued to expand, affecting much of southern Uganda. Significant progress has been made in controlling the disease, particularly in north-eastern parts of the country, which were some of the first to be affected (Thresh et al., 1994) but the epidemic continues to spread in southern and western districts. The principal control method has been the deployment of resistant varieties.

Sweetpotato virus disease (SPVD) is correspondingly the most serious viral disease of sweetpotato (*Ipomoea batatas* [L.] Lam.) in Uganda (Bashaasha et al., 1995), although its distribution and impact on sweetpotato production have scarcely been studied until recently. Aritua et al. (1998) provide information on the disease for only two of the country's (at that time) 45 districts, Mpigi and Soroti (Figure 1). SPVD results from co-infection of *Sweetpotato chlorotic stunt virus*, transmitted by *B. tabaci* and *Sweetpotato feathery mottle virus*, transmitted by the aphid *Myzus persicae* (Sulzer) (Gibson et al., 1998).

In order to enhance understanding of whiteflies and whitefly-transmitted viruses on cassava and sweetpotato in Uganda, surveys were conducted

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between November 1997 and January 1998. It was anticipated that the data collected would be useful in helping researchers in Uganda monitor the development of the CMD epidemic, target control interventions more effectively and, in the longer term, develop sound integrated pest management (IPM) strategies for cassava and sweetpotato.

Figure 1. Cassava- and sweetpotato-growing areas surveyed for whitefly incidence in Uganda.

Surveys were carried out in 12 districts of Uganda (Figure 1) by multi-disciplinary teams from the National Agricultural Research Organisation and the International Institute of Tropical Agriculture (IITA). Districts were chosen to represent major cassava and sweetpotato producing areas. Farmers' fields of cassava 3-5 months after planting and of sweetpotato 3 months after planting were selected at regular intervals along main roads traversing each district. Data were collected according to the standardized survey methods, except that the number of fields varied between the districts because the survey areas were selected on the basis of agro-ecological differences rather than administrative boundaries (Table 1).

Increased Biological Understanding

Whitefly species and abundance

The survey identified two whitefly species, *B. tabaci* and *B. afer* (Priesner and Hosny). Of the 261 samples identified on cassava, 95.8% were *B. tabaci* and 4.2% *B. afer*. Only *B. tabaci* was recorded from sweetpotato.

Whitefly populations on cassava (counted on the top five leaves for a single shoot of each of 30 plants per sampled field) were highest in the south-western districts of Masaka and Rakai and lowest in Masindi (the north-western limit of the survey area) and Tororo (on Uganda's eastern border with Kenya), with mean abundance ranging from 0.7 to 13.1 per top five leaves (Table 1) (see also Figure 2 in Chapter 1.14, this volume). The pattern of whitefly populations on sweetpotato (counted by disturbing leaves and making 10 serial counts of 1 min each of whitefly adults observed) was similar to that on cassava: abundance was greatest in the central districts of Mukono and Mpigi and lowest in Masindi and Apac. Mean adult whitefly abundance ranged from 0.9 to 14.8 per minute count. In similar studies, Aritua et al. (1998) found greater whitefly abundance on sweetpotato in the southern and central zones, which are characterized by more evenly distributed rainfall than the northern and eastern zones.

The data indicate a higher whitefly population on both crops in southern and south-western Uganda than in the northern and eastern districts of the country. Assessing the significance of these differences is difficult based on records collected on a single occasion, during what appears to be the continuing spread of an epidemic. Numerous factors such as physical

Table 1. Incidence (%) of cassava mosaic disease (CMD) and sweetpotato virus disease (SPVD), disease severity and whitefly abundance on cassava and sweetpotato in selected districts of Uganda, surveyed during November and December 1997.

District	No. fields	Cassava[a]					Sweetpotato[a]		
		Whitefly counts	CMD				Whitefly counts	SPVD	
			Whitefly infection	Cutting infection	Total incidence	Severity (1-5 scale)		Incidence	Severity (1-5 scale)
Masindi	7	0.7	11 (19)	34	45	3.0	0.9	3	2.0
Apac	6	1.4	22 (140)	71	93	3.0	1.9	9	2.3
Lira	7	1.4	11 (89)	81	92	3.0	1.4	3	2.3
Iganga	6	1.6	20 (80)	64	84	2.8	11.2	16	2.1
Tororo	7	0.9	16 (73)	69	85	3.6	1.5	15	2.8
Pallisa	7	3.0	21 (28)	15	36	3.0	3.4	6	2.6
Mukono	7	3.7	13 (47)	64	77	3.0	14.8	10	2.4
Mpigi	7	4.6	17 (59)	61	78	2.8	14.7	29	2.7
Luwero	6	3.0	12 (56)	71	83	2.6	7.8	7	2.2
Mubende	7	3.4	24 (29)	7	31	2.7	3.8	9	2.9
Masaka	6	13.1	50 (91)	17	67	3.0	3.2	14	2.7
Rakai	7	11.1	44 (60)	3	47	2.7	2.0	8	2.7
Average		3.9	22 (65)	46	68	2.9	5.5	11	2.5

a. Figures are means for each district. Whitefly counts, whitefly abundance on cassava (number of whiteflies per top five leaves) and on sweetpotato (per minute count); whitefly infection, figures in parentheses transformed to multiple infection units to allow for multiple infection (Gregory, P. H. 1948. The multiple infection transformation. Ann. Appl. Biol. 35:412-417); severity of disease is measured on an ascending 1-5 scale, from low to severe.

environment, host plant health condition and natural enemies have been shown to influence whitefly populations on cassava (Legg, 1995). Nonetheless, this general geographical trend in abundance does parallel the current trend in disease severity.

Disease incidence and symptom severity

CMD was present in all cassava fields sampled and overall incidence (the average proportion of affected plants) throughout the survey was 68% (see also Figure 2 in Chapter 1.14, this volume). Disease incidence was highest in the northern districts of Apac and Lira and lowest in Mubende. Symptoms were moderate (2.6 to 3.0 on an ascending scale of 1 to 5) with the exception of Tororo District, where the disease was severe (Table 1).

The pattern of CMD incidence in Uganda indicates a north-to-south progression. In areas of epidemic expansion in the south (Masaka and Rakai Districts) the main source of infection is whitefly transmission, whereas in post-epidemic areas to the north (Masindi, Lira and Apac Districts) whitefly transmission is less evident and diseased cuttings are the main source of infection (Table 1). The relatively low CMD incidence recorded in Pallisa District is because of the National Cassava Programme's recent introduction of CMD-resistant varieties, Nase 1 (TMS 60142), Nase 2 (TMS 30337) and Nase 3 (TMS 30572), originating from the IITA breeding programme. Similarly, disease incidence and infection were relatively low for the districts neighbouring Pallisa-Kumi, immediately to the north, and Soroti, to the south.

SPVD occurred in 60 of the 80 sweetpotato fields sampled. Generally, SPVD incidence was greater in southern areas along the shoreline of Lake Victoria, with the highest incidence in Mpigi District on the northern shore of the lake, and the lowest in Masindi and Lira, further to the north. Overall, SPVD symptom severity was moderate in all the districts surveyed.

Natural enemy species and distribution

Whitefly natural enemies collected in the survey included predators and parasitoids. Few predators were collected. This was probably due to the limited time available during the survey for collecting samples, the relatively low populations of whiteflies on cassava and sweetpotato, and the difficulty of observing arthropods actually feeding on whitefly immature or adult stages.

The predator *Conwentzia africana* Meinander was recorded from 16 locations in three of the four target areas. Three species of whitefly parasitoids were identified: *Encarsia sophia* (Girault and Dodd), *Encarsia* sp. (*luteola* group) and *Eretmocerus* sp. *E. sophia* was the most widespread and numerous, occurring through most of the surveyed area (Table 2). *Eretmocerus* sp. occurred principally in south-western Uganda. *Encarsia* sp. (*luteola* group) was only identified from eastern Uganda. The absence of records of parasitoids from some of the districts surveyed, particularly those in the north, is thought to be largely because of the relatively low abundance of whiteflies recorded in these locations. Seasonal and environmental factors clearly play an important role in the abundance of whiteflies and their parasitoids, and in order to collect more comprehensive data, the surveys would have to be repeated, perhaps at 3-monthly intervals throughout the year. Little is known about the role of natural enemies in the population dynamics of *B. tabaci* on cassava and sweetpotato.

Table 2. Occurrence of parasitoids reared from mummies collected from cassava at each of the sampling sites in Uganda.

District	Parasitoid occurrence per sampling site[a]		
	Encarsia sophia	*Eretmocerus* sp.	*Encarsia* sp. (*luteola* group)
Masindi	-	-	-
Apac	-	-	-
Lira	-	-	-
Iganga	3/6	1/6	1/6
Tororo	2/7	-	-
Pallisa	2/7	-	-
Mukono	6/7	1/7	-
Mpigi	4/7	3/7	-
Luwero	-	-	-
Mubende	2/7	-	-
Masaka	1/6	3/6	-
Rakai	3/7	2/7	-

a. Dashes indicate the absence of any reared parasitoids.

Increased Socio-Economic Understanding

Farmers' assessment of whitefly-related problems

Only a small proportion of cassava and sweetpotato farmers (22%) recognized whiteflies. In contrast, the diseases CMD and SPVD were more widely recognized (CMD by 100% of cassava farmers and SPVD by 48% of sweetpotato farmers) and had specific local names in most of the locations where they were prevalent. Both diseases were commonly referred to with words describing the mottling or deformation of the leaves induced by the disease, and a number of names were common for both crops. Virtually all cassava farmers recognized CMD as a production constraint, whereas for SPVD only 35% of producers considered it a problem. Most cassava farmers (74%) considered that CMD was becoming more severe. Sweetpotato producers were similarly pessimistic about the outlook for SPVD, with 65% of the producers who recognized SPVD as a problem considering that it was becoming more severe. Farmers expressed no clear opinion as to the time of year at which whitefly/disease problems are especially severe. The picture of the spread of the CMD epidemic provided by farmers' first record of severe CMD in their fields, indicating a north-to-south progression, is in close agreement with scientific records of the expansion of the severe CMD epidemic (Otim-Nape et al., 1997; Legg and Ogwal, 1998).

Managing whiteflies and whitefly-transmitted viruses

A common response amongst farmers to the CMD problem has been to abandon production of the crop altogether. Whilst cassava farmers assessed losses to CMD at more than 75% of production, sweetpotato farmers estimated losses to SPVD to be less than 25%. Farmers attempting to control CMD and SPVD mainly used roguing, while some applied wood ash on the infected plants. Only 6.5% of farmers reported having received information on control practices through extension and/or research channels. Selection of planting material free of disease symptoms was the most widely used disease control practice for both cassava and sweetpotato, and the second most widely reported was roguing of plants that developed symptoms. However, most farmers reported the two methods to be only partially effective in controlling the diseases.

Thirteen out of 19 farmers growing resistant varieties of cassava and zero out of 11 farmers growing resistant sweetpotato varieties cited resistance as a control method. However, only about 24% of cassava farmers growing resistant varieties found their performance to be superior to that of local varieties. Assessments of the benefits of disease-resistant sweetpotato varieties were more mixed, with some farmers reporting no difference in performance between resistant and local varieties. About 20% of both cassava and sweetpotato farmers attributed differences in the disease response of the different varieties to some kind of resistance to infection, inherent in the variety. On the other hand, 28% of cassava producers and 18% of sweetpotato producers had no idea why such differences should occur. Very few farmers reported using chemical pesticides to control either CMD or SPVD. Of these farmers, almost all used dimethoate, an organophosphate insecticide, so their intention was presumably to control the whitefly vector. Most of these farmers used pesticides on the recommendation of neighbours or relatives, sprayed when symptoms were seen, and did so on one to three occasions during the production cycle. However, many reported that the approach was ineffective.

Whitefly-related crop production costs in affected areas

Since farming families produce most cassava and sweetpotato for home consumption rather than sale, it was difficult to assess the value of lost production attributable to CMD and SPVD and costs incurred in trying to manage these diseases. As mentioned above, the most widely practiced management tactics were selection of disease-free planting material and roguing, both of which involve investment of additional labour. Because labour is usually provided by members of farming households, rather than hired, this represents a time-related opportunity cost rather than a direct financial one. More focused follow-up studies are required to provide an understanding of the economic costs and benefits of managing these diseases.

Conclusions

The results of the survey indicate that farmers’ approaches to managing the whitefly-borne virus diseases, CMD and SPVD, are generally ad hoc and based on only a partial understanding of the problem. Since researchers already have a considerable body of knowledge relating to these problems that is not being applied at farm level, this suggests that information flow between farmers and researchers is weak. However, the absence of a clearly defined and well-articulated IPM approach to managing CMD and SPVD could be a contributory factor. Researchers believe that such an approach would combine the use of both local and improved resistant varieties with the selection of clean planting material and subsequent roguing of diseased plants—plant health methods that are already widely practiced.

The development and enhancement of channels of information flow from research to farm level is clearly critical to the successful adoption of IPM of whiteflies and whitefly-transmitted viruses. A number of initiatives have recently been implemented in Uganda to strengthen both the extension system and linkages between research and extension and to foster a participatory approach to working with farmers to develop appropriate and sustainable IPM strategies. It is hoped that these will improve the efficiency of the two-way flow of information between researchers and farmers in the years ahead, facilitating the more widespread adoption of IPM strategies as they become available.

Future work should focus mainly on managing CMD and SPVD, through multiplication and distribution of the existing disease-resistant varieties, and on developing, evaluating, multiplying and distributing new disease-resistant materials. Since most farmers reported the planting of clean cuttings and roguing of diseased plants to be ineffective, there is a need to find out under what conditions these measures could be both effective and adoptable by farmers. Future work should also consider the possibility of community-based phytosanitation, which offers the prospect of widespread reductions in CMD or SPVD inoculum pressure and thereby reduced rates of spread. Too little is currently known about the activity of parasitoids or other natural enemies of *B. tabaci* on either cassava or sweetpotato to offer the prospect of their immediate use within an IPM strategy. However, both introductions of exotic parasitoids and augmentation through habitat management have been used successfully in the United States to improve management of *B. tabaci* (Goolsby et al., 2000). It may be appropriate therefore to explore similar approaches to using parasitoids such as *E. sophia* for the management of *B. tabaci* as vector of CMD and SPVD, although this is always likely to be a measure of secondary importance to the major control approaches of resistant varieties and phytosanitation.

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CHAPTER 1.7

Kenya

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Introduction

Cassava mosaic disease (CMD) is caused by cassava mosaic begomoviruses (CMBs), which are transmitted by the whitefly *Bemisia tabaci* (Gennadius). Further spread may occur through farmers distributing and planting virus-infected cuttings (Harrison, 1987). The disease causes higher production losses than any other virus disease of cassava in Kenya (Bock, 1994b). Sweetpotato virus disease (SPVD) results from co-infection of sweetpotato (*Ipomoea batatas* [L.] Lam.) by two distinct viruses: *Sweetpotato chlorotic stunt virus*, transmitted by *B. tabaci*, and *Sweetpotato feathery mottle virus*, transmitted by the aphid, *Myzus persicae* (Sulzer) (Gibson et al., 1998). Although SPVD is the most prevalent viral disease of sweetpotato in farmers' fields in Kenya (Carey et al., 1998), its effects on growth and yield have yet to be quantified.

The early history of CMD in Kenya is obscure but work on cassava under the East African breeding programme in the mid-1950s is described by Doughty (1958) and resulted in the release of CMD-resistant hybrids to research stations in the country. However, farmers did not replace their local traditional Kenyan cultivars of cassava (*Manihot esculenta* Crantz) with this improved germplasm, except for 46106/27 “Kaleso”, which became widely grown in Coast Province (Bock, 1994b).

Research on CMD and its whitefly vector only became important in Kenya during the 1970s, when a major plant virology project was initiated. The work of this project initially emphasized the isolation of the putative *Cassava mosaic virus* (Bock, 1975), which was later shown to cause CMD (Bock and Woods, 1983). Subsequent epidemiological work by Bock (1983) noted that the incidence of CMD was generally high in coastal and western Kenya, where it exceeded 80% in some districts and approached 100% on individual farms but the spread of disease into initially CMD-free plantings was generally slow. Additional studies associated with this project were the description of the population dynamics of *B. tabaci* (Seif, 1982; Robertson, 1987), assessments of yield loss (Seif, 1981) and the development of control methods (Bock, 1994a). During the 1990s, a series of surveys conducted to reassess the incidence of the disease in the country indicated increased spread and incidence of CMD in farmers' fields.

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Recent epidemiological studies on CMD in the region have provided evidence of the spread of severe CMD from Uganda to western Kenya (Legg, 1999). This is associated with the spread of a novel and particularly virulent CMB (Harrison et al., 1997; Legg, 1999). Major losses have been experienced already in Western Province, and districts of Nyanza Province are now threatened. Efforts have been made to control the disease by introducing resistant material into western Kenya from the breeding program of the International Institute of Tropical Agriculture-Eastern and Southern Africa Regional Center (IITA-ESARC) in Namulonge, Uganda. Continuing work by the cassava program of the Kenya Agricultural Research Institute (KARI) emphasizes the evaluation and multiplication of the initial stock of resistant material, in collaboration with international research institutions and networks such as the East Africa Root Crops Research Network (EARRNET) (Legg et al., 1999).

Research on SPVD began during the 1950s, when Sheffield (1957) reported the presence of a severe sweetpotato mosaic disease on sweetpotato fields in East Africa. More recently, surveys have been carried out to assess the occurrence and importance of sweetpotato viruses in Kenya (Carey et al., 1998) and the Centro Internacional de la Papa (CIP) now has a major germplasm introduction and development program based in Kenya. Whilst cultivar resistance has been identified as the principal strategy for managing SPVD, little detailed research has been done on yield loss or the epidemiology of the disease.

A study was conducted in mid-1998 to identify whiteflies and whitefly-transmitted viruses prevalent in Kenya and to assess producers' knowledge of whiteflies and the diseases they transmit on cassava and sweetpotato. Three "target areas" representing the major cassava and sweetpotato growing areas in the country were selected: Coast Province, Western Province and Nyanza Province (Figure 1). Within these areas, 3 to 5-month-old cassava fields and 3-month-old sweetpotato fields were randomly selected at regular intervals along main roads.

Figure 1. Cassava- and sweetpotato-growing areas surveyed for whitefly incidence in Kenya.

Increased Biological Understanding

Whitefly species and abundance

Whitefly nymph samples were obtained from most but not all surveyed sites and, for each sample, from one to three nymphs were identified to species level. Two whitefly species, *B. tabaci* and *Bemisia afer* (Priesner and Hosny), were identified on both cassava and sweetpotato. Seventy whitefly nymph samples were identified from cassava;

of which 58 were *B. tabaci* and 12 were *B. afer*. Only 35 whitefly samples were identified from sweetpotato (attributable to the lower populations on that crop), comprising 31 *B. tabaci* and four *B. afer*. The two species occurred throughout the three target areas.

Adult whitefly populations were relatively low on both cassava and sweetpotato (Table 1). Whiteflies on cassava were more numerous in Coast Province, while whiteflies on sweetpotato were more abundant in Nyanza Province. Mean adult whitefly numbers ranged from 0.2 to 2.9 per top five leaves on cassava and 0.5 to 4.4 per minute count on sweetpotato in the surveyed regions (see also Figure 2, Chapter 1.14, this volume).

Disease incidence and symptom severity

Mean CMD incidence in the surveyed regions was 51.3% and incidence was highest in Western Province and lowest in Nyanza (Table 1). Diseased cuttings provided the main source of infection in all three target areas. CMD symptom severity was mild in the Coast and Nyanza Provinces and more severe in Western Province. SPVD incidence ranged between 3% and 53% in farmers' fields. The disease was more prevalent in Western and Nyanza Provinces than in Coast Province.

Whitefly parasitoids

The survey identified two species of aphelenid parasitoids; 15 individual parasitoids were identified, comprising 13 *Encarsia sophia* (Girault and Dodd) and two *Eretmocerus* sp. Little is known, however, about the role of natural enemies in the population dynamics of whiteflies in Kenya.

Increased Socio-Economic Understanding

Farmers' assessment of whitefly-related problems

About half of the producers surveyed (52% of cassava farmers and 46% of sweetpotato farmers) were able to recognize whiteflies, although the names they used were non-specific. Of the farmers who recognized whiteflies on either crop, 61% considered them a production problem.

Most cassava farmers (88%) recognized CMD as a disease of their crop, whilst only 58% of sweetpotato farmers recognized SPVD. The

Table 1. Incidence (%) of cassava mosaic disease (CMD) and sweetpotato virus disease (SPVD), disease severity and whitefly abundance on cassava and sweetpotato in three provinces of Kenya, surveyed during May and June 1998.

Province	Cassava[a]						Sweetpotato[a]		
	No. fields	Whitefly counts	CMD				No. fields	Whitefly counts	SPVD incidence
			Whitefly infection	Cutting infection	Total incidence	Severity			
Coast	17	2.9	19 (35.9)	37	56	2.3	15	0.5	0.4
Western	15	0.2	14 (65.9)	71	85	2.9	18	2.5	6.9
Nyanza	18	0.6	2 (2.3)	11	13	2.2	17	4.4	6.7
Average	-	1.2	11.7 (21.6)	39.7	51.3	2.5	-	2.5	4.7

a. Figures are means for each province. Whitefly counts, whitefly abundance on cassava (number of whiteflies per top five leaves) and on sweetpotato (per minute count); whitefly infection, figures in parentheses transformed to multiple infection units to allow for multiple infection (Gregory, P. H. 1948. The multiple infection transformation. Ann. Appl. Biol. 35:412-417); severity of disease measured on an ascending 1-5 scale, from low to severe.

respective diseases were recognized as a production constraint by 68% of cassava farmers and 44% of sweetpotato farmers. Most cassava farmers (82%) considered CMD to occur every year, whilst only 52% of sweetpotato farmers believed this to be the case. Farmers had different views regarding the trend of symptom severity for CMD and SPVD. Fifty-four percent of cassava farmers considered that CMD severity was increasing, whilst only 40% of sweetpotato farmers considered that SPVD was becoming more important. In both cases, a majority of cassava (72%) and sweetpotato (62%) farmers believed that climate influences disease severity. The two diseases were thought to be more severe during periods of low rainfall by 70% of cassava farmers and 44% of sweetpotato farmers. This was consistent with farmers' views that the two diseases were more severe during periods of moderate temperature.

Yield loss estimates due to CMD varied between regions. In Western Province, 21.4% of farmers estimated losses at >75%. This is understandable in view of the impact that the CMD pandemic has had in this area (Legg, 1999). Yield loss estimates were rated at between 25% and 50% by 79% of farmers in Coast Province and at only 25% by 47% of farmers in Nyanza Province. At the time of the survey, the CMD pandemic had affected only Western Province and losses recorded elsewhere were consequently less. Only 14% of sweetpotato farmers considered SPVD to be a significant source of crop loss, estimating these losses at only 25%.

Managing whiteflies and whitefly-transmitted viruses

Cassava and sweetpotato producers reported a number of management tactics, including selection of disease-free planting material, roguing of diseased plants and use of insecticides. In extreme cases, heavily infested crops were simply abandoned. The most widely used practices, however, were roguing and selection. Just over one third (38%) of cassava farmers used roguing, with 28% of farmers considering that this practice would reduce CMD incidence, 6% that it would prevent the spread of CMD and 2% that it would reduce the number of poorly growing plants. Among sweetpotato farmers, 30% used roguing, 18% believing that it would reduce SPVD incidence and 8% believing that it would prevent spread of the disease. Both cassava and sweetpotato farmers rogued infected plants before the crop was 4 months old. Selection of clean planting material, based on the absence of disease symptoms, was used as a disease management tactic by 32% of cassava farmers and 12% of sweetpotato farmers. However, even those farmers who used roguing and selection of clean cuttings considered these measures to be only partially effective in controlling the diseases.

Only one cassava farmer and none of the sweetpotato farmers mentioned chemical control as a control method. The cassava farmer used the organophosphate insecticide dimethoate, applied it more than 10 times and sprayed when whiteflies and damage were observed in the field. Few farmers reported abandoning the production of cassava (12%) or sweetpotato (2%) but many reported occasional shortage of clean planting material. None of the farmers grew improved varieties, although 42% of cassava farmers and 20% of sweetpotato farmers noted differences in response to CMD and SPVD between varieties. However, most farmers who noted differences between varieties had no idea what caused the differences. Only 4% of cassava farmers and 2% of sweetpotato farmers considered the differences to be the result of differences in levels of resistance to disease. Very few cassava (10%) and sweetpotato (4%) farmers had received any technical assistance in the management of whiteflies and whitefly-transmitted viruses on their crops. However, more than 30% of farmers of each crop were willing to monitor whitefly and disease problems if it would help in the management of either CMD or SPVD.

Conclusions

Farmers noted that CMD and SPVD are becoming increasingly important constraints to cassava and sweetpotato production in the country. Some farmers were attempting to control the diseases, mainly using roguing and selection, initiatives that in general they considered to be developed from their own knowledge. However, farmers recognized the methods employed in controlling the two diseases as being only partially effective. During this survey in 1998, no farmer reported using resistant varieties. The limited understanding that farmers demonstrated of the causes and effects of CMD and SPVD, coupled with their reliance on their "own" control methods, suggests weaknesses in the systems of information flow, both from research through extension to farmers and from farmers back to research.

During the survey conducted in this study, the impact of the pandemic of severe CMD has been captured both through the description of the unusually high incidence and severity of the disease in Western Province and through the estimates of major yield losses provided by farmers interviewed. Considerable experience in tackling this problem has been developed already in neighbouring Uganda (Otim-Nape et al., 1997) with the focus on the deployment of host plant resistance supported by some phytosanitation. In the immediate future, some of this experience clearly needs to be transferred to the situation in western Kenya where the CMD pandemic continues to expand. CMD-resistant germplasm will need to be introduced from Uganda to Kenya and evaluated in participatory trials with farmers, whilst at the same time encouraging the maintenance of the quality of planting material through the use of selection and roguing in multiplication plots. Given the evident shortcomings in the understanding of both CMD and SPVD in Kenya, participatory learning should be a key feature of any future control programs. SPVD is clearly not as acute a problem in Kenya as is CMD. However, the report by farmers that SPVD is becoming more important is a concern and future work should aim therefore at identifying resistant germplasm and evaluating this and possible phytosanitary approaches to SPVD control through participatory research with farmers.

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CHAPTER 1.8

Tanzania

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Introduction

Cassava (*Manihot esculenta* Crantz) and sweetpotato (*Ipomoea batatas* [L] Lam.) are important staple food crops, especially in the rural communities of Tanzania. The two crops have a long history of providing food security in the country, particularly during famine. However, their production is currently threatened by cassava mosaic disease (CMD) and sweetpotato virus disease (SPVD). CMD is caused by cassava mosaic begomoviruses (CMBs) transmitted by the whitefly *Bemisia tabaci* (Gennadius) and through virus-infected planting material (Harrison, 1987). On the other hand, SPVD results from co-infection by two distinct viruses, *Sweetpotato chlorotic stunt virus* transmitted by *B. tabaci* and *Sweetpotato feathery mottle virus* transmitted by the aphid *Myzus persicae* (Sulzer) (Gibson et al., 1998).

CMD was first reported in Tanzania under the name “Krauselkrankheit” (Warburg, 1894), although it was not recorded as causing serious losses until the 1920s. Between 1920 and 1960, comprehensive studies were conducted in the country, emphasizing the development of CMD-resistant varieties through a breeding program conducted at Amani in the Usambara Mountains (Jennings, 1994). Resistant varieties developed by the programme were effective in controlling CMD and restored the crop’s productivity. Data obtained in 1989 and 1990 during the first phase of the Collaborative Study of Cassava in Africa (COSCA) indicated that Tanzania had the lowest CMD-incidence (37%) of the six countries surveyed (Thresh et al., 1994). Another extensive survey, conducted between 1993 and 1994 in mainland Tanzania and the islands of Zanzibar and Pemba, rated the incidence of CMD at 28%, with infected cuttings (24%) providing the major source of infection (Legg and Raya, 1998). During the latter part of the 1990s, CMD research has been reinvigorated and has included work to assess the development of the disease in the country.

Recent epidemiological studies on CMD elsewhere in East Africa have provided evidence of the north-to-south spread of an epidemic of severe CMD, firstly within Uganda (Otim-Nape et al., 1997; Legg and Ogwal, 1998; Chapter 1.6, this volume) and subsequently into parts of western Kenya and Tanzania

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bordering Uganda (Legg, 1999). This expansion has been associated with the spread of a novel CMB variant (Harrison et al., 1997). The pandemic poses an immediate threat to cassava production in the areas of Tanzania bordering Uganda and eventually to the country as a whole. Research towards controlling the disease is the focus of continuing work by the Root and Tubers Programme of the Lake Zone Agricultural Research and Development Institute (LZARDI) based at Ukiriguru, Mwanza. The institute is supported in this effort by other international research institutions and networks, including the International Institute of Tropical Agriculture-Eastern and Southern Africa Regional Center (IITA-ESARC), the East African Root Crops Research Network (EARRNET) and the Southern African Root Crops Research Network (SARRNET). SPVD, and its effects on the production of sweetpotato, has received less attention in Tanzania. However, the disease poses a threat to the production of the crop if left unchecked.

Diagnostic surveys were conducted in mid-1998. Three target areas were chosen to represent the major cassava and sweetpotato growing areas in the country: the “lake zone” (i.e., bordering Lake Victoria), which includes Mwanza, Shinyanga, Mara and Kagera regions; the “southern coast”, which includes Lindi and Mtwara regions; and the “northern coast”, which includes Tanga, Dar-es-Salaam and Pwani regions (Figure 1). The study aimed at identifying whiteflies and whitefly-transmitted viruses and characterizing producer knowledge and responses to these problems on cassava and sweetpotato in Tanzania.

Figure 1. Cassava- and sweetpotato-growing areas surveyed for whitefly incidence in Tanzania.

Increased Biological Understanding

Whitefly species and abundance

Three whitefly species were identified on cassava: *B. tabaci*, B. *afer* (Priesner and Hosny) and *Trialeurodes ricini* (Misra). Only *B. tabaci* and *B. afer* were identified on sweetpotato. Of 303 whitefly samples collected on cassava, 175 were *B. tabaci*, 127 *B. afer* and one *T. ricini*. The 105 whitefly samples from sweetpotato comprised 89 *B. tabaci* and 16 *B. afer*. The species *B. tabaci* and *B. afer* were found in all three survey areas. *B. afer* was more common on cassava than on sweetpotato. It comprised 26% of the whiteflies in samples collected on cassava and 9% of those on sweetpotato from the lake zone, 53% on cassava and 11% on sweetpotato from the southern coast, and 45% on cassava and 27% on sweetpotato from the northern coast (see also Figure 2, Chapter 1.14, this volume).

Adult whiteflies were more abundant on cassava in the northern coast (5.6 per top five leaves) than in either the southern coast (1.1) or the lake zone (0.7). On sweetpotato, estimates of whitefly abundance were highest in the lake zone (40.1 per 1-min count) and lowest in the southern coast (2.8)

Disease incidence and symptom severity

CMD incidence was generally low in the three target areas. The highest incidence was recorded in the northern coast (33.6%) and the lowest (9.4%) in the southern coast (Table 1). The main source of infection in the lake zone (69.3% of the total infection in area) and northern coast (56.8% of the total infection in area) was cutting infection. However, whitefly-borne infection was the predominant source of infection in the southern coast. CMD symptoms were mild in the lake zone and southern coast and slightly more severe in the northern coast.

The incidence of SPVD was highest in the southern coast (12.4%), followed by the lake zone (11.3%), and lowest in the northern coast (4.2%) survey area (Table 1). SPVD symptoms were relatively mild in most of the surveyed regions, with the exception of Shinyanga region in the lake zone, where the disease was very severe (4.5). The low incidence of SPVD should enable the use of phytosanitary measures, including selection of planting material and roguing in the management of the disease.

Whitefly parasitoids

The survey identified two whitefly parasitoids, *Encarsia sophia* (Girault and Dodd) and *Encarsia* sp. (*luteola* group). *E. sophia* was by far the more widely recorded, with 43 samples identified in the southern coast and 19 in the northern coast samples, while only one sample of *Encarsia* sp. (*luteola* group) was recorded, in the northern coast. Little is known about the role of natural enemies in the population dynamics of whiteflies in Tanzania.

Increased Socio-Economic Understanding

Farmers' assessment of whitefly-related problems

A small proportion of both cassava (30%) and sweetpotato (11.7%) farmers were able to recognize whiteflies on the two crops, and even the names given to the insect were non-specific. Moreover, of the farmers who recognized the whiteflies only 16.7% (on cassava) and 3.3% (on sweetpotato) considered them a problem in the production of their respective crops.

Most cassava farmers (70%) could recognize CMD as a disease of cassava, 58% recognized it as a constraint to crop production and 68% as a problem occurring every year in their crops. In contrast, only 38% of sweetpotato farmers recognized SPVD as a disease of sweetpotato, 32% recognized it as a constraint to production and 41% as a yearly recurring problem. One third of cassava farmers considered that CMD severity was increasing, whilst a slightly smaller proportion of sweetpotato farmers considered that SPVD was becoming more important. Forty percent of cassava farmers and 32% of sweetpotato farmers believed that climate influenced the diseases; two-thirds believed that they were more prevalent during periods of low rainfall and high temperatures. Cassava farmers attributed higher losses to CMD than did sweetpotato farmers to SPVD. Using five categories (zero, quarter, half, three quarters and total

Table 1. Incidence (%) of cassava mosaic disease (CMD) and sweet potato virus disease (SPVD), disease severity and whitefly abundance on cassava and sweet potato in selected regions of Tanzania, 1998.

Survey area	Region	Cassava[a]						Sweet potato[a]			
		No. fields	Whitefly counts	CMD				No. fields	Whitefly counts	SPVD	
				Whitefly infection	Cutting infection	Total incidence	Severity			Incidence	Severity
Lake zone	Kagera	6	2.2	0 (0)	30.7	30.7	2.6	5	18.0	20.0	3.1
	Mara	5	0.1	1.3 (1.4)	7.3	8.6	2.6	6	55.9	5.3	2.8
	Shinyanga	5	0.4	0.7 (0.9)	20.7	21.4	2.6	4	25.3	2.0	4.5
	Mwanza	4	0.2	3.3 (4.1)	18.0	21.3	2.4	5	61.1	18.0	3.1
	Mean		0.8	1.2 (1.7)	19.8	21.0	2.6		40.1	11.5	3.3
Southern coast	Lindi	6	1.0	2.8 (2.8)	0	2.8	2.8	9	2.9	8.3	2.1
	Mtwara	14	1.2	8.1 (9.2)	7.9	16.0	2.5	11	2.7	16.4	2.4
	Mean		1.2	6.5 (7.1)	5.5	12.0	2.7		2.8	12.8	2.3
Northern coast	Pwani	7	8.9	15.4 (18.0)	6.7	22.1	3.0	13	1.8	9.2	2.7
	DSM[b]	3	4.3	14.4 (16.3)	4.4	18.8	3.0	5	5.0	3.3	2.0
	Tanga	10	3.6	13.7 (29.5)	46.3	60.0	3.1	2	5.7	0	2.0
	Mean		5.8	14.4 (21.0)	26.2	40.6	3.0		3.1	6.8	2.5

a. Figures are means for each region. Whitefly counts, whitefly abundance on cassava (number of whiteflies per top five leaves) and on sweet potato (per minute count); whitefly infection, figures in parentheses transformed to multiple infection units to allow for multiple infection (Gregory, P. H. 1948. The multiple infection transformation. Ann. Appl. Biol. 35:412-417); severity of disease measured on an ascending 1-5 scale, from low to severe.

b. DSM, Dar es Salaam.

loss) to assess yield loss attributable to either CMD or SPVD, 20% of the cassava farmers believed that CMD causes at least 50% yield loss, while 23% considered the disease to cause only 25% yield loss. Among the sweetpotato farmers, over 50% considered that SPVD caused only 25% yield loss.

Managing whiteflies and whitefly-transmitted viruses

Both cassava and sweetpotato farmers attempted to control CMD and SPVD by roguing and selecting disease-free planting material. Roguing was used for disease management by 23% of cassava farmers and 5% of sweetpotato farmers. Farmers growing cassava gave three reasons for roguing diseased plants: reducing the spread of the disease (18%), poor growth (3%) and preventing disease (2%). However, only 5% of sweetpotato farmers considered that roguing would reduce SPVD. Most cassava farmers rogued infected plants before the crop was 4 months old. For sweetpotato, there was no specific period during which roguing was done. Seventy-three percent of cassava farmers compared with 47% of sweetpotato farmers selected clean planting material, with absence of disease symptoms as the selection criterion for the two diseases. In both cases, however, farmers considered the two principal methods to be only partially effective in managing the diseases.

Only one sweetpotato farmer and none of the cassava farmers mentioned chemical control as a management tactic. The sweetpotato farmer used the organophosphate pesticide dimethoate, applied 3 times, when whiteflies and damage were observed in the field. Few farmers reported abandoning the production of cassava (10%) and sweetpotato (3%) but many reported occasional shortage of clean planting material. The use of host plant resistance as a management option was mentioned by only 2% of cassava farmers but not mentioned at all by sweetpotato farmers. Only 30% of cassava farmers and 22% of sweetpotato farmers noted differences in response to CMD and SPVD between varieties. Those that did so mainly attributed it to differences in levels of resistance to disease. In general, very few cassava (8%) and sweetpotato (2%) farmers had received any technical assistance in the management of whiteflies and associated virus diseases on either crop. However, most farmers were willing to change the planting dates of their crop and monitor whitefly and disease problems if it would help in the management of either disease.

Conclusions

Both diseases covered in this study are becoming more prevalent in Tanzania (Chapters 1.13 and 1.14, this volume) and therefore measures to reduce their impact are needed urgently.

Farmers are more aware of CMD than of SPVD. However, their knowledge of management options for the two diseases is weak, implying poor communication among researchers, extension workers and farmers. Farmers’ knowledge of these diseases should be enhanced through participatory training, while researchers, farmers and other stakeholders should work together to develop and validate cost-effective disease and vector management strategies and encourage their wider adoption by producers.

Varieties that are resistant to CMD and SPVD, acceptable to farmers and well adapted to local conditions are urgently needed. The development, farm-level evaluation and wider distribution of improved varieties of cassava and sweetpotato should be strengthened and expanded.

The whitefly parasitoid *E. sophia* occurred frequently in the areas surveyed, and studies elsewhere in East Africa have shown that parasitoids can cause *B. tabaci* nymph mortality rates of up to 50%. The deployment of resistant varieties and phytosanitation measures probably will be the most readily applicable CMD and SPVD management measures in Tanzania. However, there may be scope in the future for improved management of the whitefly vector through the development and application of measures designed to conserve and augment the activity of whitefly natural enemies. The farmer field school (FFS) approach is being widely used in Tanzania, and FFS in root crop growing areas may provide an ideal opportunity to strengthen farmers' knowledge of CMD and SPVD and develop a holistic, integrated approach to their management, incorporating all possible components including host plant resistance, cultural methods and biological control.

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CHAPTER 1.9

Malawi

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Introduction

Cassava mosaic disease (CMD) is caused by cassava mosaic begomoviruses, transmitted by the whitefly *Bemisia tabaci* (Gennadius) and through virus-infected planting material (Harrison, 1987). The disease has gained significance as a major constraint to production in many areas of Malawi where cassava (*Manihot esculenta* Crantz) is grown, especially in the lowlands, where conditions are warmer; infection reportedly exceeds 90% in some plantings (Nyirenda et al., 1993).

A survey was conducted in three target areas selected to represent the major cassava growing areas in Malawi: the central lakeshore (Salima and Nkhotakhota, at 500-800 m altitude); the northern lakeshore (Rumphi and Nkhata Bay, 500-800 m); and the central plateau (Dedza, Lilongwe and Dowa, 1000-1500 m) (Figure 1). The key objectives of the study were to identify whiteflies and whitefly-transmitted viruses and to characterize producer knowledge of these problems on cassava in Malawi. Although assessments of sweetpotato virus disease (SPVD) also were planned initially, surveys were done at a time (July-September 1998) when crops of sweetpotato (*Ipomoea batatas* [L.] Lam.) mostly had been harvested, so disease and vector incidence could not be assessed adequately.

Figure 1. Areas surveyed for whitefly incidence and cassava mosaic disease in Malawi.

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Increased Biological Understanding

Whitefly species and abundance

Two whitefly species, *B. tabaci* and *B. afer* (Priesner and Hosny), were identified on cassava. Altogether, 212 whitefly samples were collected on the crop. Of these, 65 were *B. tabaci* and 147 were *B. afer*. The relative abundance of the two species varied among survey areas. *B. afer* was especially abundant in the central mid-altitude plateau, where it comprised 97% of the whitefly samples collected. The adult whitefly population did not vary significantly among the regions surveyed (Table 1).

Disease incidence and symptom severity

The incidence of CMD ranged from 11.4% in the central plateau survey area to 62.2% for the central lakeshore (Table 1). The use of diseased cuttings was the main source of infection in all three survey areas. Symptoms of CMD were moderately severe in each of the survey areas and averaged 2.8 overall.

Whitefly parasitoids

Only one species of whitefly parasitoid, *Encarsia sophia* (Girault and Dodd) was identified and this was from the northern lakeshore.

Increased Socio-Economic Understanding

Farmers' assessment of whitefly-related problems

Forty-four percent of the farmers interviewed were able to recognize whiteflies on the crop but only 32% considered them a problem. Most farmers (61%) were able to recognize CMD as a disease of cassava and gave it a range of local names. Less than half the farmers (42%) recognized the disease as a problem to cassava production and most (64%) noted that it occurs yearly. Only 22% believed that CMD was becoming more severe, with 1997 noted as the year of most severe disease symptoms. About one quarter (27%) of the farmers believed the disease to be affected by climate. There was no consensus on the effect of rainfall on CMD but 39% of farmers

Table 1. Cassava mosaic disease (CMD) incidence (%), disease symptoms and whitefly counts in selected areas of Malawi, 1998.

Target area	Provinces/ districts	No. fields	Whitefly counts	CMD[a]			
				Whitefly infection	Cutting infection	Total incidence	Severity
Central lakeshore	Salima and Nkhotakota	20	1.5	27.5 (54.7)	34.7	62.2	2.8
Northern lakeshore	Rumphi and Nkhata Bay	9	1.0	19.6 (34.4)	32.6	52.2	2.7
Central plateau	Dedza, Lilongwe and Dowa	12	1.3	3.3 (3.7)	8.1	11.4	3.0
	Mean		1.3	16.8 (25.4)	25.1	41.9	2.8

a. Figures are means for each region. Whitefly counts, whitefly abundance on cassava (number of whiteflies per top five leaves); whitefly infection, figures in parentheses transformed to multiple infection units to allow for multiple infection (Gregory, P. H. 1948. The multiple infection transformation. Ann. Appl. Biol. 35:412-417); severity of disease measured on an ascending 1-5 scale, from low to severe.

considered that the disease was most severe during periods of high temperature. Farmers' estimates of the losses attributable to CMD varied considerably, ranging from "none" to "total crop loss".

Managing whiteflies and whitefly-transmitted viruses

Only 7.3% of farmers attempted to control CMD; 2.4% rogued, 3.7% selected for healthy planting material at planting, while some 1.2% de-topped CMD-infected plants to reduce plant shoots with disease symptoms in the field. Farmers who selected their planting material mainly used the absence of disease symptoms as the selection criterion. Farmers who rogued gave three reasons for doing so—prevention of disease spread (7%), reduction of disease (7%) and removal of poorly growing plants (5%). Most of the roguing was done before the crop was 4 months old. Only 5% of farmers considered selection and 7% considered roguing as highly effective in controlling the disease. Some farmers (10%) reported the occasional shortage of clean planting material, with 5% reporting abandoning production of the crop in 1990 and 1992. This may have been associated with the severe damage caused by the cassava mealybug, *Phenacoccus manihoti* Matile-Ferrero, at this time. Improved varieties, primarily developed for their superior yield and agronomic characteristics, were grown by only 5% of the farmers, who considered their performance to be better than that of local varieties. Very few farmers (5%) had received any technical assistance in the management of whiteflies. However, a large proportion (40%) of farmers were willing to monitor whitefly and disease problems if it would help in the management of the disease.

Conclusions

Prospects for managing CMD successfully are good in most locations sampled, since the use of diseased cuttings was the main source of infection. However, despite the majority of farmers recognizing the disease as a production constraint to cassava, their knowledge of the disease was weak. The incidence of CMD was relatively high in the lakeshore survey areas and, given that moderate symptoms were observed, it is likely that significant yield losses are being experienced. Given that there appears to be very limited CMD spread due to the activity of whiteflies, and that *B. afer*, a non-vector, is the predominant cassava whitefly species in Malawi, phytosanitation may provide an effective approach to CMD management. However, before an appropriate regime for the use of roguing and the selection of CMD-free planting material can be proposed, basic information will be required on yield losses and rates of infection in the major cassava-growing areas for the most commonly grown varieties. These results should be used to develop a programme of participatory evaluation with farmers of both phytosanitation and new varieties. This should be closely linked with an educational programme targeted at increasing farmers' knowledge of CMD and the whitefly vector and of the range of possible management approaches.

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CHAPTER 1.10

Madagascar

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Introduction

Cassava mosaic disease (CMD), caused by cassava mosaic begomoviruses transmitted by the whitefly *Bemisia tabaci* (Gennadius) and through virus-infected planting material (Harrison, 1987), is the most important viral disease of cassava (*Manihot esculenta* Crantz) in Madagascar. The disease was first reported in Madagascar in 1932, although it was of only minor importance at this time (François, 1937). Epidemics were first reported in 1934, from an area to the west of Lake Alatroa on the central plateau and, between this time and the end of the 1930s, the epidemic spread to cover all cassava-growing areas of the island (Cours, 1951). Symptoms in local varieties were very severe and cassava cultivation was abandoned on a wide scale. The impact was so great that the government ordered urgent attention to be given to the problem (Frappa, 1938) and a major programme for the development of resistant varieties was initiated (Cours, 1951; Cours-Darne, 1968). The first of these varieties was widely disseminated in the mid-1940s. Since this time, CMD appears to have become a problem of relatively minor significance.

Sweetpotato virus disease (SPVD) results from co-infection of sweetpotato (*Ipomoea batatas* [L.] Lam.) by whitefly-transmitted *Sweetpotato chlorotic stunt virus* and aphid-transmitted *Sweetpotato feathery mottle virus* (Gibson et al., 1998). SPVD has not been reported as a major problem in Madagascar, although the country is one of the largest producers of sweetpotato in Africa.

In order to provide a basic understanding of the incidence of whiteflies, CMD and SPVD in Madagascar, a study was conducted in four survey areas that represent the major cassava and sweetpotato growing areas: Antananarivo (central mid-altitude plateau), Fianarantsoa (eastern humid coast), Toliara (south-western semi-arid coast) and Mahajanga (northern sub-humid coast) (Figure 1). This initial phase of the study aimed at documenting the status of whiteflies and whitefly-transmitted viruses and at describing producer knowledge of these problems on cassava and sweetpotato, with a view to prioritizing further actions and eventually developing integrated management strategies to tackle the most serious problems identified. The survey was carried out during the months of June and August 1998.

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Figure 1. Cassava- and sweetpotato-growing areas surveyed for whitefly incidence in Madagascar.

Increased Biological Understanding

Whitefly species and abundance

Two whitefly species, *B. tabaci* and B. *afer* (Priesner and Hosny) were identified on both cassava and sweetpotato. Of 415 whitefly samples collected on the two crops, 118 were *B. tabaci* and 297 were *B. afer*. Adult whitefly populations on cassava were greatest in the Antananarivo survey area and lowest in Mahajanga (Table 1, see also Figure 3 in Chapter 1.14, this volume). Whiteflies were very scarce on sweetpotato.

Disease incidence and symptom severity

The highest disease incidence (71%) was recorded in Fianarantsoa, while the lowest (31%) was recorded in Antananarivo (Table 1). The planting of infected cuttings was much more important than whitefly transmission as a source of infection and comprised 86% of total infection. CMD symptom severity was relatively mild in Antananarivo but was severe elsewhere.

SPVD was of very low incidence or absent, except in the Toliara survey area, where an incidence of 18% was recorded. SPVD symptoms were severe in Antananarivo but very mild where the disease occurred elsewhere.

Increased Socio-Economic Understanding

Farmers' assessment of whitefly-related problems

Thirteen percent of cassava farmers but only 3% of sweetpotato farmers recognized whiteflies. Scarcely any of these farmers considered whiteflies to be a problem on either crop. Almost one fifth (19%) of cassava farmers were able to recognize CMD as a disease of cassava and gave the disease a range of local names, whereas no sweetpotato farmers recognized SPVD. Only 6% of cassava farmers considered CMD as a production constraint to cassava. They noted that the disease occurs yearly and that it was becoming more severe. Climate affects CMD and SPVD incidence according to 30% of cassava farmers and 5% of sweetpotato farmers. In particular, 19% of cassava farmers noted that disease is more severe during periods of little rain and high temperature but sweetpotato farmers did not report this effect. None of the farmers could estimate the level of yield loss attributable to either disease. The lack of recognition of CMD, first as a disease and second as a constraint to production, is

Table 1. Incidence of cassava mosaic disease (CMD) and sweetpotato virus disease (SPVD), disease severity and whitefly abundance on cassava and on sweetpotato in selected regions of Madagascar, 1998.

Province	Cassava[a]						Sweetpotato[a]			
	No. fields	Whitefly counts	CMD				No. fields	Whitefly counts	SPVD	
			Whitefly infection	Cutting infection	Total incidence	Severity			Incidence	Severity
Antananarivo	35	7.1	5.6 (7.8)	25.5	31.0	2.6	11	0.01	2.6	3.3
Fianarantsoa	22	4.3	7.1 (21.9)	63.9	71.0	3.5	15	0	2.4	2.0
Toliara	12	5.9	6.4 (11.2)	39.7	46.0	3.2	7	0.10	17.6	2.3
Mahajanga	42	2.5	6.9 (11.0)	34.0	41.0	3.3	6	0	0	-
Mean		5.0	6.5 (11.6)	40.8	47.3	3.1	-	0.03	5.7	2.5

a. Figures are means for each province. Whitefly counts, whitefly abundance on cassava (number of whiteflies per top five leaves) and on sweetpotato (per minute count); whitefly infection, figures in parentheses transformed to multiple infection units to allow for multiple infection (Gregory, P. H. 1948. The multiple infection transformation. Ann. Appl. Biol. 35:412-417); severity of disease measured on an ascending 1-5 scale, from low to severe.

surprising, particularly given the history of previous CMD epidemics in Madagascar and the severity of the disease symptoms observed.

Managing whiteflies and whitefly-transmitted viruses

Selection of clean planting material was the only disease management approach used—as reported by 40% of cassava farmers and 13% of sweetpotato farmers. Sixteen percent of cassava farmers and 3% of sweetpotato farmers used the absence of disease symptoms as selection criterion but both considered the method to be only partially effective. Only 27% of cassava farmers and 3% of sweetpotato farmers were able to recognize differences between varieties of their respective crops and even fewer (only 2% of cassava farmers and no sweetpotato farmers) could describe or characterize these differences. Cassava farmers growing improved varieties noted that their performance was only slightly better than the local varieties. Only 4% of cassava farmers and no sweetpotato farmers received technical assistance in whitefly or disease management. None of the farmers considered that environmental factors such as certain patterns of weather gave them any idea about what whitefly populations or disease levels would result and most were unwilling to change the planting dates of their crops even if it might help reduce the effects of whiteflies and/or disease. The farmers' reluctance is understandable, however, given the relatively short rainy season in much of Madagascar that leaves them little scope to adjust planting dates.

Conclusions

The incidence of CMD was low in many farmers' fields, compared with figures reported from countries on the African mainland, at least in three of the four survey areas. Infection was mainly attributable to the use of diseased planting material. However, the high severity of the symptoms in plantings where the disease does occur suggests that farmers are probably sustaining significant yield losses. It would be valuable to quantify yield losses in local cultivars being grown in each of the target areas. Such an exercise should be carried out with the full participation of farmers and could be used simultaneously to introduce new germplasm and evaluate its performance and acceptability.

The sharp disparity in SPVD incidence between Toliara and the other three survey areas is difficult to explain. Although the abundance of whitefly adults was greatest in this target area, whitefly populations were generally very low compared to those recorded in other countries. In addition, at the time the survey was conducted, *B. afer*, a non-vector, was the predominant species on both cassava and sweetpotato. More detailed epidemiology and population dynamics studies investigating changes in abundance of the two whitefly species and disease spread over time would be required in order to identify the reason(s) behind this difference.

Both cassava and sweetpotato farmers had inadequate knowledge of the principal diseases and the options available for managing them. A participatory research and training effort would enhance farmers' knowledge and would help researchers understand the constraints to farmers' adoption of resistant varieties and other potential management tactics. It would be valuable also to continue to monitor the incidence of the two diseases in the country and strengthen the national breeding program, to compare local sources of resistance with those available elsewhere and so develop resistant varieties acceptable to farmers.

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CHAPTER 1.11

The Diversity of Cassava Mosaic Begomoviruses in Africa

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Introduction

Cassava mosaic disease (CMD) is the most widespread and economically important disease of cassava (*Manihot esculenta* Crantz) in tropical Africa. A disease of cassava was first described in East Africa in the nineteenth century (Warburg, 1894) and cassava mosaic, as it came to be called, was subsequently reported as spreading throughout the cassava-growing areas of Central and West Africa (Calvert and Thresh, 2002). In more recent times, CMD has been reported as occurring at varying levels of incidence throughout the cassava belt of Africa and losses have been estimated at between 12% and 25% of total production (Thresh et al., 1997).

In 1983, the first sequence of a geminivirus to be determined and published was that of *African cassava mosaic virus* (ACMV) (Stanley and Gay, 1983). Later, this virus was included in the genus *Begomovirus* of the family *Geminiviridae* (Briddon and Markham, 1995), consisting of viruses with bipartite genomes transmitted by the whitefly *Bemisia tabaci* (Gennadius). For some time, ACMV was thought to be the only geminivirus species associated with CMD, despite the suggestion by Harrison and Robinson (1988), based upon serological analysis, that CMD was regionally distinct. Some 10 years after the publication of the sequence of ACMV, Hong et al. (1993) characterized a further CMD-associated geminivirus. Although based upon only the sequence of the DNA A genomic component (and therefore lacking any corroborative information on infectivity, since this is determined by the B genome component) this virus was given the name *East African cassava mosaic virus* (EACMV). A further geminivirus species has been found since in southern Africa and named *South African cassava mosaic virus* (SACMV) (Berrie et al., 1998).

During the 1990s, an unusually severe and damaging form of CMD spread rapidly through Uganda and the wider East African region and came to be referred to as the CMD “pandemic” (Otim-Nape et al., 1997; Legg, 1999). A novel recombinant geminivirus was shown to be consistently associated with the pandemic (Zhou et al., 1997), commonly in dual infections with ACMV that gave rise to the severest symptoms (Harrison et al., 1997). The so-called Uganda variant has been shown to be a recombinant between

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EACMV and ACMV, with ACMV providing an approximately 400 bp fragment of the coat protein gene; this virus is therefore best described as EACMV-Uganda (EACMV-Ug). Subsequent studies have identified DNA B components for EACMV-Ug, have confirmed the occurrence in Uganda of EACMV and have demonstrated infectivity through the generation of infectious clones (Pita et al., 2001).

At the beginning of this project it was apparent that the different begomovirus species and strains associated with CMD did not occur necessarily in different regions of Africa. It was therefore a major objective of the work to survey the diversity of cassava mosaic geminiviruses in the main cassava growing areas of Africa and determine their geographical distribution.

Methods and Materials

Cassava is normally propagated by stem cuttings and national collaborators therefore collected stem samples from CMD-infected plants during their project surveys, and shipped these to the John Innes Centre (JIC), UK. The material was planted in compost in containers (7.6 cm diameter) and maintained at about 25 °C (± 5 °C) in glasshouses, with supplementary lighting between October and April.

Samples of nucleic acids for polymerase chain reaction (PCR) analysis were extracted from 0.1 g samples of infected leaf tissue using the Nucleon Phytopure plant DNA extraction kit (Amersham) essentially as described by the manufacturer. The main modification of the procedure was that liquid nitrogen was used in place of dry ice. Samples were stored at –200 °C and used directly in the PCR reactions.

The PCR procedure and the primers utilized have been described previously (Briddon and Markham, 1994). Typically 1 µL, or 1 µL of a 10-fold dilution, of the DNA extract was used in the amplification reactions. Reaction volumes of 100 µL were used and amplification products were cleaned further by phenol extraction and ethanol precipitation before use in the restriction fragment length polymorphism (RFLP) analysis.

Typically 10 µL (of the original 100 µL reaction volume) of the PCR product was used for restriction enzyme digestion in a 100 µL reaction volume following the manufacturers' (Gibco-BRL) instructions. After a 4-hour digestion, RFLP products were resolved on 1% agarose gels in TBE buffer stained with ethidium bromide. For most cases this sufficed to determine the restriction pattern. However, for a few samples the amplification was poor. This necessitated blotting of the gel to Hybond membranes (Amersham) and probing with radioactively labeled probes. As a rule, all gels were blotted to Hybond for subsequent analysis.

Restriction patterns with particular viruses were predicted from published sequences (ACMV: Stanley and Gay, 1983; Morris et al., 1990. EACMV: Hong et al., 1993. EACMV-Ug: Zhou et al., 1997).

For each of the virus species encountered in the restriction mapping procedures, and any viruses with restriction maps that did not conform to known patterns, full length DNA A genomic components were cloned for sequence analysis. Clones were produced by PCR amplification with specific abutting, non-overlapping primers (Briddon et al., 1993). The sequences of these primers were obtained by cloning the PCR products produced with universal primers into pGem T-Easy vectors (Promega). Sequences were determined by dideoxynucleotide chain termination sequencing using the PCR-based BIG DYE kit (Perkin Elmer Cetus) and specific internal primers (Gibco-BRL). Reaction products were resolved on an ABI 377 automated sequencer. Sequence information was stored, assembled and analysed using Version 7 of the program library of the Genetics Computer Group.

Results

Table 1 summarizes the analysis of the samples. About 13% of samples re-grew initially without symptoms, although 14% of these (2% of the total) did eventually show symptoms after more than 3 months. Less than 1% recovered after initially showing symptoms. Table 2 shows the viruses

Table 1. Summary of analysis of cassava cuttings collected during field surveys. Sampled plants were all showing symptoms when collected.[a]

Country	Total no. of stems	Dead	Re-grown healthy	Re-grown infected	Healthy > infected	Infected > healthy	DNA extract	PCR DNA-A
Tanzania	80	9	0	71	0	0	39	26
Kenya	45	15	0	30	0	2	14	5
Uganda	126	28	11	87	0	0	65	24
Zimbabwe	1	0	0	1	0	0	1	1
Malawi	23	19	2	2	0	0		
Madagascar	193	1	39	153	1	2	30	18
Benin	96	46	7	43	5	2	35	15
Nigeria	68	21	27	20	7	0	25	12
Ghana	4	0	0	4	0	0	4	0
Cameroon	32	24	4	4	0	0	4	0
Total[b]	671	163 (24)	90 (13)	418 (62)	13 (14)	6 (1)	217 (51)	101 (47)

a. “Healthy” indicates plants showing no disease symptoms after about 3 months of re-growth. “Healthy > infected” indicates plants that initially showed no symptoms on re-growth but developed disease later; “Infected > healthy” indicates the converse. Other data: not sampled 39 (9%); yet to be identified 68 (16%).

b. ( ) Percentage of totals; for DNA extract ( ) percentage of successful viral DNA extractions made from cassava mosaic disease samples; for PCR DNA-A ( ) percentage of DNA A products successfully amplified from viral DNA.

Table 2. Summary of virus species isolated following restriction mapping using *Mlu*1, *Dra*1 and *Eco*R1 on polymerase chain reaction products following the use of universal primers to amplify *Begomovirus* genome component A.

Country	Virus species[a]					Total
	ACMV	EACMV	EACMV-Ug	SACMV	Dual	
Tanzania	3	5			1	8
Kenya	1	3			1	4
Uganda	6	2	15		3	23
Zimbabwe				1		1
Madagascar	12	4				16
Benin	12	3				15
Nigeria	8					8
Totals[b]	42 (56)	17 (23)	15 (20)	1	[5]	75

a. ACMV, *African cassava mosaic virus*; EACMV, *East African cassava mosaic virus*; EACMV-Ug, EACMV-Uganda; SACMV, *South African cassava mosaic virus*; Dual, combinations.

b. ( ) percentage of total; [ ] included in total as single infections.

identified from the samples analysed: ACMV comprised 56%, EACMV comprised 43% (of which 53% were EACMV and 47% EACMV-Ug) and 7% were dual infections (of which all except one were combinations of ACMV and EACMV-Ug). Samples from West Africa comprised less than one-third of the total identified (31%) but of these only 13% were EACMV, while this virus accounted for 56% of East Africa samples.

Full-length clones of the DNA A components of CMD-associated begomoviruses were obtained from isolates originating from Uganda and from Zimbabwe. The sequence of the clone originating from Uganda shows this to be highly similar to the sequence of EACMV-Ug reported by Zhou et al. (1997). The DNA A component of this virus is essentially EACMV with an approximately 450 bp insertion of an ACMV sequence in the centre of the coat protein gene. The restriction pattern of the virus originating from Zimbabwe was entirely novel, with no counterpart in the published literature. Sequence analysis of this clone showed it to have most similarity to SACMV (Berrie et al., 1998). The sequence published by Berrie et al. (1998) covers only the coat protein gene of SACMV to which the sequence of the Zimbabwe clone shows 86% similarity. In the absence of the full sequence of SACMV we must provisionally conclude that the Zimbabwe virus is distinct from, but most closely related to, SACMV. Efforts are now under way to obtain the DNA B genomic components of these two viruses for infectivity studies. Attempts also are continuing to produce full-length clones of EACMV and ACMV originating from Madagascar. Although amplification of these viruses has been achieved, it has not yet proven possible to clone them.

Discussion

The results obtained during this screening project indicate that a PCR/RFLP-based approach is able consistently to identify the viruses associated with CMD and identify species or strains that differ significantly from those previously reported. It is unclear whether a protocol based solely upon PCR amplification with diagnostic primers for each virus species/strain could achieve the same.

It is surprising, based on experience of related viruses, to find the genetic stability which is evident from the very low variability detected both in the sequence analysis of clones and from the RFLP analysis. However, this stability makes RFLP analysis a particularly useful diagnostic tool for these viruses. This method also enables plants that are dual-infected to be correctly diagnosed because the overlaid RFLP patterns are evident. Possibly the major drawback to the technique is that high-quality samples of nucleic acids are essential but these may be difficult to extract from cassava. This was most evident in the case of the dried cassava leaf samples sent to JIC from Africa, from which no successful PCR amplification was achieved. The reason for this failure remains unclear. It is possible that sample degradation occurs under tropical (hot and humid) conditions; fungal growth was particularly evident on some of the leaf samples. This problem has been encountered previously where even samples dried under optimal conditions were not suitable for DNA extraction and subsequent begomovirus PCR amplification. However, the cassava leaf samples collected from the glasshouse and dried immediately yielded amplification products more than 1 year later, after storage at room temperature but in a dry atmosphere.

It is clear from the recent reports of new strains/species of begomoviruses identified in association with CMD that the diversity of these agents is greater than initially expected. The procedures utilized for the analysis of the diversity of begomoviruses infecting cassava across Africa were devised to be all encompassing. The use of so-called "universal primers", designed with degeneracy so as to amplify all whitefly-transmitted begomoviruses, ensures that all possible begomoviruses in a particular sample are amplified. The use of specific primers in an analysis like this would risk missing possible new viruses, or mutants of viruses, which have been described previously. The universal primers used (Briddon and Markham, 1994) are well characterized and have been able thus far to amplify all begomoviruses against which they have been tested.

The use of RFLP analysis on PCR-amplified DNA allows several diagnostic restriction enzymes to be used on a single sample to identify a virus. The initial identification of a virus species/strain by a particular restriction pattern was subsequently confirmed by sequence analysis of a full-length clone. Full characterization of a clone requires infectivity studies on cassava (Briddon et al., 1998). Our work has shown that the use of alternate hosts, such as *Nicotiana benthamiana* Domin (*Solanaceae*), can lead to selection of mutants with atypical biological characters (Liu et al., 1997) and thus is not appropriate for diagnostic studies.

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CHAPTER 1.12

Serological Analysis of Sweetpotatoes Affected by Sweetpotato Virus Disease in East Africa

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Introduction

Infection of sweetpotato (*Ipomoea batatas* [L.] Lam.) by viruses is a major cause of yield reduction worldwide. At least 13 viruses are reported to infect sweetpotato naturally (Moyer and Larsen, 1991). Most of them are insect-transmitted, mainly by whitefly or aphid species. In East Africa, symptoms of sweetpotato virus disease (SPVD) were first reported on sweetpotato in 1945 (Hansford, 1945). However, the presence of viruses was not demonstrated until 1957 when Sheffield (1957) associated two viruses with sweetpotato plants having virus-like symptoms, virus A being aphid-transmitted and virus B being whitefly-transmitted. Virus A was later identified as *Sweetpotato feathery mottle virus* (SPFMV) but the identity of the whitefly-transmitted virus remained unclear. Subsequent efforts to determine the range of viruses occurring in East African sweetpotato crops confirmed SPFMV as the most frequently found virus but also revealed the presence of *Sweetpotato mild mottle virus* (SPMMV), *Sweetpotato latent virus* (SPLV) and *Sweetpotato chlorotic fleck virus* (SPCFV) (Carey et al., 1998). SPFMV has a worldwide distribution, is readily spread by aphids such as *Myzus persicae* (Sulzer) in a non-persistent manner but probably is not spread by seed transmission (Cadena-Hinojosa et al., 1981a). Various strains of SPFMV have been differentiated, mostly by symptoms produced on certain sweetpotato cultivars. In USA, two strains are recognized—the russet crack strain or internal cork strain, which causes internal necrosis of the storage roots of some cultivars, and the common strain, which causes no such symptoms. In East Africa, SPFMV-infected sweetpotato plants typically exhibit no symptoms on either roots or foliage. SPMMV is considered to be whitefly-borne (Hollings and Stone, 1976).

The whitefly-transmitted component virus of SPVD has been named as *Sweetpotato chlorotic stunt virus* (SPCSV) largely on the basis of the symptoms of West African isolates expressed in the indicator plant *Ipomoea setosa* Ker Gawl. (Schaefers and Terry, 1976). SPCSV is synonymous (Gibson et al., 1998) with Sweetpotato sunken vein virus (Cohen et al., 1992) and SPVD-associated closterovirus. An East African strain of SPCSV (SPCSV-SEA) was identified using monoclonal antibodies (MABs) prepared against an Israeli isolate of SPCSV as well as against bacterially

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expressed coat protein of a Kenyan isolate (Hoyer et al., 1996; Gibson et al., 1998). Subsequent analyses using enzyme-linked immunosorbent assays (ELISA) led to the description of two distinct serotypes: SEA1 as originally described from Kenya, and SEA2 (Alicai et al., 1999). In East Africa, SPCSV causes only moderate stunting and yellowing or purpling of the lower and middle leaves when it occurs alone (Gibson et al., 1998). Co-infection of SPFMV and SPCSV is very common and the severe symptoms of SPVD result from a synergistic interaction between SPCSV and SPFMV. SPVD is the most important disease of sweetpotato in Africa (Geddes, 1990). Depending on the genotype, SPVD symptoms comprise leaf strapping, mottling, vein clearing, puckering and stunting of sweetpotato plants (Schaefers and Terry, 1976).

The respective distributions of SPMMV, SPCSV and its synergistic partner, SPFMV, are still poorly understood in the major sweetpotato growing regions of East Africa, although it has long been known that these viruses can cause large yield losses (Mukiibi, 1977). The growing importance of sweetpotato for food security in Africa has increased international exchange of sweetpotato germplasm and thus increased the need to identify and establish the distribution of the common viruses occurring on the crop. Such information is an important prerequisite for the development and appropriate release of cultivars resistant to the prevailing viruses/ diseases.

The following work investigates the distributions of SPMMV, SPCSV and its serotypes, and SPFMV in Uganda, Kenya and Tanzania.

Collection and Analysis of Samples

Project partners carried out field surveys according to a common protocol. Additional information on the timing and coverage of the surveys are given in the respective country papers in the present volume (Chapters 1.6, 1.7 and 1.8). The authors analysed samples collected by project partners as follows: those from Uganda were tested fresh at Namulonge Research Institute; dried samples from Kenya and Tanzania were analysed at the Biologische Bundesanstalt für Land und Forstwirtschaft (BBA), Braunschweig, Germany. The samples were analysed using polyclonal antisera or monoclonal antibodies to SPCSV, SPFMV and SPMMV using triple antibody sandwich (TAS), double antibody sandwich (DAS), or nitrocellulose membrane (NCM) ELISA.

More than 200 sweetpotato leaf samples were collected from plants showing possible symptoms of viral infection. Field symptoms observed consisted of leaf strapping, yellowing, purpling, mottling, vein clearing and stunting of sweetpotato plants. Since incidence varied among and within the three countries, different numbers of samples were collected from each country and target area.

Reactions of the Diseased Samples

Table 1 shows the numbers of samples by country and target area that tested positive for SPFMV, SPSCV-SEA and/ or SPMMV. Both SPFMV and SPCSV occurred throughout the sampled areas. Overall, SPFMV was most frequently detected, in 151 out of the 243 samples. It was most frequently

Table 1. Results of enzyme-linked immunosorbent assay (ELISA) tests using antiserum / monoclonal antibodies to viruses of sweetpotato for sweetpotato leaf samples collected in Uganda, Kenya and Tanzania.

Country	Survey area	Number of positive samples[a]		
		SPFMV 1	SPCSV (SEA) 2	SPMMV 3
Uganda	Northern	12/38	14/38	0/36
	Eastern	13/36	14/36	0/36
	Central	26/38	34/38	2/38
	Western	19/40	38/40	7/40
	% frequency	46	66	6
Kenya	Coast	2/4	1/4	0/4
	Western	9/12	5/12	4/12
	Nyanza	9/9	5/9	1/9
	% frequency	80	44	20
Tanzania	N. Coast	17/21	5/21	0/21
	S. Coast	23/23	8/23	0/23
	Lake zone	21/22	16/22	7/22
	% frequency	92	44	11

a. Data are number of positive samples / number of samples tested. SPFMV 1, *Sweetpotato feathery mottle virus*: negative control, healthy *Ipomoea setosa*; positive control, SPCSV-Ky38 in *I. setosa*. SPCSV (SEA) 2, East African strain of *Sweetpotato chlorotic stunt virus*: negative control, healthy sweetpotato; positive control, SPFMV-46b in *Nicotiana tabacum* L. SPMMV 3, *Sweetpotato mild mottle virus*: negative control, healthy sweetpotato; positive control, SPMMV-DDR (German Democratic Republic). Values are considered as positive where the mean readings were at least two times greater than the mean of the healthy controls.

detected in samples from Tanzania (92%), slightly less frequently (80%) in those from Kenya and least frequently (46%) in samples from Uganda. One hundred and forty samples reacted positively to MABs specific to the East African strain of SPCSV. Frequencies of detection were 66% in samples from Uganda, 44% in samples from Tanzania and 44% from Kenyan samples. SPMMV was detected only in samples collected from areas around Lake Victoria in Uganda (6% incidence), Kenya (20%) and Tanzania (11%) (Table 1). Co-infections of SPFMV + SPCSV were more frequent (32% of samples) than the occurrence of either SPFMV (22%) or SPCSV (16%) alone. This association was expected because the two viruses co-exist synergistically (Schaefers and Terry, 1976). All combinations of viruses occurring together in samples also were detected but more rarely (Table 2).

Some samples did not react positively to any of the viruses tested, even though they appeared to show virus symptoms in the field. Low virus concentration and the irregular distribution of SPFMV in sweetpotato plants are frequently cited as obstacles to the reliable use of ELISA for virus detection in sweetpotato (Cadena-Hinojosa and Campbell, 1981b). Other possible explanations are that the presence of high concentrations of phenols, latex or other inhibitors adversely affected the reagents used in these tests and that tests on samples from Tanzania and Kenya were done on dried leaf samples stored for various lengths of time. Moreover, symptoms of viral infection can be difficult to recognize under certain circumstances and may have led to the collection of some uninfected samples, particularly in locations where the viruses occur less frequently, as was

Table 2. Single and multiple occurrences of viruses of sweetpotato in leaf samples from three East African countries.

Country	Nothing detected	Sweetpotato viruses[a]						
		SPFMV alone	SPCSV alone	SPMMV alone	SPFMV + SPCSV	SPFMV + SPMMV	SPCSV + SPMMV	SPFMV + SPCSV + SPMMV
Uganda	36	15	40	1	54	0	1	5
Tanzania	0	30	0	0	29	1	0	6
Kenya	2	9	0	1	9	2	1	1
Total	38	54	40	2	92	3	2	12
%	16	22	16	1	38	1	1	5

[a] SPFMV, *Sweetpotato feathery mottle virus*; SPCSV, *Sweetpotato chlorotic stunt virus* and SPMMV, *Sweetpotato mild mottle virus*.

the case in northern and eastern regions of Uganda. Alternatively, since all samples tested were selected on the basis of apparent virus symptoms, the lower rates of detection of virus in samples from more easterly areas may indicate the presence there of viruses other than SPCSV, SPFMV or SPMMV (i.e., viruses for which no tests were carried out).

In Uganda, the presence of two serotypes of SPCSV was confirmed using two MABs (Hoyer et al., 1996), both serotypes reacting with one (MAB 2G8) but only SEA2 reacting with the other (MAB 6D12) (Alicai et al., 1999). Serotype SEA2 was detected in 66 of the 96 SPCSV-positive samples, while SEA1 was detected in 30 of the SPCSV positive samples. Serotype SEA2 was found in all districts except for Tororo, Masindi and Iganga, although it was found most frequently in the central and western survey areas. Serotype SEA1 was distributed more evenly across the country and, in contrast to the SEA2, occurred more

Table 3. Distribution of two *Sweetpotato chlorotic stunt virus* (SPCSV) serotypes in sweetpotato leaf samples from 12 districts of Uganda.

Target area	District	No. positive samples		SPCSV serotype	
		MAB 2G8	MAB 6D12	SEA2	SEA1
Northern	Apac	4	1	3	1
	Lira	4	1	3	1
	masindi	5	5	0	5
	Total			6	7
Eastern	Tororo	1	1	0	1
	Palisa	8	6	2	6
	Iganga	5	5	0	5
	Total			2	12
Central	Luwero	11	0	11	0
	Mpigi	9	2	7	2
	Mukono	11	4	7	4
	Total			25	6
Western	Masaka	13	3	10	3
	Rakai	12	0	12	0
	Mubende	13	2	11	2
	Total			33	5

frequently in the northern and eastern survey areas (Table 3). These results confirm the heterogeneity of SPCSV, at least at the serotype level. Further studies would therefore be useful to investigate the diversity of this group of viruses and clarify any implications for disease management.

Conclusions

SPCSV is a common whitefly-borne virus in sweetpotato throughout East Africa, often occurring together with SPFMV, with which it acts synergistically to produce the severe disease SPVD. In contrast, SPMMV is largely restricted to the Lake Victoria region of East Africa.

Differences in the distribution of two SPCSV serotypes have been confirmed. Both SEA1 and SEA2 are widely distributed throughout Uganda but SEA1 is more prevalent in northern and eastern Uganda and SEA2 is more prevalent in central and western Uganda. These areas correspond with areas of lesser and greater whitefly abundance and slower and faster rates of spread (Aritua et al., 1998) but whether this relationship is causal is unclear.

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CHAPTER 1.13

Factors Associated with Damage to Sweetpotato Crops by Sweetpotato Virus Disease

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Introduction

Sweetpotato virus disease (SPVD) is the most destructive disease of sweetpotato (*Ipomoea batatas* [L]) in Africa (Geddes, 1990). It is caused by a combined infection of the aphid-borne *Sweetpotato feathery mottle virus* (SPFMV) and the whitefly-borne *Sweetpotato chlorotic stunt virus* (SPCSV) (Schaefers and Terry, 1976; Gibson et al., 1998b). Infection with SPFMV alone causes no obvious symptoms in African sweetpotato varieties (Gibson et al., 1997), while infection with SPCSV alone causes only moderate stunting, yellowing or purpling of middle and lower leaves and some yield loss (Gibson et al., 1998b). By contrast, SPVD causes severe plant stunting and small, distorted leaves with either chlorotic mosaic or vein clearing (Schaefers and Terry, 1976). The yield of affected plants generally is reduced by more than 50% (Mukiibi, 1977; Hahn, 1979; Ngeve and Boukamp, 1991). It appears that SPCSV has a synergistic effect on SPFMV (Rossel and Thottappilly, 1987; Aritua et al., 1998a): field plants that are infected with SPCSV quickly develop SPVD, either through an activation of a latent SPFMV infection or through increased susceptibility to aphid-borne spread of the virus. In this manner, infection with SPCSV is the trigger for SPVD and damage by the disease has long been linked with high populations of its vector, the whitefly *Bemisia tabaci* (Gennadius) (Sheffield, 1957).

SPCSV is transmitted semi-persistently by *B. tabaci*; feeds lasting a few hours are required for both efficient acquisition and inoculation (Cohen et al., 1992). *B. tabaci* that colonize sweetpotato in Africa cannot readily colonize cassava (*Manihot esculenta* Crantz) and vice-versa (Burban et al., 1992; Legg, 1996). As a result, there may be no direct link between whitefly numbers on sweetpotato and whitefly numbers on cassava, or between transmission of SPCSV and of *African cassava mosaic virus*, even where sweetpotato and cassava are intercropped. Thus, the epidemic of cassava mosaic disease and *B. tabaci* in cassava crops in East Africa seems to have had little effect on the incidence of SPVD. This is particularly fortunate because sweetpotato often provides the most readily available alternative crop to cassava.

Sweetpotato is an important crop for subsistence farmers throughout

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Uganda, generally being grown continuously throughout the year. Planting occurs whenever soil moisture is adequate and so is concentrated during, although not exclusively limited to, the rains, which in most areas of Uganda occur from March to June and from September to November. Crops are usually maintained for at least 6 months, cropping frequently being extended for up to 1 year by piecemeal harvesting, using a stick to tease out individual mature tubers (Bashaasha et al., 1995).

SPVD has been reported to be prevalent at moderate altitudes, 50 to 600 m above sea level, in East Africa (Sheffield, 1953). However, in Uganda, it was rare in the eastern districts of Soroti, Tororo and Busia but reached damaging levels of incidence in central Mpigi District, as well as in the southern districts of Masaka and Rakai, even though all locations were at a similar mid-altitude (Aritua et al., 1998b). The main aim of this project therefore was to understand the underlying causes of the different incidences, so as to provide a sound base from which to develop management practices. Recent work (Gibson et al., 1997; Aritua et al., 1998b) identified three factors as being associated with low SPVD incidence:

(1) Whiteflies were rare on sweetpotato in some localities (for example, the eastern districts of Uganda);

(2) The predominant sweetpotato variety is resistant; and

(3) Farmers select planting material from symptomless parents.

Our work therefore focused on links between SPVD incidence and (a) whitefly numbers, (b) varietal resistance and (c) farmers' plant health management practices.

Methods and Results

Monthly surveys of whiteflies and SPVD were carried out at 10 farmers' fields around each of six towns in Uganda (Soroti, Busia, Iganga, Namulonge, Kanoni and Masaka), from January 1998 to April 1999. All the sites are at about the same altitude and lie roughly along a south-east/north-west axis. Sweetpotato fields, 4 to 8 months old, were chosen while travelling along a rural road or path around each town, stopping at intervals of about 1 km or at the first field observed thereafter, different plantings being sampled each month. Whitefly numbers in each field were assessed by counting the number of whitefly adults observed while turning over leaves for a 1-minute period. Ten different parts of each crop were sampled in this manner. A more detailed account of the sampling method is provided in Aritua et al. (1998b). SPVD was assessed by counting the proportion of affected plants among 30 plants selected at random along a V-shaped track through each field.

Whiteflies were relatively few at Soroti and Busia except in February 1998 (Figure 1) and, in accord with this, SPVD was also rare there (Figure 2). Similarly, there were slightly more of both whiteflies and SPVD at Iganga. Although farmers' fields at Namulonge, Kanoni and Masaka all had relatively large and similar numbers of whiteflies, SPVD at Namulonge averaged only 2% and at Masaka, 6%, reaching 18% at Kanoni. Whitefly numbers were relatively high at all sites from January to April (inclusive). At those sites where SPVD was common, its incidence remained relatively low until February, consistent with there being a latent period of about 1 month (Gibson et al., 1998a) between infection by the whiteflies and

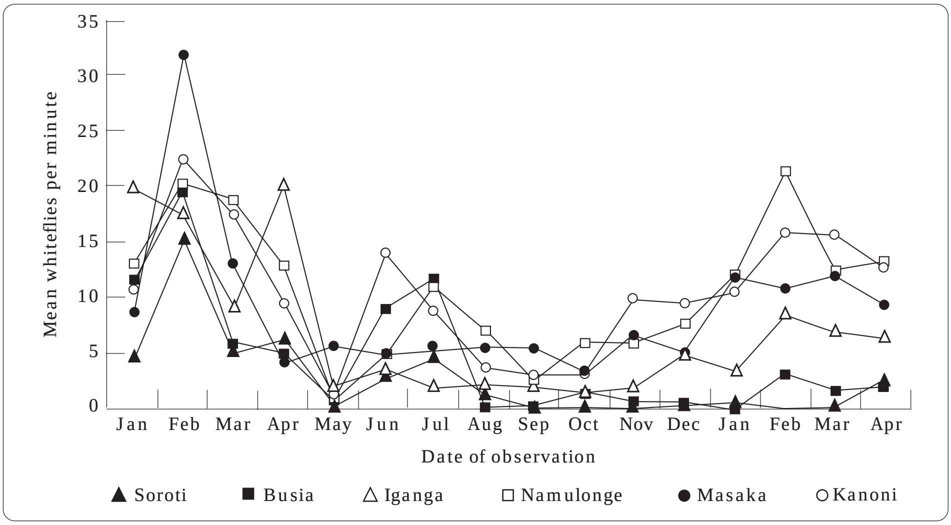

Figure 1. Mean whitefly numbers in six districts of Uganda (January 1998-April 1999). Each data point represents the mean for 10 fields.

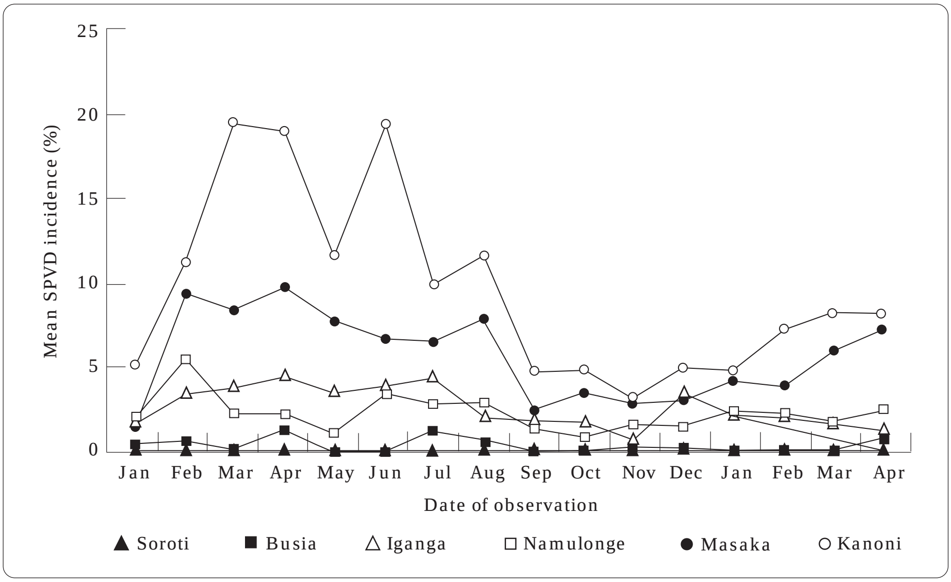

Figure 2. Sweetpotato virus disease (SPVD) incidence in farmers' fields in six districts of Uganda (January 1998-April 1999).

appearance of SPVD. The incidence of SPVD then remained relatively high until August, partly perhaps as a result of continuing infection but probably also because of the survival of affected plants.

From the point of view of understanding the relationship between vector dynamics and disease incidence over time, sampling farmers' fields had the disadvantages that they were not necessarily planted with the same cultivars and that the planting material already might have been infected and, to a varying degree, with SPVD. In order to eliminate these variables, three on-farm trials were planted at each of the three most contrasting sites, Soroti, Namulonge and Kanoni, using planting material from Namulonge Agricultural and Animal Production Research Institute (NAARI) farm, with one series of plantings in the second rains of 1997 (Aritua et al., 1999) and another in the second rains of 1998. Whitefly numbers and SPVD incidence were monitored monthly, from January to June, using the same procedures as in the surveys of farmers' fields. In both series, the results were very similar to those obtained by monitoring farmers' fields:

(1) No SPVD and few whiteflies (results not shown) were found at Soroti;

(2) Whiteflies were abundant at both Namulonge and Kanoni (Table 1);

(3) But SPVD increased in incidence rapidly only at Kanoni (Figure 3); and

(4) There was no correlation between whitefly numbers and final SPVD incidence (Table 1).

An index of the level of local inoculum for each site at Namulonge and Kanoni was also calculated from "area of each field of sweetpotato within 100 m of each trial" × "incidence of SPVD in it"¸ "distance to the trial plot". Results are tabulated (Table 1) for the first series of trials in which there was more virus spread. The value for local inoculum correlated closely with SPVD incidence at both sites, suggesting that the greater spread of SPVD at Kanoni was due to the greater incidence of SPVD in nearby farmers' fields. Examination of records of farmers' crops in the two localities also revealed that a highly SPVD-resistant variety called New Kawogo predominated at Namulonge and the surrounding villages and this may explain why SPVD was rare in the farmers' fields there. At Kanoni, more susceptible varieties predominated in farmers' fields (Aritua et al., 1999).

Links between SPVD incidence, crop data and farmers' management

Table 1. Relationship between sweetpotato virus disease (SPVD) incidence and (1) whitefly counts, (2) local inoculum and (3) whitefly numbers × local inoculum.

Farm	SPVD incidence[a]	Whiteflies[b]	Local inoculum[c]	Whiteflies × inoculum
Namulonge A	4.0	11.4	0	0
Namulonge B	1.5	13.2	100	1320.0
Namulonge C	0	14.0	120	1680.0
Kanoni D	18.5	10.5	1043	10951.5
Kanoni E	36.0	13.4	2298	30793.2
Kanoni F	23.5	9.4	1519	14278.6
Correlation[d]	-	-0.284	0.990	0.969
P	-	N.S.	< 0.001	< 0.001

a. SPVD incidence (%): final assessment (June) in each on-farm trial.
b. Whiteflies: mean monthly (January-June) whitefly counts from each on-farm trial.
c. Local inoculum: index calculated from SPVD incidence × area of field ÷ distance from field for fields within 100-m radius of trial plots.
d. Correlation of SPVD incidence with data in corresponding column.

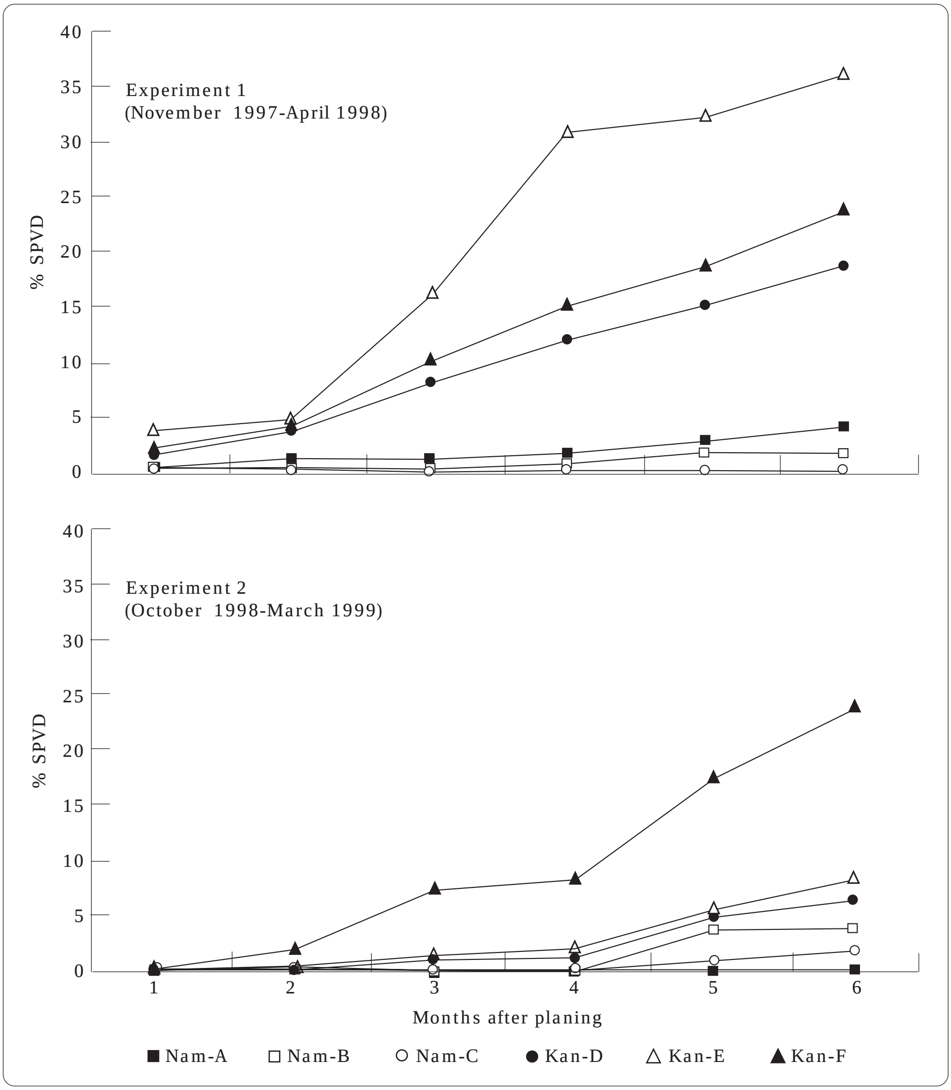

Figure 3. Progress of sweet potato virus disease (SPVD) in initially disease-free sweetpotato planted during the second rains of 1997 (Experiment 1) and the second rains of 1998 (Experiment 2) at farms at Namulonge (Nam-A, Nam-B, Nam-C) and Kanoni (Kan-D, Kan-E, Kan-F).

practices also were examined by farm surveys and farmer interviews in 1998-99 (for methods, see Gibson et al., 2000), focusing particularly on localities where SPVD was unusually prevalent, notably Bukoba and Karagwe sub-districts of Kagera District in Tanzania, and Rukungiri and Mpigi Districts in Uganda (Table 2). Most farmers, particularly those in Tanzania not exposed as yet to the bitter lessons of the cassava mosaic epidemic (which began in Uganda), did not realize that SPVD involved a virus spread by whiteflies, many considering it to be caused by too much sun. Even so, most farmers attempted to control SPVD, predominantly through the use

Table 2. Importance to farmers of different measures that they already are using to manage sweetpotato virus disease (SPVD) (adapted from Gibson et al., 2000).

Locality	Percentage of farmers[a] giving the rank indicated to a control measure									No. of farmers	
	Symptomless planting material			Roguing			Resistant cultivars			Controlling SPVD[b]	Interviewed
	1st	2nd	3rd	1st	2nd	3rd	1st	2nd	3rd		
Tanzania[c]											
Bukoba	100	0	0	0	33	0	0	0	0	15 (75)	20
Karagwe	80	0	0	20	10	0	0	0	0	10 (36)	28
Uganda											
Rukungiri	82	14	0	11	50	7	7	14	18	44 (88)	50
Mpigi	88	10	0	2	10	27	10	54	7	41 (82)	50
Soroti	100	0	0	0	17	17	0	17	0	6 (12)	50
Overall	87	9	0	7	28	13	6	25	9	116 (60)	198

a. Among those claiming to use specific measures to control SPVD.
b. Number of farmers (in parentheses, percentage of farmers among those interviews) claiming to use specific control measures for SPVD.
c. Farmers in other districts in Tanzania were not questioned on this subject.

of plant health management tactics (Table 2). In particular, most farmers carefully selected their planting material from unaffected parent plants. A few also removed (rogued) diseased plants from young crops. However, farmers often planted new crops close to old diseased crops, even though incidence of SPVD and the distance between crops were significantly ($P < 0.05$) and negatively correlated. Farmers also appeared to make no attempt to remove diseased plants from old and abandoned crops even when they were close to newly planted fields. This is consistent with farmers lacking knowledge that SPVD is caused by an insect-transmitted virus.

Although most farmers knew of SPVD-resistant varieties, few ranked resistance as a valuable management practice (Table 2), apparently because most resistant varieties had a poor and/or late yield. Despite this, they ranked new varieties with superior yield characteristics and resistance as their top requirement.

Conclusions

Our results confirm that SPVD varies considerably in incidence among sites and that part of the variation in incidence is associated with differences in the numbers of *B. tabaci* infesting the crop. Thus, the explanation for the rarity of SPVD in Soroti and Busia seems to be that whiteflies are rare throughout much of the year (Figure 1) but this still leaves unanswered the question of why fewer whiteflies should be present in Soroti and Busia than in Mpigi District. Our investigation was too limited to identify positively the reason(s) for this. However, a major difference between Soroti and Busia Districts and Mpigi District is that the first rains in Soroti and Busia Districts occur later and the second rains earlier than in Mpigi District. This leads to a more prolonged dry season between the second and the first rains in Soroti and Busia, during which time most vegetation dries out, natural bushfires are common and the resulting natural vegetation predominantly consists of low-growing bushes and grasslands. Rainfall being more evenly spread throughout the year in Mpigi, vegetation continues to grow year-round and such fires rarely occur. As a result, sweetpotato crops in this region often occur in locations sheltered by tall trees and other vegetation.

High incidences of SPVD were associated with:

(1) Large average numbers of whiteflies found on the sweetpotato crop during the year;

(2) High peak whitefly populations, which occurred during the hot dry season (January-March); and

(3) Relatively large amounts of SPVD-affected plants nearby.

The importance of local inoculum was shown directly by our on-farm field trials but was indicated also by survey data showing that SPVD incidence increased as sweetpotato cropping intensity increased. A requirement for local inoculum is consistent with SPCSV being only semi-persistently transmitted and with *B. tabaci* being a relatively weak flier. Using pesticides to reduce the population of the whitefly vector is highly unlikely to be a useful tactic in SPVD management since, even setting aside environmental risks, human health concerns and the tendency of whiteflies to develop resistance to insecticides, the crop has too low a value to justify the investment in insecticides. Choice of appropriate varieties is unlikely to be effective in reducing vector populations because earlier work has not identified major differences in the numbers of whiteflies on different sweetpotato varieties (Aritua et al., 1998a). However, local inoculum could be reduced by:

(1) Planting disease-free cuttings (already used by most farmers);

(2) Roguing plants that develop symptoms (used by a few farmers early in the season);

(3) Removal of crop debris (both roots and foliage as both re-grow readily and are often diseased) from old fields (not currently used);

(4) Separation of new crops from old diseased ones (not currently used); and

(5) Increased use of resistant varieties (favoured only in localities where acceptable resistant varieties were available).

Our work also suggests that it may be particularly important to apply plant health management tactics during the hot dry season because this is when the whitefly vectors are most abundant. However, it is important to emphasize that, although they may seem obvious, none of these strategies have been tested yet in field trials, to ensure that they are effective and adoptable by farmers, and so cannot be officially recommended. Furthermore, our farmer interviews indicated that most farmers do not have a sound understanding of the cause and source of SPVD. Some of these control practices might therefore appear to farmers to be inappropriate. Consequently, both participatory research and training programs will be essential if plant health management strategies are to be developed and widely adopted.

Farmers in areas severely affected by SPVD gave highest priority to the need for superior, virus-resistant varieties (Table 3). Our work showed that resistant varieties are valuable not only because they suffer less SPVD damage but also because their widespread cultivation reduces incidence of SPVD in remaining stands

Table 3. Aspects of sweetpotato virus disease (SPVD) management on which farmers in severely affected localities would most like to receive assistance (adapted from Gibson et al., 2000).

Locality	Percentage of farmers[a] giving a particular rank to the indicated management tactic									No. of farmers	
	Superior, resistant cultivars			Technical information			Chemical control			Requesting[b]	Interviewed
	1st	2nd	3rd	1st	2nd	3rd	1st	2nd	3rd		
Bukoba	100	0	0	0	0	0	0	0	0	14 (70)	20
Rukungiri	60	21	0	23	6	2	17	15	0	48 (96)	50
Overall	69	16	0	18	5	2	13	11	0	62 (89)	70

a. Among those requesting specific measures to manage SPVD.

b. Number of farmers (in parentheses, percentage among those interviewed) requesting assistance with management of SPVD.

of susceptible varieties. Although farmers also gave a low ranking to the use of their currently available resistant varieties (Table 2), this was because most farmers perceived their resistant landraces to have major flaws. The higher yielding, resistant varieties such as those selected at NAARI in Uganda and at the Agricultural Research Institute in Ukiriguru, Tanzania, taking account of farmer preferences, should overcome this obstacle.

Acknowledgements

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CHAPTER 1.14

Conclusions and Recommendations

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Introduction

The first phase of the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project (see Introduction to this volume) provided a unique opportunity to assess whitefly and whitefly-borne virus problems in a diverse range of cropping systems across the tropics and to obtain a broad range of diagnostic information as a basis for further concerted action. Sub-Project 4, Whiteflies as virus vectors in cassava and sweetpotato in sub-Saharan Africa, had the ambitious target of establishing collaborative research linkages among more than 15 partner institutions and implementing diagnostic surveys in nine African countries. Activities began in September 1997 and were completed in mid-1999. This chapter reviews the results obtained, considers the experiences gained in setting-up, implementing and completing the diagnostic phase of research, and discusses the implications for subsequent work on enhancing the management of whitefly-borne diseases of cassava (*Manihot esculenta* Crantz) and sweetpotato (*Ipomoea batatas* [L.] Lam.).

Increased Biological Understanding

Whitefly species and abundance

Field data collection and the examination of whitefly nymph specimens collected by project partners during surveys of cassava and sweetpotato in participating countries provided the most comprehensive information ever obtained for whiteflies on these crops in Africa. More than 2000 specimens were identified and four species were recorded. The two species that have been reported previously on cassava, *Bemisia tabaci* (Gennadius) and *B. afer* (Priesner and Hosny), were the only ones occurring widely on cassava and sweetpotato and were recorded from most locations. Two species were identified that have not been reported previously from cassava. These were the greenhouse whitefly *Trialeurodes vaporariorum* Westwood, identified from two locations in Ghana and Nigeria, and T. *ricini* (Misra) identified from a single site in the Lake Victoria zone of Tanzania. Whilst the occurrence of late instar nymphs of these two species does suggest

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colonization of cassava, the very low frequency of occurrence means that they are of little significance.

B. tabaci and *B. afer* both occurred on cassava and sweetpotato, although *B. afer* was relatively less frequent on sweetpotato and in Uganda was not recorded from this crop. There was an important geographical zonation in the relative abundance of the two species (Figures 1-3). Whilst *B. tabaci* predominated in countries in the equatorial belt (Ghana, Benin, Nigeria, Cameroon, Uganda and Kenya), the two species occurred at similar frequencies in Tanzania, to the south, and *B. afer* predominated in the most southerly countries, Malawi and Madagascar. The reasons for this pattern of occurrence are not clear, particularly since no single environmental factor appears to correlate consistently with the species ratio. In Malawi, however, there was a significant difference in the ratio between survey areas. In the two survey areas (1 and 2) along the shore of Lake Malawi (475-700 m altitude), similar numbers of the two species were recorded; whilst in survey area 3, the central plateau (1000-1500 m altitude), 97% of specimens identified were *B. afer* (Chapter 1.9, this volume).

The key difference between the lakeshore and plateau environments is temperature and it seems likely that the low temperatures that occur in the mid-altitude environments of southern Africa are unfavourable for *B. tabaci*. This possibility is supported by published information indicating that the development period of this species is significantly increased (Gerling et al., 1986) and fecundity significantly reduced (Azab et al., 1971) at temperatures below 15 °C. More detailed ecological studies and molecular characterization of the two species will be required to clarify these issues, particularly since the current surveys were conducted only at one time in the year and therefore may not be fully representative. It is important to highlight, however, that *B. afer* has never been shown to transmit viruses. The distinction between the two species therefore becomes important if errors are not to be made in describing relationships between adult whitefly populations and the spread of viruses causing cassava mosaic disease (CMD) or sweetpotato virus disease (SPVD).

Geographical patterns of variation in abundance of cassava whiteflies were partly confounded with the different patterns of distribution of the two species (Figures 1-3). Although survey teams were trained to differentiate the adult stages of the two species, it was not always possible for them to do so during counting. Some patterns in distribution were apparent, however. In West Africa, where *B. tabaci* was the dominant species, abundance was generally greater in the more humid rainforest and transition forest ecozones. Conversely, populations were smaller in the drier northern savannah zones. This is a pattern that has been recognized before, both in the Ecologically Sustainable Cassava Plant Protection (ESCaPP) surveys of 1993-94 (IITA, 1998) and through earlier studies of CMD epidemiology in Ivory Coast (Fauquet et al., 1987; 1988). In East Africa, two centres of whitefly abundance were recognized: the northern shoreline of Lake Victoria, in Uganda, and the northern Tanzania/southern Kenya coast. As for West Africa, these are also the areas in the region where humidity is greatest. It is notable, however, that abundance of *B. tabaci* in the "Lake Crescent" agro-ecological zone in Uganda, comprising the southern districts of Iganga, Mukono,

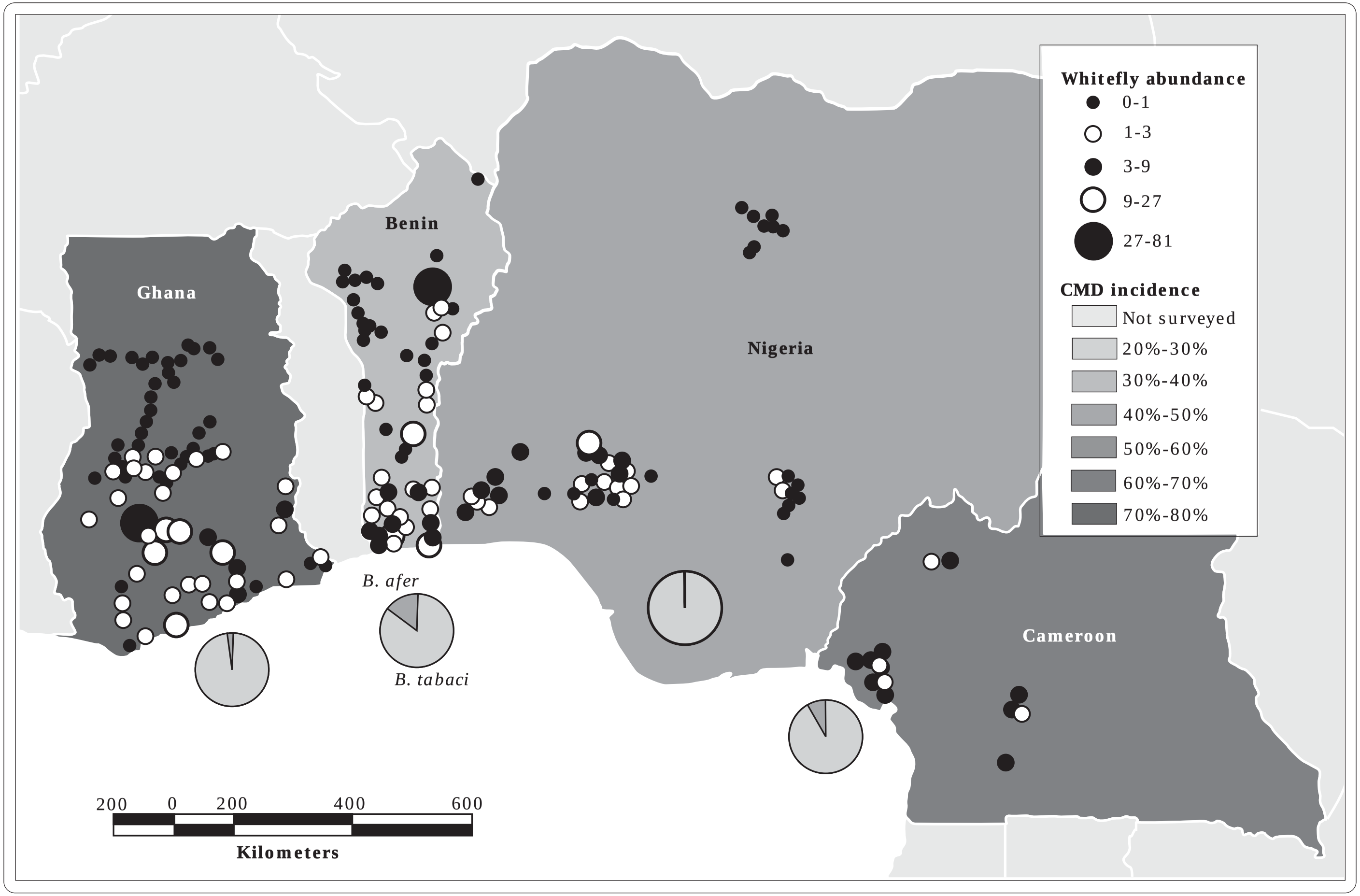

Figure 1. Whitefly abundance, cassava mosaic disease (CMD) incidence and *Bemisia tabaci*/*B. afer* ratio for Sub-Project 4 countries in West Africa.

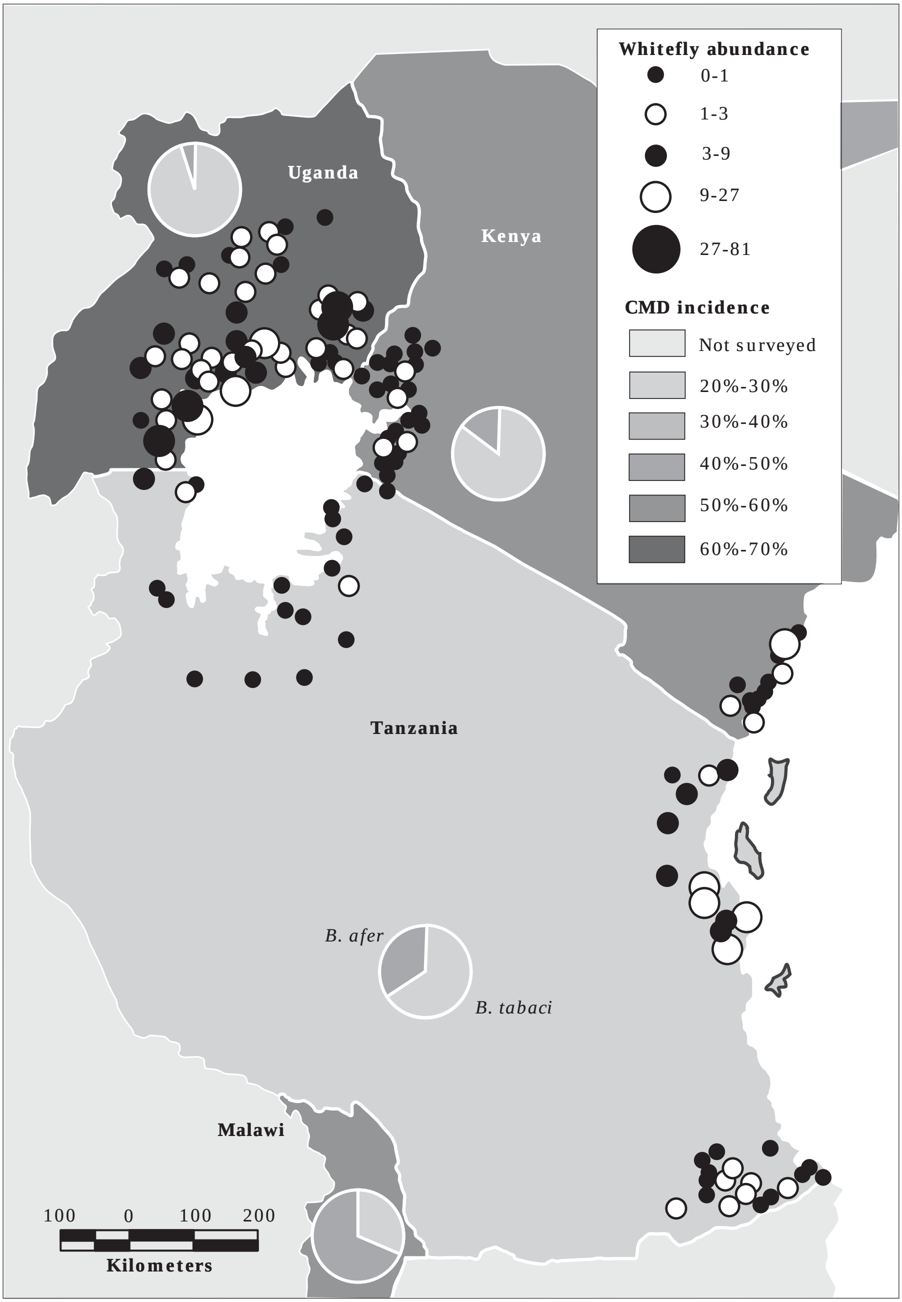

Figure 2. Whitefly abundance, cassava mosaic disease (CMD) incidence and *Bemisia tabaci*/*B. afer* ratio for Sub-Project 4 countries in East Africa.

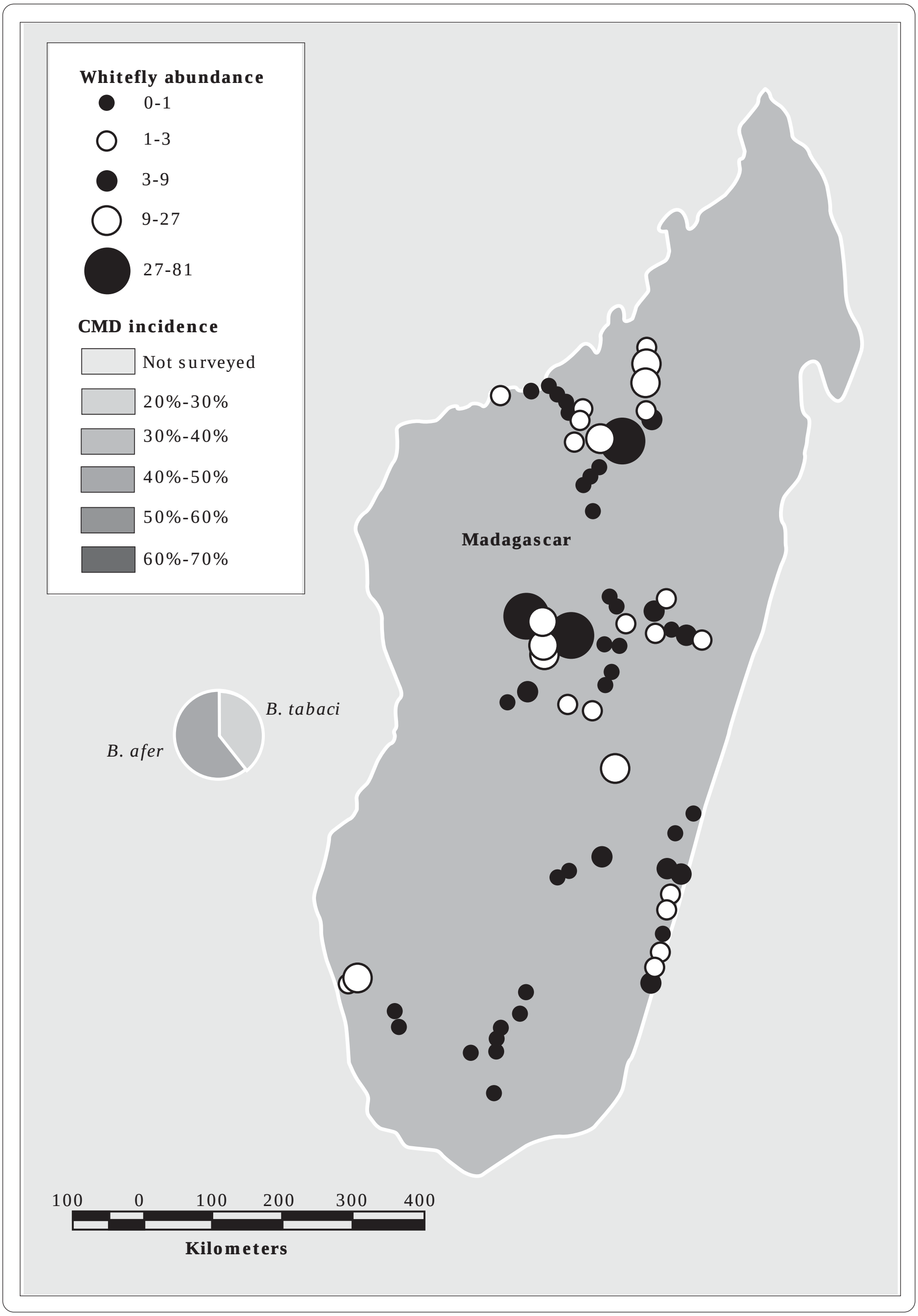

Figure 3. Whitefly abundance, cassava mosaic disease (CMD) incidence and *Bemisia tabaci*/*B. afer* ratio in Madagascar.

Mpigi, Masaka and Rakai, is recorded as having increased significantly during the second half of the 1990s (Legg, 1995; Otim-Nape et al., 1997; Legg and Ogwal, 1998). This is now thought to have resulted from a synergistic interaction with an unusually severe form of CMD, which spread through Uganda during this period (Colvin et al., 1999) and continues to spread in the East and Central African region (Legg, 1999). The interaction has occurred in all agro-ecologies in Uganda but it remains to be seen if the same will occur in the drier agro-ecologies to the south, in Tanzania. In Madagascar, there was no clear pattern of variation in abundance (Figure 3), although abundance in the southern central areas was lower than elsewhere.

Variation in abundance of sweetpotato whiteflies was much greater than for cassava whiteflies (Table 1). Very high populations were recorded around the shores of Lake Victoria, whilst whiteflies were not recorded at all from many locations in Madagascar. It is likely that seasonal effects can partly explain this degree of variation, since sweetpotato is grown for a much shorter period (typically 4-6 months) than cassava (typically 10-24 months). Thus, although cassava crops may be available throughout the year, in drier areas, sweetpotato cultivation outside the main growing season is confined to very small areas used for the maintenance of planting material. Associated with this, there is evidence showing that *B. tabaci* on sweetpotato colonize a wide range of crop plants and weed species, in contrast to *B. tabaci* on cassava, which appear to be largely restricted to cassava (Burban et al., 1992; Legg et al., 1994).

Whitefly-transmitted viruses in cassava

Stem samples of CMD-diseased cassava were sent from all nine principal participating countries to the John Innes Centre (JIC) for diagnostics and characterization. Chapter 1.11 (this volume) reports the results of this work. Until 1996, it was considered that two cassava mosaic begomoviruses were occurring in Africa, with largely disjunct distributions (Swanson and Harrison, 1994). *African cassava mosaic virus* (ACMV) was reported to occur in much of West, Central and southern Africa, and *East African cassava mosaic virus* (EACMV) was said to be restricted to coastal East Africa, Madagascar, Malawi and Zimbabwe. More recent diagnostic surveys have shown that EACMV is not restricted to East Africa (Offei et al., 1999; Ogbe et al., 1999), that the distributions of EACMV and ACMV overlap (Ogbe et al., 1996; 1997) and that mixed virus infections occur (Harrison et al., 1997). Results from the TWF-IPM Project (Chapter 1.11, this volume) have extended our understanding further by establishing the first record of EACMV from Benin in West Africa; identifying a novel begomovirus from cassava in Zimbabwe, which appears to be closely related to *South African cassava mosaic virus* (Rey and Thompson, 1998); and highlighting the occurrence of virus mixtures in East Africa. Dual infections are a relatively uncommon occurrence (< 10%) and it is significant that all these were recorded from the part of East Africa affected by the pandemic of unusually severe CMD (Otim-Nape et al., 1997; Legg 1999; Chapter 1.1, this volume).

Although virus diagnoses have been carried out on only a limited number of samples, a relationship is apparent between the viruses affecting cassava plants and the disease caused. In West Africa, diversity seems to be limited and symptoms of disease are generally mild to moderate (Table 1). In East and southern Africa, by contrast,

Table 1. Summary of data on the incidence of cassava mosaic disease (CMD), sweetpotato virus disease (SPVD) and abundance of *Bemisia tabaci*, from Sub-Project 4 diagnostic surveys.

Country	Survey area	Fields[a]	CMD (%)[b]	Wfinf[c] (miu)	CMD sev.[d]	Cassava WF[e]	SPVD (%)[f]	SP WF[g]
Uganda	North	20 (20)	77	48.0	3.0	1.2	5.0	1.40
	East	20 (20)	68	46.0	3.1	1.8	12.0	5.40
	South/Central	20 (20)	79	53.0	2.8	3.8	15.0	12.40
	West	20 (20)	48	55.0	2.8	9.2	10.0	3.00
Kenya	Coastal Province	17 (15)	56	36.0	2.3	2.9	0.5	0.50
	Western Province	15 (18)	85	64.0	2.9	0.3	6.0	2.30
	Nyanza Province	18 (17)	11	2.3	2.2	0.5	7.0	4.90
Madagascar	Central plateau	35 (11)	31	7.8	2.6	7.1	3.0	0.01
	Humid coast	22 (15)	71	11.0	3.5	4.3	2.0	0
	Dry coast	12 (7)	46	11.0	3.2	5.9	18.0	0.10
	Semi-humid coast	42 (6)	41	11.0	3.3	2.6	0	0
Tanzania	Lake zone plateau	20 (20)	21	1.7	2.6	0.7	11.0	40.10
	Semi-humid coast	20 (20)	12	7.1	2.7	1.2	13.0	2.80
	Humid coast	20 (20)	39	21.0	3.0	5.8	6.0	3.10
Malawi	Central lakeshore	20	62	55.0	2.8	1.5	-	-
	Northern lakeshore	9	52	34.0	2.7	1.0	-	-
	Central plateau	12	11	3.7	3.0	1.3	-	-
Ghana	Coastal savannah	10	76	44.0	2.2	11.8	-	-
	Rainforest	25	69	23.0	2.2	1.5	-	-
	Transition forest	25	74	54.0	2.5	1.0	-	-
	Guinea savannah	20	68	44.0	2.4	0.3	-	-
Benin	Transition forest	28	26	3.4	2.1	4.2	-	-
	Wet savannah	19	43	0.5	2.1	0.7	-	-
	Dry savannah	13	49	3.4	2.1	3.4	-	-
Nigeria	Rainforest	25	83	3.9	2.4	3.1	-	-
	Transition forest	25	32	2.7	2.1	2.6	-	-
	Wet savannah	20	56	0	2.4	1.0	-	-
	Dry savannah	10	45	2.4	2.2	0.3	-	-
Cameroon	South-west	26	54	21.0	2.2	4.1	-	-
	North-west	16	55	21.0	2.4	2.4	-	-
	Centre-south	28	73	5.6	2.3	3.1	-	-

a. Fields: Figures in parentheses indicate number of sweetpotato fields sampled.
b. CMD %: Total incidence of CMD, calculated as the average percentage of plants showing symptoms of virus disease from 30 plant samples recorded in each dataset (i.e., one field).
c. Wfinf (miu): Incidence of plants infected with CMD whitefly in the current season, transformed to allow for multiple infection units (Gregory, P. H. 1948. The multiple infection transformation. Ann. Appl. Biol. 35: 412-417).
d. CMD sev.: Severity of CMD symptoms, based on an arbitrary 1-5 scale, where 1 indicates low and 5 indicates high severity of symptoms.
e. Cassava WF: Mean whitefly abundance for cassava recorded as the average number of adult whiteflies counted on the five uppermost leaves of 30 plant samples recorded in each field.
f. SPVD %: Total incidence of SPVD, calculated as the average percentage of plants showing symptoms of virus disease from 30 plant samples recorded in each field.
g. SP WF: Mean whitefly abundance for sweetpotato recorded as the average number of adult whiteflies counted in ten 1-minute counts made at random in the sampled field.

complexity appears greater, dual infections are more prevalent and severe disease is more widespread, although in most instances (4/5) the dual infections were in areas affected by the East African CMD pandemic (Legg, 1999). An association between virus infection and epidemiology is similarly apparent (Table 1). In East Africa, current-season whitefly-borne infection is greatest in Uganda, where the "Uganda variant" of EACMV (EACMV-Ug) was reported (Chapter 1.11, this volume) and where EACMV-Ug/ACMV mixtures were common, as reported earlier by Harrison et al. (1997); whilst in Nigeria, where only ACMV was reported, current-season whitefly-borne infection was very infrequent.

The incidence of CMD varied considerably within sub-regions. In West Africa, disease incidence ranged from 36% (Benin) to 72% (Ghana); whilst in East and southern Africa, it ranged from 21% (Tanzania) to 68% (Uganda). In all countries, the proportion of diseased plants that had arisen from planting diseased cuttings (cutting infection) was greater than that derived from current season whitefly-borne infection. Many factors influence CMD incidence, including the virus(es) infecting the plant, cultivar response to the virus(es), vector abundance and transmission efficiency, the physical environment, and cropping practices (including vector or disease management measures). Most of these factors remain poorly defined for many of the participating countries, although the surveys have provided quantitative data relating to some aspects. In this respect it would be useful to compare the epidemiology of CMD between regions, using common cultivars for reference, as proposed by Legg et al. (1997).

Whitefly-transmitted viruses in sweetpotato

Sweetpotato leaf samples with symptoms of virus disease from Uganda, Kenya, Tanzania and Zambia were analysed. Chapter 1.12 discusses in detail the results of virus diagnoses from Uganda, Kenya and Tanzania, and Kaitisha and Gibson (1999) discuss those from Zambia. The East African strain (S_{EA}) of *Sweetpotato chlorotic stunt virus* (SPCSV) occurred throughout the region and, in Uganda, Aritua et al. (Chapter 1.12, this volume) have discussed distributions for the two distinct serotypes (S_{EA1} and S_{EA2}) described by Alicai et al. (1999). The other whitefly-borne virus identified from project surveys was *Sweetpotato mild mottle virus* (SPMMV), although this was restricted to areas around the shores of Lake Victoria (Chapter 1.12, this volume). SPVD, which results from co-infection of sweetpotato plants with SPCSV and the aphid-borne *Sweetpotato feathery mottle virus* (SPFMV), occurred throughout areas sampled. The disease was most frequent in south-western Uganda, the Lake Victoria zone of Tanzania, south-eastern coastal Tanzania and the semi-arid south-western part of Madagascar. Diagnostic tests related SPVD to the co-occurrence of SPCSV and SPFMV for each of these regions (Chapter 1.12, this volume). No samples from Madagascar were tested.

Whitefly natural enemies

The agreed survey protocol for the TWF-IPM Project provided guidelines for the collection of parasitoids, predators and entomopathogens during the course of diagnostic surveys. Limitations in the methods proposed, and the short period of time available for natural enemy collection at each sampling site, meant that the collection of natural enemies was less comprehensive than had been anticipated. More intensive and targeted surveys would be required for the adequate characterization of predators and entomopathogens in particular.

Survey teams in most countries collected parasitized whitefly mummies and reared adult parasitoids from them. The parasitoids were subsequently mounted and identified by Dr. Mohammed Ali Bob of the International Center of Insect Physiology and Ecology (ICIPE). Dr. A. Polaszek of the British Museum (Natural History) verified representative specimens. Five species of parasitoid were recorded: *Encarsia sophia* (Girault and Dodd), *Encarsia lutea* (Masi), *Encarsia mineoi* Viggiani, *Encarsia* sp. (*luteola* group) and *Eretmocerus* sp. (Table 2). Only two species, *E. sophia* and *Eretmocerus* sp., were widely distributed, parasitizing whiteflies on both cassava and sweetpotato. *E. sophia* was an order of magnitude more common that *Eretmocerus* sp., although it is considered that easy recognition of the former, resulting from the black coloration of the mummy from which it emerges, may have partially biased the sampling. Whilst over 30 species of parasitoid have been described as parasitizing *B. tabaci* worldwide (Gerling, 1986), only limited information is available about *Bemisia* parasitoids in Africa, and virtually no information relating to cassava or sweetpotato (Fishpool and Burban, 1994). Whilst this component of the TWF-IPM Project's diagnostic phase had deficiencies, it is nevertheless considered that the information obtained has provided an important baseline from which to develop future studies. Key issues that will need to be more fully addressed include the variation in parasitoid diversity between regions, of which there was evidence in this study, and the role (if any) of the different parasitoid species in the population dynamics of *Bemisia* spp. and the epidemiology of CMD.

Increased Socio-Economic Understanding

Farmers' assessment of whitefly-related problems

There was a clear relationship between farmers' assessments of CMD and SPVD and the prevalence and severity of the diseases as described from the field data collection. In Uganda, all farmers both recognized CMD and considered it a problem for their cassava production. In the countries of West Africa, on the other hand, although typically about 70% of farmers were able to recognize CMD, less than 50% considered it a problem. A smaller proportion of sweetpotato farmers recognized SPVD but, where it was recognized, most farmers

Table 2. Aphelinid parasitoids identified from diagnostic surveys of Sub-Project 4.

Country	*Encarsia sophia*	*Eretmocerus* sp.	*Encarsia* sp. (*luteola* group)	*Encarsia lutea*	*Encarsia mineoi*
Uganda	62	46	3	-	-
Kenya	13	2	-	-	-
Tanzania	62	-	1	-	-
Ghana	4	-	-	-	-
Benin	16	2	-	-	-
Nigeria	557	21	4	7	5
Cameroon	324	38	-	-	-

considered it a problem. Much smaller proportions of farmers were able to recognize whiteflies and many expressed surprise when informed of their role as disease vectors. There were significant differences between countries in this respect, however, with the extremes being Madagascar, where no farmers reported recognizing whiteflies, and Benin, where 70% recognized them.

Farmers had names for whiteflies and whitefly-transmitted diseases in all countries, but in many cases these were non-specific, translating simply as “insect” or “disease”. More specific names were commonest for CMD, which was often described using anthropomorphic disease equivalents such as “leprosy”. Names for SPVD were often exactly the same as those used for CMD. Where such names are in common usage, potential exists for their use, perhaps with minor modification, in training and extension programs in the future. Estimates of loss to the respective diseases were most frequently in the range of 25%-75% for cassava and 25% or less for sweetpotato, although most survey teams considered that these were likely to be overestimates, particularly in West African countries, where disease symptoms (in this case only CMD) were typically mild. Participatory yield loss studies would be helpful in many of the surveyed countries to evaluate losses scientifically and reconcile such estimates with farmer perceptions. The shortcoming of this and other responses emphasized the need to link producer interviews with the collection of more objective field data.

Managing whiteflies and whitefly-transmitted viruses

There were widely divergent responses on the use of vector and disease management measures. In Ghana and Madagascar, less than 5% of farmers reported doing anything at all to control CMD. This result was particularly anomalous for Ghana, since many farmers reported relatively high losses. Management practices were most widely reported in Uganda and Cameroon, where most farmers indicated the use of at least one practice. The most cited methods of disease management were the phytosanitary techniques of roguing, and selection of disease-free stems. This has to be set against the predominance of the use of diseased cuttings as planting material in almost all areas surveyed. Selection of disease-free planting material similarly was cited widely as a management tactic but the health status of stems was normally less important as a criterion for planting material selection than were agronomic characters.

Although it appears that farmers over-estimated their use of roguing and selection of healthy cuttings, they under-reported use of disease-resistant varieties. Use of cassava varieties recommended for resistance to CMD was most widespread in Cameroon, where 48% of farmers reported growing variety “Agric” in the knowledge that it was resistant to CMD. In Uganda, more than 15% of sweetpotato farmers were found to be growing SPVD-resistant varieties but none of these realized that this was a method of managing SPVD.

Very few farmers reported using pesticides to control whiteflies or whitefly-borne viruses. Occasionally, sweetpotato farmers reported pesticide use but this was more likely to be for the purpose of controlling other insect pests such as the sweetpotato butterfly *Acraea acerata* Hewitson. Chapter 1.13 (this volume) gives a more detailed assessment of the approaches of sweetpotato farmers to SPVD control. Few farmers reported having received any technical assistance with respect to the control of either CMD or SPVD and, as a result, most considered that the management methods being used were their own. The only exception to this was Cameroon, where 39% of farmers reported that they had received technical support relating to whiteflies and whitefly-borne diseases.

Recommendations

The overall picture that emerges from this assessment of farmers' awareness of whiteflies and whitefly-borne diseases in cassava and sweetpotato and their approach to disease management is one of weak understanding of the nature of the problem and a correspondingly haphazard implementation of management tactics. Much of the responsibility for this lies with the research (both national and international) and extension systems. Researchers have failed to provide a well-defined and well-tested strategy for managing CMD and SPVD. Moreover, weakness in the extension systems of most of the participating countries has meant that even where management recommendations have been defined, these are not reaching the majority of farmers (as is particularly the case for the control of whitefly-borne diseases of cassava and sweetpotato in Uganda). In order to address this situation, key objectives for future work on whiteflies and whitefly-borne diseases, for both cassava and sweetpotato, should be to encourage researchers, extension agents and farmers to work together to:

(1) Define exactly where disease management is needed and where not;

(2) Develop effective, clearly defined and readily implemented vector and disease management methods (which based on current knowledge are most likely to include choosing disease-resistant varieties, selecting disease-free planting material and/or roguing of diseased plants); and

(3) Facilitate the adoption of these methods by using participatory training and all media channels available to improve the access of farmers to current knowledge.

Sub-Project 4 of the TWF-IPM Project has been successful in achieving part of the first objective. Following the completion of the diagnostic phase, there is a clearer understanding of the distribution of whitefly-borne viruses, what their impact is in terms of disease incidence and epidemiology and how farmers are responding to these diseases. The project also has been uniquely successful in obtaining comparable information of this type from nine of the major cassava-producing countries of Africa and four of the major sweetpotato-producing ones; these countries together account for more than 50% of African production of each crop. In achieving this, the project has established a network of researchers working on whiteflies and whitefly-borne disease of root crops in sub-Saharan Africa. In order to complete the first objective and to begin to address the second, the following issues, a number of which have been highlighted in the country chapters, need to be addressed.

(1) How severe are yield losses in the CMD-diseased local landraces that still dominate cassava production in Africa? Knowledge of this is particularly critical, for example, in Ghana, where farmers report high losses yet appear to make little attempt to control them. A realistic estimate of current yields and yield losses would help direct research and extension efforts more effectively.

(2) What are the conditions under which tactics for improving plant health, especially selection of disease-free cuttings and roguing of diseased plants, are most likely to be effective?

(3) Where is host plant resistance required as a foundation for sustainable management of cassava and/or sweetpotato (as compared with a strategy focusing on improving the husbandry of land races) and are particular virus/vector/environmental characteristics common to these areas?

(4) Where conditions demand the use of host plant resistance, as has been the case in Uganda during the pandemic of severe CMD, what are the constraints to the adoption of newly developed CMD-resistant varieties?

(5) Are there alternative, novel approaches to crop health management that can be used to supplement existing methods? Although both biological control and host plant resistance are regarded conventionally as being more appropriate for use against direct pests, rather than virus vectors, is there scope for their deployment?

(6) Where are successes currently being realized in CMD and/or SPVD management and how can experience from these situations be incorporated into research and extension efforts elsewhere?

The second phase of the TWF-IPM Project proposes to address key research and implementation issues such as those indicated above. In the formulation of this vitally important phase of the project, attention needs to be paid to collaborative mechanisms in addition to technical issues; the first phase, described in this volume, has provided some important lessons in this regard.

(1) The coordination structure developed during the first phase has provided an effective means by which a global-scale project can be executed. Effective interaction, facilitated by electronic communication, is vital for the efficient working of this structure.

(2) At the outset of the next phase of the project, all partners at sub-project level should participate in an introductory meeting, during which opportunities are provided for critical review of the proposed work plans and budgets, and for training in additional skills required for project implementation. This was not done during the first phase and, as a result, it was difficult to consistently train partners in national research systems and to provide all partners with an appreciation of the continental and global contexts of the project.

(3) The sub-project should budget for annual review meetings for all principal partners. If resources are limited, the number of partners should be restricted to allow full participation of each.

(4) If a large number of national research system partners are to be included, as was the case in the first phase of Sub-Project 4, the task of co-ordination and provision of research support, executed by the co-ordinating international centre, should be divided between major regions covered. In the first phase of Sub-Project 4, the International Institute of Tropical Agriculture (IITA) provided co-ordination and support through

offices in both West and East Africa. This division of responsibility was considered to be a key factor in the successful implementation of the sub-project although such a division demands an effective exchange of ideas and information between the regions.

Whiteflies continue to pose a tremendous challenge to farmers, researchers and development workers in Africa, as well as in the wider global context. However, significant successes are being realized in meeting this challenge both in terms of characterizing the problems and in developing strategies to address them in a sustainable manner. In the African context, at stake are the livelihoods of many millions of producers, who depend on cassava and sweetpotato as sources of cash income, and consumers, both rural and urban, who depend on these crops to provide affordable staple foods. It is to be hoped, therefore, that initiatives to strengthen management of whitefly-transmitted virus diseases of these two major food crops, including the second phase of the TWF-IPM Project, will be able to attract a level of support commensurate with the magnitude of the problem.

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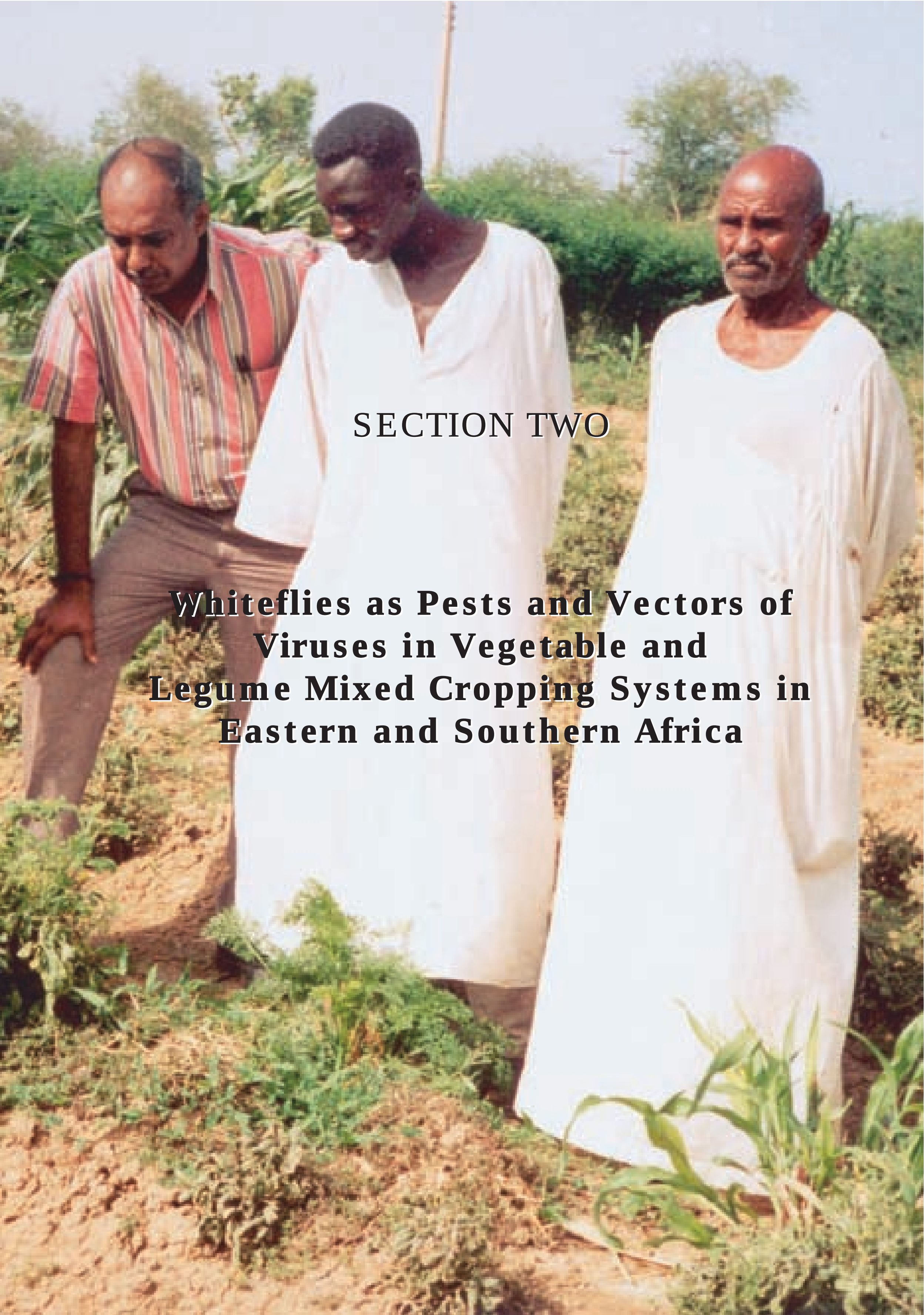

SECTION TWO

Whiteflies as Pests and Vectors of Viruses in Vegetable and Legume Mixed Cropping Systems in Eastern and Southern Africa

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CHAPTER 2.1

Introduction

Mohamed Ali Bob, *Rebbeca Raini* and
*Bernhard Löhr**

In some parts of Africa such as in the Sudan and Tanzania the countryside resembles a graveyard, not through the ravages of civil war or natural disaster but because of one pest, the whitefly *Bemisia tabaci* (Gennadius), which is sucking the life out of Africa's crops. *Bemisia*, which was originally only a problem in industrial crops such as tobacco (*Nicotiana tabacum* L.) and cotton (*Gossypium hirsutum* L.) is now much more widespread and despite all efforts to control this pest, it has stayed one step ahead of man's actions against it. *Bemisia* has done this not only by developing resistance to many of the chemicals commonly used but also by diversifying its tastes and its habitat to a much wider range of staple crops such as cassava (*Manihot esculenta* Crantz) and common bean (*Phaseolus vulgaris* L.) to high-value horticultural crops such as tomato (*Lycopersicon esculentum* Mill.) and melon (*Cucumis melo* L.). On-farm surveys in the region have shown that the number of producers who have abandoned their crops because of tomato leaf curl diseases is on a sharp increase. In the Sudan especially, whiteflies have forced entire areas out of production, depriving producers of food and cash income.

Selection of Target Crops and Target Countries

Tomato is one of the most widely cultivated vegetable crops in the region, predominantly grown by small-scale producers for fresh consumption (GTZ, 1995). National scientists have ranked *B. tabaci* as the most important pest/ vector in tomato in Tanzania and Kenya, and the second most important in Malawi after red spider mite (Varela and Pekke, 1995). In the Sudan, *B. tabaci* is considered the crop pest of highest economic importance (Eveleens, 1983; Ahmed et al., 1987).

Knowledge Available on Whitefly-Related Problems

Despite these opinions, no quantitative documentation was available on the economic importance of whiteflies as pests and/or vectors in vegetable cropping systems in the countries of this sub-project prior to its commencement. There was no clear map of the areas affected in these countries and related information on insecticide use and the cost of control in the affected areas. Knowledge on whitefly-transmitted viruses in vegetables, the host range and natural enemies was also lacking in most partner countries before this project began. Information on the status of whitefly resistance to insecticides was

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available only from the Sudan (see Chapter 2.2).

Although the history of whitefly research in the Sudan goes back to the 1930s (Kirkpatrick, 1931), a substantial collection of grey literature on whitefly and whitefly-transmitted viruses from local stations/project reports in the Sudan revealed that the research has focused almost entirely on whitefly-related problems in cotton production. Whitefly-related problems in vegetable production appear more recent, for example, *Tomato yellow leaf curl virus* (TYLCV) was first identified in 1965 (Yassin and Nour, 1965). Nevertheless, research on whitefly-related problems in vegetable crops has been limited.

In Tanzania, TYLCV was first identified in 1990 (Czosnek et al., 1990). Later, Chiang et al. (1997) identified a different virus, named ToLCV-Tz, which has complicated the scenario in Tanzania. No further documentation on whitefly-related problems in vegetable crops is available from this country. In Kenya and Malawi, documentation on whitefly-related problems is only available on citrus (*Citrus* spp. L.) and cassava.

Planning and Co-ordination

In order to form the basis for the diagnostic activities for the first phase, a planning and methodology workshop was held, with the participation of scientists from the national teams, at the International Center of Insect Physiology and Ecology (ICIPE) in April 1997. A detailed work plan for the first phase of the project was developed, methodologies were discussed and views and experiences were shared among the participants.

A progress meeting held at the ICIPE followed this up in June 1998. Progress of the two African sub-projects was presented, experiences shared and priority research areas for Phase 2 were considered. The participating co-ordinators and national teams made 10 presentations on whitefly research.

Finally, a research agenda for Phase 2 was outlined at a joint planning workshop held at the ICIPE in September 1999. Research priorities were thoroughly discussed and priority setting was carried out with full participation of all national scientists of Sub-Project 4, co-ordinators and collaborators from advanced research institutions.

Continent-Wide Appeal

Additionally, the formation of an African Whitefly and Geminivirus Network was spearheaded by the sub-project at the 12th Meeting of the African Association of Insect Scientists held in June 1997 in Stellenbosch, South Africa. Thirty-two researchers from 12 countries in Africa participated and expressed interest in participation in a regional information exchange network led by ICIPE.

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CHAPTER 2.2

Sudan

*Gassim Dafalla** and *Musa Ahmed***

Introduction

Geographical context

Diagnostic surveys were conducted during 1997-99 following the methodology agreed among project partners, and covering Gezira, Butana, Dindir, Hasaheisa, Kamlin, Managil, Umulgra, White Nile, El Jabalain, Um Rawaba, Er Rahad, Dongola, Merewe, Gedaref and Nahr el Rahad in central, eastern, northern and western Sudan (Figure 1). Most of the surveyed sites are low lying (< 500 m above sea level). The climate is hot and dry for most of the year, with temperatures in summer reaching more than 40 °C and relative humidity less than 10%, and irrigation is commonly used. The driest of the survey areas are Dongola and Merewe in the Northern State. Gedaref region and some areas in Kordufan Province are at a somewhat higher altitude (> 500 m above sea level) and receive enough rainfall for rain-fed vegetable production.

Figure 1. Areas surveyed for whitefly incidence in the Sudan.

The emergence of **Bemisia tabaci** *as a pest and virus vector*

The whitefly species *Bemisia tabaci* (Gennadius) is considered to be the economically most important crop pest in the Sudan (Eveleens, 1983; Ahmed et al., 1987). In addition to the direct damage this whitefly causes through feeding, it disseminates a number of viruses that affect fibre and vegetable crops grown in the country.

Research on *B. tabaci* and whitefly-transmitted viruses (WTVs) in Sudan started in the late1920s, when recurrent outbreaks of C*otton leaf curl virus* (CLCuV), transmitted by *B. tabaci*, threatened to end cultivation of cotton (*Gossypium hirsutum* L.) in the Gezira (Kirkpatrick, 1931). Since then, a considerable research effort has been focused on *B. tabaci* and WTVs in

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cotton, leading to the introduction of CLCuV-resistant varieties in the Gezira in the late 1960s. However, research on whitefly-related problems in vegetable crops has been limited. Vegetables are severely affected by WTVs and the incidence of WTVs in a given season constitutes the major factor determining the success or failure of tomato (*Lycopersicon esculentum* Mill.), cucurbits, okra (*Abelmoschus esculentus* [L.] Moench), pepper (*Capsicum annuum* L.) and common bean (*Phaseolus vulgaris* L.) in that season. It is estimated that, on average, 75% of the fruit yield of tomato, watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai), musk melon (*Cucumis melo* L.) and okra is lost each year to WTVs.

Yassin and Nour (1965) first reported *Tomato leaf curl virus*, transmitted by *B. tabaci*. Later investigations showed that the virus from Sudan is similar to *Tomato yellow leaf curl virus* (TYLCV) from the East Mediterranean region. In recent years, severe yellowing symptoms were observed in watermelon and melon grown in many parts of the country, similar to those described for the *B. tabaci*-transmitted *Watermelon chlorotic stunt virus* (WmCSV) in Yemen. CLCuV not only affects cotton but also has been reported by many workers to cause heavy damage in okra in most parts of the country. Both cotton and okra belong to the mallow family. According to some workers (cited by Dafalla and Sidig, 1997) the whitefly-transmitted African cassava mosaic disease is still the most destructive in cassava in Western Equatoria Province in southern Sudan.

At producer level, research efforts have not been reflected sufficiently in the successful management of *B. tabaci* and WTVs, especially in vegetables. In the absence of such management strategies, the problem has become more complicated and has defied vegetable producers' efforts to overcome it and secure a return for their efforts. The need to design viable integrated control strategies, based on a strong scientific foundation and knowledge of the field situation, has emerged as a major challenge for research.

Little information was available on the whitefly-related problems of vegetable production in Sudan prior to the commencement of the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project. Many findings from the survey conducted in the first, diagnostic phase of the project are new and add substantially to our knowledge base on biological and socio-economic aspects of the whitefly and WTV problems of vegetable cropping systems in Sudan.

Increased Biological Understanding

Characterization of begomoviruses and whitefly biotypes

Samples of whitefly adults and nymphs were collected mostly from tomato, with a few from lablab (*Lablab purpureus* [L] Sweet) and Nalta jute (*Corchorus olitorius* L.), on 80 farms. One hundred and fifty specimens were processed and mounted on 76 slides and sent to the International Center of Insect Physiology and Ecology (ICIPE) for identification. All the mounted nymph specimens were identified as *B. tabaci*.

In an experimental field at the University of Gezira, severe silver leaf symptoms were noticed on squash (*Cucurbita pepo* L.), heavily infested with whiteflies. These symptoms have been associated elsewhere with the

B. tabaci B-biotype (Shapiro, 1995) but molecular characterization of these whitefly specimens is still pending. In general, some variability has been noticed in the patterns of behaviour and efficiency in virus transmission, which also may be circumstantial evidence of the existence of different *Bemisia* biotypes.

Host plants on which whiteflies were found to reproduce, identified in the course of surveys in Sudan, belonged to the following families: *Amaranthaceae*, *Asclepiadaceae*, *Asteraceae*, *Brassicaceae*, *Commelinaceae*, *Convolvulaceae*, *Cucurbitaceae*, *Euphorbiaceae*, *Lamiaceae*, *Leguminosae*, *Malvaceae*, *Solanaceae*, *Tiliaceae* and *Verbenaceae* (Table 1).

Many of the WTVs in vegetables in Sudan have been well characterized but more information on the inter-seasonal variation of virus incidence and on the distribution and economic importance of various WTVs in the different regions is needed. The field surveys revealed that TYLCV, WmCSV, *Okra leaf curl virus* and *Pepper leaf curl virus* are the most widespread and economically important WTVs in Sudan. Tomato, watermelon, melon, okra, pepper and common bean were among the most affected crops.

Other viruses detected include Tomato vein thickening virus, which causes another important disease of tomato in Sudan and *Bean mild mosaic virus* (BMMV). *Cucumber vein yellowing virus* (CVYV) and a virus inducing potyvirus-like yellowing have been observed in cucurbits but the latter is yet to be identified. In legumes, a geminivirus and a *Closterovirus* that affect cowpea (*Vigna unguiculata* [L.] Walp.) and common bean have been observed and are yet to be identified. Symptoms of another putative WTV, whose identity is yet to be established, were also noticed on pepper.

Some weeds, *Acalypha indica* L., *Datura stramonium* L. and *Solanum coagulans* Forskål, were identified as TYLCV reservoirs. Many other non-cultivated host plants of *B. tabaci* may act as WTV reservoirs. Continuous cultivation of tomato and other solanaceous vegetables, coupled with the abundance of alternative hosts for vector and viruses, is very likely to contribute to the build up of disease problems.

Disease incidence and symptom severity

The Gezira Irrigation Scheme is the area with the most severe and persistent WTV disease problems in the country. Incidences of 80-100% were frequently recorded in Gezira, Hasaheisa, Managil, Umelgura, Dindir and Butana. In half of the tomato fields in the Gezira Province, 100% TYLCV incidence was recorded. *B. tabaci* was found to be more abundant in the southern part of Gezira that receives less rainfall than the northern region. High whitefly populations and TYLCV incidence were observed in Central Sudan, significantly lower populations and TYLCV incidences (1%-9%) were observed in Nahr el Rahad (Eastern State), Dongola and Merewe (Northern State), and in Umm Rawaba and Er Rahad (Western State). No TYLCV symptoms were observed in Gedaref Province (Babiker, Khor-Garab, El Ramla, Basunda, Doka) and the Abu Habil Basin. Vegetable gardens along the Blue Nile River were once the sole suppliers of tomato outside the period suitable for rain-fed cultivation. This is no longer the case because of severe TYLCV epidemics in recent years.

The overall impression provided by the surveys is that *B. tabaci* does not

Table 1. Reproductive host plants of *Bemisia tabaci* (Gennadius) in the Sudan.

Family	Species	Common name
Crops		
Brassicaceae	*Raphanus sativus* L.	Radish
Cucurbitaceae	*Citrullus lanatus* (Thunb.) Matsum & Nakai	Watermelon
	Cucumis melo L.	Melon
	Cucumis sativus L.	Cucumber
	Cucurbita pepo L.	Squash
Euphorbiaceae	*Manihot esculenta* Crantz	Cassava
Leguminosae	*Cajanus cajan* (L.) Millsp.	Pigeon pea
	Cassia alexandrina Mill.	Senna
	Lablab purpureus (L.) Sweet	Lablab
	Lens culinaris Medik. subsp. *culinaris*	Lentil
	Phaseolus vulgaris L.	Common bean
	Vicia faba L.	Broad bean
Lamiaceae	*Ocimum basilicum* L.	Basil
Malvaceae	*Abelmoschus esculentus* (L.) Moench	Okra
	Gossypium hirsutum L.	Cotton
	Hibiscus cannabinus L.	Kenaf
Solanaceae	*Capsicum annuum* L.	Pepper
	Lycopersicon esculentum Mill.	Tomato
Tiliaceae	*Corchorus olitorius* L.	Nalta jute
Non-cultivated hosts		
Amaranthaceae	*Achyranthes aspera* L.	
	Amaranthus tricolor L.	
Asclepiadaceae	*Leptadenia* sp.	
Asteraceae	*Ageratum conyzoides* L.	
	Bidens pilosa L.	
	Sonchus sp.	
Commelinaceae	*Commelina benghalensis* L.	
Convolvulaceae	*Ipomoea cordofana* Choisy	
Euphorbiaceae	*Acalypha indica* L.	
	Euphorbia aegyptiaca Boiss.	
	Euphorbia heterophylla L.	
Leguminosae	*Rhynchosia hirta* (Andrews) Meikle & Verdc.	
Malvaceae	*Abutilon pannosum* (G. Foster) Schltdl.	
Solanaceae	*Datura stramonium* L.	
	Solanum incancum L.	
	Solanum nigrum L.	
Verbenaceae	*Lantana camara* L.	

cause major problems outside of areas where cotton is cultivated. For example, in the Abu Habil Basin, where no cotton is grown, tomato is typically produced without whitefly infestation. Similarly, OLCV is present in many parts of the country where cotton is grown, whereas WmCSV is present in almost all melon production areas.

Natural enemy species

Seventeen collections of whitefly parasitoids and predators were made and preliminary identification made. The most common parasitoids of *B. tabaci* were found to be *Encarsia lutea* (Masi) and *Eretmocerus mundus* Mercet. The following natural enemies that were found in the vegetable-based systems in the country also could be involved in regulating *B. tabaci* populations: the coccinellids *Coccinella undecimpunctata* L., *Hippodamia variegata* (Goeze), *Cheilomenes sulphurea* (Olivier) and *Scymnus* sp.; the lacewings *Chrysoperla pudica* (Navás) and *Chrysoperla* spp.; pirate bugs, *Orius* sp.; other predatory true bugs *Campylomma* sp. and spiders.

Increased Socio-economic Understanding

Farmers' assessment of whitefly-related problems

More than 95% of vegetable production in the surveyed areas is carried out on small-scale holdings of 2 to 5 ha. Among tomato producers in Sudan, 40% farmed their own land, 38% used rented land, while 22% were squatters. Virtually all farmers interviewed (99%) were men. Almost all the producers interviewed (94%) regarded tomato as the most profitable of the horticultural crops they could grow and most (74%) have cultivated tomato for more than 5 years. The varieties of tomato most widely grown in Sudan were Moneymaker, Strain B, Peto 86, Pearson, Early pack and Ace. Most producers (65%) bought their tomato seed from the local market, while 28% used their own seed and 7% imported seed from other countries. Other commonly grown vegetable crops include cucurbits (especially melon and cucumber), okra, common bean, pepper, eggplant (*Solanum melongena* L.), sweetpotato (*Ipomoea batatas* [L.] Lam.), radish (*Raphanus sativus* L.), onion (*Allium cepa* L.), carrot (*Daucus carota* L. subsp. *sativus* [Hoffm.] Arcang. var. *sativus* Hoffm.) and leafy vegetables. Most producers (82%) practised crop rotation.

Whiteflies and/or WTVs were found to be the greatest cause of concern to most tomato producers in the survey. Other destructive pests reported were *Helicoverpa armigera* (Hübner), *Liriomyza sativae* Blanchard, *Scrobipalpa* sp. and *Keiferia lycopersicella* (Walshingham).

Most tomato producers in Sudan (75% overall, including all those in Central Sudan) were able to recognize whiteflies and even more (89%) recognized TYLCV. However, 65% of them did not know that TYLCV and *B. tabaci* were inter-related. The majority (84%) believed that whiteflies and/or TYLCV were serious production constraints in their farms. Whereas 65% of the producers reported that TYLCV was a problem, only 35% reported that both the whitefly and the disease were problematic, and none reported a problem with whitefly alone. In contrast, most producers in Gedaref, Northern and North Kordufan Provinces, where TYLCV incidence was very low (0%-9%), could not recognize whiteflies or TYLCV nor did they have local names for them.

All producers who recognized whiteflies and TYLCV had names for both and in most cases the producers gave very specific names for the problems they cause. Local names for whiteflies include *asala*, *biadah*, *dubbana*, *dubbana beida*, *zubaba* and *zubaba beida*. These literally mean "flies" or "white flies", except *asala*, which refers to the honeydew that the insects secrete. Names given to the TYLC include *hurug* (burning), *saratan* (cancer) and *karmata*, *karmasha* or *kurmut* (all of which allude to leaf deformation).

Historically, Sudan has been regarded as one of the countries worst hit by whitefly problems, especially in the cotton production system. This scenario has changed recently and the whitefly problem in cotton seems to have become less acute. Highly effective insecticide treatments, often containing insect growth regulators, have reduced the breeding success of whiteflies on cotton. Nonetheless, the whitefly has maintained its status as a highly injurious pest of tomato, cucurbits, pepper, okra and common bean, and the numbers of producers who have abandoned tomato cultivation because of TYLCV are on the increase.

Farmers' perceived yield loss to whitefly and/or TYLCV in tomato was on average 62%. According to the survey, 26% of producers, mainly in Gezira, reported total yield loss, 38% reported losing three-quarters of their yield, 10% reported losing half, 10% reported losing one-quarter of their yield and 16%, mainly from Gedaref and North Kordufan, reported no loss. Of the producers interviewed, 34% (most of them in Gezira State) reported abandoning their tomato crops in at least 1 year because of whitefly/TYLVC problems and most reported that this occurred in 1997. The perceived losses can be compared usefully with the recorded incidence of TYLCV symptoms. Among all surveyed tomato producers, 92% had some virus symptoms in their tomato crop and 50% had more than 25% incidence. It should be emphasized that 31% of the tomato producers had an 80% TYLC incidence in their tomato crops and these outbreaks were mainly confined to the greater Gezira. TYLC was significantly less severe (1%-9%) in western, northern and eastern states, and absent in Gedaref and the Abu Habil Basin. Most tomato producers (65%) believe that they have whitefly/TYLCV problems every year. The majority of producers (86%) believe that there is a direct relationship between the climate and the incidence of whiteflies and/or whitefly-transmitted disease, with 63% of producers associating high incidence with hot and dry seasons, while only 9% believe that they have the problem all year round.

Estimation of disease incidence and yield losses

The damage associated with *B. tabaci* and WTVs is always more serious in summer crops, especially during the hot season from March to August (during which there are three planting periods: "early summer" in March, "late summer" in May and the "main planting season" in July) and total failure of the tomato crop due to TYLCV is common. This is despite the lower prevalence of the vector in the summer season. Whitefly populations build up during the winter (October-January) and peak in December-January. Up to 100% infection by TYLCV can occur during a mild winter but less yield depression is associated with these winter infections.

In 1998, tomato grown during the May-July season suffered severe attack by TYLCV and this crop failed completely. Watermelon likewise suffered severe losses from WmCSV. An exceptionally heavy rainy season from mid-August to mid-October, however, seemed to alter the whitefly situation later in the year, with population levels dropping sharply and tomato crops grown during October-November being comparatively less affected by TYLCV. A field experiment conducted at the University of Gezira revealed that early infection by TYLCV reduced the total number of flowers per plant, percent fruit setting and the quality of fruits. The infection resulted in 67% reduction in the number of mature fruits and 84% reduction in the weight of marketable fruits per plant. Severe infection resulted in 86% reduction in the number of green mature fruits, 87% reduction in the number of ripe fruits and 93% reduction in the weight of marketable fruits per plant. Flower bud initiation substantially decreased as the infection became more severe or when infection occurred early in the plant's growth cycle but the proportion of flower abortion and fruit setting was not significantly affected. Affected plants had reduced foliage cover and fruits were exposed to direct sunlight, which caused white blotches (sunscald) and reduced their market value. The results show that TYLCV infection can affect all yield components, from flower production through to the quantity and quality of fruits.

WmCSV is less important economically than TYLCV but is considered to be the most important single cause of high yield loss of cucurbits in eastern and central Sudan and has affected the country's melon exports seriously in recent years. Crops affected include watermelon, musk melon and, to a lesser extent, snake melon (*Cucumis melo* L. subsp. *melo* var. *flexuosus* [L.] Naudin) and squash. Melon producers incur a 64% loss in revenue because of WmCSV. The virus is prevalent in Gezira, Upper Atbara River and southern Sinnar, where fresh market varieties such as Charleston Grey, Congo Red and Sugar Baby are grown. Watermelon in large areas of Southern Blue Nile, Shuwak and Dinder inland delta were totally devastated by WmCSV in1998-99, and up to 80% yield losses occurred in Galia melon (a variety of musk melon) grown for export in Khartoum and Gezira States. However, this virus is only sporadically important in Kordufan and western Sudan where local strains of watermelon are grown for seed production and water storage. WmCSV incidence of 82% was observed on Galia melon grown for commercial purposes at the Gezira University farm in 1997. Complete absence of "netting" on the fruit surface was evident in some severely affected plants, while the healthy ones produced fruits entirely covered with netting (an important quality that correlates positively with sweetness of the fruit). The severely affected plants gave 1.3 fruits per plant, while healthy plants gave 2.1 fruits per plant. The average incidence of WmCSV in Galia melon from eight sites in the Selait Irrigation Scheme (Kordufan Province) was 70% and data from these sites indicated a positive correlation between the level of virus incidence and yield loss.

OLCV restricts okra production in Sudan. Symptoms of the virus appeared to be more severe during the late winter and early summer growing seasons, with Gezira being the worst hit area. The virus had less effect on local varieties of okra. BMMV may constitute a potential hazard, especially in the newly established export-oriented vegetable production area in Khartoum State. This virus is less important and is considered a late season disease in northern Sudan.

Pesticide use

The cost of controlling whiteflies and WTVs accounts for more than 30% of the total production costs of tomato in Sudan. A small proportion of producers (4%) spent as much as US$300-400 per hectare on pesticide treatment. Other producers estimated their pest control costs at US$200-299 per hectare (14% of producers), US$100-199 per hectare (21% of producers), US$50-99 per hectare (27% of producers) and US$0-49 per hectare (34% of producers). The costs of chemicals and their application vary greatly among sites.

Only 10% of producers reported receiving technical advice or information regarding the management of pest and disease problems. Forty percent of producers received advice from other vegetable producers or neighbours, 30% from commercial sales agents, while 20% had not received any advice but applied insecticides according to their own judgement. The absence of an effective extension service has left the farmers to make their own decisions on whitefly/WTV management. The constraints on farmers' knowledge of whitefly management can be summarized as:

(1) Low level of education and limited knowledge of modern techniques of vegetable production;

(2) Poor ability to choose the right crop and a suitable variety of that crop;

(3) Poor knowledge on pesticide usage and related hazards; and

(4) Inability to monitor market trends as a basis for deciding what to grow and when to grow it.

Because of the lack of extension services in vegetable farming, chemical control of the whitefly remains the principal pest management strategy. The great majority of tomato producers in the country (80%) used insecticides, mostly pyrethroids and organophosphates, to combat the whitefly/TYLCV problem on their farms. The most popular pesticides applied by the producers were fenpropathrin, fenvalerate and cypermethrin (pyrethroids), omethoate, malathion, dimethoate and chlorpyriphos (organophosphates), methomyl and carbaryl (carbamates), and endosulfan (organochlorine).

Most producers believed that the pesticides were indispensable in tomato farming. A significant proportion of producers (30%) made more than 10 insecticide applications per season, 26% made 9 or 10 applications, and a few producers applied pesticides as many as 24 to 30 times per season. Ten percent of producers practiced crop rotation to combat the problem. A few farmers used cultural control methods such as burying of infected plants and inter-cropping with repellent crops such as coriander (*Coriandrum sativum* L.) and fenugreek (*Trigonella foenum-graecum* L.). Others claimed that directly sown tomato, that is, without transplanting, was less affected by TYLCV. Ten percent of producers, mainly in Gedaref, Western State and Northern State did not practice any sort of management of the problem.

Lack of knowledge of the proper use of pesticides was evident in all areas surveyed. Farmers were found to be ignorant of the judicious use of pesticides and alternative whitefly control methods. Some tomato growers overused chemicals, while others did not spray until symptoms of damage were observed, and subsequent intensive chemical application failed to save the crop. Producers were ignorant of the interval required between the last spraying and harvest, and some even used the taste or smell of the pesticides to judge their effectiveness. Lack of protective clothing and equipment exposed the farmers to serious health hazards. Inappropriate chemicals such as fungicides (propiconzale) and others without any labels were being used in an attempt to control the whitefly and WTV problem. Recommended dosages were not followed and virtually all vegetable producers mixed one tomato sauce tin, containing about 50 mL, of any insecticide, irrespective of its formulation or active ingredient, with 4 gallons (18 L) of water. A few producers used twigs to apply pesticide, instead of a knapsack sprayer, resulting in poor coverage and greater loss of pesticides. Most producers (67%) applied insecticides as a preventive measure, 17% applied insecticides when they observed whitefly/TYLCV damage, while 8% applied insecticides according to calendar.

Vegetable growers in Gezira and Rahad illegally obtain part of their supply of chemicals from the stocks of cotton schemes. These include highly toxic "cocktails" of insecticide, exclusively recommended for use on cotton. Their experience of severe whitefly outbreaks in previous seasons leads most of them to use excessive quantities of these chemicals. As a result, environmental contamination, occupational health hazards and production costs are on the increase. In addition, the overuse of chemicals to control whitefly seemed to have resulted in the build-up of previously secondary pests such as *H. armigera*, *L. sativae* and *Scrobipalpa* sp. Nevertheless, several producers suffered complete loss of even the treated crop.

Several researchers (e.g., Ahmed et al., 1987; Dittrich et al., 1990) have documented the resistance status of *B. tabaci* in cotton to various insecticides in the Sudan. They reported that *B. tabaci* showed high levels of resistance to many organophosphates and synthetic pyrethroids and moderate resistance to carbamates and organochlorines. The specific levels of resistance reported vary depending on the source of the samples, the history of insecticide application in that area, the time of the year when bioassays were done and the sensitivity of the whitefly population used. However, the reports were primarily based on dose responses of whiteflies in the cotton monoculture system. Pesticide resistance of whiteflies in the mixed vegetable cropping systems in the Sudan has not been documented.

Sudan is the only country in East Africa that has adopted IPM as its official crop protection policy. The government's position is well defined in a recent publication entitled "Sudan Country Strategy Note, 1997-2001: Partnership towards Sustainable Human Development". The document emphasizes that the government of the Sudan is pursuing an integrated program for environmentally sustainable development. However, implementation of the policy is minimal, particularly, in the case of vegetable farming, because of farmers' ignorance of alternatives to chemical control, which in turn is mainly attributed to the lack of effective extension services in this sector. In the absence of alternatives, chemical control of the whitefly therefore remains the principal strategy for managing WTVs.

Strengthened Research Capacity

Sudan's participation in the diagnostic phase of the TWF-IPM Project has provided a substantial body of knowledge covering various aspects, biological and socio-economic, of the whitefly/WTV problems in vegetable-based cropping systems in the country. Sudan's participation in the planning workshop in April 1997 and the co-ordination meeting in June 1998 provided national researchers with a chance to learn from, and share ideas and experiences with, scientists from other countries. A Ph.D. project on "Distribution of whitefly species (and biotypes) and their natural enemies in vegetable-based cropping systems in Sudan" was undertaken in collaboration with the University of Gezira under the auspices of the project.

Conclusions

The work conducted during the diagnostic phase of the TWF-IPM Project has provided, for the first time, a biological and socio-economic characterization of the problems linked to *B. tabaci* in the vegetable cropping systems of Sudan. The studies have provided a better understanding of the biology of this important vector, the distribution of its natural enemies, the epidemiology of the viruses it transmits and the socio-economic implications of virus disease problems for producers.

Farmers viewed the *B. tabaci*/TYLCV problem as the most important constraint on tomato production. The areas identified as hot spots for TYLCV include Gezira, Managil, Umelgura and Hasaheisa Provinces, all of which are characterized by irrigated production and are within cotton production areas. The main problems associated with *B. tabaci* in vegetable production systems in Sudan are:

(1) Tomato: transmission of TYLCV and TVTV, and direct feeding by *B. tabaci* during early crop stages;

(2) Watermelon and musk melon: transmission of WmCSV, CVYV, and a virus inducing potyvirus-like yellowing symptoms;

(3) Okra: transmission of OLCV, sooty mould induced by secretion of honeydew by *B. tabaci*, and direct feeding damage;

(4) Peppers: transmission of TYLCV; and

(5) Vegetable legumes: symptoms of a begomovirus and closterovirus observed on cowpea and common bean but transmission by *B. tabaci* is not yet confirmed.

The surveys documented alarming misuse of pesticides on vegetables. The information gained can help to sensitize policy makers in Sudan to the alarming magnitude of the whitefly and WTV problems and to the dangers inherent in the associated misuse of pesticides. The knowledge gained has provided a sound foundation for developing a future research agenda. The following priorities are recommended:

(1) Research should be conducted on the following IPM components:

  (a) Evaluation of available TYLCV-resistant or -tolerant tomato varieties for immediate incorporation into IPM plans;

  (b) Screening of exotic tomato varieties for resistance to *B. tabaci* and/or TYLCV;

  (c) Breeding for resistance against *B. tabaci* and WTVs, while utilizing resistant strains of crops;

  (d) Evaluation of alternative and novel pest and disease management methods, including the use of botanicals, biological control and cultural methods; and

  (e) Investigation of the efficiency of natural enemies in vector control.

(2) Available IPM options should be tested and refined, with the close participation of producers, under on-farm conditions; participatory research should be complemented with intensive training of producers and other stakeholders.

(3) Resistance of whiteflies to insecticides commonly used in vegetable farming should be

monitored, using a standardized methodology in Sudan and elsewhere to enable valid comparisons to be made.

(4) Appropriate techniques to evaluate insecticide residues in fruits and vegetables should be developed and applied.

(5) The whiteflies observed inducing silverleaf in Gezira for the first time in 1998 should be characterized using molecular techniques and biological assays.

The recommended research will provide a platform for the development and adoption of sustainable production practices for vegetables in Sudan, leading to improved well-being for producers and consumers.

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CHAPTER 2.3

Kenya

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Introduction

Geographical context

Field surveys covering 94 farms were conducted in 1997-98 following the methodology agreed among project partners. The farms represented: Kirinyaga, Thika, Muranga, Kiambu, and Nairobi Districts in Central Province; Machakos, Athi River, Meru, Kitui, and Kibwezi Districts in Eastern Province; Kwale, Kilifi, and Taita Taveta Districts in Coast Province; Kajiado and Naivasha Districts in Rift Valley Province; Kisii, Migori, Homa Bay, and Siaya Districts in Nyanza Province; and Vihiga and Busia Districts in Western Province (Figure 1). The districts were selected based on the prevalence of the cultivation of tomato (*Lycopersicon esculentum* Mill.). Table 1 gives the mean annual temperatures and precipitation for each of the surveyed regions.

In the surveyed regions, tomato is planted throughout the year with the exceptions noted below. Eastern Province is a region of semi-arid lower mid-altitude land becoming transitional semi-humid towards Central Province; here, tomato is not planted in July.

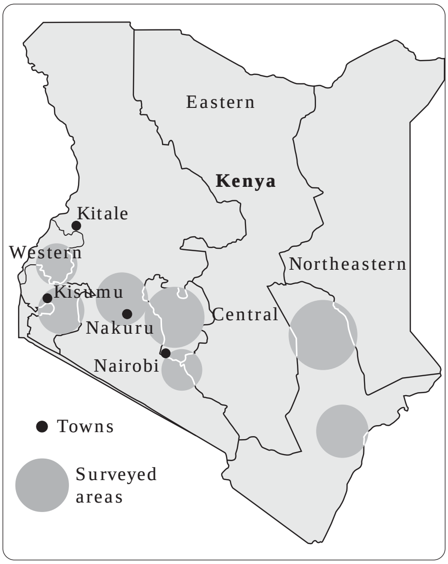

Figure 1. Tomato-growing areas surveyed for whitefly incidence in Kenya.

Table 1. Mean annual temperatures and precipitation for the surveyed regions of Kenya.

Region / Province	Mean min.-max. temperature (°C)	Mean precipitation (mm)
Eastern	15-26	728
Rift Valley	14-27	591
Central	13-25	1166
Coast	21-30	872
Western and Nyanza	16-29	1289

Whitefly surveys took place between November and March of 1997-98 and 1998-99. Rift Valley Province consists

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of transitional lower to higher mid-altitude land. In the surveyed areas of Kajiado and Naivasha Districts in the Rift Valley, tomato is not planted in January and July. Whitefly surveys were carried out in September 1998 and March 1999. Central Province consists of sub-humid highlands; whitefly surveys were carried out here between November and March in 1997-98 and in 1998-99. In the Coast Province, a region of sub-humid coastal lowlands, tomato is not planted from November to January. Whitefly surveys were carried out in July 1998. In Western and Nyanza Provinces (sub-humid to humid upper mid-altitude land and lower highlands) tomato is not planted in January. Whitefly surveys in this region were carried out in February and November 1998.

El Niño rains devastated tomato production in Kenya from September 1997 to June 1998. Whiteflies were scarce during this period.

The emergence of **Bemisia tabaci** *as a pest and virus vector*

Tomato is one of the most widely cultivated vegetable crops in East Africa, predominantly grown by small-scale producers for fresh consumption (GTZ, 1995), while in Kenya in particular, vegetable-growing both for local consumption and for export provides an increasingly important source of income to farmers. In 1995, Kenyan national scientists participating in a regional workshop ranked the whitefly *Bemisia tabaci* (Gennadius) as the most important pest/vector in tomato (Varela and Pekke, 1995). This whitefly has become an important production constraint to crops of the families *Solanaceae*, *Leguminosae*, *Malvaceae*, *Cucurbitaceae* and *Euphorbiaceae*, particularly in the semi-arid Eastern Province of Kenya. Extension workers and scientists have reported informally the occurrence of several whitefly-transmitted viruses: *Tomato yellow leaf curl virus* (TYLCV), *Cowpea mild mottle virus* (CPMMV), *Sweetpotato mild mottle virus* (SPMMV) and *Watermelon chlorotic stunt virus* (WmCSV).

Despite the perceived importance of these horticultural crops and their virus disease problems in Kenya, previously no systematic attempt has been made to document their distribution and incidence. Virtually all work related to whiteflies conducted in Kenya has focused on cassava mosaic disease (CMD), its prevalence in the country, its temporal and spatial distribution and associated yield loss (Seif, 1982; Bock, 1987). Some research also had been conducted on the citrus woolly whitefly *Aleurothrixus floccosus* (Maskell), which is reported to have become a common pest of citrus (*Citrus* spp. L.) as well as coffee (*Coffea arabica* L.) and guava (*Psidium guajava* L.) in Kenya (Löhr, 1997). In addition, the whitefly parasitoids, *Eretmocerus mundus* Mercet. and *Encarsia sublutea* (Silvestri), have been recorded in Kenya (Dr. Abdurabi Seif, personal communication, 1998). Kenya's participation in the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project provided an opportunity to gain an overview of the geographical range and economic importance of whiteflies, whitefly-transmitted viruses and whitefly natural enemies in the country—information that would serve as a sound basis for planning further research and pest management efforts.

Increased Biological Understanding

Characterization of begomoviruses and whitefly biotypes

During the surveys in Kenya, 325 collections of whitefly adults and nymphs were made. From these, 549 specimens of whitefly nymphs were mounted on 310 slides and identified to species level. Most of the whiteflies collected were *B. tabaci* (57%), while *Trialeurodes vaporariorum* (Westwood) represented 34%. *B. afer* (Priesner and Hosney) were occasionally found (7%) in fields of cassava (*Manihot esculenta* Crantz) adjacent to tomato fields. A number of other whitefly species (2%) also were encountered, including *Trialeurodes ricini* (Misra), *A. floccosus*, *Siphoninus phillyreae* (Haliday) and *Bemisia* spp. Table 2 shows the composition of whitefly species based on the identified specimens from each Province.

Disease incidence and symptom severity

Whiteflies in the surveyed farms were found breeding on host plants that belong to the following families: *Acanthaceae*, *Amaranthaceae*, *Asteraceae*, *Commelinaceae*, *Convolvulaceae*, *Cucurbitaceae*, *Euphorbiaceae*, *Lamiaceae*, *Leguminosae*, *Malvaceae*, *Rutaceae*, *Solanaceae* and *Verbenaceae* (Table 3). Surprisingly, whiteflies were found breeding only occasionally on tomato in Kenya. In Muranga, Kiambu and Mwea Districts in Central Province, it was difficult to find a single whitefly nymph in a whole tomato field. In these areas, whiteflies, mainly *T. vaporariorum*, were found breeding on crops such as common bean (*Phaseolus vulgaris* L.) and cassava and on non-cultivated plants, most significantly *Euphorbia heterophylla* L., in all areas surveyed. However, in Kihara (Nairobi) and Kigumo (Muranga) in Central Province and in Nguruman in Rift Valley Province, whiteflies, mainly *T. vaporariorum*, were found breeding readily on tomato.

Host plant feeding preference and status as reproductive hosts (oviposition and offspring survival) of *B. tabaci* was studied in field cages at the International Center of Insect Physiology and Ecology (ICIPE), Nairobi, in 1997 using adult insects collected from a tomato field nearby. Among the four host plants studied, common bean was the most preferred host plant, followed by lablab (*Lablab purpureus* [L.] Sweet), cowpea (*Vigna unguiculata* [L.] Walp.) and okra (*Abelmoschus esculentus* [L.] Moench). There were significant differences between plants with regard to the parameters tested. These results concur with observations in the field during the survey.

Table 2. Species composition (%) of whitefly samples from surveyed regions of Kenya.

Province	Species composition[a]					
	B.t.	T.v.	B.a.	T.r.	A.f.	S.p.
Eastern	61	29	6	0	0	4
Rift Valley	20	80	0	0	0	0
Central	35	60	4	1	0	0
Coast	79	0	21	0	0	0
Western	90	0	7	3	0	0
Nyanza	79	6	5	0	10	0

[a] B.t., *Bemisia tabaci*; T.v., *Trialeurodes vaporariorum*; B.a., *Bemisia afer*; T.r., *Trialeurodes ricini*; A.f., *Aleurothrixus floccosus*; S.p., *Siphoninus phillyreae*.

Table 3. Reproductive host plants of whitefly species encountered during surveys in Kenya.

Whitefly species	Crops		Non-cultivated hosts	
	Family	Species	Family	Species
Bemisia tabaci (Gennadius)	Convolvulaceae	*Ipomoea batatas* (L.) Lam.	Acanthaceae	*Asystasia schimperi* T. Anders.
	Cucurbitaceae	*Citrullus lanatus* (Thunb.) Matsum & Nakai	Amaranthaceae	*Achyranthes aspera* L.
		Cucurbita pepo L.		*Achyranthes sicula* L.
		*Momordica char*antia L.	Asteraceae	*Ageratum conyzoides* L.
	Euphorbiaceae	*Manihot esculenta* Crantz		*Aspilia mossambicensis* (Oliv.) Wild
	Leguminosae	*Crotalaria* sp.		*Bidens pilosa* L.
		Lablab purpureus (L.) Sweet		*Emilia discifolia* (Oliv.) C. Jeffrey
		Vigna unguiculata (L.) Walp.		*Galinsoga parviflora* Cav.
		Crotalaria sp.		*Gutenbergia cordifolia* Benth. ex Oliv.
				Tithonia diversifolia (Hemsl.) A. Gray
		Phaseolus vulgaris L.	Commelinaceae	*Commelina benghalensis* L.
	Lamiaceae	*Ocimum basilicum* L.	Convolvulacea	*Ipomoea tricolor* Cav.
	Malvaceae	*Abelmoschus esculentus* (L.) Moench		*Jacquemontia tamnifolia* (L). Griseb.
		Gossypium hirsutum L.	Euphorbiaceae	*Euphorbia heterophylla* L.
	Solanaceae	*Capsicum annuum* L.	Leguminosae	*Rhynchosia hirta* (Andrews) Meikle & Verdc.
		Lycopersicon esculentum Mill.	Lamiaceae	*Leonotis nepetifolia* (L.) R. Br.
		Solanum melongena L.		*Ocimum kilimandsharicum* Guerke
			Malvaceae	*Sida acuta* Burm f.
			Solanaceae	*Datura stramonium* L.
				Nicandra physalodes (L.) Gaertn.
			Verbenaceae	*Lantana camara* L.
			Asteraceae	*Tithonia diversifolia* (Hemsl.) A. Gray
Trialeurodes vaporariorum (Westwood)	Cucurbitaceae	*Cucurbita* spp.	Euphorbiaceae	*Euphorbia heterophylla* L.
		Momordica charantia L.		
	Leguminosae	*Phaseolus vulgaris* L.		
	Malvaceae	*Abelmoschus esculentus* (L.) Moench		
	Solanaceae	*Lycopersicon esculentum* Mill.		
		Solanum melongela L.		
Bemisia afer (Priesner and Hosney)	Convolvulaceae	*Ipomoea batatas* L. Lam.		
	Leguminosae	*Phaseolus vulgaris* L.		
	Malvaceae	*Gossypium hirsutum* L.		
			Asteraceae	*Tithonia diversifolia* (Hemsl.) A. Gray
Trialeurodes ricini (Misra)	Solanaceae	*Lycopersicon esculentum* Mill.	Lamiaceae	*Leonotis nepetifolia* (L.) R. Br.
Aleurothrixus floccosus (Maskell)	Rutaceae	*Citrus* spp.		
Siphoninus phillyreae (Haliday)				Unidentified tree

Samples of 10 plants per field, on average, collected from all surveyed areas, were squashed on nylon membranes and sent to the John Innes Centre, UK, for hybridization. Hybridization signals varied from strong to very weak, probably because of differences in the degree of nucleic acid homology. This could suggest different strains of begomoviruses or different begomoviruses altogether. The signal strength also may have been influenced by the sampling method, because the virus titre may have been low in the particular tissues sampled or at that stage of infection.

At Kibwezi, begomovirus symptoms were observed on sweet pepper (*Capsicum annuum* L.) and a low incidence of WmCSV (about 5%) was recorded on watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai). A variety of mosaic symptoms that need further investigation also were noticed in pepper and watermelon. There were symptoms that suggest that tomato in this region should be tested for potyviruses such as *Potato virus Y* (PVY). Aphid infestation levels were very high at Kibwezi and some of the symptoms observed could be a result of aphid-borne viruses. High incidence and severe symptoms of CMD were observed in Mombasa and Kilifi Districts (Coast Province) and in Kitui District (Eastern Province). The symptoms appeared more severe than those observed in western Kenya. Whether these symptoms are caused by infections of *East African cassava mosaic virus* (EACMV) or are associated with the occurrence of the Ugandan variant needs to be ascertained.

Leaf curl symptoms were found during the survey on non-cultivated host plants. These were *Achyranthes aspera* L., *E. heterophylla* and *Nicandra physalodes* (L.) Gaertn. These plants are therefore likely to act as reservoirs of TYLCV. Severe begomovirus symptoms also were observed on a weed identified as *Gutenbergia cordifolia* Benth. ex Oliv. *E. heterophylla* was found to be a major reproductive host of both *B. tabaci* and *T. vaporariorum* at any site where whitefly infestation occurs and this plant species is present. Therefore, the role of *E. heterophylla* in survival of the disease and the vector outside the crop-growing season needs to be investigated.

Natural enemy species

Forty whitefly parasitoid samples were obtained from the surveys and preliminary identification was carried out. *Encarsia sophia* (Girault and Dodd) was found to be the predominant species, while *Eretmocerus* sp. was encountered in the Central Province. *Encarsia formosa* Gahan was only collected from Mwea in the Central Province. At Nguruman, Rift Valley, the level of parasitization appeared higher than at other survey sites in Kenya.

Some of the 24 predators collected were found feeding on the whitefly pupae. Most were coccinnellids, while a few were spiders and mites but none were identified to species level. *Macrolophus caliginosus* Wagner, bugs known to prey on *T. vaporariorum* in other parts of the world, were collected at several locations in Nguruman (Rift Valley Province), Kibwezi (Eastern province) and Mwea (Central Province).

Geographical range of whitefly and whitefly-transmitted virus infestation

Areas identified as “hot spots” for TYLCV infection include Kibwezi, Kitui, Machakos, Athi River (Eastern Province) and Naivasha (Rift Valley Province). Low incidence of TYLCV symptoms was observed in all other surveyed provinces with the exception of Western and Nyanza Provinces, where no TYLCV symptoms were observed.

B. tabaci was found to be the most common whitefly species in the country (Table 3). *T. vaporariorum* was common in the highlands and was encountered frequently in the Central, Rift Valley and Eastern Provinces. *T. vaporariorum* was not recorded from the Coastal and Western Provinces. The abundance pattern and economic importance of *B. tabaci*/TYLCV differed significantly between provinces. It was found to attain particularly high epidemic incidence (>25% of the tomato with TYLCV symptoms) in Eastern Province (Kibwezi 30%-100%, Kitui 30%-100%, Machakos and Athi River 15%-30%) and in Rift Valley Province (Naivasha 15%-40%), where whiteflies and TYLCV were of great concern to the tomato producers. This suggests that the impact of the *B. tabaci*/TYLCV complex is generally more severe in the dry areas.

In Coast Province, where the weather is hot and humid throughout the year, *B. tabaci* constituted 79% of the collected whitefly specimens. However, only a moderate incidence of TYLCV symptoms (5%-25%) was observed. In Western Province, *B. tabaci* constituted 90% of the collected whitefly specimens and in Nyanza Province, 79% (weather is also hot and humid throughout the year) but no TYLCV symptoms were observed on tomato, other vegetable crops, or weeds, and collected samples were negative when hybridized using a probe to the original Israeli isolate of TYLCV. In Western and Nyanza Provinces, most producers plant tomato as a secondary crop after the main staple cereal crops, maize (*Zea mays* L.) and sorghum (*Sorghum bicolor* [L.] Moench). The *B. tabaci* population at Kibwezi (Eastern Province) is usually very high from December to early March (hot and dry season) but the whiteflies virtually disappear from April to August (long rainy season followed by cold season), despite the presence of the same crops in the irrigated fields throughout the year.

High *T. vaporariorum* populations were observed in Nguruman (Rift Valley Province) where the underside of leaves of tomato, common bean and eggplant (*Solanum melongena* L.) could be found covered with adults and nymphs. In this area, the infestation level only drops during a brief rainy season, around April-May. A similar level of infestation by the same whitefly species was observed in the relatively cooler area of Kihara (Central Province). Direct feeding damage by *T. vaporariorum* was observed in both Central Province (Kihara and Mwea) and Rift Valley Province (Nguruman), where it was the predominant whitefly species.

Since completion of the survey, very high whitefly infestations have been reported in the cut-flower and ornamental plants on export-oriented flower and vegetable farms around Lake Naivasha (Rift Valley Province). The highest TYLCV incidences were observed at altitudes ranging from about 500 to 1000 m altitude (Table 4). *B. tabaci* was the dominant whitefly species up to an altitude of about 1700 m, above which *T. vaporariorum* became dominant.

Increased Socio-economic Understanding

Farmers' assessment of whitefly-related problems

Smallholders dominate vegetable production in Kenya and account for 70% of the total production (Mulandi, 1998). Producers in the survey areas

Table 4. Maximum incidence (% of sampled plants showing disease symptoms) of *Tomato yellow leaf curl virus* (TYLCV) encountered and species composition (%) of whitefly samples collected at different altitudes in Kenya.

Altitude[a] (m)	TYLCV incidence	Whitefly species composition[b]						
		B.t.	T.v.	T.r.	B.a.	S.p.	A.f.	B. spp.
20-270	8	72	6	0	14	8	0	0
520-770	100	81	6	0	13	0	0	0
770-1020	80	33	65	2	0	0	0	0
1020-1270	30	66	25	1	8	0	0	0
1270-1520	15	81	16	0	1	0	0	2
1520-1770	27	77	9	2	5	0	7	0
1770-2020	39	43	53	0	4	0	0	0

a. No survey was conducted in the 270-520 altitude range.
b. B.t., *Bemisia tabaci*; T.v., *Trialeurodes vaporariorum*; T.r., *Trialeurodes ricini*; B.a., *Bemisia afer*; S.p., *Siphoninus phillyreae*; A.f., *Aleurothrixus floccosus*; *B.* spp., *Bemisia* spp.

were surveyed using agreed protocols. Most of the interviewed producers (75%) used their own land, while 13% used rented lands and 12% had other arrangements. The great majority of producers (80%) were men.

Tomato is one of the most important vegetable crops in the country. Virtually all interviewed producers (98%) regarded tomato as their most profitable vegetable crop and 58% of the producers interviewed have cultivated tomato for more than 5 years. Main varieties of tomato grown in the country were Moneymaker and Cal-J, while other varieties include Roma, Marglobe, Petomech, Early Beauty, Mecheast, Bonny Best, Heinz, Floradade, Marmande, Ponderosa and Hotset. However, no tomato variety grown in Kenya was known to be resistant or tolerant to TYLC. Most producers (75%) bought their tomato seed from the local market, while 20% used their own seed and 5% reported buying directly from the seed companies. Other vegetable crops include kale (*Brassica oleracea* L.), common bean, sweet pepper, cabbage (*Brassica oleracea* var. *capitata* L.), eggplant, sweetpotato (*Ipomoea batatas* [L.] Lam.), okra, bitter gourd (*Momordica charantia* L.), melon (*Cucumis melo* L.) and onion (*Allium cepa* L.). Most producers (94%) practised crop rotation.

Fifty-five percent of the interviewed producers listed whiteflies and/or associated virus diseases among their principal pests and diseases and ranked these problems, on average, as their third-most-important problem in tomato. For tomato producers in Kenya, late blight *Phytophthora infestans* (Mont.) de Bary is the main cause of concern as a yield-limiting factor. Other pest and disease problems were early blight (*Alternaria solani* [Ell. and Mart.] Jones and Grout), red spider mites (*Tetranychus urticae* Koch.), bollworms (*Helicoverpa armigera* [Hubner]), leaf miners (*Lyriomyza* spp.), bacterial wilt (*Erwinia tracheiphila* [Smith]), aphids, cutworms (*Spodoptera litura* [Fabricius]), nematodes (Rhabditida: Steinernematidae & Heterorhabditae), end rot (*Godronia cassandrae* f. *vaccinii*), thrips (*Thrips palmi* Karny) and rats (*Rattus* sp.).

Most producers (88%) were able to recognize whiteflies, while only 64% of producers were able to recognize the symptoms of TYLCV and only 1% knew that whiteflies and TYLCV were inter-related. Although only about half of the producers (55%) listed whiteflies and associated viruses among their principal pests and diseases, 79% of interviewed producers believed that whiteflies cause problems in the crops on their farms. Among these producers, 50% attributed the problems to both whiteflies and TYLCV, 47% attributed the problem to whiteflies only, while only 3% mentioned TYLCV alone.

Areas where producers were not able to recognize whiteflies were mainly in Western Province (Funyula, Mokonja and Buhuma), Nyanza Province (South Ugenya and Nyabondo) and Coast Province (Shimba Hills and Kikambala), where whiteflies were scarce. Most producers in these localities also could not recognize TYLCV symptoms and were not aware of its association with the vector. Some thought that TYLCV was either a soil-borne or seed-borne problem, while some confused it with blight.

Most producers who recognized whiteflies and TYLCV symptoms had names for both but in most cases the producers gave non-specific names for whiteflies (e.g., “insects”) and for TYLCV (e.g., “disease”). Most of the names given to the whiteflies literally mean “flies” or “small insects” and local names include e*bichuni* (insect), *keguneta*, *kimbulutwa* (butterfly), *kudni* (insect), *mbuu* (mosquito), *ngaturia*, *okogataa* (mites), *oulolo* (white moulds), *rwagi rweru* (white mosquito), *twihuruta tweru* (small white butterflies), *ume* (aphids), *umuu* (moulds), *vipuvute vidogo* and *obororo* (plant fleas). Names given to the disease problem mean “leaf curling”, “folding” or “stunted plant growth” and local names include *chana matawi* (combing of leaves), *gathuri* (stunted old man), *gikware*, *gukunja mathangu* (folding of leaves), *kanyaria* (the devastating disease), *kugogonyara* (curly) and *mbaa* (blight).

Estimation of disease incidence and yield losses

Among the surveyed farms, 55% had TYLCV symptoms in their tomato crop. However, very low incidences (1% infection) were found in most parts of Central, Eastern and Coastal Provinces. Incidence of TYLCV symptoms of more than 30% was recorded in 11 out of 94 farms. These were in Kibwezi (3), Kitui (3), Machakos/Athi River (1) and Naivasha (4). High incidence (80%-100%) was only recorded in two farms in Eastern Province, in Kibwezi and Kitui areas. The incidence in Kibwezi resulted in a total loss of the tomato crop. Severe leaf curling plus subsequent yield depression of nearly 100% in sweet pepper was also observed in one experimental plot in Kibwezi. Despite the presence of *B. tabaci* in western Kenya (Western and Nyanza Provinces), no symptoms of TYLCV infection were recorded in this part of the country.

On average, producers’ estimate of the yield loss in tomato production was 30%. According to the survey, 1% reported a total yield loss, 5% reported three-quarter yield loss, 26% reported half yield loss and 50% of the producers reported one-quarter yield loss due to the whitefly/TYLCV complex. The problem was more serious in drier areas where irrigation was practised and this could be because of the presence of TYLCV reservoirs and the virus vector. Eighteen percent of the producers interviewed, mainly from Western Province, thought that their tomato crops did not suffer any yield loss because of the whitefly/TYLCV complex. However, 8% of producers interviewed reported abandoning their tomato crops in at least 1 year because of the problem. These were from Eastern, Rift Valley, Central and Coast Provinces. Half of them reported that the loss occurred in 1997.

Seventy-nine percent of tomato producers believe that they encounter the whitefly/TYLCV complex every year. Almost one-third of the producers (30%) believed that the incidence and severity of whitefly-transmitted viruses, especially in tomato, was exceptionally high in early 1997 before the El Niño phenomenon. Most (89%) also believe that there is a direct relationship between the seasonal weather changes and the whitefly/disease incidence. Seventy-five percent of producers reported that the period from December to March (hot and dry season) was the time when they experience the most serious whitefly/TYLCV problem, while few believed that they have the problem all year round. Producers gave estimation of the costs involved (see below).

Costs estimated by producers in control of whitefly/TYLCV complex per hectare of tomato

US$	% producers
0-49	13
50-99	17
100-199	30
200-299	17
300-400	23

Pesticide use

Almost half of the producers (43%) received advice and recommendations on the use of insecticides from technical advisors, 37% from other producers, neighbours or family members, 10% from pesticides salesmen, while 10% claimed to use chemicals on their own initiative. Most producers (85%) made their own decision on what, when and which insecticides to apply, while only 10% relied on the technical advice they received. Half of the producers (48%) applied insecticides as a preventive measure, 25% applied insecticides when they observed whitefly/TYLCV damage, while 18% applied insecticides routinely according to the calendar. The high level of insecticide usage coupled with prophylactic chemical spraying could easily lead to the whiteflies developing resistance, which would hamper their control.

Because of the lack of virus-resistant tomato cultivars, producers resort to the use of pesticides to combat the whitefly/TYLCV complex. The use of pesticides in most production areas in Kenya has increased, even though it is hazardous and sometimes ineffective and uneconomical. Eighty-six percent of tomato producers used insecticides, mostly pyrethroids and organophosphates, to combat the whitefly problem on their farms. However, 9% of the producers did not practice any sort of control against the problem, while 4% practised crop rotation as a means of combating it. Some producers used a *Bacillus thuringiensis* formulation (Dipel) for control of whiteflies. One producer in Mwea (Central Province) reported using ash, in addition to commercial chemicals, to manage the whitefly/TYLCV complex on his farm. Another producer in Kihara (Central Province) used a home-prepared blend of botanicals and a detergent to manage the whitefly problem but apparently with little success. Chemical control was in many cases directed against blight (i.e., using fungicides). The most commonly used insecticides for control of the whitefly/TYLCV complex in the country were pyrethroids (lambda-cyhalothrin and cypermethrin) and organophosphates (dimethoate and diazinon). Other commonly used insecticides include alpha cypermethrin (pyrethroid), amitraz (amidine), malathion, fenitrothion, and omethoate (organophosphates) and endosulfan (organochlorines). Acaricides such as dicofol, which has little effect on insects (EXTOXNET, 1998), were also commonly applied against whiteflies. The use of fungicides such as metalaxyl, copper sulphate and Mancozeb (ethylene bisdithiocarbamate), in an attempt to combat TYLCV was very common. Almost half the producers (48%) made between one and four insecticide applications per crop season to control the whitefly/TYLCV complex, while only 8% applied nine to ten applications. Most producers in Nyanza and Western Provinces did not apply insecticides or practice any sort of control measure against the whitefly/TYLCV complex.

There was lack of knowledge on proper use of pesticides. Some tomato growers overused chemicals, while others did not spray until symptoms of damage were observed, and subsequent intensive chemical application failed to save the crop. In many cases, producers continued spraying on a crop already severely affected by the virus, using a higher dosage in the hope of curing the plants. Producers were ignorant about the safety period between the last spraying of the crop and date of sale or consumption. Some producers said they did not care about the pre-harvest interval as long as the produce looked good for sale. Some of the local agrochemical suppliers bought large volumes of pesticides and measured out small volumes according to the financial capability of the buyers. The application of inappropriate pesticides and dose rates was common and led to control failure. Chemicals without any labels were usually bought from local dealers.

For the first time in Kenya, information was generated on whitefly resistance to insecticides in selected areas of the country. Assessment of the resistance of *B. tabaci* and *T. vaporariorum* to methomyl (a carbamate), cypermethrin and bifenthrin (pyrethroids) using both glass-vial and leaf-dip bioassays was carried out at Kibwezi, Kitui (Eastern Province), Nguruman (Rift Valley Province) and Mwea and Kihara (Central Province). Generally, *T. vaporariorum* was found to be more resistant to the three insecticides than was *B. tabaci*. Of the three insecticides used in the tests, cypermethrin was the most commonly used in the survey farms and bifenthrin the least. The resistance levels deduced from the analysis showed that both *B. tabaci* and *T. vaporariorum* were more resistant to cypermethrin than to either bifenthrin or methomyl. The results obtained suggest that repeated exposure of whiteflies to specific insecticides could have selected them for resistance to the chemicals. However, this was not a universal conclusion because there were some deviations from the general trend. For instance, a 6.8-fold resistance to bifenthrin was detected at Kihara where the insecticide had reportedly never been used. Whiteflies from Kajiado were the most resistant to cypermethrin, giving a resistance factor value (RF) of 87 in the leaf-dip method, while those from Nairobi were the most resistant to methomyl.

Strengthened Research Capacity

The present work has substantially improved our understanding of the whitefly problem in the major vegetable production areas of Kenya. Quantitative information on whiteflies and natural enemy species diversity and the occurrence of the whitefly-transmitted diseases, mainly TYLCV, as well as the socio-economic understanding of the producers' perception and practices relating to management of the whitefly/TYLCV complex, has provided Kenyan researchers with a solid background for setting a research agenda for further work, some of which may be carried out under Phase 2 of the TWF-IPM Project (see Introduction to this volume). The first, diagnostic, phase of the project also contributed to capacity building through a planning workshop and a mid-term meeting for all participating scientists, training of one M.Sc. thesis student in assessment of insecticide resistance and training of one B.Sc. student in host plant preference studies.

Conclusions

The following areas in Kenya have been identified as hot spots for future research on whiteflies and whitefly-transmitted viruses: Kibwezi, Kitui, Machakos/Athi River and Naivasha. The survey revealed that the prevalent whitefly species in the country are *B. tabaci*, *T. vaporariorum*, *B. afer*, *T. ricini*, *A. floccosus* and *S. phillyreae*. The fact that 55% of the surveyed farms had TYLCV symptoms indicates a potential for whitefly/TYLCV problems in Kenya. The major constraint in producers' knowledge on the whitefly/TYLCV complex was a complete lack of understanding (99% of farmers) of the connection between the disease and the vector.

To minimize the devastating effects of whitefly-transmitted viruses in tomato, sustainable vector- and disease-management options need to be developed, tested and deployed, including the use of resistant and/or tolerant cultivars, as well as cultural and biological control practices. The development of such options needs to be based on, and supported by, appropriate research. In addition, we need to understand the source of whitefly infestations and the reasons behind the striking between-field variability in whitefly population levels that is often observed among fields in close proximity to each other. For example, very heavy whitefly infestation was observed in two adjacent farms at Kihara but not in any of the other nearby farms. The contrast could have been due to the insects' feeding preference, because the two farms had eggplant and squash (both preferred hosts) that were not on the other farms but this possibility would need to be investigated further. We need to understand why there is low or no incidence of TLCV in western Kenya, despite the prevalence of the vector. The potential of local isolates and commercial formulations of entomopathogenic fungi, as well as parasitoids and predators, for control of whiteflies could be evaluated. There is also a need to look at other whitefly-transmitted viral diseases and at the role of weeds as reservoirs of begomoviruses.

There is a need for training of entomologists and virologists in areas of detection, identification and characterization of whitefly-transmitted viruses, whiteflies and their natural enemies, as well as training in epidemiological research methodologies. The continued involvement of scientists from ICIPE and the Asian Vegetable Research and Development Center (AVRDC), who have provided technical support during the first phase of this project, would be helpful. Collaborative work involving the network of regional stations of the Kenya Agricultural Research Institute (KARI) should be extended.

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CHAPTER 2.4

Tanzania

*Ignas Swai** and *Simon Slumpa***

Introduction

Geographical context

The following regions were covered in the surveys conducted during the 1997-99 period: Arusha, Babati, Arumeru, Dodoma, Kongwa, Mpwapwa, Njombe, Iringa, Mwanga, Rombo, Moshi, Hai, Same, Mkuu, Mbeya, Vwawa, Tukuyu, Morogoro, Tanga, Lushoto and Zanzibar (Figure 1). Surveys followed the methodology agreed among the project partners.

The surveyed areas can be grouped into distinct eco-climatic zones (Table 1). The northern highlands cover the Arusha and Kilimanjaro regions and Usambara Highlands in the Tanga region. The zone experiences two rainy seasons: short rains (*vuli*) from November to December and long rains (*masika*) from March to May. The main dry season is from June to September. The coastal lowlands include such areas as Muheza, Tanga, Ruvu, Dar-es-Salaam, Kibaha and Zanzibar Island. The rainfall pattern is the same as for the northern highlands but the lowlands are hotter. The Central Plateau includes Dodoma and western parts of Morogoro regions, where the rainfall pattern is variable and unpredictable. The Morogoro region is transitional and receives higher rainfall than the Dodoma region. Dodoma experiences only one rainy season, around November to January, and production of vegetables in this zone is seasonal. Irrigation is common in both Morogoro and Dodoma.

Figure 1. Regions covered in the 1997-99 survey, Tanzania.

The emergence of **Bemisia tabaci** *as a pest and virus vector*

The whitefly species *Bemisia tabaci* (Gennadius), the vector of *Tomato leaf curl virus* (ToLCV) and *Tomato yellow*

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Table 1. Mean annual temperature, precipitation and altitude range of the surveyed regions in Tanzania.

Region (Province)[a]	Mean min.-max. temperature (°C)	Mean precipitation (mm)	Altitude range (m)
Arusha	11-21	913	485-1660
Dodoma	16-29	556	329-378
Iringa	14-25	633	396-561
Kilimanjaro (Same)	14-26	1130	244-1600
Kilimanjaro (Moshi)	16-26	549	120-850
Mbeya	10-27	1132	424-536
Morogoro	19-30	854	450-900
Tanga (Lushoto)	19-30	1006	366-549
Zanzibar	21-30	1436	20-60

a. Same is on the windward side and Moshi, the leeward side, of Mt. Kilimanjaro.

leaf curl virus (TYLCV), is ranked as the most important insect pest of tomato (*Lycopersicon esculentum* Mill.) in Tanzania (Varela and Pekke, 1995). *B. tabaci* also transmits other viral diseases in common bean (*Phaseolus vulgaris* [L.]), cowpea (*Vigna unguiculata* [L.] Walp.), sweetpotato (*Ipomoea batatas* [L.] Lam.) and cassava (*Manihot esculenta* Crantz), while the woolly whitefly *Aleurothrixus floccosus* (Maskell) is widespread in citrus (*Citrus* spp. L.) growing areas. No information was available on the geographical range of whitefly species and their socio-economic significance in Tanzania prior to the country's participation in the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project.

ToLCV was first reported in Tanzania in 1990 (Czosnek et al., 1990). Additional survey studies conducted by the Asian Vegetable Research and Development Centre (AVRDC)-Africa Regional Program in Tanzania from 1994-97 showed that several tomato samples with typical leaf curl symptoms did not hybridize with the Egyptian or Israeli ToLCV-DNA probes. Polymerase chain reaction (PCR) analysis performed with the above tomato samples indicated that in addition to TYLCV, a virus different from all previously characterized tomato geminiviruses of the Old World occur in Tanzania and was tentatively named *Tomato leaf curl virus-Tanzania* (TLCV-Tan; Chiang et al., 1997), now ToLCV-Tz.

Increased Biological Understanding

Characterization of begomoviruses and whitefly biotypes

During the 1997-99 period, 179 samples of whitefly adults and nymphs were collected from vegetable cropping systems in Tanzania, of which 407 nymph specimens were histologically processed and mounted on 213 slides. Identification of whiteflies conducted by the International Center of Insect Physiology and Ecology (ICIPE) revealed that *B. tabaci* was the most common whitefly species, representing 78% of the mounted specimens. *B. afer* (Priesner and Hosney) represented 8% and *Trialeurodes vaporariorum* (Westwood), 7%. Other whitefly species made up 7% of the specimens and included *Trialeurodes ricini* (Misra), *B. hirta* Bink-Moenen, *Orchamoplatus citri* (Takahashi), *Tetraleurodes andropogon* (Dozier) and *Aleurothrixus floccosus* (Maskell).

The reproductive host plants of whiteflies identified from the surveys in Tanzania belong to the following families: *Amaranthaceae, Asteraceae, Commelinaceae, Convolvulaceae, Cucurbitaceae, Euphorbiaceae, Lamiaceae, Leguminosae, Myrtaceae, Rutaceae, Solanaceae* and *Verbenaceae*. Table 2 gives the scientific names of the host plant species on which each whitefly species was found to be reproducing. Whiteflies were abundant on common bean but no whitefly-transmitted disease symptoms were observed.

ToLCV symptoms were observed in *Achyranthes aspera* L., *Euphorbia heterophylla* L. and *Nicandra physalodes* (L.) Gaertn. Nono-Womdim et al. (1996) identified TYLCV, the causative virus in these hosts. These non-cultivated host plants are widespread in the country and may serve as major reservoirs of TYLCV.

Disease incidence and symptom severity

Areas identified as “hot spots” for ToLCV incidence in Tanzania include Morogoro, Dodoma, Kilimanjaro (especially Same) and Arusha (Figure 1). Fifteen percent of the surveyed farms had 100% ToLCV incidence in their tomato crop. It should be highlighted that all surveyed farms in Morogoro (Dakawa, Kariakoo, Kipela, Kibundi, Kiruka and Bigwa) had 100% TLC incidence. Up to 100% of the crop also were reported to be affected in Kilimanjaro (Marwa, Bangalala and Same) and Dodoma (Mbalala, Chikula and Mpwapwa). Nine percent of the surveyed farms had ToLCV incidences ranging from 70% to 99%. These were in Arusha (Baraa), Kilimanjaro (Mijongomeni, Kivulini and Mkolowoni) and Dodoma (Msolota). Ten percent of the surveyed farms had ToLCV incidences ranging from 50% to 69%, while 65% of the farms had incidences ranging from 1%-19%. The lowest incidences of ToLCV symptoms, 1%-2%, were observed in the southern part of Tanzania (Iringa, Mbeya, Tukuyu and Njombe) and in Zanzibar. There were no distinct ToLCV-free zones found within the surveyed areas of Tanzania. ToLCV incidence was highest during the summer months (hot season), from December to March. Yield loss due to ToLCV depends on the susceptibility of the tomato cultivar, the crop stage at the time of infection and environmental conditions (Nono-Womdim et al., 1999).

Twenty-three tomato accessions, including progenies of crosses between *Lycopersicon chilense* Dunal LA 1969 and cultivated tomato, were field-screened for resistance to ToLCV by AVRDC (Nono-Womdin et al., 1999). Two commercial F_1 hybrids, Fiona and Tyking, had the highest level of resistance. Plants of these varieties were symptomless and they did not hybridize with Israel TYLCV-DNA probe. The varieties PSR-403511 and PSR-407111 were also resistant to ToLCV but hybridization tests showed that a few symptomless plants contained ToLCV DNA. The following progenies of crosses between *L. chilense* LA 1969 and *L. esculentum* had 30%-60% resistant plants: Chilytlc 94-1, 94-2, 94-4, 94-5, 94-6 and MultichilTLC. Tests on two commonly grown commercial tomato varieties, Moneymaker and Roma, showed 100% susceptibility.

Natural enemy species

Samples of whitefly parasitoids and predators were collected during the surveys. Preliminary identification of the sampled parasitoids showed that most were *Encarsia sophia* (Girault and Dodd), while *Eretmocerus* sp. was collected on only two occasions. A number of predators, coccinellids and predatory bugs were also collected in the vicinity of whitefly populations in the survey fields.

Table 2. Reproductive host plants of whitefly species encountered during surveys in Tanzania.

Whitefly species	Crops		Non-cultivated hosts	
	Family	Species	Family	Species
Bemisia tabaci (Gennadius)	Convolvulaceae	*Ipomoea batatas* (L.) Lam.	Amaranthaceae	*Achyranthes aspera* L.
	Euphorbiaceae	*Manihot esculenta* Crantz		*Amaranthus* sp.
	Leguminosae	*Desmodium* sp.	Asteraceae	*Ageratum conyzoides* L.
		Vigna unguiculata (L.) Walp.		*Bidens pilosa* L.
		Phaseolus vulgaris L.		*Galinsoga parviflora* Cav.
	Myrtaceae	*Psidium guajava* L.	Commelinaceae	*Commelina* sp.
	Solanaceae	*Lycopersicon esculentum* Mill.	Euphorbiaceae	*Euphorbia heterophylla* L.
		Solanum tuberosum L.	Leguminosae	*Tephrosia* sp.
			Lamiaceae	*Leonotis nepetifolia* (L.) R. Br.
			Verbenaceae	*Lantana camara* L.
Bemisia afer (Priesner and Hosney)	Euphorbiaceae	*Manihot esculenta* Crantz		
	Leguminosae	*Phaseolus vulgaris* L.		
Trialeurodes vaporariorum (Westwood)	Leguminosae	*Desmodium* sp.	Euphorbiaceae	*Euphorbia heterophylla* L.
		Vigna unguiculata (L.) Walp		
		Phaseolus vulgaris L.		
Trialeurodes ricini (Misra)	Cucurbitaceae	*Citrullus lanatus* (Thunb.) Matsum & Nakai	Amaranthaceae	*Achyranthes aspera* L.
	Euphorbiaceae	*Manihot esculenta* Crantz	Asteraceae	*Galinsoga parviflora* Cav.
	Leguminosae	*Desmodium* sp.		
		Phaseolus vulgaris L.		
Bemisia hirta Bink-Moenen			Euphorbiaceae	*Euphorbia heterophylla* L
Orchamoplatus citri (Takahashi)				Plant unidentified
Tetraleurodes andropogon (Dozier)				Plant unidentified
Aleurothrixus floccosus (Maskell)	Rutaceae	*Citrus* spp.		

Geographical range of whitefly and whitefly-transmitted virus infestation

B. tabaci was the most common whitefly species throughout the country (Table 3). *B. afer* was common in cassava in the areas of Iringa, Arusha, Mbeya and Kilimanjaro (Moshi). *T. vaporariorum* was encountered in Arusha, Dodoma, Iringa, Kilimanjaro (Same), Mbeya and Tanga (Lushoto) but not in Morogoro, Kilimanjaro (Moshi) or Zanzibar. *T. ricini* was common in Arusha, Iringa and Mbeya. *A. floccosus* was collected only from citrus. The few specimens identified as *B. hirta* require additional confirmation.

Up to 100% ToLCV incidence was found at altitudes ranging from 260 to 1060 m, while ToLCV incidence of up to 70% was encountered as high as 1460 m altitude (Table 4). *B. tabaci* was the prevalent whitefly species up to an altitude of 1260 m, while the two *Trialeurodes* species mentioned above were prevalent at higher altitudes (Table 4).

Table 3. Species composition (%) of whitefly samples collected in regions of Tanzania.

Province[a]	Species composition[b]							
	B.t.	B.a.	T.v.	T.r.	B.h.	O.c.	T.a.	A.f.
Arusha	48	18	8	14	2	0	0	10
Dodoma	90	2.5	2.5	5	0	0	0	0
Iringa	37.5	25	25	12.5	0	0	0	0
Kilimanjaro (Same)	96	2	2	0	0	0	0	0
Kilimanjaro (Moshi)	89	11	0	0	0	0	0	0
Mbeya	40	17	33	10	0	0	0	0
Morogoro	99	0	0	0	0	1	0	0
Tanga (Lushoto)	91	2	5	2	0	0	0	0
Zanzibar	83	0	0	0	0	0	17	0

a. Same is on the windward side and Moshi, the leeward side, of Mt. Kilimanjaro.
b. B.t., *Bemisia tabaci*; B.a., *B. afer*; T.v., *Trialeurodes vaporariorum*; T.r., *T. ricini*; B.h., *B. hirta*; O.c., *Orchamoplatus citri*; T.a., *Tetraleurodes andropogon*; A.f., *Aleurothrixus floccosus*.

Table 4. Maximum incidence (% of sampled plants showing disease symptoms) of tomato leaf curl (TLC) encountered and species composition (%) of whitefly samples collected at different altitudes in Tanzania.

Altitude (m)	Maximum TLC incidence	Whitefly species composition[a]							
		B.t.	T.v.	T.r.	B.a.	A.f.	B.h.	T.a.	O.c.
0-60	4	83	0	0	0	0	0	17	0
60-260	100	91	9	0	0	0	0	0	0
260-460	100	88	3	4	5	0	0	0	0
460-660	100	66	18	7	9	0	0	0	0
660-860	100	84	0	0	14	0	2	0	0
860-1060	100	44	0	0	0	56	0	0	0
1060-1260	50	75	13	12	0	0	0	0	0
1260-1460	70	0	0	100	0	0	0	0	0
1460-1660	15	14	43	0	43	0	0	0	0

a. B.t., *Bemisia tabaci*; T.v., *Trialeurodes vaporariorum*; T.r., *T. ricini*; B.a., *B. afer*; A.f., *Aleurothrixus floccosus*; B.h., *B. hirta*; T.a., *Tetraleurodes andropogon*; O.c., *Orchamoplatus citri*.

Increased Socio-economic Understanding

Farmers' assessment of whitefly-related problems

Tomato is one of the most important vegetables cultivated in Tanzania. Most (79%) of the tomato producers interviewed used their own land for production, while 17% used rented land. A vast majority of the producers (92%) were men. Almost all the producers (96%) regarded tomato as their most profitable vegetable crops and 62% of producers have been involved in vegetable farming for more than 5 years. Although tomato production has increased in the last 5 years to meet increased demand for processing and fresh markets, the average yields remain low at about 10-14 tons per hectare.

The main tomato varieties cultivated in Tanzania are Marglobe, Moneymaker and Roma VF, all of which are highly susceptible to several viral diseases. Most producers (60%) obtain planting material from the local market, 36% use their own seeds and 4% get planting material from other tomato producers. Other commonly grown vegetable crops include cowpea, pea (*Pisum sativum* L.), common bean, cabbage (*Brassica oleracea* L. var. *capitata* L.), onion (*Allium cepa* L.), Irish potato (*Solanum tuberosum* L.), eggplant (*Solanum melongena* L.), sweet pepper (*Capsicum annuum* L.), okra (*Abelmoschus esculentus* [L.] Moench), sweetpotato, *Amaranthus* spp., watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai), melon (*Cucumis melo* L.), cucumber (*Cucumis sativus* L. var. *sativus*) and other cucurbits. Whiteflies also attack most of these crops. The great majority (94%) of the producers interviewed practice crop rotation.

Producers ranked their pest and disease problems in tomato and associated vegetable crops in Tanzania (in order of importance) as follows: late blight (*Phytophthora infestans* [Mont.] de Bary), TLC, fruit borers (*Helicoverpa armigera* [Hübner]), whiteflies (Homoptera: Aleyrodidae), aphids (Aphididae), *Fusarium* wilt, early blight (*Alternaria solani* [Ell. and Mart.] Jones and Grout), red spider mite (*Tetranychus urticae* Koch.), diamond back moth (*Plutella xylostella* [L.]), angular leaf spot (*Pseudomonas syringae* pv. Lachrymans), bruchids (Coleoptera: Bruchidae), bacterial leaf spot (*Xanthomonas campestris* pv. *visicatoria*), leaf miner (*Lyriomyza* spp.), beanfly (*Ophiomyia* spp.), cutworm (*Spodoptera litura* [Fabricius]), powdery mildew (*Leveillula taurica*), black rot (*X. campestris* pv. *campestris*), fruit fly (*Drosophila melanogaster*), bacterial canker (*Clavibacter michiganensis*), bean rust (*Uromyces phaseoli*), *Tomato mosaic virus*, *Septoria* leaf spot (*Septoria lycopersici*), nematodes (*Meloidogyne* spp.), *Cucumber mosaic virus*, thrips (Thysanoptera: Thripidae), cabbage sawfly (*Nematus* spp.) and soft rot (*Erwinia carotovora*).

Most producers (90%) were able to recognize the whitefly and 84% also recognized ToLCV. Only 11% of the interviewed producers knew that whiteflies and ToLCV were interrelated. None of the interviewed producers from Morogoro (where the disease incidence was 100% on all surveyed farms) knew the cause of ToLCV; they did not have names for the whiteflies other than "insects", nor did they have precise names for ToLCV. Most producers (75%) believed that the whiteflies caused problems on their farms. Almost half of them (43%) believed that both whiteflies and ToLCV caused problems, whereas 30% attributed the problems to ToLCV only and 27% to whiteflies only.

Local names given to whiteflies included *chawa* (lice), *inzi weupe* (white flies), *kibanda, kifizi, kipe weupe, kipepeo* (small butterflies), *kurukury, mbuu, msubi weupe, mvumuu, ndaka, sughru* (small flying insects), *sunhuu, suru, tukorokotwa, wadudu* (insects) and *wadudu weupe* (white insects). The local names given to ToLC included *rasta* (dreadlocks), *ugonjwa wa kukunja* (curling disease), *ukoma* (leprosy), *dume* (sterile), *ghojo, kibangi, kudulala, kutu* (rust), *bondia, majani, masai, mdamango, mwanga bondia, ngofu, ngumi* (boxer), and *kibangi* and *kobe* (tortoise).

Estimation of disease incidence and yield losses

Among the surveyed farms, 99% had TLC symptoms in their tomato crop and 34% had ToLCV incidences above 25%. Common symptoms include leaf curling, yellowing, chlorosis of leaf margins, leaf distortion, reduction in leaf size, shortening of the internodes, stunting, excessive branching and flower abscission (Nono-Womdim et al., 1999). The average perceived yield loss in tomatoes due to the whitefly/ToLCV complex was 45%. Some producers (9%) reported a total yield loss, 13% reported three-quarters yield loss, 32% reported half yield loss, 42% reported one-quarter yield loss and only 5% of the producers reported no yield loss.

Most of the producers interviewed (74%) believed that they had whitefly and/or virus problems every year and 26% reported that the most severe problems occurred in 1997. Most producers (94%) also believed that there is a relationship between climate and whitefly/ToLCV occurrence. The majority (90%) believe that the problem is worst during the hot season, between December and March, while the problem is less pronounced between August and December. The producers mostly attributed the increase in severity to weather changes, particularly long dry spells. Other factors mentioned by some producers were ineffective insecticides, resistance to insecticides and old crop remains (acting as a source of infestation and virus reservoir).

Producers gave estimation of the costs involved in crop protection measures (see below). The costs were mainly incurred in the purchasing of chemical insecticides and labor for their application.

Costs estimated by producers in control of whitefly/TYLCV complex per hectare of tomato

US$	% producers
0-49	40
50-99	18
100-199	26
200-299	9
300-400	7

Pesticide use

One-third of producers (35%) received recommendations on management practices from technical advisors, 35% from other tomato producers, 3% from sales agents, 10% from others and 17% relied on their own judgement. Among those producers who applied insecticides, the decision on what insecticides to apply and when was made by the producer in 93% of the cases, while 6% relied on technical advice.

Use of pesticides was the most common option for managing the whitefly/ToLCV problem and was practiced by 76% of those interviewed. Cultural control was practiced by 11% of the producers, while 13% did not practice any control. The pesticides most commonly applied by vegetable producers in Tanzania for control of whitefly/disease problems on their farms are profenofos, chlorpyrifos, fenitrothion, dimethoate, pirimiphos-methyl, chlorpyrifos and diazinon (organophosphates); deltamethrin, lambda-cyhalothrin and fenvalerate (pyrethoids); thiodan and endosulfan (organochlorines); and methomyl (carbamate).

Producers who made 10 insecticide applications per season to manage the whitefly/ToLCV complex accounted for 21% of those interviewed. Those who made from five to eight applications comprised 31% of interviewees and a similar percentage made from one to four applications. However, 18% reported that they did not apply insecticides. Most producers (62%) applied insecticides as a preventive measure, 26% made the application when they observed damage and 6% made the application according to the calendar.

Strengthened Research Capacity

This survey has substantially improved our understanding of the magnitude of the whitefly-associated problems in the major vegetable production areas of Tanzania. The work conducted under this first, diagnostic, phase of the TWF-IPM Project also has strengthened collaboration between the national agricultural research organization and international research institutions such as ICIPE and AVRDC.

Conclusions

TYLCV is the most important whitefly-transmitted viral disease in tomato cropping systems in Tanzania. The disease causes significant yield loss in tomato production in various parts of the country. The lowlands of Morogoro and Dodoma were identified as the principal “hot spots” for ToLCV, where incidences of symptoms of 100% were frequently encountered. Moderately high incidences were also observed in the Arumeru Highlands near Arusha and Kilimanjaro.

Commercial cultivars of tomato grown in Tanzania are susceptible to TYLCV. Some breeding lines from the African Regional Program (ARP) of AVRDC at Arusha have shown significant tolerance to TYLCV. AVRDC-ARP in collaboration with the Horticultural Research and Training Institute (HORTI-Tengeru) have begun efforts to screen a broad array of tomato germplasm and incorporate the resistance genes into cultivated tomato varieties.

Further work in Tanzania, under the second phase of the TWF-IPM Project, should focus on evaluation of germplasm for use in breeding for resistance to TYLCV and ToLCV, and development of IPM options for the management of the whitefly vector.

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CHAPTER 2.5

Malawi

Harriet Thindwa and *Patric Khonje**

Introduction

Geographical context

Malawi lies between latitude 9° 22' and 17° 7' South, and between longitudes 32° 40' and 35° 55' East. The climate is characterized by three main seasons: cool and dry from May to August, warm and dry from September to November, and warm and wet from December to April. The 5-month rainy season (November to March) begins earlier in southern and central regions of the country. Annual rainfall ranges from 600 mm in the lower Shire Valley and the Karonga Lake Shore Plains to over 3000 mm in high-elevation areas. Temperatures range between 20 °C and 35 °C but may approach and surpass 40 °C in the Rift Valley areas during October and November (Mkanda et al., 1995).

Surveys were carried out in the main areas of the country growing tomato (*Lycopersicon esculentum* Mill.) during the period 1997 to 1999. Areas surveyed were Mzuzu, Mzimba, Nkhata Bay, Salima, Lilongwe, Dedza, Ntcheu and Thyolo (Figure 1).

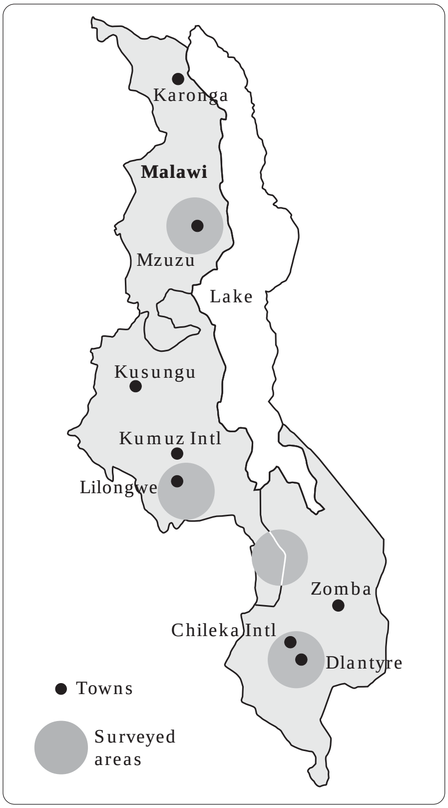

Figure 1. Tomato-growing areas surveyed for whitefly incidence in Malawi.

The emergence of **Bemisia tabaci** *as a pest and virus vector*

In Malawi, the whitefly species *Bemisia tabaci* (Gennadius) has been reported as the vector of cassava mosaic disease (Swanson and Harrison, 1994). Varying

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levels of infestation by the citrus woolly whitefly *Aleurothrixus floccosus* (Maskell) also have been recorded in regions of the country growing citrus (*Citrus* spp. L.) (Löhr, 1996).

Although whiteflies were suspected of transmitting viral diseases in vegetable crops, especially tomato, no reliable information on this subject was available from Malawi prior to the survey conducted under the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project. The occurrence of whitefly species, their geographical distribution and their natural enemies in the vegetable cropping system also were unknown.

Increased Biological Understanding

Characterization of begomoviruses and whitefly biotypes

Selection of survey sites and other methodologies followed the standards agreed with other project partners. Questionnaires were used to gather information from vegetable producers. The incidence of plants affected by tomato yellow leaf curl was estimated on their farms, whiteflies were sampled for species identification and parasitoids and suspected natural enemies were collected.

During the surveys, 130 collections of adults and nymphs of whiteflies were made. Of these, 159 specimens were processed and mounted on 67 slides. Preliminary identification carried out at the International Center of Insect Physiology and Ecology (ICIPE) revealed that *Bemisia afer* (Priesner and Hosney) is the most prevalent species in the country, accounting for 67% of samples identified. Of the remaining specimens, *B. tabaci* comprised 30%, and *Trialeurodes vaporariorum* (Westwood), *T. ricini* (Misra) and *Aleyrodes proletella* (Linnaeus) 1% each. The species composition of samples from the various areas surveyed is reported in Table 1.

Table 1. Species composition (%) of whitefly samples collected in tomato-producing regions of Malawi.

Province	Species composition[a]				
	B.a.	B.t.	T.v.	T.r.	A.p.
Dedza	56	34	5	0	5
Lilongwe	86	14	0	0	0
Mzimba	86	14	0	0	0
Mzuzu	100	0	0	0	0
Nkhata Bay	53	47	0	0	0
Ntcheu	55	15	10	20	0
Salima	33	67	0	0	0
Thyolo	54	42	0	0	4

a. B.a., *Bemisia afer*; B.t., *B. tabaci*; T.v., *Trialeurodes vaporariorum*; T.r., *T. ricini*; A.p., *Aleyrodes proletella*.

The reproductive host plants from which whitefly nymphs were collected belong to the families Solanaceae, Euphorbiaceae, Leguminosae and Verbenaceae (Table 2). *B. tabaci* was found breeding on tomato in only a few areas.

Symptoms of tomato yellow leaf curl were observed in most of the surveyed sites. Samples of about 10 plants per field were collected from all the affected farms, squashed on nylon membranes and sent to the John Innes Centre (JIC) for hybridization. The results confirmed the presence of *Tomato yellow leaf curl virus* (TYLCV).

Disease incidence and symptom severity

Areas identified as "hot spots" (high incidence of TYLCV) include Dedza, Mzimba, Thyolo and Ntcheu (Figure 1). The highest incidences of TYLCV were observed at altitudes ranging from 1085 to 1485 m (Table 3).

Table 2. Reproductive host plants of whitefly species recorded in Malawi.

Whitefly species	Crops		Non-cultivated hosts	
	Family	Species	Family	Species
Bemisia afer (Priesner and Hosney)	Euphorbiaceae	*Manihot esculenta* Crantz		
	Leguminosae	*Phaseolus vulgaris* L.		
Bemisia tabaci (Gennadius)	Euphorbiaceae	*Manihot esculenta* Crantz	Euphorbiaceae	*Euphorbia heterophylla* L.
	Leguminosae	*Mucuna deeringiana* (Bort.) Merr.	Verbenaceae	*Lantana camara* L.
		Phaseolus vulgaris L.		
	Solanaceae	*Lycopersicon esculentum* Mill.		
Trialeurodes vaporariorum (Westwood)	Leguminosae	*Phaseolus vulgaris* L.		
Trialeurodes ricini (Misra)	Leguminosae	*Phaseolus vulgaris* L.		
Aleyrodes proletella (Linnaeus)	Leguminosae	*Mucuna deeringiana* (Bort.) Merr.		

Table 3. Maximum incidence (%) of *Tomato yellow leaf curl virus* (TYLCV) and species composition (%) of whitefly samples encountered at increasing altitudes in Malawi.

Altitude (m)	TYLCV incidence[a]	Whitefly species composition[b]				
		B.a.	B.t.	T.v.	T.r.	A.p.
485-685	14	31	56	0	0	13
685-885	14	38	63	0	0	0
885-1085	7	64	27	0	0	9
1085-1285	52	79	21	0	0	0
1285-1485	49	59	24	5	12	0
1485-1685	5	66	23	6	0	5

a. A TYLCV incidence of 94% was observed in one farm in the highest altitude range (1485-1684 m), while other farms in this altitude range had incidences of 0%-5%.

b. B.a., *Bemisia afer*; B.t., *B. tabaci*; T.v., *Trialeurodes vaporariorum*; T.r., *T. ricini*; A.p., *Aleyrodes proletella*.

The survey revealed that *B. afer* is the predominant whitefly species in most parts of Malawi (Table 1). This species was found on cassava (*Manihot esculenta* Crantz), common bean (*Phaseolus vulgaris* L.) and unidentified weeds but not on other vegetable crops. *B. tabaci* was also common in most parts of the country (Table 1). *pT. vaporariorum* was collected from Dedza and Ntcheu, while *T. ricini* was collected from Ntcheu only and *A. proletella* from Dedza and Thyolo. High numbers of nymphs were collected from Nkhata Bay, Dedza and Ntcheu, the latter two being the only regions from which more than three whitefly species were identified. On farms in Salima, very few whitefly nymphs could be found for identification and the region had relatively low TYLCV incidences (0%-11%). *B. tabaci* was the predominant whitefly species up to an altitude of about 900 m, while *B. afer* became more abundant at higher altitudes (Table 3).

Natural enemy species

Nine collections of whitefly parasitoids and 10 of predators were made during the survey. Among these were *Encarsia sophia* (Girault and Dodd), coccinellids (Coleoptera: Coccinellidae) and predatory bugs (Heteroptera: Anthocoridae).

Increased Socio-economic Understanding

Farmers' assessment of whitefly-related problems

Most of the producers interviewed (86%) grow tomato on their own land, while 12% rent the land. The majority of producers (88%) were men. Tomato was regarded as the most profitable vegetable crop by 98% of the producers. Only 27% of them have been growing tomato for more than 5 years. Tomato varieties commonly grown in Malawi include Moneymaker, Red Khakhi, Vercles and Heinz. The tomato seed was bought from the local market by 35% of the producers, 33% used their own seed (from the previous season), 21% obtained seed from the Agriculture Trading Company (ATC) and 11% used imported seeds. Tomato is grown both under rain-fed and irrigated conditions.

Other common vegetable crops in the country include common bean, cabbage (*Brassica oleracea* var. *capitata* L.), Irish potato (*Solanum tuberosum* L.), mustard (*Sinapis alba* L. subsp. *alba*), onion (*Allium cepa* L.), pumpkin (*Cucurbita* spp.), rape (*Brassica napus* L. var. *napus*), sweetpotato (*Ipomoea batatas* [L.] Lam.), turnip (*Brassica rapa* L. subsp. *rapa*), okra

(*Abelmoschus esculentus* [L.] Moench), pepper (*Capsicum annuum* L.), eggplant (*Solanum melongena* L.), and carrot (*Daucus carota* L. subsp. *sativus* [Hoffm.] Arcang. var. *sativus* Hoffm.). Cassava is also a major staple crop. Most producers (94%) practice crop rotation.

The main pests and diseases recorded on tomato, ranked according to importance, were late blight (*Phytophthora infestans* [Mont.] de Bary), red spider mites (*Tetranychus* spp.) TYLCV, whiteflies, bacterial wilt (*Ralstonia solanacearum*) and cutworms (*Spodoptera litura* [Fabricius]).

Eighty-three percent of producers could recognize whiteflies and 82% could recognize TYLCV. None of the producers seemed to know the interrelationship between TYLCV and whiteflies and confused TYLCV symptoms with red spider mite damage, aphid damage, heavy rain damage, fungal diseases, nitrogen deficiency and soil-borne diseases. Most of those interviewed (83%) thought that whiteflies and TYLCV caused problems on their farms. Almost half of the producers (46%) complained about the whitefly only, 17% of the disease only and 37% complained about both the disease and the whitefly.

Local names given to whiteflies include *msambe*, aphids, flies, pests, vegetable lice, white aphids, cobweb, *chinoni*, white lice, white ants, mites, small flies, lice and white butterflies. Local names given to TYLCV included blight, leprosy, *chisaka* (bushy top), *kaligwiti* (bushy top), crinkled leaves, *katungana*, *ciguduli*, bunchy top, *cicsaka*, *kafumbata* and malformation.

Estimation of disease incidence and yield losses

Among the surveyed farms, 75% had TYLCV symptoms in their tomato crop. However, only 6% of the farms had incidences of TYLCV symptoms above 25%, while 16% of the farms had incidences ranging from 10% to 25%, and 53% of the producers had incidences ranging from 0.5% to 9%. The highest incidence (94%) was reported on a farm in Dedza.

On average, the producers' estimate of yield loss in tomato due to the whitefly/TYLCV complex was 49%. According to the survey, 14% reported total yield loss, 14% reported three-quarter yield loss, 35% reported half yield loss, 29% reported one-quarter yield loss and 9% did not report any losses due to the whitefly/TYLCV complex. Those who reported incurring total loss were from the Dedza and Ntcheu Provinces, and a single producer from Mzimba. Producers who reported abandoning tomato growing, at least once, due to the problem made up 21% of those interviewed; 38% of them abandoned their farms in 1997.

Most producers (68%) believed that they had whitefly/TYLCV problems every year. The most severe attack was in 1997 according to 43% of the respondents. These producers were from Ntcheu (Kasamba), Dedza (Kalilombe, Chimlambe, Jere), Nkhata Bay (Kashonga, Chipayika), Salima (Matumba), Mzimba (Chizuminja) and Mzuzu (Kaboko). Nearly all producers (96%) believe there is a direct relationship between the climate and whitefly/disease incidence. There was no obvious consensus among the producers as to what weather conditions trigger whitefly and TYLCV outbreaks. The whitefly/TYLCV problems were reported to be worst from January to June and again from September to October. Some producers

(40%) believe that the whitefly/TYLCV incidence is higher when it is dry and hot, while others (30%) said they experience most problems when it is wet and cold. The opinions of the remaining producers were scattered between the remaining combinations of dry, moderate, wet and hot, moderate, and cold weather.

Producers gave estimation of the costs involved in crop protection measures (see below).

Costs estimated by producers in control of whitefly/TYLCV complex per season of tomato production

US$	% producers
0-49	26
50-99	16
100-199	19
200-299	14
300-400	9
>400	16

Many producers (42%) had not received recommendations on management practices for whiteflies. Those who had received advice obtained it from various sources: 34% received recommendations from technical advisors, 15% from other vegetable producers, 6% from family members or neighbours and 3% from sales agents. Most producers (90%) managed the whitefly/virus problem by means of synthetic pesticides, 4% by cultural means and 6% practiced no control at all. Other control methods such as legislative measures, natural insecticides (e.g., botanicals and bio-pesticides), biological control and the use of resistant varieties, were not reported.

Pesticide use

The decision on what insecticide to apply and when was made by the producer himself in 58% of the cases, by technical advisors (29%), sales people (3%) or a family member (2%), while 7% did not use insecticides on their farms. Half the producers (52%) applied insecticides as a preventive measure, while 33% applied them when damage was observed and 5% applied insecticides according to the calendar. Few producers (5%) reported making more than 10 insecticide applications per season, while 4% made 9-10 applications, 37% of the producers made 5-8 applications, 40% made 1-4, and 14% applied no insecticides to manage the whitefly/disease problem.

The insecticides most widely used in the management of the whitefly/TYLCV problem in Malawi include: fenitrothion, malathion and dimethoate (organophosphates); cypermethrin and lambda-cyhalothrin (pyrethroids); carbaryl and sevin (carbamates). Farmers were ignorant of the fact that chemicals with the same active ingredient often are sold under various trade names and they therefore risk using the same active ingredient throughout, rather than alternating with others. The use of fungicides was common because producers confused TYLCV symptoms with blight. Miticides such as dicofol were used also in an attempt to control whiteflies but EXTOXNET (1998) reports that this chemical has little effect on insects.

Strengthened Research Capacity

Scientists from ICIPE trained and accompanied the national scientists during the beginning of the survey. The project has equipped national scientists with a better understanding of the severity of whitefly-related problems in the country’s vegetable production. New information on the status of whiteflies and the whitefly-transmitted viruses in tomato now is available. Understanding our producers' perceptions and practices in relation to whitefly problems has been a significant achievement of this project.

Conclusions

Whitefly/TYLCV "hot spots" in Malawi include Dedza, Mzimba, Thyolo and Ntcheu. In both Dedza and Ntcheu, several farmers reported having abandoned their tomato crops at least once due to whiteflies/TYLCV. For this reason, these two areas should be targeted for further research in the second phase of the project. *B. afer*, followed by *B. tabaci*, is the prevalent whitefly species in most parts of the country and the most widespread parasitoid was *E. sophia*.

Although many producers recognized whiteflies and TYLCV, they often confused TYLCV symptoms with those of other pest and disease or nutrient deficiencies. Producers also were confused about the climatic conditions that trigger outbreaks of whiteflies and TYLCV.

Incidences of TYLCV in Malawi were not as high as in neighbouring Tanzania. Only 6% of the surveyed farms had TYLCV incidence above 25%. Although TYLCV incidences were generally low, the disease was encountered in all surveyed areas, indicating that the whitefly/TYLCV complex is a potential threat to Malawi's vegetable production.

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CHAPTER 2.6

Tomato Yellow Leaf Curl Virus and Tomato Leaf Curl-like Viruses in Eastern and Southern Africa

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Introduction

Tomato (*Lycopersicon esculentum* Mill.) is one of the most popular vegetables in eastern and southern Africa. It is a high-value crop, providing a good source of income to small-scale farmers. Tomato is consumed both in fresh and processed forms. The processing industry often provides good rural employment opportunities and products for export.

The productivity of tomato in Africa, particularly in eastern and southern Africa, is unfortunately among the world's lowest. For example, the mean yield per hectare of tomato in southern African countries ranges from 1.5 to 14 tons per hectare as compared to the world average of 25 tons per hectare based on the 1989 Food and Agriculture Organization (FAO) production estimates. Several fungal, bacterial and viral diseases have drastically hampered the cultivation of tomato in eastern and southern Africa (AVRDC 1996; 1997; 1998). Among viral pathogens, *Tomato yellow leaf curl virus* (TYLCV) and tomato leaf curl-like viruses account for most losses in farmers' fields (unpublished results). However, adequate information on the epidemiology of TYLCV and associated viruses in eastern and southern Africa remain scanty.

This chapter refers to the emergence of TYLCV and associated viruses in tomato in eastern and southern Africa and describes the development of our understanding of their identification and management.

Impact and Assessment Losses

Over the past decade, TYLCV and leaf curl-like viruses have caused significant losses for tomato farmers in eastern and southern Africa. Yield losses depend on the growing season, tomato cultivars and growth stage at which plants become infected. Recent on-farm surveys indicated that the average perceived yield losses in tomato were up to 40% in Malawi, 50% in Kenya, 75% in Tanzania and 100% in Sudan (CIAT, 1998). In Sudan, yield losses due to TYLCV have been reported to vary in different regions (Yassin and Nour, 1965; Makkouk et al., 1979). Moreover, Ioannau and Iordanou (1985) indicated that losses due to TYLCV vary between 50% and 82% depending on the time of infection.

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In East Africa, TYLCV was first reported in the Sudan (Yassin and Nour, 1965). The disease has since spread to Tanzania (Czosnek et al., 1990; Nono-Womdim et al., 1996), Malawi and Zambia (AVRDC, 1996). Recent survey studies have indicated that this virus occurs in Namibia and Swaziland (AVRDC, 1998), and in Kenya and Uganda (Table 1). A similar disease caused by a distinct virus, tentatively named "*Tomato leaf curl virus-Tanzania*" (TLCV-Tan; now accepted as ToLCV-Tz) (Chiang et al., 1997) has been reported in the central region of Tanzania (Nono-Womdim et al., 1996).

Table 1. Occurrence of *Tomato yellow leaf curl virus* (TYLCV) in eastern and southern Africa.

Country	Year of survey[a]		
	1996	1997	1998
Kenya	16/43	23/81	-
Tanzania	-	73/271	162/344
Uganda	-	5/35	10/58

a. Number of infected samples/number of tested samples on which squash blot hybridization tests were performed using a specific DNA probe for TYLCV from Israel (TYLCV-Is).

Tomato plants infected with TYLCV exhibit severe and varied symptoms. Common symptoms consist of leaf curling, yellowing, chlorosis of leaf margins, leaf distortion, reduction in leaf size, shortening of the internodes or stunting. In some cases, infected plants show excessive branching and flower abscission. Symptoms of TYLCV vary with strain, tomato cultivars, plant age at the time of infection and environmental conditions. In eastern and southern Africa, epidemics of TYLCV occur during the summer months.

Diagnosis

In east and southern African countries, TYLCV is primarily identified by the presence of symptoms that consist of yellowing, leaf curling and stunting. However, visual diagnosis is not useful for the implementation of effective control strategies since the symptoms are very clear on old plants that are already fruiting.

Selected monoclonal antibodies developed against *African cassava mosaic virus* (ACMV) or *Indian cassava mosaic virus* (ICMV) have been used to detect different isolates of TYLCV (Macintosh et al., 1992; Givord et al., 1994). These antibodies also were used to identify TYLCV isolates from Tanzania (unpublished results). It has been shown that many whitefly-transmitted begomoviruses are serologically related (Roberts et al., 1984). Therefore, serological tests for identification of TYLCV are unspecific and do not differentiate strains or different leaf curl viruses.

Nucleic acid hybridization assay is commonly used for the detection of TYLCV. Many cDNA probes prepared against different strains of TYLCV are now available (Czosnek et al., 1990; Kheyr-Pour et al., 1991). Some of these probes currently are being used to identify leaf curl isolates from eastern and southern Africa.

Etiology

TYLCV belongs to the Geminiviridae family and has small geminate particles measuring about 20 × 30 nm (Harrison et al., 1977; Czosnek et al., 1988). The genome of TYLCV consists of a circular single-stranded DNA encapsulated by a single capsid protein (Navot et al., 1991). Many isolates from eastern and southern Africa are closely related to TYLCV from Israel (TYLCV-Is).

TYLCV has been reported to have a wide host range, including plant species belonging to Acanthaceae, Asteraceae, Canicaceae, Euphorbiacea, Leguminosae, Malvaceae, Oxalidaceae, Pedaliaceae, Plantaginaceae and Solanaceae (Singh and Reddy, 1993). In Tanzania, TYLCV was detected in three newly identified hosts, *Achyranthes aspera*, *Euphorbia heterophylla* and *Nicandra physalloides* (Nono-Womdim et al., 1996). These weeds are widespread and serve as a major reservoir of TYLCV.

TYLCV is transmitted by the whitefly *Bemisia tabaci* (Gennadius), not by seeds. In four African countries, Kenya, Malawi, Tanzania and Sudan, several populations of *B. tabaci* have been collected from TYLCV-infected tomato samples and characterized (CIAT, 1998).

TYLCV diseases exhibit various symptoms indicating the existence of different strains. Preliminary identification studies conducted in 1994 showed that several tomato samples from Tanzania, with typical leaf curl and yellowing symptoms, did not hybridize with the Egyptian and/or Israel DNA probes (Nono-Womdim et al., 1996). Two of these Tanzanian leaf curl samples were positive for begomovirus by polymerase chain reaction (PCR) using a primer which specifically amplifies part of the replicase (*AC1*) open reading frame (ORF), the intergenic region and the coat protein (*AV1*) ORF of whitefly-transmitted begomoviruses. Furthermore, the nucleotide sequence of the PCR fragment was compared with the sequences of several distinct begomoviruses, including TYLCV-Is, TYLCV-Th, TYLCV-Sar, TYLCV-Ind, etc. that infect tomato, as well as with the sequence of ACMV, ICMV and *Mung bean yellow mosaic virus* (MYMV). It was found that the Tanzanian isolate shows less than 80% nucleotide identities with these other begomoviruses (Chiang et al., 1997). Since it has been proposed that two isolates of the same begomovirus species have nucleotide sequence greater than 90%, the Tanzanian begomovirus, TYLCV-Tz, is different from all previously characterized begomoviruses from the Old World. Recent survey studies have shown that TYLCV-Tz is now widespread in Tanzania (Table 2). A few leaf curl samples from Malawi have shown a weak positive reaction in squash blot hybridization with the DNA probe of TYLCV-Tz. These results indicate that this virus, or another closely related begomovirus, might be present in this country.

Recently, another study conducted on the identification of leaf curl viruses of tomato indicated the presence of a newly identified virus in Uganda that has 73% homology with TYLCV-Is and 78% homology with ToLCV-Tz. The leaf curl virus from Uganda is tentatively named ToLCV-Ug (Charles Sskyewa, personal communication, 1999).

These findings indicate the presence of at least three different leaf curl viruses of tomato in eastern and

Table 2. Preliminary results on the distribution of tomato leaf curl disease in Tanzania using two specific DNA probes against tomato yellow leaf curl virus-Egypt (TYLCV-Eg) and *Tomato leaf curl virus*-Tanzania (ToLCV-Tz).

Region	Disease incidence[a]	
	TYLCV-Is	ToLCV-Tz
Arusha	13/40	39/40
Dodoma	79/113	96/113
Kilimanjaro	12/30	29/30
Morogoro	34/70	60/70
Zanzibar	5/29	6/29

a. Number of positive samples in DNA hybridization tests/number of samples with leaf curl symptoms.

southern Africa. The causes of this new emergence of leaf curl viruses in Africa have not been investigated yet.

Management

Several management strategies to control TYLCV and associate viruses have been reported (Singh and Reddy, 1993; Polston and Anderson, 1997). These strategies include cultural practices, control of the insect vector and the use of resistant varieties. In eastern and southern African countries, the implementation of cultural practices such as use of virus-free seedlings, reduction of virus inoculum (roguing), elimination of sources of infection and intercropping to control TYLCV has not been performed at farmers' field level. Many African farmers still cannot recognize TYLCV in order to apply these cultural practices for management. However, the use of insecticides, primarily to control whiteflies and not TYLCV, is very common in eastern and southern Africa (CIAT, 1998).

Experiments have been conducted since 1995 at AVRDC Africa Regional Program to identify tomato varieties with resistance to TYLCV (Table 3). The varieties Fiona and Tyking are resistant to TYLCV in Tanzania. Plants of these species are symptomless and TYLCV is not detected by DNA hybridization. Generally, resistance to TYLCV is quite specific. For instance, tomato cultivars with resistance or tolerance to TYLCV from the Old World are susceptible to tomato leaf curl viruses from the Western Hemisphere (Polston and Anderson, 1997). The situation seems to be different in Africa. In recent field screening tests, Fiona, Tyking and TY52, which are resistant to TYLCV, were found to resist ToLCV-Tz as well. Such lines will provide an efficient control strategy for the management of TYLCV and ToLCV-Tz since both viruses occur in mixed infections in Tanzania.

Table 3. Field screening of *Lycopersicon* accessions for resistance to *Tomato yellow leaf curl virus*.

Lycopersicon accessions	Final disease incidence (%)
Chepertylc C-1	96
4-2	95
FI-3	100
J-7	100
L-7	100
Sin-3	100
Siv-5	100
Siv-6	100
Columbian 36	100
EC-104395	100
Jackal	55
LA 3214	100
LA 3216	95
Lignon C8-6	100
PSR-403511	5
PSR-407111	9
Roza	100
Ty-20	54
8476	75
Fiona	0
Tyking	0
MoneyMaker	100
Roma	100

Conclusion

This chapter summarizes recent outbreaks of TYLCV and leaf curl-like viruses in eastern and southern Africa. The virus was first reported in Sudan in the 1960s, where it has been a threat for several decades. Then, in 1990, it was found in Tanzania. To date, TYLCV is widespread in many countries of the subregion. It is not well known whether TYLCV has occurred in these countries from some time ago, as in Sudan, or whether it has spread southwards from Mediterranean countries.

Until 1997, TYLCV was the only begomovirus reported to cause disease on tomato in eastern and southern Africa. Two additional viruses, namely

ToLCV-Tz and ToLCV-Ug, which are different from TYLCV, have been reported recently (Chiang et al., 1997; Charles Sskyewa, personal communication, 1999). Recent surveys indicate that many tomato leaf curl samples do not hybridize with the available cDNA probes in nucleic acid hybridization tests. This may indicate the existence of additional African indigenous leaf curl viruses.

In Tanzania, tomato varieties with resistant to TYLCV under field conditions have been identified. These resistant tomatoes are hybrid varieties that many farmers in eastern and southern Africa cannot afford.

At AVRDC, a breeding programme has been initiated aimed at incorporating the TYLCV resistance derived from *Lycopersicon chilense* (Zakay et al., 1991) into cultivated tomato. However, until open-pollinate varieties with resistance to TYLCV become available, there is a need to develop alternative control strategies for African farmers that will include control of the insect vector and cultural practices.

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CHAPTER 2.7

Conclusions and Recommendations

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Results of Country Studies

Bemisia tabaci (Gennadius) research in the eastern African region has been focused so far on cotton (*Gossypium hirsutum* L.) in Sudan and on cassava (*Manihot esculenta* Crantz) in Kenya, Tanzania and Malawi. In recent years, vegetables have become the crops most severely affected by *B. tabaci*-transmitted viruses. Little attention has been directed to vegetables therefore little information was available prior to the commencement of the System-wide Tropical Whitefly Integrated Pest Management (TWF-IPM) Project in this region. For the first time, information about critical aspects of *B. tabaci* characterization on the vegetable-based cropping system has been reported from the eastern Africa region.

Characterization of begomovirus and whitefly biotypes

B. tabaci is the most dominating whitefly species in the region as a whole. All specimens of whitefly collected in Sudan were *B. tabaci*; in Tanzania, *B. tabaci* made up 78% of the collected specimens and in Kenya, 65%. The species *Trialeurodes vaporariorum* (Westwood), *Bemisia afer* (Priesner & Hosney), *Siphoninus phillyreae* (Haliday), *Aleurothrixus floccosus* (Maskell), *Trialeurodes ricini* (Misra), *Orchamoplatus citri* (Takahashi), *Tetraleurodes andropogon* (Dozier) and *Aleyrodes proletella* (Linnaeus) were occasionally encountered in Malawi, Kenya and Tanzania, of which the last four species were registered for the first time in Tanzania. *Bemisia hirta* spec. nov. was encountered once but is subject to confirmation. *A. floccosus* was encountered on citrus. In the East African highlands, *B. tabaci* was the dominating whitefly species from sea level up to 1300-1700 m above sea level, above which *T. vaporariorum* and *T. ricini* dominated. In southern Africa (Malawi), *B. tabaci* only dominated up to 900 m above sea level, above which *B. afer* was the dominating species.

Disease incidence and symptom severity

The Phase 1 survey revealed silver leaf symptoms on squash (*Cucurbita pepo* L.) in Sudan. Elsewhere, these symptoms are associated with *B. tabaci* biotype-B as the causal pest. This is the first observation of silver leaf symptoms in the region and may indicate that this feared biotype-B has reached Sudan. Characterization of the *B. tabaci* specimens collected at the site where the silver leaf symptoms were found is underway and the biotype-B is yet to be confirmed.

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B. tabaci was found widespread on tomato (*Lycopersicon esculentum* Mill.), pepper (*Capsicum* spp. L.) and eggplant (*Solanum melongena* L.), in the *Solanaceae*; watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai), melon (*Cucumis melo* L), squash, cucumber (*Cucumis sativus* L. var. *sativus*) and bitter gourd (*Momordica charantia* L.), in the *Cucurbitaceae*; common bean (*Phaseolus vulgaris* L.), lablab (*Lablab purpureus* [L] Sweet), *Leguminosae* family; cassava (*Euphorbiaceae*); okra (*Abelmoschus esculentus* [L.] Moench), cotton, sweetpotato (*Ipomoea batatas* [L.] Lam.), *Desmodium* and basil (*Ocimum basilicum* L.), in the *Labiateae*. In addition to these families, *B. tabaci* also was found frequently on wild plant species within the families of *Acanthaceae*, *Amaranthaceae*, *Asteraceae*, *Commelinaceae* and *Verberaceae*.

Whiteflies in association with *Tomato yellow leaf curl virus* (TYLCV) had been studied in the Sudan (Dafalla and Sidig, 1997) and *Tomato leaf curl virus* (ToLCV) symptoms in Tanzania had been found due to a unique strain named ToLCV-Tz (Chiang et al., 1997). Since tomato was known as one of the most profitable crops, and whiteflies had been ranked as the most/second most important pest/vector in tomato (Varela and Pekke, 1995), tomato was selected as the target crop in an extensive survey on whiteflies and whitefly-transmitted viruses (WTVs) in vegetable production in the region.

Tomato yellow leaf curl (TYLC) symptoms have been identified as caused by TYLCV in Sudan, Kenya and Malawi. TYLC/tomato leaf curl (ToLC) symptoms were found in more than 93% of the surveyed fields of tomato in Sudan and Tanzania, in 72% in Malawi and 55% in Kenya. More than 25% TYLC incidence was found in 50% of the fields in Sudan, 34% in Tanzania, 11% in Kenya and 6% in Malawi. In the East African highlands, high ToLC incidence (>70%) was found from 500-1000 m above sea level in Kenya but from 50-1500 m above sea level in Tanzania. TYLC symptoms are particularly severe during the hot seasons (December to March in Tanzania and Kenya; March to September in Sudan).

Reservoirs for TYLCV in the wild vegetation were found in plant species belonging to the families of *Solanaceae* and *Euphorbiaceae*, in particular, and in *Amaranthaceae*.

In Sudan, the experienced team surveyed WTVs in all commonly grown vegetable crops. TYLCV in tomato is by far the most economically important *B. tabaci*-transmitted virus in Sudan. Next ranks *Watermelon chlorotic stunt virus* (WmCSV) and *Okra leaf curl virus* (OLCV). TYLCV and WmCSV are particularly severe during the hot season growing periods (March-September) and OLCV during the late winter and early summer (January-March). Other severe WFTV were *Tomato vein thickening virus* (TVTV) in tomato, *Cucumber vein yellowing virus* (CVYV) and a yellowing-inducing poty-like virus not fully identified in watermelon and musk melon (*Cucumis melo* L.). Less important WTVs, yet representing a potential hazard, are TYLCV in pepper, WmCSV in snake melon (*Cucumis melo* L. subsp. *melo* var. *flexuosus* [L.] Naudin) and squash, a begomovirus and a closterovirus not yet fully identified in *Vigna* sp. and common bean (*Phaseolus vulgaris* L. var. *vulgaris*), *Bean mild mosaic virus* (BMMV) and *Pepper leaf curl virus* (PepLCV).

The African Regional Program (ARP) of the Asian Vegetable Research and Development Center (AVRDC) has identified three tomato accessions

(Fiona, Tyking and TY52) resistant to TYLCV and ToLCV-Tz. Under heavy disease pressure, these accessions are symptomless and the viruses are not detected by DNA hybridization. Furthermore, two tomato accessions (PSR-403511 and PSR-407111) were identified as tolerant to TYLCV; only 5%-9% disease incidence was detected under heavy disease pressure.

Natural enemy species

Encarsia sophia (Girault and Dodd) was the predominant whitefly parasitoid in Kenya, Tanzania and Malawi, but was not encountered during the survey in the Sudan. *Encarsia lutea* (Masi) and *Eretmocerus mundus* Mercet. were predominant in the Sudan but not in the other countries, although *Eretmocerus* spp. were occasionally encountered in Kenya and Tanzania. *Encarsia formasa* (Gahan) was encountered in the highlands of Kenya. Predators of coccinelids, lacewings, bugs, spiders and mites were found adjacent to whitefly populations and their identification at species level is yet to be confirmed.

Estimation of disease incidence and yield losses

In Sudan, the “hot spots” for *B. tabaci* and transmitted viruses are the arid and irrigated cotton-growing areas in Gezira and Khartoum States, at 400-500 m, where the problem is severe throughout the year but peaking in March to September. Where cotton is not grown, the incidence is less. In Tanzania, the hot spots for *B. tabaci* and TYLC are transitional areas in Morogor, Dodoma, Kilimanjaro and Arusha, ranging from 100-1450 m. In Kenya, the hot spots are semi-arid to transitional areas in Eastern Province (Kibwezi, Kitui and Machacos) and Rift Valley (Naivasha), 700-1200 m. In Malawi, the hot spots are in southern areas of the Lake Malawi region (Dedza, Mzimba, Thyolo and Ntcheu), 600-1500 m. In Tanzania, Kenya and Malawi the problem is most severe during the hot dry season, December to March. Irrigation in the semi-arid and arid regions allows for continuous cultivation of crops favoured by *B. tabaci* and transmitted viruses, which guarantees continuous availability of reproductive hosts during the year.

In hot spot areas throughout the region, yield losses due to *B. tabaci* and transmitted viruses ranged between 50% and100%. As an average of all surveyed tomato fields, the yield losses due to *B. tabaci* and TYLC were 49% in Malawi, 45% in Tanzania and 30% in Kenya. In Sudan, the survey revealed that an average of 62% fruit yields of tomato, watermelon, musk melon and okra are lost annually due to *B. tabaci*-transmitted viruses. WmCSV was implicated in up to 80% yield losses of export Galia melon in Khartoum and Gezira States during the 1998-99 season, and completely wiped out the production in southern Blue Nile, Shuwak and Dinder inland delta. In the region in general, more than 65% of producers reckoned that they had whitefly/disease problems in their tomato crop every year and 1997 was the year with the most severe attack (prior to the onset of the El Niño rains). On average, producers in the region spend US$145 per hectare combating whiteflies.

Farmers’ assessment of whitefly-related problems

In Sudan, farmers ranked *B. tabaci* and its transmitted viruses as the most economically and most damaging crop protection problem in tomato in the country. In Tanzania, farmers ranked *B. tabaci* and its transmitted viruses as a second problem in tomato after bacterial blight. In Kenya and Malawi, *B. tabaci* and its transmitted viruses ranked less important. Nevertheless, 79% of the interviewed farmers in Kenya found that the problems were on the increase compared with 74% in Tanzania, 68% in Malawi and 65% in Sudan.

It was generally believed that the whitefly/disease complex is most damaging during dry and hot seasons (December through March in Tanzania and Kenya, March through August in the Sudan). In Malawi, however, controversial opinions on the seasonality of outbreaks were found among the interviewed producers. Interestingly, in the Sudan, TYLCV incidence was most severe during the hot summer months despite less prevalence of the vector, whose population peaked during the cooler winter months (December through January).

Pesticide use

Eighty three percent of the farmers interviewed in the region applied pesticides in their tomato crops. The percentage of farmers making more than nine pesticide application per tomato cropping season was 56% in Sudan, 21% in Tanzania, 9% in Malawi and 8% in Kenya. The use of synthetic insecticides was the most prevailing control method and in most cases the producer alone made the decision on what insecticide to use and when (average 84%). Half of the producers sprayed insecticides as a prevention measure and 25% of them would start spraying after damage had been observed. Five to six sprayings per season was practiced on average but in the Sudan, 56% of the interviewed farmers made nine or more applications per season. Pyrethroids and organophosphates were the most commonly used insecticides in the region.

Only 35% of the interviewed producers in Sudan, 11% in Tanzania, 1% in Kenya and none in Malawi knew about the inter-relationship between whiteflies and ToLCV symptoms. In Morogoro in Tanzania, where 100% ToLCV incidence was found on all the surveyed farms, none of the producers knew about its inter-relationship with whiteflies. In Malawi, almost all producers confused TYLC symptoms with other damage such as damage from red spider mite, aphids, heavy rain, fungal diseases, nitrogen deficiency and soil-borne diseases.

Sudan is the only country in the region that has adopted IPM as its official policy of crop protection aimed at combating the whitefly problem. Government support for IPM is well defined in the recent publication, Sudan Country Strategy Note, 1997-2001: Partnership towards Sustainable Human Development. The document emphasizes that the Government of the Sudan is in pursuit of an integrated program for environmentally sustainable development. However, there is at present a laxity in implementation and in cultural and legislative measures. There are no restrictions on the number of pesticide applications. Sudan has no extension service at all, whereas technical advisors from extension services have visited about one-third of the interviewed producers in the other countries.

Strengthened Research Capacity

The TWF-IPM Project has helped in increasing the capacity in whitefly research by training one M.Sc. student from Kenya and one Ph.D. student from Sudan. With assistance/input from the Institute of International Education (IIE), London, and the John Innes

Centre (JIC), project scientists were trained in conventional taxonomy and DNA techniques. A short-term attachment also was completed on the effect of host plants on the oviposition and survival of the sweetpotato whitefly. After long fruitless efforts, finally cultures of *Bemisia* and *Trialeurodes* were successfully established at the International Center of Insect Physiology and Ecology (ICIPE). The Centre, AVRDC and the national partners are planning work on transmission studies and host suitability studies in Phase 2. The hot spot areas of whitefly problems in partner countries have been identified. Besides confirming the local occurrence of several known natural enemies of whiteflies, additional genera/species of predators/parasitoids have been identified. Perceived on-farm losses due to tomato geminiviruses and the average cost to control whitefly/TYLC per hectare of tomato in one season were documented.

ICIPE has extended assistance in the identification of whitefly species and natural enemy collections from surveys across nine countries in Africa to the subproject on whiteflies on cassava and sweetpotato, led by the International Institute of Tropical Agriculture (IITA). Likewise, ICIPE has extended assistance in the identification of whiteflies and natural enemies to countries that were not participating officially in this project, for example, Uganda and Senegal. The Center has established a reference collection for African whiteflies in the past 2 years from 10 countries within the framework of the two African whitefly subprojects. This is in connection with a collection of about 2000 samples of whiteflies and more than 4000 nymph specimens mounted on permanent slides, in addition to about 400 whitefly parasitoids and predators collected. It is hoped that this collection will further help understand whiteflies in future studies and training.

This project demonstrates how effective collaboration can result in proper identification of research agendas and their correct implementation. This has been possible only because of the joint efforts of the national programmes and the collaborators in a well-coordinated approach. The joint efforts of the ICIPE scientists and the national teams have further strengthened ICIPE's partnership with national programmes in Africa. This mode of collaboration and the complementation between ICIPE, AVRDC, JIC and national agricultural research systems scientists has helped link up the expertise of these centres and the local knowledge and experiences of the national programmes. The adoption of a standardized methodology and being able to modify it to suit specific requirements has simplified work and made it possible to compare results and share experiences.

Recommendations

As a result of a joint planning workshop in September 1999, the partners in Sub-project 3 (Whiteflies as vectors of viruses in vegetable and mixed cropping systems in eastern Africa) made the following recommendations for the second project phase.

It was proposed that Phase 2 should include Uganda in addition to Sudan, Tanzania and Kenya. The focus should remain on *B. tabaci* as vector of viral diseases. The target cropping system in Kenya and Uganda will remain tomato. Target cropping systems will include both tomato and watermelon in Tanzania and tomato, cucurbits and okra in the Sudan. There is a need to continue the characterization of whiteflies and WTVs in target cropping systems. This characterization comprehends both characterization at molecular level, including bio-typing of whiteflies, and biological characterization of both whiteflies and WTVs with emphasis on host range, host preferences and interaction between the whiteflies and the viruses.

The aim is to continue strengthening the global whitefly network set up by the TWF-IPM Project. Through this network, scientists from the region will enjoy the information flow of knowledge and participation in experimental research in order to boost the whitefly research in their respective countries with new knowledge and disseminate new control methods to the national extension services and vegetable producers. Initiatives taken to create an African Whitefly Network should continue.

More evidence must be recorded on yield depression due to whiteflies and their transmitted viruses. Geographical information on agricultural regions, major crops, target crops, whitefly/virus hot spots, topographical and climatic data, crop seasonality and spatial patterns of cropping systems must be collected in order to apply geographic information systems to the information created.

There is an urgent need for the immediate development of IPM components viable under the socio-economic conditions in the region. Selection, multiplication and dissemination of tomato varieties resistant/tolerant to whitefly and/or ToLCVs were given first priority in the development of the IPM component. AVRDC should gather and multiply a “package” of resistant germplasm from all over the world and distribute it to the national partners for screening, selection, multiplication and dissemination.

Development of physical barriers for vector control was given second priority in developing the IPM component. Different physical screening methods to prevent early invasion of whiteflies should be gathered from all over the world and tested as physical control measures under the socio-economic and climatic conditions in each partner country.

Testing rational use of conventional and non-conventional insecticides was given third priority in developing the IPM component. Producers urgently need alternative control methods in order to meet the new requirements of pesticide residue-free products for the export market. Regional monitoring of insecticide resistance of whiteflies will provide useful information to convince policy makers of the need for developing alternative control methods. Non-conventional and indigenous methods from all over the world such as oil, non-phytotoxic soaps, botanicals and ash should be gathered in a “package” and tested in partner countries by means of a standardized methodology that will allow comparisons across ecosystems. Evaluation of the effect of both conventional and non-conventional control methods on natural enemies and biological control agents such as parasitoids and entomopathogens will reveal information on their potential environmental impact and their suitability in organic farming.

Understanding of the mechanism behind effective cultural practices in order to define appropriate cropping sequences also was put on the agenda. Such studies may require several years and experimental cropping seasons and may not provide immediate applicable information for producers as the target stakeholders. The priority of such studies was therefore set to follow the above IPM components. Such studies would evaluate the effect of diversity-based cropping in time and space, planting dates, planting density, ground cover, roguing, in-field weed management, crop residue management and micro-environment modification.

Studies on biological control of whitefly vectors were given less priority. Since low vector populations can create a severe virus outbreak, it is believed that biological control may have low impact in the control of virus outbreaks driven by the movement of whitefly vectors. It was decided to start gathering cases of successful legal control methods and related documentation. When sufficient evidence has been provided through the project work, the aim is to start reinforcing official regulation and inspection for area-wide management, including planting date, harvesting date, crop free periods, destruction of crop residues, irrigation cut-off, etc.

The research capacity in the partner countries must be strengthened through training of national scientists in detection, identification and characterization of whiteflies, WTVs, natural enemies of whiteflies and epidemiology. Furthermore, yearly information exchange and co-ordination meetings will be needed.

When feasible, producers must be actively involved in the development of the IPM component, including the brainstorming, planning, realization and evaluation of experiments. Training handouts on rational control of whiteflies and WTVs must be produced for the extension service and producers, and farmers' organizations in hot spot areas must be sensitized, in particular. Decision support tools must be available for policy makers and news media must be used currently as the project develops.

Acknowledgements

The ICIPE-coordinated subproject has derived valuable input in geminivirus characterization from JIC in the UK and from AVRDC in Tanzania. Dr. James Legg (IITA, Uganda) offered valuable support in linking with the IITA-led whitefly subproject on cassava/ sweetpotato systems. The project also has received valuable assistance from Dr. Andrew Polaszek (CAB International Bioscience, UK Centre) and Dr. Eddie Ueckermann of the Plant Protection Research Institute (PPRI), South Africa in the identification of some whitefly natural enemies. This book is a joint product of all partners and the ICIPE co-ordinating unit scientists.

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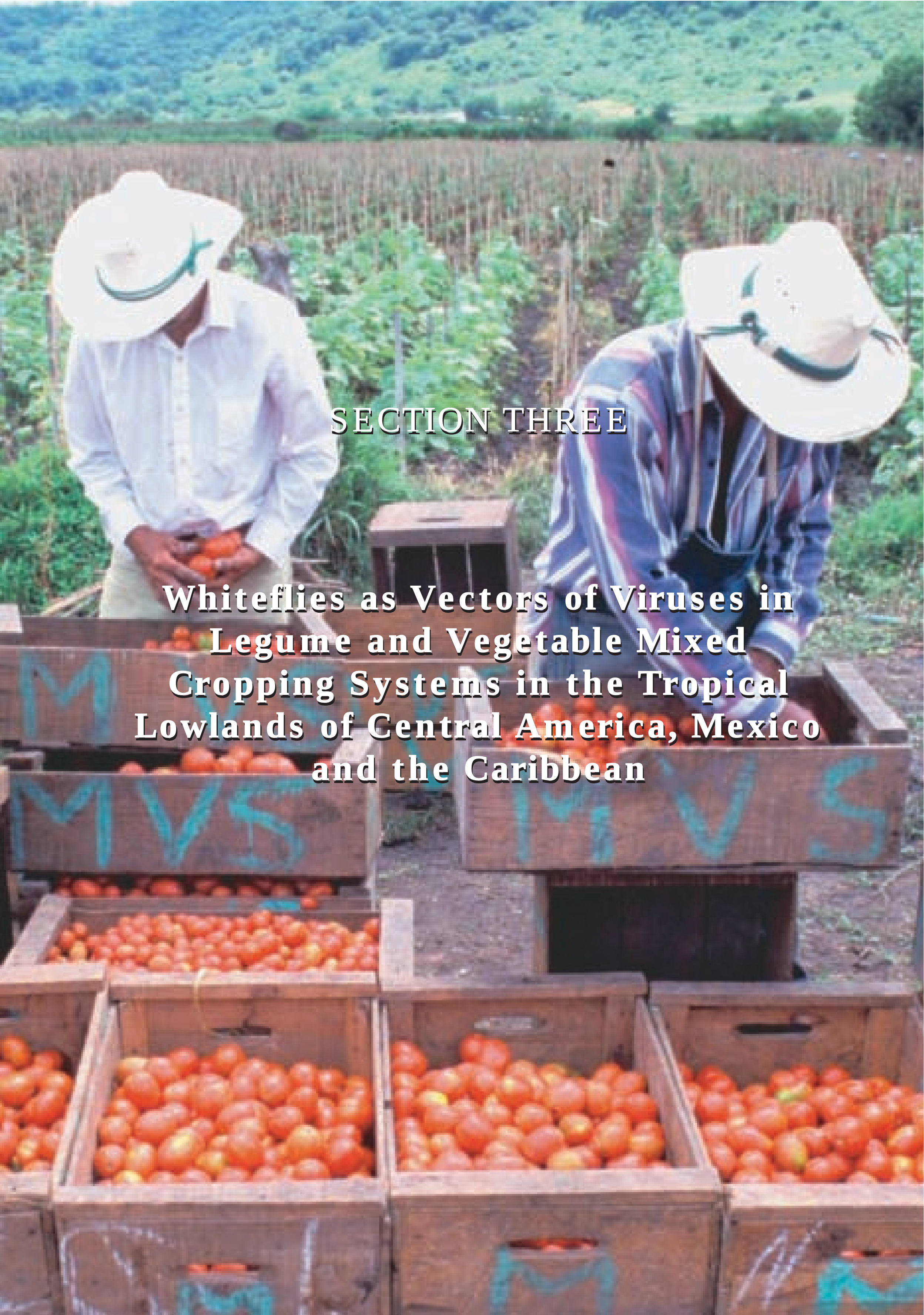

SECTION THREE

Whiteflies as Vectors of Viruses in Legume and Vegetable Mixed Cropping Systems in the Tropical Lowlands of Central America, Mexico and the Caribbean

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CHAPTER 3.1

Introduction

*Francisco Morales**

Establishing an International Network

Organizing an international network on whitefly-transmitted viruses in a region as extensive as Central America, Mexico and the Caribbean is, for various reasons, no easy task. First, whitefly-borne viruses have affected more than a dozen countries and commercial crops in this region. Second, most of these countries lack the necessary research infrastructure or resources to study plant viruses. Third, most of the research conducted in the region has been focused on the whitefly *Bemisia tabaci* (Gennadius) (Homoptera: Aleyrodidae) as a pest and only rarely as a vector. And, finally, communication among researchers in the region remains difficult. However, this was the first task facing partners in Sub-project 2 of the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project. Sub-project 2, Sustainable integrated management of whiteflies as pests and vectors of plant viruses in the tropics, is led by the Centro Internacional de Agricultura Tropical (CIAT) on behalf of the Systemwide Programme on Integrated Pest Management.

Fortunately, a Regional Action Plan for Whitefly Management had been created in 1992 under the initiative of various international institutions such as the Centro Agronómico Tropical de Investigación y Enseñanza (CATIE) in Costa Rica, the Pan American Agricultural School at Zamorano in Honduras and the Instituto Interamericano de Cooperación para la Agricultura (IICA). The network has promoted annual meetings of regional scientists interested in controlling this important pest, mainly under the coordination of CATIE. Another important network that operates in the region is the Programa Cooperativo Regional de Frijol para Centro América, México y El Caribe (PROFRIJOL), financed by the Swiss Government to promote the production of common bean, *Phaseolus vulgaris* L. (Leguminosae), in the region. A main production problem of common bean in the lowlands and the mid-altitude valleys of Central America, Mexico and the Caribbean has been *Bean golden yellow mosaic virus* (Geminiviridae: Begomovirus), a disease caused by a whitefly-transmitted virus. Both PROFRIJOL and CIAT have given considerable attention to this problem since 1978 through a collaborative research agreement. The existence of these networks greatly facilitated the task of organizing an international network to focus specifically on viruses transmitted by the whitefly *B. tabaci* in legume and horticultural crops in the region.

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To further strengthen this network, Sub-project 2 selected three subregional coordinators and three advanced research institutions with previous research experience in each of the three subregions. The first subregion selected was Central America and the advanced research institution chosen to provide technical support was the Plant Pathology Department of the University of Wisconsin, Madison, USA. The subregional coordinator was Dr. Luko Hilje, an entomologist at CATIE, and the responsible scientist for the University of Wisconsin was Dr. Douglas Maxwell. Subregion 2 was Mexico, and its support research institution was the University of Arizona, in Phoenix, USA. The subregional coordinator was Dr. Rafael Rivera Bustamante of the Centro de Investigaciones y Estudios Avanzados (CINVESTAV) in Irapuato Mexico, and the responsible scientist for the University of Arizona was Dr. Judith K. Brown. Subregion 3 was the Caribbean, and its support institution was the University of Florida, Bradenton, USA. The subregional coordinator was Dr. Colmar Serra, of the Instituto Superior de Agricultura (ISA), Dominican Republic, and the responsible scientist at the University of Florida, Dr. Jane Polston. Later on during the development of the project, Drs. Hilje and Serra stepped down from their respective coordination roles and the coordinator of Sub-project 2, Dr. Francisco Morales, assumed these responsibilities.

Assessing Socio-economic and Environmental Impact

The large number of countries included in Sub-project 2 precluded the implementation of socio-economic surveys in each country. Nevertheless, four countries in Central America—Guatemala, El Salvador, Honduras and Costa Rica—were selected to conduct these studies, based on the following considerations. First, national institutions were available that were willing to conduct the surveys. Second, a predominant small-scale farming system could be identified in the three countries and study areas selected. Third, a truly diversified agriculture, including legumes and horticultural crops, could be found in the study areas selected. These conditions were not satisfied in most of the areas surveyed in Mexico or the Caribbean region, with the exception of Cuba, which was excluded on the basis of other logistical constraints.

Another consideration was that, in Mexico and the Caribbean region, statutory measures to counter whitefly and virus epidemics have been successfully implemented. The national program scientists who have had the responsibility of implementing the regulatory measures will discuss the case of the Dominican Republic (Chapter 3.11, this volume).

Emergence of Whitefly-Transmitted Viruses

A number of historical, economic and biological factors have been identified as being associated with the emergence of whitefly-borne viruses in legumes and horticultural crops in the region covered by Sub-project 2. These will be discussed in the individual country papers and in the concluding chapter of this section (Chapter 3.14, this volume). However, in introducing this section, two crosscutting trends can be highlighted. First, the presence of whitefly-transmitted viruses is frequently associated with the cultivation of large-scale commercial or non-traditional horticultural crops, especially for export. Second, the disintegration of national agricultural research programs throughout the study region has had a negative impact on the management of whitefly-related production problems. Basically, the lack of technical assistance to control the whitefly vector has left producers in the hands of pesticide vendors. As a consequence, pesticide abuse has resulted, with serious biological and environmental consequences.

The economic situation in Latin America has been deteriorating since the late 1970s, when the oil crisis generated a gigantic debt for most countries in the region. Possessing only an incipient industrial capacity, most countries had to resort to the export of non-traditional crops such as soybean (*Glycine max* [L.] Merr.) and vegetables. This strategy produced complex changes in the region's agricultural environment that national agricultural research programs in Central America, Mexico or the Caribbean have not addressed. The cropping systems involved have been difficult to analyse in this project because the production losses caused by whitefly-transmitted viruses have forced growers to abandon and replace crops frequently. Nevertheless, the Sub-project partners have been able to document the rapid turnover of crops in whitefly-affected areas and to analyse some of the consequences.

Looking Ahead

A wealth of information, characterizing problems associated with whitefly-transmitted viruses affecting legumes and horticultural crops in Central America, Mexico and the Caribbean was accumulated during this preliminary phase of the TWF-IPM Project. Considerable progress was made in the use of a geographic information system to analyse the data collected, and in the creation of an interactive system to allow potential users to access these data. The amount of data collected across this diverse region considerably exceeded expectations and further analysis will be required to extract all the valuable information it contains. However, when this task is complete, the knowledge gained will provide a sound foundation for developing and implementing sustainable management strategies for whiteflies and whitefly-borne viruses in the region. Figure 1 shows the areas affected by whitefly-transmitted viruses.

In the following chapters, the reader will find a brief description of the different historical, geographic, biological and socio-economic factors that contributed to the emergence of the whitefly *B. tabaci* as a major pest and vector of viruses (referred to here as geminiviruses or, more specifically, as begomoviruses). Concluding chapters draw together results regarding the biotypes and reproductive hosts of *B. tabaci* in this region and, based on a synthesis of all results, make recommendations for further research and implementation directed towards achieving more effective and sustainable management of whiteflies and whitefly-borne virus diseases in the region.

Acknowledgements

The following chapters were written by the senior author based on results obtained at the Plant Virology Laboratory (CIAT, Cali, Colombia) from samples collected by the senior author or collaborators in participating countries, as well as on the available literature and personal observations and experience in the region. Hence, co-authors are not responsible for

Figure 1. Regions in the Neotropics affected by whitefly-transmitted viruses (shaded areas).

omissions or errors found in these chapters. Data provided by collaborating national scientists are dutifully acknowledged in the corresponding chapters. Several national scientists and students collaborating in the project conducted socio-economic studies. Most of the survey data presented have been published as theses by their main authors, also included as co-authors in the corresponding chapters. Special acknowledgements are due to the professional personnel of the CIAT Plant Virology Laboratory, who processed most of the samples of whiteflies and viruses analysed in this extensive survey: Claritza Gómez, Raúl Sedano, Ana Karine Martínez, Ana Cecilia Velasco, Mauricio Castaño, José Arroyave, Iván Lozano, Natalia Villareal and Maritza Cuervo. Special thanks are due to Ms. María del Pilar Hernández, CIAT's whitefly taxonomist, for examining all the whitefly specimens collected during this investigation. Last but not least, we recognize the work of Dr. Lee Calvert, who supervised the molecular characterization of *B. tabaci* biotypes and *Trialeurodes vaporariorum* (Westwood) (Homoptera: Aleyrodidae) specimens collected during this investigation.

CHAPTER 3.2

Mexico

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Introduction

Geographical context

Mexico is the largest country in Middle America, with an area of 1,958,201 km². The country can be divided into different regions, the largest being the Central Plateau, which extends from the United States border in the north to the Isthmus of Tehuantepec in the south. At its northern end, the plateau is about 1320 m above sea level, rising to more than 2650 m south of Mexico City, the country's capital. This Central Plateau is divided into two parts—the Mesa del Norte extends from the United States border to near San Luis Potosí, and the Mesa Central from San Luis Potosí to just south of Mexico City. The Mesa Central is a less arid, higher and flatter plateau than the Mesa del Norte. Although the Central Plateau contains many agricultural basins and valleys, the altitude limits the survival and activity of the whitefly *Bemisia tabaci* (Gennadius). Instead, other whiteflies such as *Trialeurodes vaporariorum* (Westwood) predominate. The latter species can become a major pest on some crops, including common bean (*Phaseolus vulgaris* L.), tomato (*Lycopersicon esculentum* Mill.), potato (*Solanum tuberosum* L.) and other horticultural crops grown above 900 m altitude; but it is not a vector of begomoviruses. Figure 1 shows the main agricultural regions affected by whitefly-transmitted begomoviruses.

The Central Plateau is flanked by the Sierra Madre, two mountain ranges running west and east of the plateau. To the south is the Balsas Depression, a hot, dry and broken area with numerous small basins. Further south is the Mesa del Sur, with small isolated valleys, 1300 m to 1650 m above sea level. The Valley of Oaxaca is the largest and most densely populated of these valleys. The Southern Highlands consist of a series of mountain ranges and plateaux, including the Sierra Madre del Sur. The Isthmus of Tehuantepec is a narrow stretch of lowland (up to about 330 m altitude) with a hilly central area. The presence of *B. tabaci* south of the Central Plateau has not been well documented and it seems that *T. vaporariorum* may predominate as a pest of agricultural significance. The Chiapas Highlands are an extension of the mountain ranges of Central America. The Sierra de Soconusco runs along the Pacific Coast. The Grijalva river valley leads

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Figure 1. The main agricultural regions affected by whitefly-transmitted begomoviruses, Mexico.

northwest into the Tabasco Plain, an extension of the Yucatán Peninsula, bordering the Gulf of Mexico. This peninsula consists of flat, limestone terrain, seldom exceeding 160 m above sea level. The northernmost coastal area of the Yucatán Peninsula is an important horticultural area and has been affected by *B. tabaci* and different begomoviruses transmitted by this vector.

The main horticultural areas of Mexico affected by *B. tabaci* are the coastal lowlands that lie east and west of the Central Plateau. The Gulf Coastal Plain extends some 1400 km from the Texas border to the Yucatán Peninsula. The triangular northern portion is more than 160 km wide near the US border, narrowing southwards until it encounters the Sierra Madre Oriental close to the sea, north of Tampico. This is a swampy area with several lagoons and is called the Zona Lagunera. South of this constriction, the Gulf Coastal Plain runs, narrow and irregular, all the way to the Yucatán Peninsula. The narrower Pacific coastal lowlands begin near the Mexicali Valley in the north and end, also some 1400 km to the south, near Tepic, Nayarit. Parts of these arid regions have been irrigated since the 1930s and support intensive cropping systems of traditional and non-traditional (export) crops. Finally, Baja California is an isolated strip of arid land, about 1280 km long and 160 km wide. Most of the eastern side of this peninsula is mountainous, reaching elevations of almost 3000 m and sloping downwards towards the west.

The emergence of **Bemisia tabaci** *as a pest and virus vector*

The first report of *B. tabaci* as a vector of plant viruses in Mexico, in the early 1950s, is linked to the production of cotton (*Gossypium hirsutum* L.) in the Valley of Mexicali, Baja California (Cárdenas et al., 1996). The disease observed was probably “cotton leaf crumple”, caused by a begomovirus transmitted by *B. tabaci*, previously observed in the United States near the Colorado River Valley (Brown, 1994). Around that time, *B. tabaci* was also associated with a disease of tomato (*Lycopersicon esculentum* Mill.), called ‘chino del tomate’, in the Valley of

Culiacán, Sinaloa, in north-western Mexico (Gallegos, 1978). In 1962, *B. tabaci* was observed attacking cotton in the Soconusco region of the southern state of Chiapas (Cardenas et al., 1996).

In 1978, *B. tabaci* caused considerable damage to horticultural crops in the Huasteca region, including parts of the states of Hidalgo, Tamaulipas, San Luis Potosí and Veracruz, where the Gulf Coastal Plain tapers to form a narrow strip that reaches all the way to the northernmost point of the Yucatán Peninsula. A disease believed to be *Bean golden yellow mosaic virus* (BGYMV) was observed for the first time in the north-western state of Sinaloa, in 1974 (López, 1974). When this disease occurred later (1990) in the state of Sonora, it was shown to be caused not by BGYMV but by a distinct virus, *Bean calico mosaic virus* (BCaMV) (Brown et al., 1990). BCaMV induces more striking golden mosaic symptoms, which eventually resemble a bleaching effect. The virus is not closely related to BGYMV but rather to *Squash leaf curl virus* (SLCV), a virus previously described attacking cucurbits in California (Flock and Mayhew, 1981). BGYMV emerged on the Gulf Coast of Veracruz in 1977. In 1979, it affected common bean plantings in Las Huastecas and in 1980 affected the same crop in the state of Chiapas (López-Salinas, 1994). Finally, in 1980, *B. tabaci* reached the Yucatán Peninsula, particularly the state of Yucatán, where it caused severe yield losses to horticultural crops as a pest and virus vector (Díaz-Plaza et al., 1996).

Most of the subsequent dissemination of whitefly-transmitted begomoviruses in Mexico has been associated with the boom in horticultural and other export crops that took place in the 1980s. The cultivation of melon (*Cucumis melo* L.), soybean (*Glycine max* [L.] Merr.), tomato and peppers (*Capsicum* spp. L.), has created conditions favourable for the reproduction of *B. tabaci* away from its traditional hosts such as cotton. These non-traditional export crops are currently planted in *B. tabaci*-affected regions such as the north-west, Las Huastecas and Yucatán. Among the most important viruses affecting horticultural crops in Mexico may be mentioned: Tomato chino (leaf crumple) virus, described in Sinaloa in 1970 (Gallegos, 1978); Serrano golden mosaic virus, in north-eastern Mexico (Sánchez et al., 1996) and Texas pepper virus (Stenger et al., 1990), both of which are regarded as *Pepper golden mosaic virus* (PepGMV); (Brown et al., 1993); *Pepper Huasteco yellow vein virus* (PHYVV) in pepper and tomato in Las Huastecas and Sinaloa (Hou et al., 1996); and “El Tigre” virus of pepper in Tamaulipas (Brown, 1989). In Yucatán, chilli pepper (*Capsicum* spp.), tomato, squash (*Cucurbita* spp. L.), melon, and watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai) have been attacked since the 1980s by *B. tabaci* and the begomoviruses this whitefly species transmits (Díaz-Plaza et al., 1996).

More recently, *Tomato mottle virus* (ToMoV), a begomovirus originally described from Florida, United States, was detected in the Yucatán Peninsula, also affecting tomato (Garrido and Gilbertson, 1998). In addition, PHYVV was found in Habanero varieties of chilli pepper and in pepper, and PepGMV was found in Habanero. PHYVV had already been detected infecting Jalapeño varieties of chilli pepper in the neighbouring state of Quintana Roo (Díaz-Plaza et al., 1996). Meanwhile, an Old World begomovirus, *Tomato yellow leaf curl virus* (TYLCV) has made its appearance in the Yucatán Peninsula (Ascencio-Ibáñez et al., 1999). This virus has caused yield losses in the

Caribbean estimated at millions of US dollars and thus poses an evident threat to agriculture in Mexico.

Advances in Biological Research

The participation of Mexican researchers in Sub-project 2 of the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project provided an opportunity to gain an updated and more systematic view of the status of whitefly-borne viruses in the country. The Centro de Investigaciones y Estudios Avanzados (CINVESTAV), located at Irapuato, Guanajuato, was selected as the lead institution in Mexico. This Centre conducts research on begomoviruses affecting horticultural crops in Mexico. Dr. Rafael Rivera-Bustamante conducted surveys in collaboration with Dr. Irineo Torres-Pacheco, co-ordinator of the horticulture program of the Instituto Nacional de Investigaciones Forestales y Agropecuarias (INIFAP) at that time.

In the course of this project, samples were taken from crop plants showing begomovirus-like symptoms at a range of sites in major horticultural production areas of Mexico (Table 1) and assayed using monoclonal antibodies (Cancino et al., 1995). In addition, a wide-ranging survey was conducted of viruses affecting tomato and chilli in Mexico (Table 2). The results obtained demonstrate that begomoviruses can attack the main

Table 1. Analyses of plants affected by begomovirus-like diseases in Mexico.

Location	Plant (year)	Begomovirus[a]		Other viruses
		MAB-BS	MAB-GA	
Los Mochis, Sinaloa	Common bean (93)	+	-	-
Los Mochis, Sinaloa	Common bean (93)	+	-	-
J. Rosas, Guanajuato	Tomato	+	-	-
Dzemul, Yucatán	Squash	+	-	-
Dzemul, Yucatán	Chilli	-	-	-
Dzemul, Yucatán	Chilli	-	-	-
Dzemul, Yucatán	Chilli	+	-	-
Los Mochis, Sinaloa	Common bean	+	-	-
Los Mochis, Sinaloa	Common bean	+	-	-
Los Mochis, Sinaloa	Common bean	+	-	-
Los Mochis, Sinaloa	Squash	+	na	-
Los Mochis, Sinaloa	Squash	+	na	-
Los Mochis, Sinaloa	Squash	+	na	-
Culiacán, Sinaloa	Common bean	+	na	-
Culiacán, Sinaloa	Common bean	+	na	-
Culiacán, Sinaloa	Common bean	+	-	-
Culiacán, Sinaloa	Common bean	+	-	-
Culiacán, Sinaloa	Soybean	+	na	-
Culiacán, Sinaloa	Soybean	+	na	-
Culiacán, Sinaloa	Squash	+	na	-
Culiacán, Sinaloa	Squash	-	na	-
Etchojoa, Son.	Common bean	+	-	-

a. MAB-BS, a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses; MAB-GA, a monoclonal antibody used to detect the original Middle American isolates of *Bean golden yellow mosaic virus*-Guatemala; and na, not analysed.

Table 2. Virus survey of tomato and chilli fields in 22 states of Mexico.

Sample	Plant	Locality[a]	State	N	W	V1[b]	V2[b]	V3
1	Sida	Chetumal	Q. Roo	18 30	88 15	TMV	TEV	-
2	Chilli	Chetumal	Q. Roo	18 30	88 15	PHYVV	TPV	-
3	Malvaceae	Campeche	Campeche	19 50	90 26	TMV	TEV	-
4	Weed	Uxmal	Yucatán	20 25	89 42	TMV	-	-
5	Chilli	Campeche	Campeche	19 50	90 26	PHYVV	-	-
6	Chilli	Sayula	Jalisco	19 50	103 30	TPV	-	-
7	Chilli	Sayula	Jalisco	19 50	103 30	TEV	-	-
8	Tomato	Sayula	Jalisco	19 50	103 30	TMV	PHYVV	-
9	Squash	Sayula	Jalisco	19 50	103 30	PHYVV	-	-
10	Euphorbia	Sayula	Jalisco	19 50	103 30	TPV	-	-
11	Sida	Sayula	Jalisco	19 50	103 30	PHYVV	-	-
12	Chilli	Tecomán	Colima	19 01	103 55	TMV	PHYVV	-
13	Chilli	Tecomán	Colima	19 01	103 55	TMV	-	-
14	Chilli	Tecomán	Colima	19 01	103 55	PHYVV	-	-
15	Chilli	Tecomán	Colima	19 01	103 55	TPV	-	-
16	Weed	Tecomán	Colima	19 01	103 55	TPV	-	-
17	Chilli	S. Ixcuintla	Nayarit	21 50	105 10	PHYVV	-	-
18	Chilli	S. Ixcuintla	Nayarit	21 50	105 10	TPV	-	-
19	Tomato	S. Ixcuintla	Nayarit	21 50	105 10	TPV	-	-
20	Chilli	Apaseo Alto	Guanajuato	20 35	100 25	PHYVV	-	-
21	Weed	Celaya	Guanajuato	20 32	100 49	TMV	-	-
22	Malvaceae	Los Angeles	R. Lagunera	25 32	103 30	TMV	-	-
23	Common bean	Los Angeles	R. Lagunera	25 32	103 30	TMV	-	-
24	Tomate	CIAN	R. Lagunera	25 32	103 30	TMV	-	-
25	Melon	CELALA	R. Lagunera	25 32	103 30	TMV	-	-
26	Tomate	CELALA	R. Lagunera	25 32	103 30	TMV	-	-
27	Chilli	Ciudad Isla	Veracruz	18 5.1	95 36	TPV	-	-
28	Chilli	Martinez dlt	Veracruz	20 5.0	97 5.0	PHYVV	TPV	-
29	Chilli	Papantla	Veracruz	20 30	97 12	PHYVV	-	-
30	Chilli	Papantla	Veracruz	20 30	97 12	PHYVV	TPV	-
31	Chilli	Poza Rica	Veracruz	20 33	97 26	PHYVV	-	-
32	Melampodium	Veracruz	Veracruz	19 9.5	96 6.4	TMV	CMV	TEV/TPV
33	Chilli	Guegovela	Oaxaca	16 46	96 49	TEV	-	-
34	Datura	S.L.Cacaote	Oaxaca	17 7.9	96 47	TPV	-	-
35	Tomato	Guegovela	Oaxaca	16 46	96 49	TEV	-	-
36	Chilli	Pabellón	Aguascal.	21 55	102 15	CMV	-	-
37	Chilli	Jalpa	Aguascal.	21 55	102 15	TEV	-	-
38	Malvaceae	Jalpa	Aguascal.	21 55	102 15	TEV	-	-
39	Chilli	Jaral Progr.	Aguascal.	20 30	101 6	TEV	TMV	CMV
40	Chilli	Jaral Progr.	Aguascal.	20 30	101 6	TEV	TMV	-
41	Chilli	Jaral Progr.	Aguascal.	20 30	101 6	TEV	CMV	-
42	Chilli	Paracuaro	Guanajuato	20 15	100 48	TEV	CMV	-
43	Chilli	Paracuaro	Guanajuato	20 15	100 48	TEV	CMV	-
44	Malvaceae	S. Luis Paz	Guanajuato	21 33	100 32	TMV	-	-
45	Malvaceae	S. Luis Paz	Guanajuato	21 33	100 32	TEV	-	-
46	Tomato	Yautepec	Morelos	18 50	99 11	PHYVV	-	-
47	Physalis	Yautepec	Morelos	18 50	99 11	PHYVV	-	-
48	Chilli	Yautepec	Morelos	18 50	99 11	PHYVV	TPV	-
49	Tomato	Metztitlán	Hidalgo	20 38	98 39	PHYVV	TPV	-
50	Chilli	Metztitlán	Hidalgo	20 38	98 39	PHYVV	-	-
51	Chilli	Salitral	S.L. Potosí	22 53	102 02	PHYVV	TPV	-
52	Tomato	Salitral	S.L. Potosí	22 53	102 02	PHYVV	-	-
53	Sida	Salitral	S.L. Potosí	22 53	102 02	PHYVV	-	-
54	Chilli	Apaseo	Querétaro	20 35	100 25	PHYVV	-	-

a. CIAN and CELALA are experiment stations in the Lagunera region of Mexico.

b. TMV, *Tobacco mosaic virus*; TEV, *Tobacco etch virus*; PHYVV, *Pepper Huasteco yellow vein virus*; TPV, *Texas pepper virus* (= *Pepper golden mosaic virus*); CMV, *Cucumber mosaic virus*.

horticultural crops grown in the country: common bean, tomato, chilli and squash. Additionally, a begomovirus was detected in soybean plants in Sinaloa State. Soybean is a host for *B. tabaci* in South America but its role in Mesoamerica as a reproductive host for this whitefly seems to have been secondary to that of other crops such as cotton and some horticultural crops. Nevertheless, *B. tabaci* reportedly destroyed over 5000 ha of soybean in the Valle del Fuerte, Sinaloa, in 1994 (INIFAP, 1995).

As noted above, golden mosaic symptoms in common bean in northern Mexico are caused by BCaMV, which is related to SLCV, a cucurbit virus originally recorded from the United States. In this project, we surveyed three key locations in the north-western region of Mexico—Etchojoa in Sonora, and Los Mochis and Culiacán in Sinaloa state—and took samples of common bean plants affected by symptoms of the disease. A partial molecular characterization of the viral isolates collected (Table 3) shows that BCaMV is still present in common bean plantings in north-western Mexico, specifically in Los Mochis. This isolate is still closely related to an isolate of BCaMV collected in the same locality in 1986. The sample from Culiacán represents a begomovirus closely related to BCaMV but already evolving into a new species (as indicated by a sequence homology of less than 90%). Perhaps of even greater interest is that the common bean begomovirus from Sonora is still identifiable as an isolate of the original SLCV, constituting the first direct evidence of the evolution of this virus.

Selected whitefly samples were also collected on different crops in north-western Mexico. Table 4 presents the

Table 3. Comparative sequence homologies (%) of common bean begomoviruses from north-western Mexico (Sonora and Sinaloa) and other whitefly-transmitted begomoviruses.

Virus isolate	ORF[a]	Common bean begomoviruses[b]				
		BGYMV-GA	BCaMV	SLCV	BCaMV-MO	BDMV
Etchojoa, Sonora	*AC1*	57.0	77.2	96.0	76.8	61.0
	AV1	72.2	75.4	97.8	74.9	74.2
Los Mochis, Sinaloa	*AC1*	69.8	89.7	70.5	88.3	68.8
	AV1	74.4	92.2	76.1	92.2	73.9
Culiacán, Sinaloa	*AC1*	74.3	83.0	69.1	81.8	74.6
	AV1	73.0	86.1	79.2	85.7	74.2

[a] ORF, open reading frame; *AC1*, replicase; and *AV1*, capsid protein.

[b] BGYMV-GA, *Bean golden yellow mosaic virus*-Guatemala; BCaMV, *Bean calico mosaic virus*; SLCV, *Squash leaf curl virus*; MO, Mochis; BDMV, *Bean dwarf mosaic virus*.

Table 4. Identification of *Bemisia tabaci* biotypes from Sinaloa, northern Mexico.

Locality	Crop	Biotype A	Biotype B
Los Mochis	Common bean	X	-
Los Mochis	Common bean	X	-
Los Mochis	Common bean	-	X
Los Mochis	Common bean	-	X
Los Mochis	Common bean	-	X
Culiacán	Eggplant	X	-
Culiacán	Eggplant	X	-
Culiacán	Eggplant	X	-
Culiacán	Eggplant	X	-
Culiacán	Eggplant	X	-

results of the molecular biotyping of the whiteflies collected. As can be observed, there is a mixed population of biotypes A and B of *B. tabaci*.

Finally, the project supported work conducted by INIFAP researcher, Genovevo Ramírez-Jaramillo, based in Mérida, Yucatán, on “the bio-ecology of the whitefly *B. tabaci* in the Yucatán Peninsula.” This work—which also included the Government of the State of Yucatán, the Secretary of Agriculture, Livestock and Rural Development, the Regional Research Centre for the South-east and the Fundación Produce de Yucatán—involved developing a geographical information system (GIS)-based model to predict the probability of whitefly and begomovirus attack.

According to the classification of Toledo (1989), Mexico can be divided into five major ecological zones. Interestingly, the northernmost portion of the Yucatán Peninsula belongs to the Dry Tropics (*Trópico Seco*), the same ecological zone as the northern Pacific Plains, where the *B. tabaci* thrives. In this dry zone, the annual precipitation ranges between 600 and 1500 mm and the mean annual temperature is over 20 °C. Although Yucatán is not one of the country’s main horticultural regions, in 1992 this peninsula cultivated over 11,000 ha of Habanero and Jalapeño chilli pepper, watermelon and tomato (listed in declining order of production area). However, the agriculture of the peninsula includes a diversity of other crops, including species that can act as reproductive hosts of *B. tabaci* such as soybean and cotton of which over 500 ha of each are grown.

The GIS-based model for whitefly and virus outbreaks drew on three components of a database developed by INIFAP: (1) a digital elevation model, (2) a climate database and (3) a soils database. For the biological parameters, a mean monthly temperature of 23-27 °C and annual rainfall of less than 50 mm were chosen as defining areas with a high probability of *B. tabaci* infestation. Whitefly population dynamics in Yucatán were influenced to a large extent by the amount of rainfall but only minimally by the temperature, which remains within a range that is favourable for whitefly development (Figure 1). Thus, the peak populations of *B. tabaci* occurred in the months of May and June, following 4-5 months (December-April) of low precipitation (0-18 mm/month). Based on all of these parameters, risk assessment maps for the Yucatán Peninsula were produced. The assessments will allow recommendations to be issued regarding the most appropriate planting dates for those crops most likely to be affected by *B. tabaci*, both as a pest and vector of plant viruses.

Socio-economic Analysis

The results of the biological surveys conducted during the course of the project show that whitefly-transmitted viruses and *B. tabaci* affect several crops of social and economic importance throughout Mexico, with the exception of the Central Highlands, where other whitefly species predominate.

In the Mexicali Valley, Baja California and the San Luis Río Colorado region of Sonora, *B. tabaci* attacked fields of cotton (damaging some 14,000 ha), summer melon (causing the loss of 1500 ha) and sesame (*Sesamum indicum* L.) (damaging 7500 ha) in the 1991-92 season, causing nearly US$10,000,000 in production losses (López, 1996). Some 3000 ha of Serrano chilli and tomato crops were lost between 1992 and 1995 in the central region of San Luis Potosí State, because of damage by *B. tabaci* and begomoviruses. Other crops colonized by *B. tabaci* in this region were common bean, cotton, *tomatillo* (*Physalis ixocarpa* B.), squash, melon, watermelon, cucumber (*Cucumis sativus* L. var. *sativus*), sweetpotato (*Ipomoea batatas* [L.] Lam.) and sunflower (*Helianthus annuus* L.) (Hernández et al., 1996).

B. tabaci caused severe yield losses to various horticultural crops, especially melon, in the Comarca Lagunera, Durango, in 1993 (Morales et al., 1996). *B. tabaci* attacked melon, chilli and cotton in the state of Coahuila, northern Mexico. The investigation that was conducted at the time showed that melon was an excellent host, cotton was a good host and chilli did not adequately support the reproduction of *B. tabaci* (Nava and Riley, 1996). Sonora has been one of the states in northern Mexico most affected by infestations of *B. tabaci* on various crops. An epidemiological study in the Valley of the Yaqui indicated that one of the main reproductive hosts of the whitefly in the valley was soybean (Pacheco, 1996). The incidence of whitefly-transmitted begomoviruses in common bean plantings in the valley of El Fuerte, Sinaloa, caused yield losses estimated at 20%-30% in the entire region, between February and June 1994 (Salinas et al., 1996). *B. tabaci* has been increasing its populations on cucumber in the valley of Culiacán, Sinaloa, since 1993, causing significant yield losses in over 9000 ha of this crop (Avilés and Valenzuela, 1996).

In the state of Nayarit, the fall-winter cropping cycle has been reduced by 50% because of the direct and indirect damage caused by *B. tabaci* as a pest and vector of viruses in tomato, common bean, chilli and eggplant (*Solanum melongena* L.) (Ortíz et al., 1996). Viruses transmitted by *B. tabaci* have severely affected tomato plantings in the state of Guerrero. In order to harvest tomato, farmers have resorted to treating their fields twice a week with insecticides (Barajas, 1996). And in the state of Yucatán, *B. tabaci* and whitefly-borne begomoviruses have affected horticultural crops such as hot peppers, tomato, squash, melon and watermelon since the 1980s (Díaz-Plaza et al., 1996).

The social and economic impact of these pest and disease outbreaks must be enormous. In the mid-1990s, horticultural and fruit crops occupied over 1 million hectares in Mexico. About 7 million tons of produce, mainly tomato, melon and watermelon were exported (principally to the United States, Canada, Japan and Brazil). The rest, some 35 million tons of squash, melon, pepper, tomato, potato, eggplant and chillis, was absorbed by the national market for local consumption, underlining the importance of these crops to the domestic economy. The production of horticultural crops demands considerable human labour. For instance, tomato requires 271 work-days per cropping cycle as compared with only 24 for maize (*Zea mays* L.). Production costs for maize were US$430 per hectare compared with US$3800 per hectare for tomato in 1990. The marketing of horticultural products is difficult because of their perishable nature and price instability. In 1988, over 50% of the farmers producing horticultural products in Mexico had less than 5 ha of land (FIRA, 1997). Overall, the incidence of whitefly-transmitted viruses affects a large portion of Mexico's rural population, many of whom can be considered small-scale farmers.

Strengthened Research Capacity

In addition to its research activities, CINVESTAV offers training to visiting scientists from developing countries. During the course of this project, two Cuban scientists conducted research on *Taino tomato mottle virus* (TToMoV), a whitefly-transmitted begomovirus affecting tomato in Cuba (Ramos et al., 1997). The project also contributed limited funds for research on viruses of economic importance in horticultural crops to the Horticulture Program of INIFAP, and paved the way for continuing collaboration between INIFAP and the current Tropical Whitefly Project. (The state of Yucatán is the main target area for the current Phase II of the project.)

Current Status of Whitefly/Begomovirus Problems

The whitefly/begomovirus situation in Mexico is rather complex because of the size and diversity of the country—and because of the introduction or emergence of different whitefly-transmitted viruses in both the north and the south. In the north, the intensive cropping systems for non-traditional export crops share not only a common border but also common whitefly/begomovirus problems with the equally intensive agricultural systems of the south-western United States. In the south, commercial and small-scale subsistence agriculture co-exist as an extension of the cropping systems found in Central America. Thus, it is not surprising to find some North American begomoviruses in northern Mexico and some Central American begomoviruses in southern Mexico.

Whereas the begomovirus situation on common bean has been partially controlled through the use of resistant cultivars, the situation of other horticultural crops affected by whitefly-transmitted begomoviruses such as tomato, pepper, chilli and melon is not yet resolved in most regions. Nevertheless, Mexico has made considerable progress in implementing integrated pest management (IPM) measures such as biological control, physical barriers (micro-tunnels) and other relevant cultural practices, and in developing the necessary knowledge base, including, for instance, virus characterization. Mexico is also at the forefront in using GIS technology for more effective management of agricultural resources.

More research is needed to study the epidemiology of *B. tabaci* biotypes and other whitefly species in agricultural regions with altitudes above 1000 m, including those in the states of Oaxaca, Guerrero, Morelos, Mexico, San Luis Potosí, Guanajuato and Durango. Suitable IPM measures should be rapidly implemented and evaluated to reduce dependence on chemical insecticides for whitefly control.

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CHAPTER 3.3

Guatemala

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Introduction

Geographical context

Of the seven countries that comprise the isthmus of Central America, Guatemala is physically and culturally the most diverse. The highlands of the southern half of Guatemala descend southward to the Pacific Coast to form the fertile “Piedmont” or “Coffee Belt” (500-1500 m above sea level) and the hot, grass- and forest-covered lowlands known as “The Coast” (0-500 m). The northern regions of Guatemala are tropical lowlands covered by rain forest and scattered savannahs, which are only sparsely populated (West and Augelli, 1977). However, it is the south-eastern region of the country (“El Oriente”), formed by low- and mid-altitude (200-1000 m) valleys, which has suffered the worst attacks from the whitefly *Bemisia tabaci* (Gennadius) and different begomoviruses transmitted by this whitefly species. This region has drastically changed its traditional agricultural environment to join the boom in export crops, thus creating complex and fragile cropping systems. Figure 1 shows the main agricultural regions affected by whitefly-transmitted begomoviruses.

Figure 1. The main agricultural regions affected by whitefly-transmitted begomoviruses, Guatemala.

The emergence of **Bemisia tabaci** *as a pest and virus vector*

The civil war in the United States in the 1860s created an increased demand for cotton (*Gossypium hirsutum* L.). Soon, cotton produced in the Pacific lowlands of the country comprised 20% of Guatemala’s exports. This species was probably one of the first commercially cultivated hosts of the whitefly *B. tabaci*. A second boom of cotton production took place in the 1950s and

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1960s, and at this time the first viruses transmitted by whiteflies were observed in Guatemala and neighbouring countries (Gill, 1994).

Tobacco (*Nicotiana tabacum* L.) is another popular export crop that continues to be grown in Guatemala. The cultivation of tobacco in south-eastern Guatemala has been blamed also for increasing the population of the virus vector *B. tabaci* in the 1970s, thus provoking the first outbreaks of *Bean golden yellow mosaic virus* (BGYMV) in common bean (*Phaseolus vulgaris* L.) in that region of Guatemala (Morales, 1994).

In 1974, the Horticultural Research Division of the Instituto de Ciencia y Tecnología Agrícolas (ICTA) initiated extension work in the Zacapa Department with a view to producing export crops such as melon (*Cucumis melo* L.). At that time, the main production problems identified were outdated agronomic practices, inadequate cultivars and the poor quality of the produce, which could only meet the relatively low standards of the local markets. A similar industry to produce tomato (*Lycopersicon esculentum* Mill.) was also initiated in the late 1970s in south-eastern Guatemala (Gaytán, 1979). These crops provided better reproductive hosts for *B. tabaci* than either common bean or wild plant species. The populations of the vector soon soared and then new begomoviruses emerged to attack the new crops. The arrival of the B biotype of *B. tabaci*, a more aggressive and polyphagous variant of the original species, further aggravated yield losses, with the new whitefly acting as both a pest and a vector of damaging plant viruses (Dardón, 1992).

The first collaborative project that set out to tackle the new and growing threat of whitefly-borne viruses was the Programa Cooperativo Regional de Frijol para Centro América, México y el Caribe (PROFRIJOL), initiated by the Centro Internacional de Agricultura Tropical (CIAT) to address the bean golden yellow mosaic problem. This project, initiated in 1978 with support from the United Nations Development Programme (UNDP) and subsequently funded (from 1980) by the Swiss Development Cooperation (SDC), is credited with the development of several BGYMV-resistant (DOR) lines that have subsequently been widely adopted as commercial varieties in Mexico, Central America, the Caribbean and South America.

In Guatemala, the project began work under the auspices of ICTA in the south-eastern Monjas Valley, about 960 m above sea level. Until the 1970s, this valley had been planted to traditional crops, mainly common bean and maize (*Zea mays* L.). In the 1980s, horticultural crops such as tomato and broccoli (*Brassica oleracea* L. var. *italica* Plenck) made their appearance in the valley. Other non-traditional crops—pepper (*Capsicum* spp. L.), cucumber (*Cucumis sativus* L. var. *sativus*) and more recently watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai) and grape (*Vitis vinifera* L.)—have also found a niche in this valley over the latter part of the 1990s. Tobacco is a large-scale commercial crop that was first planted in the valley during the early 1970s; its production collapsed in the late 1980s, only to make a comeback in recent years.

BGYMV, appearing in the mid-1970s, was the first whitefly-transmitted virus to cause severe yield losses in the Monjas Valley (Rodríguez, 1994). Tobacco was probably the most important reproductive host of *B. tabaci*. The local common bean cultivars, Pecho Amarillo and Rabia de Gato, soon succumbed to the disease. Fortunately, the PROFRIJOL project began to show results and in due course released the first disease-resistant breeding lines: ICTA-Quetzal, ICTA-Jutiapán and ICTA-Tamazulapa (Rodríguez, 1994). The causal agent, initially believed to be *Bean golden mosaic virus* (BGMV), was characterized at the molecular level, thanks to collaborative research financed by the United States Agency for International Development and carried out at the University of Wisconsin and at CIAT. The Guatemalan virus in the early 1990s was shown to be a distinct species, different from the Brazilian isolates of BGMV (Faria et al., 1994) but with a considerable degree of homology and causing a very similar disease; it was therefore given the name *Bean golden yellow mosaic virus*. Maize is not a host of the whitefly *B. tabaci* and is not attacked by these particular begomoviruses, although maize in Africa is infected by different geminiviruses (Geminiviridae: Mastrevirus) transmitted by leafhoppers.

Advances in Biological Research

Further research, undertaken in collaboration with the University of Florida, was intended to produce monoclonal antibodies (MABs) against different isolates of BGYMV, using the Guatemalan (MAB-GA) isolate of BGYMV. At the same time, the project sought to produce a broad-spectrum monoclonal antibody (MAB-BS), capable of detecting most whitefly-transmitted begomoviruses (Cancino et al., 1995). The results shown in Table 1 indicate that these objectives were realized, in as much as MAB-BS has been able to detect all bi-partite begomoviruses encountered to date in Guatemala, but MAB-GA reacts only with the original Middle-American BGYMV isolates. The result that MAB-GA, raised against the original isolate from Monjas, does not react with isolates subsequently collected from this area suggests that some change in the antigenic properties of the virus has occurred or that a new isolate has been introduced and become established in the area. Begomoviruses are known to adapt to different host plants and vector species. It has been suggested that the changes observed in the antigenic reaction of the Monjas isolates may be associated with the displacement of the A biotype of *B. tabaci* by the B biotype, which is believed to have occurred in the 1990s.

Table 1. Antigenic properties of common bean begomoviruses from Guatemala.

Isolate	Locality	MAB-BS[a]	MAB-GA[b]
1	Monjas	+	+
2	Tecpán	+	-
3	Cuyuta	+	-
4	Monjas	+	-
5	Monjas	+	-
6	San Jerónimo	+	-
7	Quiaté	+	-

a. MAB-BS, a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses.

b. MAB-GA, a monoclonal antibody used to detect the original Middle American isolates of *Bean golden yellow mosaic virus*-Guatemala.

Characterization of whiteflies collected from sites across Guatemala confirms that the B biotype has displaced the A biotype almost completely (Table 2). This would be consistent with the hypothesis that the antigenic changes in the virus detected with the specific MAB may be related to the presence of a new vector. To explore this possibility further, the coat protein of a recent isolate of BGYMV from the locality of Monjas was partially sequenced and compared with the original isolate. Table 3 shows the comparative results obtained.

Table 2. Results of whitefly surveys in different agricultural regions of Guatemala. Data are numbers of samples that included *Bemisia tabaci* biotype A (B.t.-A), *B. tabaci* biotype B (B.t.-B) or other species of whitefly (other spp.).

Crop	Samples	Locality[a]	B.t.-A	B.t.-B	Other spp.
Melon	12	Cuyuta-Jutiapa	0	12	0
Melon	6	Río Hondo-Zacapa	0	6	0
Watermelon	10	La Fragua-Zacapa	0	10	0
Watermelon	22	L.de Retana-Jutiapa	0	2	20
Cucumber	17	Usumatlán-Zacapa	0	17	0
Cucumber	10	AMCO-Jutiapa	0	10	0
Cucumber	10	S. Agustín-Jutiapa	0	10	0
Okra	7	La Fragua-Zacapa	0	7	0
Tobacco	2	Santa Cruz-Jutiapa	0	2	0
Tomato	2	Miramar-Jutiapa	2	0	0
Tomato	1	AMCO-Jutiapa	0	1	0
Eggplant	10	San Agustín-Jutiapa	0	10	0

a. AMCO-Jutiapa, an experiment station.

Table 3. Comparative analysis of partial coat protein sequence homologies (%) among viruses associated with *Bean golden yellow mosaic virus*, comparing the isolate GA-2, collected in Guatemala during the project, with GA-1, the original isolate, and isolates from the Dominican Republic (DR), Puerto Rico (PR) and *Bean golden mosaic virus*- Brazil (BR).

Virus	GA-2	GA-1	DR	PR	BR
GA-2	100	92.7	92.3	90.0	76.0

These results show a divergence in coat protein nucleotide sequence of ca. 7.3% between the original and recent Guatemalan isolates. This difference is significant, considering that when sequences of two isolates diverge by more than 10% they are normally regarded as distinct species—as in the case of the difference observed between BGYMV-GA and BGMV-BR (see Table 3).

Table 4 shows the results of serological and other complementary diagnostic tests carried out on plant samples taken from selected crops in whitefly-affected regions in Guatemala. These results demonstrate the presence of whitefly-transmitted begomoviruses in most horticultural crops assayed, with the exception of cucumber. Tobacco is also beginning to be attacked by begomoviruses, as production of this crop increases once more. Tobacco, however, was observed to be principally affected by *Tobacco mosaic virus* (TMV) (*Tobamovirus*), a highly infectious virus that is spread by workers entering the field. A similar virus, probably *Tomato mosaic virus*, was also found infecting tomato in the south-eastern region of Guatemala, although the two viruses may be strains of TMV adapted to each crop.

The presence of *Pepper golden mosaic virus* in pepper was confirmed in samples taken from the locality of Laguna de Retana. The collaborating Plant Pathology Laboratory of the University of Wisconsin had detected this virus already in Guatemala (Douglas Maxwell, personal communication, 2003).

Socio-economic Analysis

Over half of Guatemala's total population of some 12 million people are of pure Amerindian stock. This sector of the population is concentrated

Table 4. Results of assays carried out on selected plant samples from Guatemala.

Sample	Locality	Plant	Reaction[a]		EM[b]
			MAB-BS	PTY1	
1	Zacapa	Melon	+	-	nt
2	Zacapa	Melon	-	+	nt
3	Monjas	Tobacco	+	nt	nt
4	Monjas	Tobacco	+	nt	TMV
5	El Ovejero	Tobacco	-	nt	TMV
6	L. de Retana	Tomato	+	nt	nt
7	L. de Retana	Tomato	-	-	nt
8	L. de Retana	Tomato	+	nt	nt
9	Zacapa	Tomato	-	-	nt
10	Las Conchas	Tomato	-	nt	TMV
11	Las Conchas	Tomato	-	nt	TMV
12	Las Flores	Tomato	+	nt	nt
13	L. de Retana	Tomato	+	nt	nt
14	L. de Retana	Tomato	+	nt	nt
15	Plan de la Cruz	Tomato	+	nt	nt
16	Plan de la Cruz	Tomato	+	nt	nt
17	Monjas	Tomato	+	nt	nt
18	Monjas	Tomato	+	nt	nt
19	Monjas	Tomato	+	nt	nt
20	L. de Retana	Pepper	+	nt	nt
21	L. de Retana	Pepper	-	nt	-
22	L. de Retana	Pepper	+	nt	nt
23	L. de Retana	Cucumber	-	nt	-
24	L. de Retana	Cucumber	-	nt	-

a. Positive or negative reactions to a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses (MAB-BS); reactions to a potyvirus-specific monoclonal antibody (PTY1); and nt, no tests for these samples.

b. Detection of *Tobacco mosaic virus* (TMV) by electron microscopy (EM); and nt, no tests for these samples.

in the highlands, north and west of Guatemala City, as well as in the region of Petén. South-eastern Guatemala is the region with the lowest proportion of native Americans. The presence of mestizos or *ladinos* in the *Oriente* of Guatemala explains the more dynamic and complex cropping systems in these drier lowlands, including the prevalence of non-traditional export crops.

A special study was conducted in the municipality of Monjas, Jalapa Department, where Amerindians still make up over 70% of the total population. The Monjas Valley is located at an altitude of 960 m and has an average annual precipitation of 900 mm, most of which falls between May and October. The mean temperature is 23.7 °C and the mean relative humidity is 80%. Ecologically, the Monjas Valley is classified both as dry subtropical forest and temperate subtropical humid forest. Most of the agricultural land is private property, although it may have been acquired by “right” and not through a commercial transaction.

Table 5 shows the existing cropping systems in the Monjas Valley. Maize and common bean are evidently

Table 5. Cropping systems in the Monjas Valley, Jalapa, Guatemala.

Crop	Years of planting	Irrigation	Rain-fed
Common bean	>50	-	+
Maize	>50	-	+
Tobacco	25	+	+
Tomato	15	+	-
Broccoli	10	+	-
Onion	10	-	+
Pepper	7	+	+
Cucumber	5	+	-
Sweet corn	3	+	-
Grape	3	+	-

the most traditional crops in the valley. Tobacco has been planted for over 20 years as a commercial crop but almost disappeared in the late 1970s because of market problems, coming back into favour only in the late 1990s. Onion (*Allium cepa* L.) is another crop that has been planted in the valley for over 10 years but it has never become prevalent. Broccoli has been a very popular crop in the valley in the past decade and pepper has been commercially cultivated during the past 7 years. Cucumber has been planted commercially for the past 5 years and watermelon for the past 2 years, although it has been planted for local consumption for decades in the valley. Finally, sweet corn (*Zea mays* L. subsp. *mays*) and grape have been introduced in the past 3 years.

Table 5 also shows the association between commercial crops and the use of irrigation in the Monjas Valley. Subsistence crops such as maize and common bean are planted during the rainy season. The use of irrigation demonstrates the intention of local growers to minimize risk in the case of high-value crops.

Table 6 shows the perception that growers have of the whitefly *B. tabaci*, both as a pest and virus vector in the crops listed above, and the number of pesticides used in each crop. The results clearly illustrate that pesticide abuse occurs in most of the crops affected by whiteflies. In the case of tomato, only five of the 11 pesticides used are specific to the control of whitefly; the remaining six pesticides are applied as “repellents” in the belief that the “smell” of these products repels the whitefly. The low use of insecticides on broccoli is interesting and responds to demand from foreign buyers (many of whom only allow bio-pesticides to be used).

Table 6. Growers’ perception of whitefly problems and growers’ use of pesticides in selected cropping systems of the Monjas Valley, Jalapa.

Crop	Whitefly problem	Pesticides used (no.)
Common bean	Yes	8
Maize	No	1
Tobacco	Yes	9
Tomato	Yes	11
Broccoli	Yes	1
Onion	No	-
Pepper	Yes	5
Cucumber	Yes	10
Sweet corn	No	1
Grape	No	-

In general terms, whiteflies are the main pest on common bean, tomato, broccoli and tomato. Other pests such as white grubs and lepidopterous larvae also cause significant damage to tobacco, tomato and sweet corn. Slugs and chrysomelids are perceived as important pests of common bean in the valley. Diamond-back moth, *Plutella xylostella* (Linnaeus), is also a pest of broccoli; and the corn ear worm, *Helicoverpa zea* (Boddie) causes severe damage to maize and sweet corn. The existence of insect pests other than whiteflies in the various crops grown in the Monjas Valley hampers efforts to reduce the use of chemical pesticides against whiteflies. In fact, most growers of non-traditional export crops use “cocktails”, or “bombs”, as pesticide mixtures are locally called.

Whereas growers recognize *B. tabaci* as a pest, they do not fully understand its role as a vector of viruses. However, they readily associate the bright yellowing shown by infected common bean plants with a disease (bean golden yellow mosaic). In the case of tomato, growers call symptoms expressed by begomovirus-infected plants *acolochamiento* (curling). In the case of tobacco, growers recognize the severe malformation and stunting symptoms shown by virus-infected plants. However, they cannot distinguish between whitefly-transmitted begomoviruses and TMV.

According to most growers, whiteflies are a regular pest, year after year, particularly during the summer time because of the higher temperatures and relative humidity. Growers also perceive the complex cropping systems in the valley as a negative factor because they claim that these crops represent more food sources for the whitefly. Finally, growers blame ratoon crops for the survival of whiteflies between summer and winter seasons. Growers believe in the existence of whitefly-resistant or more generally pest- and disease-resistant crops, particularly tomato, tobacco and common bean. However, the “resistance” of tomato and tobacco seems to be associated with the implementation of more successful chemical control practices in the valley. In reality, the case of common bean is a good example of genetically controlled virus resistance obtained through a well-designed breeding programme.

A general question on the ability of growers to predict seasons of high or low whitefly incidence was generally misinterpreted. Most growers responded that they were always ready to control whiteflies in seedbeds, after transplanting and during the vegetative and early reproductive phases of the crop. In fact, some growers indirectly answered the question, associating mild winters and an abundance of alternative crops with higher whitefly populations and crop damage.

Strengthened Research Capacity

The Tropical Whitefly Integrated Pest Management (TWF-IPM) Project financed two case studies conducted by ICTA agronomists, in the departments of Jalapa, Jutiapa and Baja Verapaz. These departments used to be traditional agricultural areas, devoted to the planting of food staples such as maize and common bean. The boom in export crops, however, rapidly displaced traditional crops and drastically changed the socio-economic structure of the rural communities found in these departments. The studies conducted in these regions provided information on the positive aspects (potential higher income) and negative consequences (new biotic problems and pesticide abuse) of non-traditional export crops. The first study was conducted in 15 rural communities, located in three municipalities of the departments of Jalapa and Jutiapa. The second case study was conducted in five rural communities in the municipalities of San Jerónimo, Rabinal and San Miguel Chicaj. This study helped to develop non-chemical strategies for whitefly control and generated information of value to ICTA in its attempts to adopt suitable research policies to assist small-scale farmers.

Although training in advanced laboratory techniques was not contemplated in this preliminary phase of the project, it became apparent that a number of agricultural scientists in Central America, Mexico and the Caribbean have adequate laboratory facilities to conduct such work. These scientists are becoming increasingly reluctant to act simply as “guides” or “providers” of biological samples to foreign scientists from advanced research laboratories that often visit developing countries in search of interesting scientific materials. Although the aim of this project was to conduct research in a truly collaborative manner, in which national scientists have preferential access to the results of all research undertaken, the possibility of training national scientists in advanced molecular techniques became a need. The two research areas in which advanced techniques are being implemented in this project are in the molecular identification of whitefly biotypes and of the begomoviruses they transmit.

In Guatemala, the identification of begomoviruses is being done at the University of San Carlos with the support of the Plant Pathology Laboratory of the University of Wisconsin. The molecular characterization of *B. tabaci* biotypes is being carried out at the University of El Valle, Guatemala City, which has adequate facilities to conduct research in the area of molecular biology but did not have the technology to identify whitefly biotypes. Consequently, a research assistant from the relevant laboratory received 1 week of intensive training at CIAT on the molecular characterization of *B. tabaci* biotypes and was able to carry out this work successfully upon return to Guatemala.

Thus, the project helped national scientists to conduct surveys that improved both the biological and socio-economic knowledge base on which research and implementation of agricultural policy are founded. The research sector of Guatemala was also strengthened through the training of scientists from advanced research laboratories in the country in techniques used for the identification of whitefly biotypes and the begomoviruses that affect food and export crops of social and economic importance.

Current Status of Whitefly/Begomovirus Problems

The whitefly/begomovirus problems in Guatemala are concentrated in the south-eastern agricultural regions of the country (Figure 1). This region contains most of the country’s mid-altitude valleys (at 500 to 1000 m altitude) with an average annual rainfall below 1500 mm, and temperatures between 20 and 25 ºC; these valleys are also characterized by highly diverse cropping systems, including irrigation. All of these factors favour the reproduction of *B. tabaci*. Moreover, the concentration of non-traditional export crops in this region is associated with the intensive use of pesticides, regularly used to protect these high-value crops. Pesticide abuse is another predisposing factor for the establishment of *B. tabaci* as a pest. Any crop planted in the Pacific lowlands will be severely attacked by whitefly-transmitted begomoviruses particularly because of the higher temperatures, which increase disease severity.

The current begomovirus situation is becoming more complex as new whitefly-transmitted viruses are introduced from neighbouring countries, or evolve from the indigenous virus population, or arise through genetic recombination between the existing begomoviruses that affect the diverse crops such as common bean, tomato, pepper, squash (*Cucurbita* spp. L.), melon and tobacco that are grown in this area. Only in the case of common bean has there been a sustained effort on the part of the national program and CIAT (with Swiss funding) to develop and release virus-resistant cultivars. For the remaining crops, the emphasis has been on chemical control, although large agro-export companies have adopted some IPM packages. South-eastern Guatemala provides an ideal site to conduct a case study with small-scale farmers on the implementation of practical IPM measures to combat *B. tabaci* and the begomoviruses that this species transmits to food and industrial crops in this region.

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CHAPTER 3.4

El Salvador

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Introduction

Geographical context

El Salvador lies entirely within a volcanic region but most of its territory is formed by mid to low altitude valleys (up to 1000 m). About 40% of El Salvador consists of agricultural land—the highest proportion in Latin America, where it is the smallest country but the most densely populated. Given the shortage of land, the higher slopes are dedicated to crops such as coffee (*Coffea arabica* L.). Farmers make up over 50% of the population, primarily producing subsistence crops such as maize (*Zea mays* L.) and common bean (*Phaseolus vulgaris* L.).

Cotton (*Gossypium hirsutum* L.), undoubtedly the second most important commercial crop in El Salvador in past decades (for export to Japan), is mostly found in the Pacific lowlands. Sugarcane (*Saccharum officinarum* L.) became an important cash crop at the turn of the century and a third of the sugar produced is exported to the United States. Rice (*Oryza sativa* L.) is another important crop as well as some cucurbits such as watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai) (Pastor, 1988). More recently, the production of melon (*Cucumis melo* L.), pepper (*Capsicum* spp. L.) and tomato (*Lycopersicon esculentum* Mill.) has greatly increased because of their relatively high market value; even so, some of these products continue to be imported because the local market absorbs most of the country's produce. Common bean is widely grown in El Salvador, occupying about 75,000 ha (1995). However, total common bean production does not satisfy internal demand and, in 1994-95, El Salvador imported 14,600 tons of common bean (Parada and Pérez, 1996).

Because of the relatively small size of the country, there are few ecologically distinct areas other than those defined by altitude, where the whitefly *Bemisia tabaci* (Gennadius) cannot thrive. Hence, the whitefly/begomovirus problem is distributed fairly evenly throughout the country. Figure 1 shows the main agricultural regions affected by whitefly-transmitted begomoviruses.

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Figure 1. The main agricultural regions affected by whitefly-transmitted begomoviruses, El Salvador.

The emergence of Bemisia tabaci *as a pest and virus vector*

Granillo and co-workers (1974) first reported *B. tabaci* as a vector of plant viruses in El Salvador. The first virus and whitefly outbreaks occurred in the Pacific coastal area, affecting cotton, kenaf (*Hibiscus cannabinus* L.) and common bean. As in the case of Guatemala, we find here a close association between cotton and the whitefly *B. tabaci*. Among the biotic problems that limit common bean production in the country, the main pest and disease problems are *B. tabaci* and *Bean golden yellow mosaic virus* (BGYMV). Zaumeyer and Smith (1964) first reported this virus in El Salvador. BGYMV was more commonly observed in the Pacific lowlands, reaching up to 100% incidence in most affected common bean plantings in the departments of La Libertad, La Paz, San Vicente and Usulutan. BGYMV has reached most of the adjacent mid-altitude valleys in the departments of La Libertad and Ahuchapan, and it can be found now wherever common bean is grown in the country. The introduction of new crops, particularly horticultural crops, further complicated the dynamics of whitefly populations in El Salvador. The Zapotitán Valley (La Libertad) is a good example of a traditional agricultural area where crops such as maize and common bean were largely displaced by high-value horticultural crops (e.g., tomato, pepper and cucurbits). However, these vegetable crops have been eliminated gradually from the valley because of the damage caused by whitefly-transmitted viruses.

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The first generation of black-seeded, BGYMV-resistant common bean varieties produced in Guatemala in the late 1970s made no impact on the control of BGYMV in El Salvador, because Salvadorans prefer bright red-coloured beans. It was not until 1992, when the first red-seeded bean varieties were developed and released, that bean could be cultivated again in these mid-altitude valleys. The BGYMV isolates from El Salvador are very invasive in susceptible common bean cultivars such as Rojo de Seda, the preferred common bean cultivar in the country. Nevertheless, the BGYMV isolates from El Salvador were shown to be closely related to the Middle American BGYMV isolates from Guatemala and Puerto Rico (Table 1).

Table 1. Serological reactions of selected begomoviruses from El Salvador.

BGYMV[a] isolate	Year collected	MAB-BS[b]	MAB-GA[c]
Guatemala	1992	+	+
El Salvador	1992	+	+
Puerto Rico	1992	+	+
El Salvador	1998	+	-
El Salvador	1999	+	-
El Salvador	1999	+	-

a. BGYMV, *Bean golden yellow mosaic virus*.
b. MAB-BS, a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses.
c. MAB-GA, a monoclonal antibody used to detect the original Middle American isolates of BGYMV-Guatemala.

During the course of this project, individual virus samples were collected from common bean plants showing typical BGYMV symptoms. Table 1 shows the results of the serological tests, with the broad spectrum and specific (Guatemalan) monoclonal antibodies developed for BGYMV, for BGYMV isolates collected in 1992, 1998 and 1999. The results show that, as of 1998, there have been changes in the coat protein composition of BGYMV isolates in El Salvador (no reaction with the specific MAB-GA antiserum).

In order to determine the current composition of whitefly species/biotypes in El Salvador, a collection of immature whiteflies found on economically important crops affected by begomoviruses and/or *B. tabaci* was undertaken in selected agricultural departments of the country. The results obtained (Table 2) show an interesting situation of mixed populations of biotype A and B of *B. tabaci*, biotype B being predominantly associated with cucurbits.

The next step was to investigate the proportion of whitefly-transmitted viruses in the different crops grown in whitefly-affected agricultural regions. Table 3 shows the results from this investigation. Basically, it is evident that whiteflies are not the only group of vectors transmitting viruses to horticultural crops in El Salvador. Many of the samples tested also contained potyviruses transmitted by aphids. It is interesting to note that the cucurbit locally called *pipián* (*Cucurbita argyrosperma* C. Huber subsp. *argyosperma*) had not been recorded previously in Central America as a host of whitefly-transmitted viruses. For the four cucurbit species assayed in the above survey, aphid-transmitted potyviruses appear as the main viral problem, with the exception of *pipián*. For common bean, BGYMV was present but the relatively low number of samples collected probably reflects the displacement of this food legume from areas traditionally growing common bean. The results obtained for tomato and pepper suggest a complex

Table 2. Results of whitefly biotype surveys in different agricultural regions of El Salvador.

Crop[a]	Sample	Department	B.t.-A[b]	B.t.-B[b]	Other
Tomato	1	Chalatenango	9	-	1
Chilli	2	Chalatenango	4	5	1
Watermelon	3	La Libertad	7	-	3
Loroco	4	La Libertad	-	3	7
Chilli	5	La Libertad	7	-	3
Ayote	6	Santa Ana	6	-	4
Watermelon	7	La Paz	8	1	1
Melon	8	La Paz	2	7	1
Pipián	9	La Paz	4	-	6
Tomato	10	Chalatenango	5	-	5
Tomato	11	Chalatenango	18	-	2
Watermelon	12	Santa Ana	9	-	1
Tomato	13	Ahuachapán	8	-	2
Cucumber	14	Ahuachapán	4	3	3
Ayote	15	Ahuachapán	4	4	2
Watermelon	16	Santa Ana	2	6	2

a. *Loroco* (*Fernaldia pandurata* [A. DC.] Woodson), *ayote* (*Cucurbita moschata* Duchesne), *pipián* (*Cucurbita argyrosperma* C. Huber subsp. *argyrosperma*).

b. *Bemisia tabaci* A and B biotypes.

Table 3. Serological analyses of plants showing virus-like symptoms associated with the presence of whiteflies in El Salvador.

Samples	Plant[a]	Begomovirus	Potyvirus	Other	None
45	*Pipián*	25	11	0	9
9	Watermelon	0	9	1	-
11	Cucumber	0	7	0	4
6	*Ayote*	1	2	0	3
6	Common bean	2	0	0	4
38	Tomato	0	2	2	36
19	Pepper	0	4	0	0
1	Okra	0	0	0	0
1	*Loroco*	0	1	CMV[b]	0
1	*Chilipuca* (lima bean)	1	0	0	0

a. *Pipián* (*Cucurbita argyrosperma* C. Huber subsp. *argyrosperma*), *ayote* (*Cucurbita moschata* Duchesne), *loroco* (*Fernaldia pandurata* [A. DC.] Woodson), *chilipuca* (*Phaseolus lunatus* L. var. *lunatus*).

b. CMV, cucumovirus.

virus problem in these crops. In past tests, one pepper sample and one tomato sample collected earlier in the Zapotitán Valley had tested positive for the presence of begomoviruses.

A BGYMV isolate from San Andrés, El Salvador, was selected for partial sequencing to determine the degree of molecular divergence with respect to BGYMV isolates characterized in the early 1990s. Table 4 shows the comparative results obtained with two partial regions of the BGYMV genome: the viral replicase (*AC1*) and the coat protein (*AV1*). As can be observed, the Salvadoran BGYMV-ES isolate has not changed drastically over the past decade and it can still be considered an isolate of the Middle American BGYMV species (sequence homologies above 90%). The differences observed at the coat protein level (*AV1*) probably reflect minor changes in certain aminoacids as a result of the introduction of a new whitefly biotype. Differences at the replicase level (*AC1*) may represent an evolutionary trend. The significant differences between the Middle American BGYMV species and the Brazilian BGMV species reflect their different origins or divergent evolutionary paths.

A visit to one of the most traditional agricultural areas of El Salvador, the Zapotitán Valley, showed the profound changes that have occurred in the cropping systems of this country. This irrigation district, thanks to its proximity to the capital, San Salvador, was the breadbasket of El Salvador, producing most of the basic food crops demanded by the local

Table 4. Partial sequence homology (%) between a *Bean golden yellow mosaic virus* (BGYMV) isolate from El Salvador (ES) and related BGYMV/*Bean golden mosaic virus* (BGMV) isolates characterized in the early 1990s.

ORF[a]	Isolates[b]				
	BGYMV-DR	BGYMV-PR	BGYMV-GA	BGMV-BR	BGYMV-ES
AC1	90.5	88.8	91.7	69.4	100
AV1	94.3	91.5	93.8	78.2	100

a. ORF, open reading frame; *AC1*, viral replicase gene; and *AV1*, coat protein gene.

b. DR, Dominican Republic; PR, Puerto Rico; GA, Guatemala; and BR, Brazil.

markets: common bean, maize, etc. In the past decade, this valley has experienced drastic changes in its agriculture to meet the increasing demand for higher value crops, particularly horticultural crops such as tomato, pepper and cucurbits. A recent survey of the Zapotitán Valley revealed the predominance of horticultural crops and the high incidence of viral diseases affecting them. Eight samples (Table 5) were taken selectively from different fields visited in the valley, all from plants showing symptoms usually associated with whitefly-borne viruses.

This preliminary survey, conducted in order to define priorities in Phase II of the Tropical Whitefly Project, shows that other viruses are present in the valley, besides whitefly-transmitted begomoviruses. The viruses detected by electron microscopy and with the use of the monoclonal antibody PTY1 are potyviruses transmitted by aphids and predominantly were found affecting pepper and cucumber (*Cucumis sativus* L. var. *sativus*) in this valley.

Socio-economic Analysis

A case study was conducted in El Salvador, in the San Vicente Department. The study was entitled "Farmers' perception of the whitefly *B. tabaci* as an agricultural pest: management and environmental factors affecting the incidence of the whitefly in the department of San Vicente". This department has an agricultural area of 55,340 ha, including 20,346 ha planted to basic grains—maize, sorghum (*Sorghum bicolor* [L.] Moench), common bean, sesame (*Sesamum indicum* L.) and soybean (*Glycine max* [L.] Merr.)—4015 ha are cultivated to coffee, 2844 ha are in sugarcane and the rest in horticultural and fruit crops. Among the horticultural crops grown here, *ayote* (*Cucurbita moschata* Duchesne), *pipián*, sweet pepper (*Capsicum annuum* L. var. *annuum*), hot pepper (*Capsicum* spp. L.), tomato, cucumber, melon and watermelon are the most important.

About 50% of the farmers interviewed were over 40 years old and

Table 5. Results of the analyses practiced on selected samples of horticultural crops affected by viral diseases in the Zapotitán Valley, El Salvador.

Sample	Crop	Analyses[a]					
		EM	MAB-BS	MAB-GA	PTY1	CMV	PCR
1	Pepper	+	-	-	+	nt	nt
2	Pepper	+	-	-	+	nt	nt
3	Tomato	-	+/-	-	-	nt	nt
4	Tomato	-	-	-	-	-	nt
5	Pepper	+	-	-	+	nt	nt
6	Cucumber	+	-	-	+	-	nt
7	Pepper	-	+	-	-	nt	nt
8	Tomato	-	-	-	-	nt	nt

a. EM, electron microscopy; MAB-BS, a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses; MAB-GA, a monoclonal antibody used to detect the original Middle American isolates of *Bean golden yellow mosaic virus*-Guatemala; PTY1, monoclonal antibodies to detect potyviruses; CMV, monoclonal against cucumovirus; PCR, polymerase chain reaction; and nt, not tested.

40% were between 25-40 years of age. Only 25% of the farmers interviewed had secondary (high school) education and 41% had received technical assistance in the selection of pesticides to control the whitefly problem. Pesticide salesmen are highly influential in the selection and use of insecticides for all crops. Most farmers (54%) apply insecticides with a frequency between 10-15 times per crop cycle. A significant proportion (16%) of the farmers interviewed apply pesticides more than 20 times in a single crop cycle. One hundred and forty-seven different commercial pesticides were mentioned during this survey. Sixty-two percent of the farmers interviewed mentioned preventive pesticide applications as a risk-averting practice.

Of the farmers interviewed, about 97% recognizes the whitefly problem but only 36% associates this pest with viral diseases. Farmers believe that whitefly-related problems are more severe during the dry months of the year but they cannot predict the incidence of whiteflies and viral diseases based on climatic factors. Over 60% of the respondent farmers considered that yield losses in susceptible crops range from 25%-50% because of the whitefly/ begomovirus problems.

Strengthened Research Capacity

El Salvador was undoubtedly the country where this project established more collaborative activities with different agricultural research institutions. The local coordinating entity was the national agricultural research institution, CENTA, under the co-ordination of Dr. Priscila Henríquez (biotechnology) and Ing. Carlos A. Pérez (Coordinator of Programa Cooperativo Regional de Frijol para Centro América, México y el Caribe [PROFRIJOL] activities in El Salvador). A major collaborator in this project has been the University of El Salvador, under the guidance of Prof. Leopoldo Serrano. The University has generated and collected a considerable amount of grey literature on whitefly-related production problems in El Salvador and has offered students to conduct thesis research on whitefly-transmitted viruses affecting crops of socio-economic importance in the country. The University of El Salvador covered the San Vicente Valley, another important agricultural area. A second institution of higher learning collaborating in this project was the Universidad Técnica Latinoamericana (UTLA), which also conducted some studies in the Zapotitán Valley. The Plant Health Division of the Dirección General de Salud Ambiental (DIGESA) also participated in this collaborative effort, covering the departments of Santa Ana, Ahuachapán and Sonsonate. Finally, a nongovernmental organization, the Programa Regional de Manejo Integrado de Plagas en América Central (PROMIPAC), devoted to the production of non-traditional crops, also joined the project by providing support to the various activities undertaken by the different groups. The creation of a national network was unique for Central America and represented a major local effort to address the important problem of whitefly-transmitted begomoviruses in El Salvador.

All of these institutions were provided with technical guidelines for the collection and processing of biological samples related to the whitefly problem affecting crops of socio-economic importance.

Current Status of Whitefly/Begomovirus Problems

The whitefly/begomovirus problems in El Salvador are widely dispersed in the main agricultural areas of the country. However, the incidence of these pests is higher in the plains and mid-altitude valleys. These agro-ecosystems range in altitude, from sea level to 1000 m, with average temperatures of 27 ºC and about 1600 mm of rainfall; all of these being conditions that favour the biology and reproduction of *B. tabaci*.

The displacement of traditional agriculture (e.g., maize and common bean) with non-traditional export crops (e.g., tomato, pepper and cucurbits) has aggravated the whitefly/begomovirus problem because of the availability of new reproductive hosts for *B. tabaci* and the intensive use of pesticides to protect the new high-value crops against begomoviruses.

The current situation of the whitefly/begomovirus problems in El Salvador merits an effort to implement integrated pest management strategies in the main horticultural areas of the country, mainly in the departments of San Vicente, San Salvador, La Libertad, Sonsonate and Santa Ana. The use of virus-resistant common bean cultivars has attenuated the problem in this food crop but genetic resistance is not available in any of the other crops affected by begomoviruses in El Salvador.

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CHAPTER 3.5

Honduras

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Introduction

Geographical context

The territory of Honduras consists of highly mineralized soils and mountains, which favoured the development of mining and livestock exploitations in colonial times. Unlike in other Pacific Coast valleys of neighbouring countries, there are few volcanoes contributing their fertile ashes for the benefit of the soils. The first region to be devoted to agriculture was the humid Caribbean lowlands or “Costa”, which possess fertile alluvial soils planted primarily to banana (*Musa* spp. L.). These geographic characteristics leave only the interior mid-altitude valleys (350-1000 m), for agricultural production (West and Augelli, 1977). One of the most important valleys in this mountainous region is the Comayagua, with an area of about 295 km^2. Much of the mountainous interior of Honduras supports one of the most extensive pine and oak forests in Middle America. Figure 1 shows the main agricultural regions affected by whitefly-transmitted begomoviruses.

The emergence of Bemisia tabaci *as a pest and virus vector*

The isolated situation of the interior valleys and the absence of an extensive cotton (*Gossypium hirsutum* L.) industry in the Pacific coast of Honduras probably protected this country from early outbreaks of *Bemisia tabaci* (Gennadius) and begomoviruses recorded in neighbouring countries to the north, already in the 1970s. It was not until the mid-1980s that the intensification and diversification of cropping systems in the highland central valleys coincided with the emergence of begomovirus problems in Honduras. *Bean golden yellow mosaic virus* (BGYMV) emerged around 1985. In 1989, most common bean (*Phaseolus vulgaris* L.) producing regions in Honduras were already affected by BGYMV. The most affected areas were those in the central and southern agricultural regions of the country, where annual rainfall does not surpass the 1500 mm range, particularly from May through July (Rodríguez et al., 1994).

As in the rest of Middle America, in the last decade Honduras has experienced a boom in the production of non-traditional crops such as tomato (*Lycopersicon esculentum* Mill.), pepper (*Capsicum* spp. L.) and melon (*Cucumis melo* L.). The tomato industry in the Comayagua Valley (580 m) was developed as early as 1978, with a view

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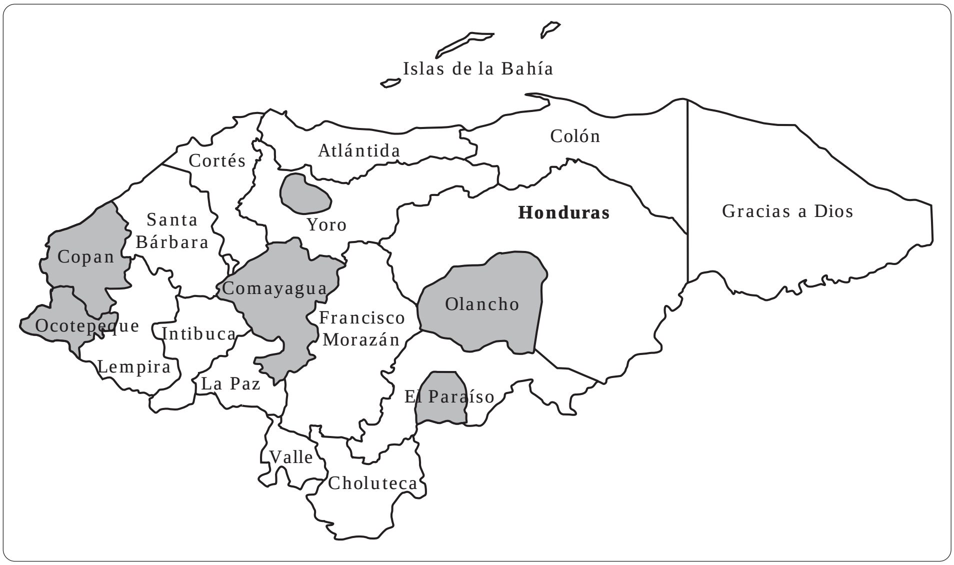

Figure 1. The main agricultural regions affected by whitefly-transmitted begomoviruses, Honduras.

to producing high-quality produce for export (Standard Fruit Company, 1978). This industry has come practically to an end now because of the persistent attacks of whitefly-borne viruses. A similar phenomenon has been recorded for the valleys of Jamastrán, El Paraíso and La Lima (El Paraíso Department), although the most affected crop in these areas is pepper. The melon industry of southern Honduras also has been brought to the point of extinction by whitefly-related problems (Caballero, 1995). These production problems have forced many growers to abandon the cultivation of susceptible food crops leaving as alternative crops such as tobacco (*Nicotiana tabacum* L.). However, tobacco is currently under attack by whiteflies and begomoviruses in neighbouring countries, particularly in Guatemala.

Advances in Biological Research

Research on whitefly-transmitted begomoviruses in Honduras has been scant. During this project, common bean plants exhibiting golden mosaic-like symptoms were tested with the monoclonal BGYMV antibodies. In these tests, the Honduran BGYMV isolate reacted with the broad spectrum monoclonal antibody (MAB-BS) but not with the Guatemalan (GA), as observed in this project for other BGYMV isolates from Guatemala and El Salvador. In a different survey, eight samples taken in the Comayagua Valley from diseased common bean, tomato, pepper, cucumber (*Cucumis sativus* L. var. *sativus*) and a new vegetable grown for export, *cundeamor* (*Momordica charantia* L.), were assayed serologically. Table 1 shows the results obtained with these samples.

Begomoviruses were detected in common bean and tomato but not in the two cucurbits tested, namely cucumber and *cundeamor*. The latter seemed to have a low incidence of a phytoplasma disease, and the cucumber sample was infected by a

Table 1. Results of various diagnostic assays performed on selected plant samples from the Comayagua Valley, Honduras.

Sample	Crop	Diagnostic assay[a]					
		EM	MAB-BS	MAB-GA	PTY1	CMV	PCR
1	Common bean	nt	+	-	nt	nt	+
2	Cucumber	+	-	nt	+	nt	nt
3	*Cundeamor*	nt	-	nt	nt	nt	nt
4	Common bean	nt	+	-	nt	nt	+
5	Common bean	-	-	-	-	-	nt
6	Tomato	nt	+	nt	nt	nt	nt
7	Tomato	nt	+	nt	nt	nt	nt
8	Tomato	nt	+	nt	nt	nt	nt

a. EM, electron microscopy; MAB-BS, a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses; MAB-GA, a monoclonal antibody used to detect the original Middle American isolates of *Bean golden yellow mosaic virus*-Guatemala; PTY1, monoclonal antibody to detect potyviruses; CMV, monoclonal antibody against cucumoviruses; PCR, polymerase chain reaction; and nt, not tested.

potyvirus, most likely transmitted by aphids. The BGYMV isolates from Honduras did not react with the MAB-BS prepared in 1993 to the Guatemalan isolate. The original Guatemalan isolate reacted to this monoclonal when used as a control in this test. This result suggests that the present Honduran BGYMV isolates also have altered their capsid protein composition, probably in response to the arrival of a more ubiquitous vector, the B biotype of *B. tabaci*.

The deoxyribonucleic acid (DNA) of the BGYMV isolates found infecting common bean was amplified by polymerase chain reaction (PCR) for cloning and partial sequencing. These results (Table 2) show that the Honduran BGYMV isolate is a strain of the BGYMV species found in Middle America. In order to test the hypothesis that the differences in nucleotide sequence homology at the coat protein level (*AV1*) may have occurred in response to the presence of the new *B. tabaci* biotype.

Eight whitefly samples from common bean, four samples from cucumber, three samples from tomato and one sample from chilli were biotyped by random amplified polymorphic DNA (RAPD) analysis. The results from these tests (Table 3) do not support the hypothesis of coat protein changes in the Honduran BGYMV isolate due to the emergence of a new whitefly biotype, because only the A biotype of *B. tabaci* biotype was detected in these samples.

Table 2. Partial sequence homology (%) between a *Bean golden yellow mosaic virus* (BGYMV) isolate from Honduras (HD) and related BGYMV/*Bean golden mosaic virus* (BGMV) isolates characterized in the early 1990s.

ORF[a]	Isolates[b]				
	BGYMV-DR	BGYMV-PR	BGYMV-GA	BGMV-BR	BGYMV-HD
AC1	91.1	89.5	92.0	66.8	100
AV1	93.8	91.8	94.2	78.0	100

a. ORF, open reading frame; *AC1*, viral replicase gene; and *AV1*, coat protein gene.
b. DR, Dominican Republic; PR, Puerto Rico; GA, Guatemala; and BR, Brazil.

Table 3. Biotyping of whitefly (*Bemisia tabaci*) samples collected from different commercial crops grown in different municipalities of two departments in Honduras.

Department	Municipality	Crop	Biotype
Comayagua	V. San Antonio	Common bean	A
Comayagua	V. San Antonio	Common bean	A
Comayagua	V. San Antonio	Common bean	A
Comayagua	V. San Antonio	Common bean	A
Comayagua	V. San Antonio	Common bean	A
Comayagua	Comayagua	Common bean	A
Comayagua	Comayagua	Cucumber	A
Comayagua	Comayagua	Cucumber	A
Comayagua	San Nicolás	Tomato	A
Comayagua	San Nicolás	Tomato	A
Comayagua	Flores	Tomato	A
Comayagua	V. San Antonio	Common bean	A
Comayagua	Flores	Common bean	A
Comayagua	Comayagua	Cucumber	A
Comayagua	Comayagua	Cucumber	A
Francisco Morazán	Cedros	Chilli	A

Socio-economic Analysis

A case study was conducted in the Comayagua Valley with the collaboration of the Escuela Agrícola Panamericana (EAP), Zamorano. The results of this study are published in a thesis (Jara, 1998). In the Comayagua Valley, more than 70% of the 100 producers surveyed had been cultivating the land longer than 5 years. The number of literate farmers in the population surveyed was 75%, of which 66% had received only primary education. Only half or less (in some areas) of the farmers surveyed had received technical assistance to control whitefly problems.

Regarding whitefly incidence, 87% of the farmers believed that whiteflies and whitefly-borne viruses increased in the warmer months of the year. About 80% of the farmers noted that whitefly populations increase in periods of low rainfall. The worst whitefly/virus epidemics in the Comayagua Valley occurred in 1989, when drought and high temperatures struck the region. These results demonstrate that most farmers are aware of the key climatic factors that determine whitefly epidemics.

Over 40% of the farmers interviewed apply insecticides on a calendar basis for whitefly control. This practice can be interpreted as a risk reduction measure, particularly in the case of tomato growers, who invest between US$2100 and $3500 per *manzana* (0.764 ha). In tomato plantings, 63% of the growers apply insecticides against whiteflies as soon as the tomato plants are transplanted, 31% apply 1 week after transplanting and only 6% wait until they see whiteflies in their fields. In the case of common bean, 12% of the farmers apply pesticides at planting time, 42% start controlling whiteflies 1 week after germination of the plants and 46% apply insecticides later on during the vegetative phase of the crop. These results demonstrate farmers' perception regarding the investment required to plant tomato (usually 10 times higher than in the case of common bean).

Regarding the likelihood of introducing non-chemical control measures based on cultural practices such as changing planting dates to avoid whitefly population peaks, about 65% of the growers were reluctant to change. This finding suggests that climatic and market factors determine planting times in Honduras.

In economic terms, 53% of the production costs for tomato and 47% for common bean are related to the chemical control of whitefly/ begomoviruses. In cucumber production, only 16% of the total costs are related to chemical whitefly control. Total production costs for tomato are up to 20 times more than for common bean or cucumber. It is evident from this study that the implementation of an integrated whitefly management package in the Comayagua Valley would significantly contribute to lessening the environmental impact of insecticides and to a significant reduction in production costs of high-value crops such as tomato and peppers.

Strengthened Research Capacity

The Tropical Whitefly Integrated Pest Management Project made possible the development of a thesis entitled "Characterization of the incidence and management of whiteflies (Homoptera:Aleyrodidae) in the valley of Comayagua, Honduras" at the Plant Protection Department, EAP-Zamorano. The case study helped acquire a better view of how small-scale farmers perceive the whitefly problem. Experts in various fields such as agronomy, entomology, virology, socio-economics and biometry developed the questionnaire, which consequently constitutes a guide for future surveys on similar field production problems.

The Plant Protection Department of the EAP sent Elsa Barrientos to the Centro Internacional de Agricultura Tropical (CIAT), Colombia, for training in molecular biotyping of *B. tabaci* and characterization of plant viruses during a 2-week period.

Current Status of Whitefly/Begomovirus Problems

Although the whitefly/begomovirus problems arrived relatively late in Honduras, most of the territory offers suitable environmental conditions for *B. tabaci* to reproduce and attack susceptible crops, either as a pest or as a virus vector. The exceptions are the northern Caribbean coast, which usually receives more than 2000 mm of rainfall, a deleterious climatic factor for *B. tabaci*. This region is the prime banana-producing zone of Honduras. The rest of the country has an average rainfall of less than 1500 mm and average temperature of 26 °C, and most horticultural regions are located between 200 and 1000 m above sea level—all favourable conditions for *B. tabaci*.

A good indicator of the presence of whitefly-transmitted viruses in a region is the occurrence of BGYMV. This virus is distributed in the departments of El Paraíso, Francisco Morazán, Choluteca, Valle, Olancho, Comayagua and Copán, marking a vast region where whitefly/begomovirus problems can emerge on susceptible crops. Undoubtedly, one of the most affected areas is the Comayagua Valley, north of the capital, Tegucigalpa. This valley is located in a drier zone (500-1000 mm) and has been planted intensively to non-traditional export crops under considerable pesticide abuse.

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CHAPTER 3.6

Nicaragua

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Introduction

Geographical context

Nicaragua is the largest country in Central America and one of its richest nations in terms of agricultural resources. Its agricultural economy follows the natural divisions of the country: the Pacific region, the northern part of the Central Highlands and the lowlands of the Caribbean. The Pacific region has been Nicaragua's centre of commercial agriculture since colonial times, when livestock and the production of indigo (blue dye) were two of the main commodities. Livestock production still remains an important activity in this region. Cotton (*Gossypium hirsutum* L.) became a highly important crop in the Pacific lowlands, between Lake Managua and the Gulf of Fonseca (Chinandega to León) and along the eastern side of Lake Managua (Tipitapa) in the early 1950s. By 1977, Nicaragua was the largest (217,000 ha) producer of cotton in Central America (Gill, 1994).

As in the rest of Central America, there are a number of fertile mid-altitude valleys in Nicaragua such as Boaco and Sébaco where a more intensive agriculture eventually developed, consisting of non-traditional crops such as tomato (*Lycopersicon esculentum* Mill.), pepper (*Capsicum* spp. L.) and other horticultural crops. Common bean (*Phaseolus vulgaris* L.) remains a major staple and consequently is grown throughout the country. However, the main regions producing common bean are the Pacific region (30% of total production), the Central Highlands (50%) and the Caribbean Plains (10%). In the highlands, the main departments producing common bean are Matagalpa, Jinotega, Estelí, Madriz and Nueva Segovia. Figure 1 shows the main agricultural regions affected by whitefly-transmitted begomoviruses.

The emergence of Bemisia tabaci *as a pest and virus vector*

Bemisia tabaci (Gennadius) first became a pest of cotton in the 1970s, which together with adverse marketing circumstances reduced the area planted to cotton to a mere 2520 ha in 1993. Interestingly, *B. tabaci* was not an insect of economic significance in the early years of cotton production in Nicaragua. The emergence of this whitefly species as a major pest of cotton followed the introduction and intensive use of pesticides on this crop in the 1960s (Gill, 1994).

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Figure 1. The main agricultural regions affected by whitefly-transmitted begomoviruses, Nicaragua.

Bean golden yellow mosaic virus (BGYMV) was first observed affecting common bean in the Pacific Coast of Nicaragua, around 1971. BGYMV is particularly severe in Estelí, Nueva Segovia, León, Chinandega and the lowlands of Matagalpa (Llano et al., 1997). Large populations of *B. tabaci* have been reported on cotton in the Pacific Coast of Nicaragua since 1952, and from tomato in the Atlantic Coast as early as 1953 (Hidalgo et al., 1975). This whitefly species was the third most important pest problem in cotton in Nicaragua in the late 1970s (Kramer, 1966). The whitefly problem became so serious that, in 1975, Nicaragua created a special commission to study *B. tabaci* and make recommendations on research and whitefly management (Hildago et al., 1975).

Mid-altitude valleys such as Sébaco (Matagalpa Department) witnessed the emergence of whitefly-transmitted viruses in 1986. By 1992, yield losses reached 100% in several tomato fields affected by what growers referred to as "*crespo*" (leaf curl). In 1991, large populations of whiteflies attacked common bean and melon (*Cucumis melo* L.) but only bean was affected by begomoviruses. Whitefly-transmitted begomoviruses also attacked pepper and tomato plantings in the Central Highlands. Tomato plantings were affected in the region of Boaco in 1991. The same year, *B. tabaci* attacked tobacco (*Nicotiana tabacum* L.) plantings in the departments of Nueva Segovia, Jinotega, Estelí, Chinandega, Masaya and Rivas. The melon industry in the departments of León, Rivas, Managua and Matagalpa was first affected by whiteflies in 1991 but the incidence of viruses was low (Sediles, 1998).

Advances in Biological Research

No records exist of serological identification of BGYMV in Nicaragua prior to 1995. In 1993, a few common bean and tomato samples were collected near the town of Santa Lucía in the Boaco Valley. The common bean samples were taken from plants showing dwarfing symptoms, rather than mosaic. These samples reacted positively with the broad spectrum monoclonal antibody that detects whitefly-transmitted begomoviruses in general (MAB-BS). The virus was later identified as *Bean dwarf mosaic virus* (BDMV) by nucleic acid hybridization methods (Zamora 1996). At that time, and from the same locality, samples collected from tomato plants affected by *crespo* disease also gave positive results in serological assays with the same monoclonal antibody. The *crespo* disease of tomato was first observed in Nicaragua in 1986. An in-depth investigation conducted in 2000 showed that tomato in Nicaragua is infected by at least four distinct begomoviruses (Rojas et al., 2000).

In 1995, three BGYMV-affected common bean samples from the north-western department of Estelí were tested with the monoclonal antibody used to detect the original Middle American isolates of BGYMV-Guatemala (MAB-GA), available at the Centro Internacional de Agricultura Tropical (CIAT). The three common bean samples reacted with the MAB-BS but only one common bean sample was recognized by the specific MAB-GA.

A survey in the departments of Estelí (Condega), Matagalpa (Apompua and Sébaco) and Managua (Managua and Pochocoape) was undertaken to collect plant samples for virus assay. Table 1 gives the results of this survey, which show a rather low incidence of whitefly-transmitted geminiviruses (GV) in the samples tested. However, the sampling was done at the peak of the rainy season, which, for the second time since Hurricane Mitch in the previous year, brought considerable amounts of rain. Rainfall in 1999 almost doubled the amount of rain recorded in 1998 in the Pacific

Table 1. Results of survey of virus-affected plants in three departments of Nicaragua.

Sample	Crop	Locality	Department	Reaction[a]	
				MAB-BS	PTY1
1-N5	Tomato	Condega	Estelí	-	-
2-N5	Tomato	Condega		+ / -	-
3-N5	Tomato	Condega		-	-
4-N5	Tomato	Condega		+	-
5-N5	Tomato	Condega		-	-
6-N5	Tomato	Condega		-	-
7-N6	Tomato	Apompua	Matagalpa	-	-
8-N6	Tomato	Apompua		-	-
9-N6	Tomato	Apompua		-	-
10-N6	Tomato	Apompua		-	-
11-N6	Tomato	Apompua		-	-
12-N6	Tomato	Apompua		-	-
13-N7	Tomato	Managua	Managua	-	-
14-N7	Tomato	Managua		-	-
15-N7	Tomato	Managua		-	-
16-N7	Tomato	Managua		-	-
17-N8	Tomato	Sébaco	Matagalpa	-	-
18-N8	Tomato	Sébaco		-	-
19-N9	Tomato	Pochocoape	Managua	-	-
20-N9	Tomato	Pochocoape		-	-
21-N9	Tomato	Pochocoape		-	-
22-N9	Tomato	Pochocoape		-	-
23-N9	Tomato	Pochocoape		-	-
24-N9	Tomato	Pochocoape		-	-
25-N10	*Pipián*	Apompua	Matagalpa	+	+
26-N10	*Pipián*	Apompua		+	+
27-N10	*Pipián*	Apompua		+	+
28-N11	Tomato	Sébaco	Matagalpa	-	-
29-N11	Tomato	Sébaco		-	-
30-N11	Tomato	Sébaco		-	-
31-N11	Tomato	Sébaco		-	-
32-N11	Tomato	Sébaco		-	-
33-N11	Tomato	Sébaco		-	-
34-N11	Tomato	Sébaco		-	-
35-N12	Tomato	Apompua	Matagalpa	-	-
36-N12	Tomato	Apompua		-	-

a. Positive or negative reactions to a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses (MAB-BS); reactions to a potyvirus-specific monoclonal antibody (PTY1).

Lowlands of Nicaragua. The presence of begomoviruses in *pipián* (*Cucurbita argyrosperma* C. Huber subsp. *argyrosperma*) is interesting, considering that this crop had not been recorded previously as an important host/reservoir of begomoviruses. However, the same observation was made in El Salvador and, furthermore, this cucurbit was also doubly infected with aphid-transmitted potyviruses in both countries.

Because of the rainy conditions that affected Nicaragua in 1998 and 1999, very few whitefly samples could be taken for analyses. Table 2 summarizes some of the preliminary results obtained. Although representing only a limited sample, results are taken from one of the main horticultural valleys of Nicaragua and, consequently, are interesting in that they show a predominance of the original biotype A.

Table 2. Results of *Bemisia tabaci* biotyping analyses performed on 15 samples of whiteflies from Apompua and Sébaco, department of Matagalpa, Nicaragua.

Sample code	Biotype A	Other
N2	4	1
N3	5	-
N4	5	-

Socio-economic Analysis

Common bean is one of the two main food staples, together with maize (*Zea mays* L.), being grown on about 140,000 ha. However, productivity is low (about 450 kg/ha) because 80% of the national production takes place on farms of less than 3 ha. The new economic policies that followed the economic crisis of the 1970s have gradually led to the displacement of traditional food crops from most of the fertile valleys in Central America in order to make room for high-value, non-traditional export crops such as tomato, pepper and melon. As a result, there is a higher demand for traditional crops such as common bean in the region. Nicaragua could take advantage of its relatively larger agricultural area to capture the increased regional demand for food staples.

Horticultural crops such as tomato and pepper have been cultivated in Nicaragua mainly to meet local demand. The area planted to tomato has doubled in the past 30 years from 350 to 750 ha, whereas in Guatemala, tomato production grew from 5000 ha in 1960 to 12,000 ha in 1970. In 1986, tomato plantings in the Sébaco Valley, Matagalpa Department, suffered unusual infestations of the whitefly *B. tabaci* and, soon after, the emergence of viral diseases associated with the whitefly outbreak. By 1991, tomato production in the Sébaco Valley had been reduced 20%-50% because of yield losses ranging between 30%-100% (Sediles, 1998). A similar situation was observed for pepper plantings, which in 1991-92, reported yield losses between 30%-50%. Melon, another non-traditional crop grown mostly for export, was beginning to experience whitefly-related problems. However, there was no significant incidence of begomoviruses in this crop.

The whitefly problem on tomato became so severe that it prompted the creation of an inter-institutional tomato group composed of CATIE/MAG-MIP–NORAD-ASDI (for full names, see Acronyms list on page 345), which organized the first national meeting of tomato producers in April 1994. Tomato growers met to discuss whitefly management strategies with the technical organizing group (Grupo

Interinstitucional e Interdisciplinario de Tomate-GIIT). In June 1995, a second meeting took place in Santa Emilia, Matagalpa, with 24 producers representing 16 tomato-growing regions of Nicaragua, where the whitefly is a limiting factor to tomato production. In these meetings, tomato farmers became aware of the consequences of abusing pesticides as well as of the existence of integrated pest management practices (live barriers, yellow traps and organic insecticides). An outcome of the meeting was the creation of the Nicaraguan Tomato Growers Association, which is responsible for the sustainability of tomato production in Nicaragua.

Strengthened Research Capacity

The Universidad Nacional Agraria (UNA) located in Managua, the capital of Nicaragua, was selected as the main collaborating institution. The national program and the Ministry of Agriculture of Nicaragua, already have the substantial financial support of international organizations. The financial support to UNA permitted a general evaluation of the whitefly situation in the six administrative regions of Nicaragua described below.

Region I includes the north-western departments of Nueva Segovia, Madriz and Estelí. In this region, there are valleys and plateaus with altitudes ranging between 500 and 1000 m and a variety of food crops such as bean, tomato, cucurbits, tobacco and pepper, which attract whiteflies. The whitefly population peak occurs in the period November-May.

Region II includes the south-western departments of León and Chinandega and agricultural lowlands below 500 m. The region is formed by the hot Pacific Plains, where cotton production took place in past decades. The whitefly *B. tabaci* became a limiting biotic problem for cotton production and the region has moved on to new crops: tomato, watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai), squash (*Cucurbita* spp. L.), pepper, melon and traditional ones as well such as common bean and tobacco. Soybean (*Glycine max* [L.] Merr.), peanut (*Arachis hypogaea* L.) and cassava (*Manihot esculenta* Crantz) are other crops infested by whiteflies in this region. Whitefly population peaks occur in January and February.

Region III consists of Managua Department and two vegetable production zones, the coastal area of Lake Managua located north of the city of Managua and the zone of Pochocuape, south of Managua. Here, whiteflies attack tomato, squash, watermelon, common bean and tobacco and most of these crops are attacked also by begomoviruses.

Region IV includes the departments of Carazo, Granada, Masaya and Rivas, which form the southernmost portion of the Pacific Lowlands. At these low altitudes, the whitefly *B. tabaci* thrives, severely attacking tomato and tobacco and, to a lesser extent, watermelon, squash, common bean, pepper and melon. The main whitefly peak occurs in January and February.

Region V is formed by the departments of Boaco and Chontales. Although this area has been devoted primarily to livestock, there are some parts such as Santa Lucía, Boaco, where bean and tomato traditionally have been planted. These crops have been affected by whitefly-transmitted begomoviruses since the mid 1980s, particularly by BGYMV (common bean) and by the *crespo* disease in the case of tomato.

Region VI is a main horticultural area of Nicaragua, particularly the Sébaco Valley, in the department of Matagalpa. The second department that forms this region, Jinotega, lies further north, bordering Honduras. The tomato crop in this region has been affected severely by whitefly-borne begomoviruses, to the point that some farmers have abandoned this crop, particularly in the southern part of the Sébaco Valley. Large whitefly populations and viral symptoms also affected cucurbits such as squash, pumpkin (*Cucurbita* spp. L.) and cucumber (*Cucumis sativus* L. var. *sativus*).

It is evident that whiteflies and *B. tabaci*-transmitted viruses affect all of the agricultural regions of Nicaragua. Nicaraguan agricultural research institutions have come together to reach affected farmers and manage the whitefly problem in a highly exemplary manner. The integrated pest management projects developed in Nicaragua with the collaboration of international and national institutions have recovered some of the most affected agricultural areas for the production of traditional (e.g., common bean) and non-traditional (e.g., tomato) crops. It is expected that the scientific knowledge generated in these projects will contribute to the implementation of more effective measures to control whiteflies and the viruses they transmit in Nicaragua and Middle America.

Current Status of Whitefly/Begomovirus Problems

Most of the whitefly/begomovirus problems in Nicaragua occur in the western half of the country, mainly because this region concentrates most of the country's population. The eastern half of Nicaragua (the Caribbean Plains) is only cultivated to a minor extent with, for example, banana (*Musa* spp. L.), rice (*Oryza sativa* L.) and oil palm (*Elaeis guineensis* Jacq.), because it is one of the rainiest regions in Central America. This climatic factor is a major deterrent for the whitefly *B. tabaci* and for the establishment of its main plant hosts. The Pacific Lowlands, on the other hand, experience several dry months without significant rainfall and thus possess all the necessary conditions for the whitefly *B. tabaci* to thrive, including the presence of good breeding hosts such as cotton. Moreover, as in the rest of Central America, most horticultural crops are located in mid-altitude (200-1000 m) valleys created by the mountain ranges that make up the central and northern regions of Nicaragua. Valleys such as the Sébaco, Matagalpa Department (450 m) constitute an example of a locality with a high incidence of *B. tabaci* and begomoviruses, because of the favourable environmental conditions for the pest and the presence of susceptible horticultural crops, some of which also act as suitable reproductive hosts for *B. tabaci*.

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CHAPTER 3.7

Costa Rica

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Introduction

Geographical context

The Central Plateau, around the capital city of San José, is one of the most intensively cultivated areas in Costa Rica. North and south of this region, the broken and volcanic topography of mountain ranges expands all the way to the northern border with Nicaragua and the southern border with Panama. East and west of the northern mountain ranges lie the Caribbean and Pacific lowlands, the least developed and inhabited regions of Costa Rica. South-eastern Costa Rica receives over 1500 mm of rain annually, which prompted the development of a thriving banana (*Musa* spp. L.) production industry towards 1880. Coffee (*Coffea arabica* L.) was introduced into Costa Rica in the 1830s and soon became a source of wealth for many farmers in the Central Plateau. Among the basic food staples, maize (*Zea mays* L.) and common bean (*Phaseolus vulgaris* L.) are important components of the Costa Rican diet, together with crops such as potato (*Solanum tuberosum* L.) and plantain (*Musa* × *paradisiacal* L.). Cotton (*Gossypium hirsutum* L.) is produced only to a limited extent in the Pacific lowlands of the Province of Guanacaste (West and Augelli, 1977; Pastor, 1988). The Central Plateau is undoubtedly one of the most affected areas in terms of whitefly-related damage. Figure 1 shows the main agricultural regions affected by whitefly-transmitted begomoviruses.

Figure 1. The main agricultural regions affected by whitefly-transmitted begomoviruses, Costa Rica.

The emergence of Bemisia tabaci *as a pest and virus vector*

The lack of suitable reproductive hosts for the whitefly *Bemisia tabaci* (Gennadius) such as cotton or soybean (*Glycine max* [L.] Merr.), together with the high rainfall over much of the Costa Rican territory, protected the

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Central Plateau's agricultural region against the early arrival of this pest. The first records of the presence of *B. tabaci* in the country originated in the 1970s, precisely in the drier lowlands of the Pacific province of Guanacaste, where an incipient cotton industry was developed in the 1960s. The first record of *B. tabaci* as a pest and vector of a plant virus *Bean golden yellow mosaic virus* (BGYMV) in common bean fields in the Central Valley, occurred in 1987. BGYMV already had been observed in the country in the 1960s (Gámez, 1970), albeit at a very low incidence. In 1988, *B. tabaci* moved on to tomato (*Lycopersicon esculentum* Mill.) in the Central Valley (Hilje, 1997). Coincidentally, the attack to common bean and tomato took place in the western areas of the Central Valley, which is the closest region to the cotton-growing areas of Costa Rica. *B. tabaci* is currently a pest and vector of viruses in common bean and tomato fields in most of the horticultural provinces of the country.

Advances in Biological Research

A BGYMV isolate from Alajuela (Central Valley) had been characterized serologically at the Centro Internacional de Agricultura Tropical (CIAT) in 1993 as a member of the Central American/ Caribbean group of BGYMV isolates. However, different begomoviruses isolated during the course of this project from BGYMV-affected bean plants in different regions of Costa Rica (Puriscal, Alajuela and Los Chiles) were shown to be serologically distinct from the original Middle American BGYMV isolates characterized in the early 1990s (Cancino et al., 1995).

A begomovirus of tomato had been isolated previously in the locality of Turrialba (Castillo, 1997) and identified as *Tomato yellow mosaic virus* (ToYMV). This virus is currently considered a tentative new species referred to as *Tomato yellow mottle virus* (ToYMoV). Another begomovirus, isolated from diseased tomato plants in Turrialba, was shown in 1998 to be related to a tomato begomovirus tentatively designated as *Tomato leaf curl Sinaloa virus*, originally isolated in north-western Mexico (Idris et al., 1999).

The Tropical Whitefly Project financed a survey of *B. tabaci* biotypes in Costa Rica, conducted under the guidance of Dr. Luko Hilje of the Centro Agronómico Tropical de Investigación y Enseñanza (CATIE), Turrialba. Table 1 shows the results of the molecular characterization analyses conducted at CIAT for some of the samples collected. These results confirmed the presence of *B. tabaci* in various crops of socio-economic importance and showed the penetration of biotype B of *B. tabaci* in different regions of Costa Rica. However, at the time of the survey, the original A biotype still predominated in the country. Some of the most heavily colonized crops were cucurbits but, unlike the case of other Central American countries, *B. tabaci* was found to colonize pepper (*Capsicum* spp. L.) in Costa Rica (Hilje, 1997; Vallejos, 1997).

Socio-economic Analysis

At the beginning of this project, a previous investigation on the economic impact of the *B. tabaci*-begomovirus pest complex in Costa Rica, and particularly on common bean, tomato and pepper, was used as ex ante data for the survey conducted during the current project (Vallejos, 1997). According to the results obtained in that investigation, the trend to replace

Table 1. Characterization of *Bemisia tabaci* biotypes in Costa Rica.

Province	Locality	Crop	Biotype
Alajuela	Orotina	Watermelon	B
Alajuela	Orotina	Cucumber	B
Puntarenas	Puntarenas	Melon	B
Puntarenas	Ticaral	Watermelon	B
Puntarenas	Ticaral	Pumpkin	A
Puntarenas	Lepanto	Common bean	A/B
Guanacaste	Carrillo	Melon	A
Guanacaste	Carrillo	Watermelon	A
Guanacaste	Bagaces	Melon	A
Guanacaste	Bagaces	Spider flower	A
Guanacaste	Bagaces	Cucurbit	A
Guanacaste	Tilarón	Tomato	A
Guanacaste	Tilarón	Chilli	A
San José	Pérez Zeledón	Snap bean	A
San José	Pérez Zeledón	Pepper	A
San José	Pérez Zeledón	Chilli	A
San José	Pérez Zeledón	Tomato	A
San José	Pérez Zeledón	Tomato	A
Heredia	not cited	Chilli	A
Heredia	not cited	Tomato	A
Heredia	not cited	Chilli	A
Heredia	not cited	Chilli	A
Limón	Guácimo	Chilli	A
Limón	Guápiles	Pumpkin	A
Alajuela	Gracia	Tomato	A
Alajuela	Gracia	Chilli	A
Alajuela	Naranjo	Chilli	A
Alajuela	Naranjo	Cucumber	A
Alajuela	S. Ramón	Tomato	A
Alajuela	Atenas	Tomato	A
Alajuela	Alajuela	Sweetpotato	A
Alajuela	Alajuela	Tomato	A
Alajuela	Alajuela	Chilli	A
Alajuela	Alajuela	Common bean	A
Alajuela	Alajuela	Cucumber	A
Cartago	Turrialba	Chilli	A
Cartago	Cervante	Tomato	A
Cartago	Turrialba	Tomato	B
Cartago	Turrialba	Chilli	A
Cartago	Cartago	Tomato	A
Cartago	Cartago	Chilli	A
Cartago	Paraíso	Tomato	B
Cartago	Paraíso	Chilli	B

basic food crops such as common bean for high-value crops such as tomato and pepper was quite apparent and still continues in Costa Rica. As a result, common bean and other traditional crops are increasingly planted in marginal soils, with an expected fall in the productivity of these crops.

Yield losses due to whitefly-transmitted viruses were 37% for common bean, 22% for tomato and 9% for pepper. The whitefly *B. tabaci* could reproduce on pepper without causing apparent damage to this host plant. Regarding the economic feasibility of these crops, tomato and chilli (*Capsicum* sp.) were the most profitable crops, producing over US$6300/ha. The net return for common bean was US$145/ha.

Strengthened Research Capacity

Costa Rica, and specifically CATIE, has been a leader in the collection and dissemination of information on integrated pest management (IPM) strategies to control whitefly pests. Dr. Luko Hilje of CATIE has co-ordinated these information exchanges through an international network created in 1992. Annual workshops have been organized in different Latin American countries to update and discuss whitefly-related problems and the most suitable IPM measures implemented to date. The Systemwide Tropical Whitefly (TWF)-IPM Project has given its support to this effort by promoting complementary activities, particularly in the area of molecular characterization of whiteflies and begomoviruses. With the funds provided by this project, Dr. Hilje was able to attend the International Workshop on *Bemisia* and Geminiviruses held in San Juan, Puerto Rico, in June 1998. His presence in this meeting was important for maintaining the Latin American whitefly network active and for planning future activities. Additionally, the funds provided to CATIE allowed Dr. Hilje to conduct a countrywide survey of the whitefly/begomovirus-affected regions in Costa Rica.

A short study fellowship was awarded to Ing. Agr. Guillermo Sibaja Chinchilla of the Costa Rican Ministry of Agriculture and Livestock, Plant Health Division. Mr. Sibaja came to CIAT, Colombia, for intensive training on the characterization of *B. tabaci* biotypes.

Current Status of Whitefly/Begomovirus Problems

Whiteflies and begomoviruses in Costa Rica mainly affect common bean and tomato crops, particularly in the Central Valley, where most of the horticultural crops are grown. Another area where the whitefly problem is gaining momentum is the “Pacífico Seco” (Dry Pacific Region) in Guanacaste Province. Rainfall in this region ranges from 1220 to 2000 mm annually, with an average temperature of 20 to 25 °C. These are not optimal conditions for *B. tabaci* but the occurrence of a prolonged dry period from December through April favours the build up of whitefly populations and transmission of viruses to susceptible horticultural crops. Fortunately, most of the Costa Rican territory receives over 1500 mm of annual rainfall, which maintains whitefly populations depressed most of the year. These climatic factors and various IPM measures implemented in Costa Rica, including the use of resistant varieties in the case of common bean, have contributed significantly to the attenuation of the whitefly/begomovirus problems in this country.

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CHAPTER 3.8

Panama

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Introduction

Geographical context

Most of the Panamanian territory receives tropical rains in excess of 2500 mm/yr, a climatic factor known to drastically reduce whitefly populations. Nevertheless, during the first (1992) workshop on whiteflies in Central America and the Caribbean, held in Costa Rica, Panama was already present (Zachrisson and Poveda, 1992). According to the report presented by the Panamanian delegate at that meeting, the emergence of whiteflies as agricultural pests in Panama was associated with the expansion of non-traditional crops, mainly melon (*Cucumis melo* L.), for export to the United States. Three horticultural areas were developed in the late 1980s: the Pacific Coast of the province of Panama; the central provinces of Coclé, Herrera and Los Santos (the largest); and the western province of Chiriquí, near the Costa Rican border (Zachrisson and Poveda, 1992; Delgado, 1994). These regions have in common the presence of a well-defined dry period and below-average precipitation during the year. Figure 1 shows the main agricultural regions affected by whitefly-transmitted begomoviruses.

The emergence of Bemisia tabaci *as a pest and virus vector*

The presence of *Bemisia tabaci* (Gennadius) on tomato (*Lycopersicon esculentum* Mill.) was first observed in the Azuero Peninsula in 1983. However, it was not until 1991, when unusually dry climatic conditions favoured the reproduction of *B. tabaci*, that this whitefly species became a pest in the central provinces of Panama. *B. tabaci* has been observed to reproduce in eggplant (*Solanum melongena* L.), sweet pepper (*Capsicum annuum* L. var. *annuum*) and chilli pepper (*Capsicum* spp. L.). In 1992, the silver leaf symptom was observed on squash (*Cucurbita* spp. L.) attacked by *B. tabaci,* which suggested the presence of biotype B in Panama. The western horticultural zone, located in Chiriquí Province, is at a higher elevation and receives more rainfall than the other two horticultural zones of Panama. The problem pest in this area is not so much *B. tabaci* as *Trialeurodes vaporariorum* (Westwood), a whitefly species that attacks horticultural crops at altitudes above 1000 m (Zachrisson and Poveda, 1992).

In Panama, common bean (*Phaseolus vulgaris* L.) is cultivated in

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Figure 1. The main agricultural regions affected by whitefly-transmitted begomoviruses, Panama.

Chiriquí Province, at altitudes between 550 and 1000 m. *Bean golden yellow mosaic virus* (BGYMV) was observed affecting common bean in Panama prior to 1970 (Gámez, 1970) but this disease never has been a major production problem of common bean in Panama.

Advances in Biological Research

A whitefly-borne geminivirus affecting tomato was described in Panama in 1998 (Engel et al., 1998). The virus, named *Tomato leaf curl virus*, was shown in this project to have a high degree of nucleotide sequence similarity when compared to *Tomato yellow mosaic virus* (ToYMV) from Venezuela. The virus had been described in Panama as a strain of *Potato yellow mosaic virus*, a misnomer for ToYMV. Although this virus may be a new species, the name "*Tomato leaf curl virus*" is not appropriate because it is already given to a distinct begomovirus species that only exists in the Old World (Asia). Therefore, the Panamanian virus should be renamed.

A molecular characterization of *B. tabaci* biotypes from the central horticultural zone of Azuero was conducted at the Centro Internacional de Agricultura Tropical (CIAT), with samples provided by Ing. Agr. José Angel Guerra of the Instituto de Investigación Agropecuaria de Panamá (IDIAP) (Table 1). Results demonstrate the coexistence of biotypes A and B of *B. tabaci* in the central provinces of the Azuero Peninsula. This is an interesting situation that needs to be monitored in order to study the outcome of the interaction between these two biotypes in a horticultural zone characterized by mixed cropping systems.

Socio-economic Analysis

Because Panama was not originally included in this project, no socio-economic information was collected for this country. However, it is known that whitefly-transmitted geminiviruses do not affect common bean severely in Chiriquí Province.

Between 1991 and 1994, about 6000 tons of tomato were lost to whitefly problems in Panama. During the 1997-98 tomato planting season, 2800 tons were lost because of the effect of *B. tabaci*, both as a pest and

Table 1. Molecular characterization of *Bemisia tabaci* biotypes from Azuero, Panama.

Sample	Crop	Biotype
1	Tomato	A/B
2	Melon	A
3	Melon	B
4	Melon	A/B
5	Melon	A
6	Tomato	B
7	Tomato	B
8	Tomato	B
9	Tomato	B
10	Tomato	A/B
11	Melon	A/B
12	Melon	B
13	Melon	B
14	Melon	B
15	Melon	B
16	Melon	B
17	Melon	B
18	Melon	A/B

vector of plant viruses. Whereas large-scale tomato growers absorbed most of the losses, these pests affected a number of small-scale producers in the province of Los Santos.

Strengthened Research Capacity

Considering that Panama was not included officially in the project, an agreement was made with the Red Colaborativa de Investigación y Desarrollo de Hortalizas para América Central, Panamá y República Dominicana (REDCAHOR), a horticultural research network operating in the region, to train Panamanian researchers. The network financed the training at CIAT of two national scientists: an entomologist (Ing. José Guerra) and a virologist (Dr. Orencio Fernández) from IDIAP. These scientists were trained in the molecular characterization of *B. tabaci* biotypes and the use of monoclonal antibodies for the identification of begomoviruses.

Current Status of Whitefly/Begomovirus Problems

Although Panama is one of the rainiest countries in the world, with an average annual rainfall above 2000 mm, there are less humid (1000-2000 mm) coastal regions around the Gulf of Panama, where horticultural production has increased significantly in the last decades. The main horticultural area is located in the Azuero Peninsula, where most of the whitefly/begomovirus problems have emerged in past years. Whitefly populations increase in the drier months of the year: January-March. Panama is in the process of expanding the production of non-traditional export crops and consequently this country should take measures to control the whitefly problem before it becomes unmanageable.

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CHAPTER 3.9

Haiti

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Introduction

Geographical context

Haiti is a mountainous country; 40% of its territory is located at altitudes above 490 m. The mountain ranges alternate with fertile valleys, the largest being the Plaine du Nord. Annual rainfall varies from 500 in the north-west to 2500 mm in the eastern/southern highlands. Agriculture accounts for one-third of the gross domestic product but subsistence farming predominates. Maize (*Zea mays* L.; 250,000 ha), sorghum (*Sorghum bicolor* [L.] Moench; 160,000 ha), common bean (*Phaseolus vulgaris* L.; 100,000 ha) are the main staples, with rice (*Oryza sativa* L.) and sweetpotato (*Ipomoea batatas* [L.] Lam.) also being important components of the Haitian diet. However, most of the food consumed in Haiti has to be imported. Coffee (*Coffea arabica* L.) is the main export crop (145,000 ha), followed by sugarcane (*Saccharum officinarum* L.), sisal (*Agave sisalana* Perrine ex Engelm.) and cacao (*Theobroma cacao* L.). French-Creole is spoken by 90% of the population and only 10% speak French. Horticultural crops have become increasingly important in Haiti, mainly as an extension of the agricultural export business in the Dominican Republic. Figure 1 shows the main agricultural regions affected by whitefly-transmitted begomoviruses.

The emergence of Bemisia tabaci *as a pest and virus vector*

The whitefly *Bemisia tabaci* was first regarded as a serious pest in Haiti, in the early 1980s. Common bean, tomato (*Lycopersicon esculentum* Mill.), eggplant (*Solanum melongena* L.), pepper (*Capsicum* spp. L.), cowpea (*Vigna unguiculata* [L.] Walp.) and lima bean (*Phaseolus lunatus* L. var. *lunatus*) are the main crops affected by this insect pest (Donis and Prophete, 1997). As a vector, *B. tabaci* already had been observed to transmit *Bean golden yellow mosaic virus* (BGYMV) in 1978 (Balthazar, 1978).

Advances in Biological Research

The main agricultural areas of Haiti were surveyed and plant samples analysed to determine the importance of whiteflies and whitefly-borne viruses. Table 1 shows the different regions visited and the results obtained from the tests practiced with samples collected. Of whitefly-transmitted

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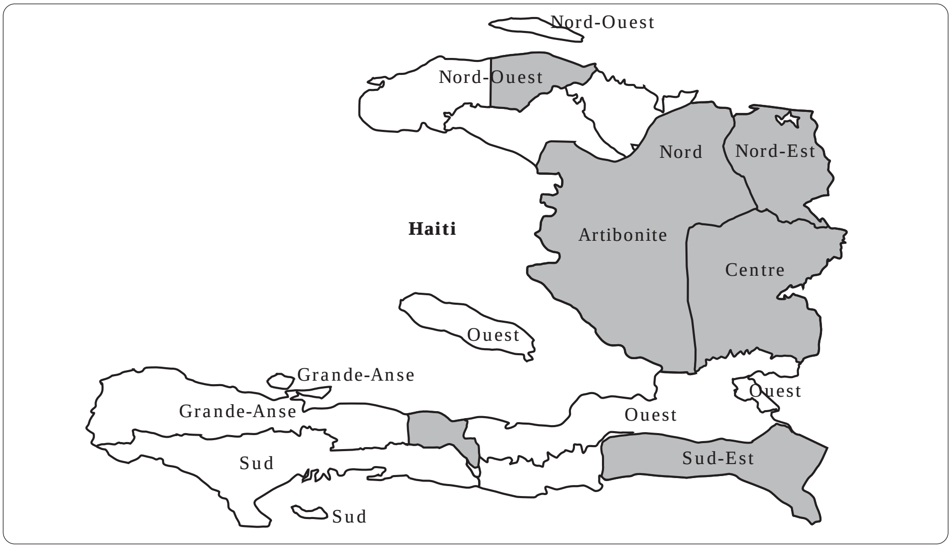

Figure 1. The main agricultural regions affected by whitefly-transmitted begomoviruses, Haiti.

begomoviruses, BGYMV was present in common bean and *Tomato yellow leaf curl virus* (TYLCV) in tomato. As in the case of the Dominican Republic, the specific monoclonal antibody (MAB-GA), which recognizes BGYMV isolates from Central America and southern Mexico, detected the Haitian isolate of BGYMV. This isolate was partially sequenced and compared to the rest of BGYMV isolates from the Caribbean and Central America characterized to date. Table 2 shows the results of these comparative analyses.

As suggested by the serological assay using the Guatemalan (GA) specific MAB, the Haitian BGYMV

Table 1. Results of serological and polymerase chain reaction (PCR) assays of selected samples collected in Haiti.

Sample	Locality	Plant	Reaction[a]			
			MAB-BS	MAB-GA	PTY1	TYLCV
1	Cul-de-Sac	Cowpea	-	nt	-	nt
2	Passereine	Tomato	-	nt	-	+
3	Passereine	Tomato	-	nt	-	+
4	Passereine	Tomato	-	nt	-	+
5	Passereine	Cowpea	-	nt	+	nt
6	Passereine	Common bean	+	+	-	nt
7	Terrier Rouge	Tomato	-	nt	+	+
8	Terrier Rouge	Tomato	-	nt	-	+
9	St Raphael	Tomato	-	nt	-	+
10	St Raphael	Tomato	-	nt	-	+
11	St Raphael	Pepper	-	nt	+	nt

a. Positive or negative reactions to a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses (MAB-BS); a MAB used to detect the original Middle American isolates of *Bean golden yellow mosaic virus*-Guatemala (MAB-GA); a potyvirus-specific monoclonal antibody (PTY1); and PCR detection of *Tomato yellow leaf curl virus*; and nt, no test.

Table 2. Comparative nucleotide sequence homology (%) between a begomovirus isolated from common bean in Haiti and previously sequenced common bean begomoviruses.

Region	BG[a] Haiti	Common bean begomoviruses[b]				
		BGYMV Dominican Republic	BGYMV Guatemala	BGYMV Puerto Rico	BGMV Brazil	BDMV Colombia
Coat protein (*AV1*)	100	90.2	92.0	89.5	68.3	68.7
Replicase (*AC1*)	100	87.5	90.9	88.9	77.6	78.8

a. BG, begomovirus.

b BGYMV, *Bean golden yellow mosaic virus*; BGMV, *Bean golden mosaic virus*; and BDMV, *Bean dwarf mosaic virus*.

isolate assayed is similar to the Central American and Caribbean BGYMV isolates but different from the South American bean begomoviruses, *Bean golden mosaic virus* and *Bean dwarf mosaic virus*. Few of the tomato, pepper and cowpea samples collected in Haiti contained aphid-transmitted potyviruses. All tomato samples were shown by polymerase chain reaction (PCR) to be infected by TYLCV. The composite whitefly sample collected from eggplant resulted in the detection and identification of biotype B of *B. tabaci*. Although preliminary, these results suggest that the whitefly and begomovirus problems in the Hispaniola Island (Dominican Republic and Haiti) are similar.

Socio-economic Analysis

Whitefly-transmitted begomoviruses have been observed to cause significant or even total yield losses in common bean and susceptible horticultural crops in Haiti. In the case of common bean, the cultivation of BGYMV-resistant varieties (available through the Programa Cooperativo Regional de Frijol para Centro América, México y el Caribe [PROFRIJOL] project) and the cultivation of common bean at altitudes above 1000 m should reduce the incidence of BGYMV. The same strategy could be adopted for begomovirus-susceptible horticultural crops, that is, growing vegetables above 1000 m. For valleys below this altitude, where *B. tabaci* thrives, cultural practices such as the implementation of a period free of certain crops that act as reproductive hosts for the whitefly should be adopted following the model implemented in the Dominican Republic. Although there are tomato varieties resistant to TYLCV, these are commercial varieties that small-scale farmers cannot afford. The production of high-value crops (e.g., tomato, pepper and eggplant) by small-scale farmers could greatly benefit from the adoption of integrated pest management (IPM) practices such as the use of micro-tunnels and biological insecticides or mild soaps.

Strengthened Research Capacity

Two collaborating scientists from the Centre de recherche et de documentation agricoles-Ministère de l’agriculture des ressources naturelles et du développement rural (CRDA-MARNDR, Damien), participated in the field survey conducted in Haiti. These scientists were invited to attend international workshops in Central America and to acquire new whitefly/ begomovirus characterization techniques at the Centro Internacional de Agricultura Tropical (CIAT) but official and administrative problems prevented their participation in these activities. Nevertheless, as collaborators in this project, these researchers have benefited from the information generated to date.

The visit to Haiti was also helpful for establishing closer links between the PROFRIJOL project, CIAT and Haiti. As a result, two bean breeders are now actively working to deploy BGYMV-resistant bean varieties in Haiti, and the United States Agency for International Development (USAID) has financed a new project to work on sustainable agricultural practices, including hillside technologies and food security issues in Haiti.

Current Status of Whitefly/Begomovirus Problems

The importance of *B. tabaci* as a pest and vector of plant viruses is expected to decrease in the near future as new bean cultivars possessing resistance to BGYMV are introduced by the USAID-funded project. The introduction of TYLCV-resistant tomato varieties from the Dominican Republic should also reduce the impact of TYLCV in Haiti. However, poverty issues will limit the adoption of resistant cultivars, particularly in the case of tomato. The low economic capacity of most small-scale farmers practicing subsistence agriculture also precludes the use of effective but expensive insecticides (e.g., imidacloprid) to control whiteflies and whitefly-transmitted viruses. The low levels of insecticidal action of cheaper pesticides encourages pesticide abuse, with the consequent development of insecticide resistance in *B. tabaci* populations. Finally, Haiti might benefit from its broken topography to avoid the attack of *B. tabaci* by planting susceptible crops at altitudes above 1000 m.

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CHAPTER 3.10

Cuba

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Introduction

Geographical context

About 25% of Cuba's territory is mountainous, with three distinct mountain ranges running east-west: the eastern (Sierra Maestra), central (*alturas*) and western (Cordillera de Guaniguanico) ranges. The remaining land is composed of extensive plains and basins. Cuba has a semi-tropical climate with two seasons: dry from November through April and rainy from May through October. The mean annual temperature is 26 °C, with a 23-28 °C range. The average precipitation is 1380 mm. All these conditions favour the dissemination of whiteflies, particularly during the dry season of the year, particularly in the regions marked in Figure 1.

Horticultural products have been traditional commodities in Cuba, particularly those referred to as *viandas* and basic grains. These crops include tomato (*Lycopersicon esculentum* Mill.), squash (*Cucurbita* spp. L.), cucumber (*Cucumis sativus* L. var. *sativus*), cabbage (*Brassica oleraceae* L.), sweetpotato (*Ipomoea batatas* [L.] Lam.), eggplant (*Solanum melongena* L.) and common bean (*Phaseolus vulgaris* L.). Vegetable production has become increasingly important to support a rapidly expanding tourist industry in Cuba.

The emergence of Bemisia tabaci *as a pest and virus vector*

The emergence of *Bemisia tabaci* (Gennadius) as a pest is probably linked to the advent and intensive use of agricultural pesticides soon after World War II. *B. tabaci* was reported as a pest of tobacco (*Nicotiana tabacum* L.) and a vector of plant viruses of common bean in the mid 1970s (Blanco and Bencomo, 1978). But it was not until 1989 that *B. tabaci* became a major production problem of tomato and common bean, as a vector of begomoviruses (Murguido et al., 1997). Currently, this whitefly species attacks tomato, common bean, squash, cucumber, melon (*Cucumis melo* L.), cabbage and eggplant throughout Cuba.

By 1990, the new B biotype of *B. tabaci* already had been introduced into Cuba and surrounding Caribbean islands (Brown, 1994). Between 1991 and 1993, the silver leaf syndrome induced by this new biotype was observed on squash (Murguido et al.,

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Figure 1. The main agricultural regions affected by whitefly-transmitted begomoviruses, Cuba.

1997). Another major production problem for Cuba and the Caribbean region occurred when and Old World virus, *Tomato yellow leaf curl virus* (TYLCV) was introduced from Israel into the Caribbean region, including Cuba (Polston and Anderson, 1997). This exotic virus has caused crop losses worth millions of dollars in the Caribbean region, and it is now known to occur in southern USA and the Peninsula of Yucatán, Mexico.

Advances in Biological Research

A main objective of the research undertaken in Cuba was to determine the composition of the whitefly population. A Cuban trainee, Ms. Yenín Hernandez of the Centro Nacional de Sanidad Agropecuaria (CENSA) conducted the tests at the Centro Internacional de Agricultura Tropical (CIAT) in Colombia, under the supervision of CIAT technical staff. Ninety-nine samples were collected from tomato plants in the localities of Quivicán and Alquizar, Havana Province. The results of the random amplified polymorphic DNA (RAPD) assay showed that 100% of the whitefly samples tested corresponded to biotype B of *B. tabaci*. Additionally, seven whitefly samples from potato (*Solanum tuberosum* L.) plants grown in Havana Province were also shown to consist of *B. tabaci* biotype B. These findings suggest that biotype B has adapted well and now predominates over biotype A of *B. tabaci* in Havana Province.

Bean golden yellow mosaic virus (BGYMV) has been an important disease of common bean in Cuba since the early 1970s (Blanco and Bencomo, 1978). The epicentre of the problem was the locality of Velasco in the western province of Holguín and it is now widely disseminated in the provinces of Holguín, Las Tunas, Guantánamo and Havana. In recent years, the incidence of BGYMV has increased in the provinces of Ciego de Avila, Holguín, Las Tunas and Camagüey.

When the monoclonal antibodies (MABs) to BGYMV were developed in 1990 (Cancino et al., 1995), the Cuban isolate of BGYMV reacted with a monoclonal antibody (MAB 2G5) produced to a Guatemalan isolate of BGYMV, which recognized all of the Middle American BGYMV isolates. By 1993, the Cuban isolates of BGYMV were not reacting with the specific MAB 2G5. As can be observed in Table 1, none of the BGYMV samples from three different provinces of Cuba reacted with the specific MAB-GA (2G5), including a BGYMV isolate from lima

Table 1. Assay of selected common bean samples with a broad spectrum and specific monoclonal antibodies prepared to *Bean golden yellow mosaic virus* (BGYMV), Guatemala, 1993.

Sample	Plant	Locality	MAB-BS[a]	MAB-GA[b]
1	Common bean	Velasco, Holguín	+	-
2	Lima bean	Velasco, Holguín	+	-
3	Common bean (Bonita 11)	Tomeguín, Matanzas	+	-
4	Common bean (Chévere)	Tomeguín, Matanzas	+	-
5	Common bean (Velasco Largo)	Tomeguín, Matanzas	+	-
6	Common bean (BAT 304)	Pulido, La Habana	+	-
7	Common bean	CIAT[c]	+	+
8	Common bean	CIAT[d]	-	-

a. MAB-BS, a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses.

b. MAB-GA, a monoclonal antibody used to detect the original Middle American isolates of BGYMV-Guatemala.

c. Sample 7, BGYMV-GA, Guatemala; CIAT, Centro Internacional de Agricultura Tropical.

d. Sample 8, healthy common bean.

bean (*Phaseolus lunatus* L.). The homologous control, BGYMV-GA, reacted with MAB 2G5 as expected. However, all of the diseased plant samples that were assayed reacted with the broad spectrum MAB (3F7) to BGYMV, which recognizes bipartite begomoviruses.

These tests were repeated in 1997, including some BGYMV-infected common bean plants from the Dominican Republic, a neighbouring Caribbean island (Table 2). The results show that the Cuban BGYMV isolates had changed their antigenic properties, whereas the Dominican BGYMV isolates had not. In order to investigate whether the failure of the MAB-GA to detect the Cuban BGYMV isolates was due to a major change in the capsid protein or putative replicase virus genes, one of the BGYMV isolates from Quivicán was partially sequenced. Table 3 shows the result of this analysis. These results suggest that the Cuban BGYMV isolate has not changed significantly in relation to the original BGYMV isolates from the region and that the Cuban isolate still can be considered a strain of BGYMV.

Table 2. Assay of selected common bean samples with a broad spectrum and specific monoclonal antibodies prepared to *Bean golden yellow mosaic virus* (BGYMV), Guatemala, 1997.

Sample	Plant	Locality	MAB-BS[a]	MAB-GA[b]
1	Common bean	Quivicán	+	-
2	Common bean	Quivicán	+	-
3	Common bean	Quivicán	+	-
4	Common bean	Dominican Republic	+	+
5	Common bean	Dominican Republic	+	+
6	Common bean	CIAT[c]	+	+
7	Common bean	CIAT[d]	-	-

a MAB-BS, a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses.

b. MAB-GA, a monoclonal antibody used to detect the original Middle American isolates of BGYMV-Guatemala.

c. Sample 6, BGYMV-GA, Guatemala; CIAT, Centro Internacional de Agricultura Tropical.

d. Sample 7, healthy common bean.

Table 3. Comparative homologies (%) between a Cuban *Bean golden yellow mosaic virus* (BGYMV) isolate and other common bean begomoviruses.

ORF[a]	BGYMV			BGMV-BR[b]	BDMV-CO[c]
	Guatemala	Dominican Republic	Puerto Rico		
AC1	91.5	91.5	89.8	71.9	69.9
AV1	91.3	91.6	91.3	81.6	78.1

a. ORF, open reading frame; *AC1*, replicase; and *AV1*, capsid protein.
b. BGMV-BR, *Bean golden mosaic virus*-Brazil.
c. BDMV-CO, *Bean dwarf mosaic virus*-Colombia.

With respect to tomato, one of the most affected crops in Cuba, three random samples were take in the locality of Quivicán, Havana Province, and tested for different pathogens (Table 4). As shown, the broad spectrum monoclonal antibody (MAB-BS) used to detect bi-partite begomoviruses did not detect any bipartite begomovirus in the tomato samples, although the observation of two of the samples in the electron microscope revealed the presence of begomovirus-like particles. This result suggested the need to test for the presence of TYLCV, a monopartite begomovirus reported to attack tomato in Cuba (Polston and Anderson, 1997). This introduced virus is best detected by polymerase chain reaction (PCR), using specific primers, kindly provided by D. P. Maxwell and M. K. Nakhla, of the Department of Plant Pathology, University of Wisconsin. The PCR assay detected the presence of TYLCV in two of the tomato samples from Quivican, Cuba.

In 1998, Dr. Gloria González of the Instituto de Investigaciones de Sanidad Vegetal (INISAV), Havana, Cuba, collected samples from symptomatic potato plants in Havana Province. As reported above, potato in Cuba is colonized also by the B biotype of *B. tabaci*. For this test, the viral nucleic acid extracted from affected potato plants was amplified by PCR using the primers developed by Rojas (1992). The amplified region (*AV1* and *AC1*) was cloned and sequenced. Table 5 shows the results of the comparative nucleotide sequence analyses conducted in reference to other whitefly-transmitted begomoviruses.

As can be observed from this comparative test, the begomovirus isolated from potato in Cuba is an isolate of *Tomato mottle Taino virus* (ToMoTV), a virus described in 1997 attacking tomato in Cuba (Ramos et al., 1997). This virus was also closely related to *Bean dwarf mosaic virus* from Colombia (93.3%), *Tomato yellow*

Table 4. Diagnostic assays practiced with three diseased tomato samples from Quivicán, Havana, Cuba.

Sample	EM[a]	MAB-BS[b]	PTY1[c]	CMV[d]	PCR-TYLCV[e]
1	Isometric	-	-	-	+
2	Isometric	-	-	-	+
3	-	-	-	-	-

a. EM, electron microscopy.
b. MAB-BS, a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses.
c. PTY1, potyviruses.
d. CMV, cucumoviruses.
e. PCR-TYLCV, DNA amplification of *Tomato yellow leaf curl virus*.

mosaic virus (ex-Potato yellow mosaic virus) from Venezuela, *Tomato mottle virus* from Florida, USA, *Sida golden mosaic virus* from Costa Rica and *Abutilon mosaic virus* (South American isolate and type species of the cluster in which the Cuban tomato virus is taxonomically placed).

Table 5. Comparative nucleotide sequence homology (%) between a begomovirus isolated from potato in Cuba and previously sequenced whitefly-transmitted begomoviruses.

Region	Virus[a]		
	ToMoTV	ToYMV	SiGMV
Coat protein (*AV1*)	98.5	84.2	85.1
Replicase (*AC1*)	97.7	83.7	82.4

a. ToMoTV, *Tomato mottle Taino virus*; ToYMV, *Tomato yellow mosaic virus*; and SiGMV, *Sida golden mosaic virus*.

Socio-economic Analysis

Cuba has a unique comparative advantage over the rest of the Latin American countries affected by whiteflies and whitefly-borne viruses, specifically, the limited use of agro-chemicals in this island. Moreover, Cuba has been able to develop and successfully implement biological control methods to combat the whitefly *B. tabaci*. One of the most effective entomopathogens produced has been *Verticillium lecanii*. This fungus is very pathogenic to the immature stages of *B. tabaci*. The production of biological control agents in Cuba constitutes an industrial activity in at least 15 provinces of Cuba, where it is being applied on a regular basis to horticultural crops affected by *B. tabaci*. In 1996, 170 tons of the fungus compound were produced in Cuba to treat 12,565 ha of affected horticultural crops. During the peak of the 1992 whitefly epidemics, 29,896 ha were treated in Cuba with this bio-control agent (Murguido et al., 1997).

Tabaquina, a concoction of tobacco leaf residues from the intensive Cuban tobacco industry, is another industrial subproduct used in Cuba to control whiteflies in a highly effective manner. Obviously, the well-planned cropping systems in Cuba also facilitate the implementation of legal measures regulating the time of planting for certain crops which act as reproductive host for the whitefly *B. tabaci* (Murguido et al., 1997).

The control of BGYMV in beans, on the other hand, is effectively carried out through the use of BGYMV-resistant bean cultivars developed in collaboration with the Programa Cooperativo Regional de Frijol para Centro América, México y el Caribe (PROFRIJOL) network and CIAT-Colombia.

Strengthened Research Capacity

The two main Cuban institutions collaborating in the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project were INISAV and the Instituto de Investigaciones de Hortícolas “Liliana Dimitroya” (LILIANA). During Phase I, a third Cuban institute, CENSA, joined the project. The sub-project’s coordinator for Cuba, Dr. Gloria González of INISAV, received funds to attend the VII Latin American and Caribbean Workshop on whiteflies and begomoviruses held on 26-30 October 1998, in Nicaragua. Dr. González also visited CIAT, Colombia, for a short but intensive training period on the characterization of whitefly-transmitted viruses.

The TWF-IPM Project contributed to the organization of an International Workshop on Begomoviruses in the Caribbean Region, held in Quivicán, Cuba on 24-29 November 1997. The project provided three keynote speakers: the coordinator, Dr. Francisco J. Morales; the past co-ordinator of the TWF-IPM Project, Dr. Pamela K. Anderson; and the main support scientist for the Caribbean region, Dr. Jane Polston of the University of Florida. Dr. Anderson explained the nature and objectives of the TWF-IPM Project; Dr. Polston delivered a talk on whitefly-transmitted viruses in the Caribbean Region and disserted on advanced techniques for the characterization of begomoviruses; and Dr. Morales gave a talk on breeding for disease resistance to whitefly-transmitted viruses.

The Whitefly Project also provided funds for the training at CIAT-Colombia of a junior scientist, Ms. Yenín Hernández, of CENSA, Cuba, on the molecular characterization of *B. tabaci* biotypes. CENSA is one of the leading institutes in Cuba in the area of Entomology.

Current Status of Whitefly/Begomovirus Problems

The whitefly situation in Cuba worsened following the invasion of the new B biotype of *B. tabaci*. However, the relatively low use of insecticides and the production of biological control agents in Cuba should maintain the whitefly problem at a manageable level. The Cuban provinces most affected by *B. tabaci* and the viruses it transmits have been Pinar del Río and Havana in the western end of the island and Holguín and Las Tunas in the eastern half of the country. The whitefly problem also has been severe in certain years in the provinces of Santiago and Guantánamo in the eastern end of Cuba. Other provinces affected have been Matanzas, Ciego de Avila, Granma, Camagüey, Villa Clara, Cienfuegos and Sancti Spiritus.

The provinces of Holguín and Las Tunas are located in one of the regions with less precipitation. Also, these provinces have high average temperatures (23.5 ºC) during the winter season (January), conditions that favour the reproduction of *B. tabaci*. The eastern end of the island concentrates the bulk of the tobacco and vegetable production of Cuba, which probably favours the development of large whitefly populations.

The use of entomopathogens, mainly the fungus *Verticillium lecanii*, constitutes a highly effective control measure against the proliferation of *B. tabaci*. Unfortunately, the production of this micro-organism does not meet the needs of the intensive agriculture practiced in Cuba. In the case of common bean, the use of varieties possessing high levels of resistance to BGYMV remains the main strategy for virus control.

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CHAPTER 3.11

Dominican Republic

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Introduction

Geographical context

The Dominican Republic comprises the eastern two-thirds of the island of Hispaniola. The country is generally mountainous, with the most prominent range being the Central Highlands (elevation up to 3175 m), the highest point in the West Indies. The Constanza Valley is a highly developed agricultural area located in the Central Highlands and is currently planted to high-value crops such as garlic (*Allium sativum* L.), onion (*Allium cepa* L.) and other horticultural crops. The Cibao Valley, in the north-west, is one of the most fertile agricultural areas in the country, where rice (*Oryza sativa* L.), maize (*Zea mays* L.), common bean (*Phaseolus vulgaris* L.), tobacco (*Nicotiana tabacum* L.) and coffee (*Coffea arabica* L.) are produced. The western part of the country is dry but rivers provide irrigation. San Juan de la Maguana is the main bean area and the Azua Valley is the main horticultural area in south-western Dominican Republic. The annual mean temperature is 25 °C and precipitation is 1346 mm. Tropical storms and hurricanes are a major threat every year (West and Augelli, 1977). Figure 1 shows the main agricultural regions affected by whitefly-transmitted begomoviruses.

The emergence of **Bemisia tabaci** *as a pest and virus vector*

Bemisia tabaci (Gennadius) was first observed attacking common bean in 1975. However, the presence of *Bean golden yellow mosaic virus* (BGYMV) in the Dominican Republic had been noticed as early as the late 1960s (Schieber, 1970). By 1988, other crops such as tomato (*Lycopersicon esculentum* Mill.), melon (*Cucumis melo* L.), eggplant (*Solanum melongena* L.), cucumber (*Cucumis sativus* L. var. *sativus*) and watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai) were under attack from *B. tabaci*. In 1991, biotype B of *B. tabaci* made its appearance in the main horticultural regions of the Dominican Republic. At the same time, the first begomovirus from the Old World, *Tomato yellow leaf curl virus* (TYLCV) was irresponsibly introduced into the Dominican Republic and the Americas. Following the introduction of a new and more aggressive biotype of the whitefly vector and an exotic begomovirus, crops such as tomato and melon practically disappeared, and the tomato processing plants had to close down in certain areas.

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Figure 1. The main agricultural regions affected by whitefly-transmitted begomoviruses, Dominican Republic.

Advances in Biological Research

The first begomovirus of economic importance to be characterized at the molecular level in the Dominican Republic was BGYMV. This virus was isolated in the early 1990s by the senior author and later shown to be molecularly and serologically related to the Guatemalan isolate of BGYMV (Faria et al., 1994; Cancino et al., 1995). The BGYMV isolate from the Dominican Republic (San Juan de la Maguana) proved to be similar to BGYMV isolates from the Caribbean (Puerto Rica) and Central America (Guatemala). The BGYMV isolate from the Dominican Republic was shown to be a different species when compared with *Bean golden mosaic virus* (BGMV) from Brazil. BGYMV affects over 20,000 ha of common bean in the Dominican Republic each year, particularly in the south-western valley of San Juan de la Maguana. The presence of a begomovirus infecting tomatoes in the Dominican Republic was first noticed in 1992 and subsequently identified as TYLCV from Israel (Polston et al., 1994; Polston and Anderson, 1997). The introduction of an exotic begomovirus from the Old World created a major pandemic throughout the Caribbean at a cost to tomato growers and tomato processing plants worth millions of US dollars in yield/export losses (Dupuy, 1998).

At the beginning of this project, in April 1997, common bean samples were collected in San Juan de la Maguana and tomato samples in Azua, in south-western Dominican Republic. The common bean samples reacted with the broad-spectrum monoclonal antibody 3F7 as well as with the monoclonal antibody prepared to the Guatemalan isolate of BGYMV. However, the tomato samples did not react against the broad-spectrum monoclonal antibody (MAB) in these tests. These tomato samples were then assayed by polymerase chain reaction (PCR) using two sets of primers (PTYIRV21/PTYIRC287 and PTYIRV21/RTYC2C1814) to TYLCV provided by Dr. M. Nakhla, Plant Pathology Department of the University of Wisconsin, Madison, WI. These primers amplified two fragments of M_r 287 and 1793 pb, respectively, demonstrating the presence of TYLCV in the tomato plants sampled in Azua.

In February 1998, six more tomato samples from Azua and one from the northern locality of Santiago were assayed with similar results (Table 1), demonstrating the endemic nature of TYLCV in the Dominican Republic. The three tomato samples (9-11) from the Centro de Investigaciones Aplicadas a Zonas Áridas (CIAZA), Azua, corresponded to TYLCV-tolerant tomato varieties (Gem-Pear, Gem-Star and UC82), which produce acceptably in this locality.

The common bean samples from the valley of San Juan de la Maguana behaved as expected for the BGYMV isolates previously detected in this locality. The presence of BGYMV in the Constanza Valley is interesting, because it is located at altitudes between 1000 and 1100 m, above the altitudinal range (0-950 m) at which *B. tabaci* is usually a problem. However, the incidence of the virus was moderate in the Constanza Valley.

Socio-economic Analysis

The agriculture of the Dominican Republic was described in the 1970s as a mixture of traditional and modern cropping systems. Traditional agriculture was equivalent to “subsistence” agriculture and included crops such as common bean, cassava (*Manihot esculenta* Crantz), rice, sweetpotato (*Ipomoea batatas* [L.] Lam.), pigeon pea (*Cajanus cajan* [L.] Millsp.) and taro (*Colocasia esculenta*

Table 1. Analyses of selected plant samples from the Dominican Republic.

Sample	Plant	Locality[a]	MAB-BS[b]	MAB-GA[c]	PTY1[d]	TYLCV[e]
1	Common bean	San Juan	+	+	-	nt
2	Common bean	San Juan	-	-	-	-
3	Common bean	Constanza	+	+	-	nt
4	Common bean	Constanza	+	+	-	nt
5	Garlic	Constanza	nt	nt	+	nt
6	Tomato	Azua	-	nt	nt	+
7	Tomato	Azua	-	nt	nt	+
8	Tomato	Azua	-	nt	nt	+
9	Tomato	CIAZA-Azua	-	nt	nt	+
10	Tomato	CIAZA-Azua	-	nt	nt	+
11	Tomato	CIAZA-Azua	-	nt	nt	+
12	Tomato	Santiago	-	nt	nt	+
13	Tomato	Santiago	-	nt	nt	+

a. CIAZA, Centro de Investigaciones Aplicadas a Zonas Áridas.
b. MAB-BS, a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses.
c. MAB-GA, a monoclonal antibody used to detect the original Middle American isolates of *Bean golden yellow mosaic virus*-Guatemala; and nt, no tests for these samples.
d. PTYI, monoclonal antibodies to detect potyviruses; and nt, no tests for these samples.
e. TYLCV, polymerase chain reaction detection of *Tomato yellow leaf curl virus*; and nt, no test for these samples.

[L.] Schott). Modern agriculture was equivalent to the “plantation” agriculture going back to colonial times, represented mostly by the extensive cultivation of sugarcane (*Saccharum officinarum* L.). In 1970, sugarcane was the main agro-industry of the country, occupying over 150,000 ha.

In 1966, a Land Reform Programme began to be implemented in the Dominican Republic. Large-scale farming in the form of co-operatives began to take place with the support of the government, the financial contribution of international agencies —for example the Agency for International Development (AID)—and the technical support of third countries. The Sisal Project, for instance, was created at that time with the help of the Government of Israel. The objective of the project was to produce tomato under irrigation in the valley of Azua. The irrigation system was initiated in 1970 with the construction of the Sabana Yegua Dam to irrigate some 18,000 ha. Thus, the Dominican Republic was one of the first countries in Latin America to look into the possibility of producing non-traditional, high-value crops for export such as tomato, chilli (*Capsicum annuum* L. var. *annuum*), melon, watermelon and eggplant. In 1970, 1600 ha of tomato were planted in the Dominican Republic. In the late 1990s, the tomato industry covered 8000 ha, employed over 6500 small-scale farmers (farms averaging less than 2.5 ha) and created jobs for 90,000 people per crop cycle (Dupuy, 1998).

The arrival of TYLCV from Israel and biotype B of *B. tabaci* brought the thriving tomato industry to a halt, causing economic losses in excess of 30 million US dollars. Table 2 shows the yield losses in the production of industrial tomato for the two main production areas, Azua and Cibao, between 1988 and 1995 (Alvarez and Abud-Antún, 1995). This crisis was managed through the adoption of legal measures that included a 3-month crop-free period to break the cycle of the whitefly vector. Additionally, virus-resistant tomato cultivars were introduced.

Table 2. Yield losses (%) caused by whitefly/ begomovirus damage to industrial tomatoes in the main tomato producing regions of the Dominican Republic.

Cropping season	Region	
	Azua	Cibao
1988-89	25	5
1989-90	45	10
1990-91	40	15
1991-92	30	15
1992-93	80	80
1993-94	95	50
1994-95	20	15

Following the implementation of legal measures against *B. tabaci*, the melon industry made a comeback. The case of common bean was not promising, because this crop was being displaced from the traditional bean-growing areas such as the valley of San Juan de la Maguana by other crops without whitefly problems and in higher demand, including cassava and sweetpotato. In 1984, there were 63,000 ha of red-seeded bean in the country and in 1994 the area planted to common bean is only 36,000 ha, almost half of the area planted 15 years ago. As a result, the country has had to import common bean, usually grain types of little demand in the country, considering that red-seeded bean is the main food staple in the diet of the Dominicans (Fundación de Desarrollo Agropecuario, 1995).

Strengthened Research Capacity

Considering that the two main horticultural regions affected by whiteflies and whitefly-transmitted geminiviruses in the Dominican Republic are Azua and Cibao, two Agricultural Engineers were contacted to collaborate in this project. They were Augusto Villar of the Secretaría del Estado de Agricultura (SEA)-CIAZA, Azua, and Emigdio Gómez of SEA-Programa Nacional de Manejo Integrado de Plagas (MIP), N-N.W. Cibao. These national programme scientists were brought to the Centro Internacional de Agricultura Tropical (CIAT) for training in the identification of *B. tabaci* biotypes and diagnosis of viruses affecting crops of socio-economic importance in their respective agricultural regions. Table 3 shows the results obtained by these two researchers at CIAT with the whitefly samples they collected in the Dominican Republic.

The two researchers were trained also in the identification of geminiviruses and other plant viruses using the enzyme-linked immunosorbent assay (ELISA) technique in conjunction with the utilization of monoclonal and polyclonal antibodies and financed by the project to attend the International Workshop on *Bemisia* and Geminiviruses, held in San Juan, Puerto Rico, June 1998. Here they presented the novel approach adopted in the Dominican Republic to control whitefly/begomovirus damage by regulatory measures designed to break the continuous populational cycle of *B. tabaci* in tropical environments.

The project also supported the activities of several researchers on horticultural crops in the Constanza Valley, in the central region of the country, in collaboration with the horticultural research network, La Red Colaborativa de Investigación y Desarrollo de Hortalizas para América Central, Panamá y República Dominicana (REDCAHOR). Two conferences were delivered on the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project and on the breeding and selection of horticultural crops for their resistance to whitefly-transmitted begomoviruses. The CIAT Bean Project is collaborating with the Dominican Republic to breed Dominican bean cultivars, mainly the Pompadour types, for resistance to BGYMV.

Table 3. Results of the analyses of *Bemisia* spp. biotypes found in the Dominican Republic on different economically important crops.

Locality	Crop	No. samples	Whitefly
Azua-CIAZA[a]	Eggplant	3	*Bemisia tuberculata*
Azua-Estebanía	Tomato	9	*Bemisia tabaci*-B
Barahona	Eggplant	3	*Bemisia tuberculata*
Azua-Arroyo Salado	Eggplant	3	*Bemisia tuberculata*
San José de Ocoa	Tomato	6	*Bemisia tabaci*-B
Santiago	Okra	3	*Bemisia tabaci*-B
Santiago	Cucumber	3	*Bemisia tabaci*-B
Santiago	Eggplant	3	*Bemisia tabaci*-B
Montecristi	Melon	6	*Bemisia tabaci*-B
Santiago	Tomato	3	*Bemisia tabaci*-B
Santiago	Okra	3	*Bemisia tabaci*-B

a. CIAZA, Centro de Investigaciones Aplicadas a Zonas Áridas.

Current Situation of Whitefly/Geminivirus Problems

The Dominican Republic constitutes a unique example of a country that implemented legal measures to minimize yield losses due to the activity of *B. tabaci* both as a pest and virus vector. Nevertheless, pesticide use remains high in most agricultural regions of the country, and the existing cropping systems favour the reproduction of *B. tabaci*. Currently, the situation in the lowlands and highlands remains under partial control, and some crops that had been taken out of production such as melon are being planted again. Another crop and good host to the whitefly *B. tabaci*, tobacco, also has been increasing its area in the country. A recent viral epidemic in tobacco in the Cibao region was diagnosed at CIAT as being caused by *Tobacco mosaic virus*.

In the case of tomato, the cultivation of varieties resistant to TYLCV should contribute to the recovery of the production of industrial tomato plantings. In the case of common bean, however, we have noticed a marked reduction in the area planted, because of the susceptibility of most local varieties to BGYMV. Moreover, TYLCV has been reported to attack common bean in other parts of the world (Navas-Castillo et al., 1999).

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CHAPTER 3.12

Reproductive Crop Hosts of *Bemisia tabaci* (Gennadius) in Latin America and the Caribbean

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Introduction

The sweetpotato whitefly *Bemisia tabaci* (Gennadius) is a vector of at least 30 begomoviruses (Geminiviridae: Begomovirus) in Latin America and the Caribbean (Morales and Anderson, 2001). As is often the case for insect vector-host relations, many of the crops that begomoviruses affect are not reproductive hosts for *B. tabaci*. A first critical step in vector entomology, then, is to answer the question. "Where is *B. tabaci* reproducing?"

Crop biomass can provide excellent breeding and feeding sources and thus give rise to large populations of *B. tabaci* (Byrne et al., 1991). We argue here that the first step to understanding regional population dynamics as the basis for area-wide management is to elucidate which crops *B. tabaci* utilizes as reproductive hosts and define their relative importance.

Crop Hosts of *Bemisia tabaci*

The most reliable sources of host plant records for *B. tabaci* are published taxonomic reports and unpublished records from museum collections. The first author has reviewed the white and grey literature as well as the arthropod collections from the United States National Museum (USNM), the Florida State Collection of Arthropods (FSCA), the British Museum of Natural History (BMNH), and the Escuela Agrícola Panamericana (EAP) in Honduras. The EAP (1992) references are unpublished records from Latin America, collected and identified by R. Caballero as the basis for development of several taxonomic keys (Caballero, 1992). Table 1 also records *B. tabaci* host records generated by the national program (national agricultural research systems [NARS]) teams as part of the extensive diagnostic surveys for Phase 1 of the Tropical Whitefly Integrated Pest Management Project (TWF-IPM). P. Hernández identified these specimens and A. Hamon verified them. Verified specimens are deposited in the insect museum at the Centro Internacional de Agricultura Tropical (CIAT) in Cali, Colombia, and the FSCA in Gainesville, Florida, USA.

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ψ British Museum of Natural History, London, UK.

Based upon the reliable published taxonomic reports, unpublished museum records and the verified specimen collected during Phase 1 of the TWF-IPM, we conclude that *B. tabaci* is utilizing at least 50 cultivated species as reproductive hosts in Latin America and the Caribbean (Table 1).

Table 1. Known crop hosts of *Bemisia tabaci* in Latin America and the Caribbean.

Crop		Country	Reference[a]
Latin name	Common name		
Abelmoschus esculentus (L.) Moench [= *Hibiscus esculentus* L.]	Okra	Cuba	Vázquez et al. (1996)
		Dom. Rep.	TWF-IPM Project
		El Salvador	USNM (1967), Lipes (1968), WF-IPM Project
		Guatemala	TWF-IPM Project
		Trinidad	BMNH (1989)
		Venezuela	Arnal et al. (1993b)
Annona muricata L.	Soursop	Cuba	Vázquez et al. (1996)
Arachis hypogaea L.	Peanut	Cuba	Vázquez et al. (1996)
		Venezuela	Arnal et al. (1993b)
Beta vulgaris L.	Beet	Cuba	Vázquez et al. (1996)
Beta vulgaris subsp *cicla* (L.) W. Koch	Swiss chard	Cuba	Vázquez et al. (1996)
Brasica rapas L. subsp. *rapa*	Turnip	Cuba	Vázquez et al. (1996)
Brassica oleracea L. var. *capitata* L.	Cabbage	Cuba	Vázquez et al. (1995)
Brassica oleracea L. var. *gemminifera* DC	Brussel sprouts	Venezuela	Arnal et al. (1993b)
Brassica oleracea L. var. *gongylodes* L.	Kohl-rabi	Cuba	Vázquez et al. (1995)
Cajanas cajan (L.) Millsp.	Pigeon pea	Cuba	Vázquez et al. (1996)
Canavalia ensiformis (L.) DC.	Jack bean, sword bean	Cuba	Vázquez et al. (1996)
Capsicum annuum L. *var. annuum*	Bell pepper	Belize	EAP (1992)
		Costa Rica	TWF-IPM Project
		Cuba	Vázquez et al. (1996)
		Honduras	EAP (1992)
		Panama	TWF-IPM Project
		Venezuela	Arnal et al. (1993b)
Capsicum spp.	Chilli peppers	Costa Rica	TWF-IPM Project
		Cuba	Vázquez et al. (1996)
		El Salvador	TWF-IPM Project
		Guatemala	TWF-IPM Project
		Honduras	TWF-IPM Project
		Nicaragua	EAP (1992)
		Venezuela	Arnal et al. (1993b)
Carica papaya L.	Papaya	Cuba	Vázquez et al. (1996)
		Guatemala	TWF-IPM Project
Cicer arietinum L.	Chick-pea	Cuba	Vázquez et al. (1996)
Citrullus lanatus (Thunb.) Matsum. & Nakai [= *Citrullus vulgaris* Schrad.]	Watermelon	Belize	EAP (1992)
		Colombia	TWF-IPM Project
		Costa Rica	TWF-IPM Project
		Cuba	Vázquez et al. (1995)
		Ecuador	TWF-IPM Project
		El Salvador	TWF-IPM Project
		Guatemala	TWF-IPM Project
		Honduras	EAP (1992)
		Panama	TWF-IPM Project
		Venezuela	Arnal et al. (1993b)
Cucumis melo L.	Melon	Belize	EAP (1992)
		Colombia	TWF-IPM Project
		Costa Rica	TWF-IPM Project
		Dom. Rep.	TWF-IPM Project
		Ecuador	TWF-IPM Project
		El Salvador	TWF-IPM Project

(Continued)

Table 1. (Continued.)

Crop		Country	Reference[a]
Latin name	Common name		
		Guatemala	EAP (1992), TWF-IPM Project
		Honduras	EAP (1992)
		Venezuela	Arnal et al. (1993a; 1993b), TWF-IPM Project
Cucumis sativus L.	Cucumber	Colombia	TWF-IPM Project
		Costa Rica	TWF-IPM Project
		Cuba	Vázquez et al. (1996)
		Dom. Rep.	TWF-IPM Project
		El Salvador	TWF-IPM Project
		Guatemala	TWF-IPM Project
		Honduras	EAP (1992), TWF-IPM Project
		Venezuela	Arnal et al. (1993b), TWF-IPM Project
Cucurbita argyrosperma C. Huber subsp. *argyrosperma*	Pipián	El Salvador	TWF-IPM Project
Cucurbita maxima Duch. Ex Lam.	Squash	Cuba	Vázquez et al. (1996)
		Venezuela	Arnal et al. (1993b), TWF-IPM Project
Cucurbita moschata (Duch. ex Lam.) Duch. ex Poir.	Ayote	Colombia	TWF-IPM Project
		El Salvador	TWF-IPM Project
		Honduras	EAP (1992)
		Panama	TWF-IPM Project
Cucurbita pepo L.	Squash	Ecuador	TWF-IPM Project
		Nicaragua	USNM (1958)
		Venezuela	Arnal et al. (1993b)
Euphorbia pulcherrima Willd. Ex Klotzch	Poinsettia	El Salvador	USNM (1967)
Fernaldia pandurata (A. DC.) Woodson	Loroco	El Salvador	TWF-IPM Project
Glycine max (L.) Merr.	Soybean	Argentina	TWF-IPM Project
		Brazil	BMNH (1974)
		Colombia	EAP (1992), TWF-IPM Project
		Cuba	Vázquez et al. (1996)
		Ecuador	TWF-IPM Project
		Honduras	EAP (1992)
		Venezuela	Arnal et al. (1993a; 1993b)
Gossypium hirsutum L.	Cotton	Argentina	Viscarrret and Botto (1996)
		Barbados	BMNH (1982)
		Colombia	EAP (1992), TWF-IPM Project
		El Salvador	USNM (1964; 1965; 1967; 1969)
		Guatemala	EAP (1992)
		Honduras	USNM (1966)
		Nicaragua	USNM (1951; 1965; 1978)
		Venezuela	Arnal et al. (1993b)
Helianthus annuus L.	Sunflower	Cuba	Vázquez et al. (1996)
		Venezuela	Arnal et al. (1993b)
Ipomoea batatas (L.) Lam.	Sweetpotato	Cuba	Vázquez et al. (1996)
		Venezuela	Arnal et al. (1993b)
Lactuca sativa L.	Lettuce	Cuba	Vázquez et al. (1996)
		Venezuela	Arnal et al. (1993b)
Lycopersicon esculentum Mill.	Tomato	Belize	EAP (1992)
		Colombia	EAP (1992), TWF-IPM Project

(Continued)

Table 1. (Continued.)

Crop		Country	Reference[a]
Latin name	Common name		
		Costa Rica	TWF-IPM Project
		Cuba	Vázquez et al. (1996)
		Dom. Rep.	EAP (1992), TWF-IPM Project
		El Salvador	USNM (1967), TWF-IPM Project
		Guatemala	EAP (1992), TWF-IPM Project
		Honduras	EAP (1992), TWF-IPM Project
		Mexico	BMNH (1992)
		Nicaragua	USNM (1952), TWF-IPM Project
		Panama	TWF-IPM Project
		Puerto Rico	BMNH (1988)
		Trinidad	BMNH (1990)
		Venezuela	Arnal et al. (1993a; 1993b), TWF-IPM Project
Mangifera indica L.	Mango	Cuba	Vázquez et al. (1996)
Manihot esculenta Crantz	Cassava	Colombia	EAP (1992), TWF-IPM Project
		Cuba	Vázquez et al. (1996)
		Dom. Rep.	TWF-IPM Project
		Ecuador	TWF-IPM Project
		Venezuela	Arnal et al. (1993b)
Musa × *paradisiacal* L.	Banana/plantain	Cuba	Vázquez et al. (1996)
		Venezuela	Arnal et al. (1993b)
Nicotiana tabacum L.	Tobacco	Cuba	Vázquez et al. (1996)
		Guatemala	TWF-IPM Project
		Nicaragua	USNM (1951)
		Venezuela	Arnal et al. (1993b)
Ocimum basilicum L.	Basil	Cuba	Vázquez et al. (1996)
Passiflora edulis Sims	Passionfruit	Venezuela	Arnal et al. (1993b)
Persea americana Mill.	Avocado	Cuba	Vázquez et al. (1996)
		Guatemala	USNM (1954)
		Venezuela	Arnal et al. (1993b)
Phaseolus acutifolius A. Gray var. *acutifolius*	Tepary bean	El Salvador	TWF-IPM Project
Phaseolus lunatus L.	Lima bean	Brazil	BMNH (1975)
		Venezuela	Arnal et al. (1993b)
Phaseolus vulgaris L.	Common bean	Argentina	USNM (1982), TWF-IPM Project
		Colombia	USNM (1974), TWF-IPM Project
		Costa Rica	TWF-IPM Project
		Cuba	Vázquez et al. (1996)
		Dom. Rep.	BMNH (1979); TWF-IPM Project
		Ecuador	TWF-IPM Project
		El Salvador	USNM (1964); TWF-IPM Project
Phaseolus vulgaris L.	Common bean	Guatemala	USNM (1973); TWF-IPM Project
		Honduras	EAP (1992); TWF-IPM Project
		Nicaragua	USNM (1974); TWF-IPM Project
		Panama	TWF-IPM Project
		Venezuela	Arnal et al. (1993a; 1993b)

(Continued)

Table 1. (Continued.)

Crop		Country	Reference[a]
Latin name	Common name		
Psidium guajava L.	Guava	Cuba	Vázquez et al. (1996)
		Nicaragua	USNM (1978)
Raphanus sativus L.	Radish	Cuba	Vázquez et al. (1995)
Sesamum indicum L.	Sesame	Cuba	Vázquez et al. (1996)
		Venezuela	Arnal et al. (1993b)
Solanum melongena L.	Eggplant	Colombia	TWF-IPM Project
		Cuba	Vázquez et al. (1996)
		Dom. Rep.	EAP (1992), TWF-IPM Project
		Ecuador	TWF-IPM Project
		Guatemala	TWF-IPM Project
		Mexico	TWF-IPM Project
		Puerto Rico	BMNH (1987)
		Trinidad	BMNH (1988)
		Venezuela	Arnal et al. (1993b)
Solanum tuberosum L.	Potato	Argentina	USNM (1982)
		Cuba	Vázquez et al. (1996)
		Honduras	EAP (1992)
		Nicaragua	EAP (1992)
		Venezuela	Arnal et al. (1993a; 1993b)
Spondias purpurea L.	Spanish plum	Cuba	Vázquez et al. (1996)
Vicia faba L.	Broad bean	Cuba	Vázquez et al. (1996)
Vigna unguiculata (L.) Walp.	Cowpea	Cuba	Vázquez et al. (1996)
		Venezuela	Arnal et al. (1993b)
Vigna unguiculata subsp. *sesquipedalis* (L.) Verdc.	Yard-long bean	Cuba	Vázquez et al. (1996)
Xanthosoma sagittifolium (L.) Schott	Malanga	Cuba	Vázquez et al. (1996)

a. For references, see list at end of chapter.
BMNH, British Museum of Natural History, data from various dates;
EAP, Escuela Agrícola Panamericana, Honduras, unpublished data;
USNM, United States National Museum, data from various dates;
TWF-IPM Project, Tropical Whitefly-Integrated Pest Management Project.

Relative Importance of *Bemisia tabaci* on Different Crop Hosts

B. tabaci is cosmopolitan in distribution, highly polyphagous and characterized by intercrop movement (Butler et al., 1986). Based on identified museum specimens, Mound and Halsey (1978) listed 317 plant species as host plants for *B. tabaci*, worldwide. However, sufficient empirical and experimental evidence exists to indicate *B. tabaci* populations do not reproduce equally among all potential crop hosts in any given geographical region. Rather, *B. tabaci* appears to develop strong host associations, resulting in host races. A host race is a population of a species that is partially reproductively isolated from other non-specific, sympatric sister populations as a direct consequence of adaptation to a specific host or habitat (Diehl and Bush, 1984; Bush, 1994). Host races are one category of biotypes, that is, two or more morphologically similar or indistinguishable taxa that differ from one another in a biologically significant way such as host preference, emergence time or some other ecological or behavioral trait (Bush, 1994).

In any given geographical area, beyond more precisely defining the range of reproductive crop hosts, it is necessary to define the relative importance, or reproductive potential, of the host plants, that is, to quantify the average number of *B. tabaci* produced per unit area. For example, intensive surveys carried out in Venezuela (Arnal et al., 1993a; 1993b) and Cuba (Vázquez et al., 1995; 1996) each identified 27 and 39reproductive crop hosts for *B. tabaci* respectively (Table 1). The next step is to identify the relative importance of each crop as a reproductive host.

Caballero and Nolasco (1995) made 289 collections of *B. tabaci* from economically important crop hosts in Belize, Costa Rica, El Salvador, Guatemala, Honduras, Nicaragua and Panama. They quantified *B. tabaci* abundance for each crop utilizing *B. tabaci* density/leaf as an indicator of abundance (Table 2).

However, comparisons of whitefly abundance on different crops is problematic because of variation in architecture, growth habits, foliage density and distribution of *B. tabaci* within the plant, among different crops and among different varieties within the same crop. Density measures on a per ground unit basis are necessary for meaningful comparisons. Recently S. Naranjo and L. Cañas (United States Department of Agriculture [USDA]- Agricultural Research Service [ARS]) have developed a protocol to quantify *B. tabaci* abundance for a range of crop plants within the same area on a per ground unit basis. This protocol is currently being tested in Arizona (Cañas, Naranjo and Ellis, in preparation) and in El Salvador (Serrano and Anderson, in preparation). Such quantification will allow us to compare the relative importance of *B. tabaci* reproductive hosts within and among zones in a country, and among different countries; to monitor fluctuations in the *B. tabaci* populations on the same crop in the same zone from year to year; and to identify newly emerging reproductive hosts for *B. tabaci*.

We cannot effectively implement crop protection tactics aimed at source reduction within an area-wide management program if we cannot prioritize and target the principal reproductive sources for *B. tabaci*.

Table 2. Relative importance of *Bemisia tabaci* crop hosts in Central America, based on leaf samples.

Ranking[a]	Crop	Common name
5	*Gossypium hirsutum* L.	Cotton
	Citrillus lanatus (Thumb.) Matsum. & Nakai	Watermelon
	Cucurbita pepo L.	Squash
4	*Lycopersicon esculentum* Mill.	Tomato
	Cucmus melo L.	Melon
	Capsicum annuum L.	Sweet pepper
3	*Nicotiana tabacum* L.	Tobacco
	Solanum melongena L.	Eggplant
	Phaseolus vulgaris L.	Common bean
2	*Glycine max* L. (Merr.)	Soybean

a. Rank 5, up to 100 nymphs per leaf, in all countries surveyed; rank 4, up to 100 nymphs per leaf, in certain countries; rank 3, up to 50 nymphs per leaf, in all countries; rank 2, up to 20 nymphs per leaf, in all countries; and rank 1, less than 10 nymphs per leaf, in all countries.

Conclusions and Recommendations

After more than a decade of epidemic events in Mexico, Central America and the Caribbean, we still do not have a clear enough understanding of the range of reproductive crop hosts for *B. tabaci*. Nor do we understand the relative importance of the *B. tabaci* reproductive hosts within a zone, for purposes of area-wide management. The question, "where is *B. tabaci* reproducing?", continues to be a basic research priority for vector entomology in the region.

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CHAPTER 3.13

Using Molecular Techniques to Analyse Whitefly Species and Biotypes in Latin America

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Introduction

When identifying and describing insect taxa, morphology has been used historically to separate species. Among many groups of insects, however, morphological characters can vary with respect to environmental factors within a single species, or be so convergent and cryptic among closely related species as to be of limited usefulness. Under such conditions, studies of their biology and molecular profiles become essential to defining species and characterizing populations. At a molecular level, protein and DNA polymorphisms can be combined with studies of biological characteristics by using one of four experimental or technological approaches: electrophoresis of allozymes, analysis of randomly amplified polymorphic DNAs (RAPDs) and nucleic acid sequence comparisons of nuclear or mitochondrial DNA markers. Here, we review the application of molecular approaches to characterizing whitefly *Bemisia tabaci* (Gennadius) populations and biotypes in Latin America.

Each method has its own characteristic advantages and disadvantages. Isozyme analyses using starch—or polyacrylamide—gel electrophoresis have been in use for several decades and are useful for processing large numbers of samples relatively inexpensively compared to DNA sequencing. However, because the technique relies on detecting enzymes, samples must be kept live or frozen in order to preserve activity. The technique is also less sensitive than DNA approaches because the underlying nucleic acid variability is usually masked.

When, in the early 1980s, changes in whitefly populations and associated begomovirus infections were first noticed, protein polymorphisms were employed to investigate natural populations of *B. tabaci*. Differences in esterase isozyme patterns were used to describe two biotypes in the Americas: a now-known-to-be native form, or biotype A, and a second form, biotype B, which exhibited high population density, wide host plant range, relatively high insecticide resistance and was capable of inducing “silverleaf” symptoms on some plants (Brown et al., 1995). *B. tabaci* biotype A was predominant in most regions of the Americas but many of these populations now have been displaced by *B. tabaci* biotype B. Other reports suggested that there might be additional biotypes in Latin America (Wool et al., 1994). Eighteen other biotypes from throughout the world

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have been described based on esterase banding patterns alone (Brown et al., 1995). Reliable morphological markers, which do not vary with ecological parameters (Mound, 1963; David and Ananthakrishnan, 1976; Mohanty and Basu, 1986) and which can distinguish between biotypes, are not known (Rosell et al., 1997).

DNA sequence comparisons can be made between individuals or populations using polymerase chain reaction (PCR) generated sequences from several markers in either the nuclear or mitochondrial genome. The latter offers some advantages because it is maternally inherited and non-recombining and contains known and predictable gene sequences for which general insect primers have been designed. Some sequences vary enough to be useful for population level analyses (e.g., the control region, parts of cytochrome oxidases I-III), while others evolve slowly enough to be more appropriate to analyses above the species level (e.g., conserved parts of cytochrome oxidase I [COI], 12S and parts of the 16S r-DNA). However, as with some nuclear markers, comparisons between mitochondrial sequences may reflect local gene evolution and not population or species evolution. Nuclear data offer a wide range of neutral markers but can be particularly difficult to design PCR primers for specific targets in poorly known or unknown genomes (e.g., introns). Thus, many studies have used variable regions surrounded by relatively conserved sequences (e.g., internal transcribed spacers or ITS of the rDNA genes) for population/species analyses, or more conserved sequences (e.g., 18S rDNA) for higher order studies.

An alternative approach to specific nuclear markers involves the use of short, random sequence PCR primers that anneal randomly in the genome and produce characteristic banding patterns of collections of PCR products when run on an agarose gel (RAPDs). Advantages here include the low cost and capability of processing large numbers of samples without the necessity of cloning or sequencing PCR products as well as no requirement for detailed sequence knowledge about the genome. Disadvantages include the fact that repeatability can be difficult, and the polymorphisms may be hard to distinguish from PCR artefacts. That is, template preparation and amplification conditions must be consistent and tightly controlled. Interpretation of negative data (absence of bands) also may have numerous explanations.

Perring et al. (1993) used RAPD patterns in combination with allozyme frequency data and mating and behavioural studies to elevate the B biotype to new species status, *Bemisia argentifolii* Bellows & Perring. In a detailed study of 20 RAPD primers, however, Gawel and Bartlett (1993) concluded that the technique was not useful for clarifying taxon status in the case of whiteflies. Campbell et al. (1994) were the first to look at a nuclear DNA sequence, albeit highly conserved, and showed that only a single unique nucleotide difference lies between *B. tabaci* biotypes A and B in the 18S gene. More recently, Frohlich et al. (1999) used variable portions of the mitochondrial COI gene and 16S rDNA sequence to evaluate *B. tabaci* populations from four continents, and concluded that biotype B is a recent introduction from the Old World to the Americas. De Barro et al. (2000), using ribosomal ITS 1, reached the same conclusion and cautioned that if the status of the B biotype were to remain species novum, a taxonomic review of all of the biotypes of *B. tabaci* would have to be made.

For this study of the whiteflies affecting crops of economic importance, a variable portion of the 16S mitochondrial ribosomal DNA was chosen for evolutionary analysis. It is more variable than the 18S mt DNA but less so than the COI gene. This allows both a comparison of distinct genera as well as closely related biotypes.

Methods and Materials

PCR, cloning and sequence analysis of a region of the 16S mitochondrial DNA

The primers 4119 (5' CGCCTGTTTAACAAAAACAT) and 4118 (5' CCGGTCTGAACTCAG ATCACGT 3') were used to amplify a region of the 16S mitochondrial DNA (Xiong and Kocher, 1991). The PCR reaction conditions were 30 cycles of 1 min at 95 °C, 50 s at 50 °C and 50 s at 72 °C, and in the last cycle the 72 °C reaction was extended to 10 min. The products were purified using the Wizard™ PCR purification columns (Promega, WI, USA) and were visualized by agarose gel electrophoresis. The PCR products were cloned into the plasmid PCR script amp SK(+)™ (Stratagene, La Joya, CA, USA). Using the ABI dye terminator kit, the sequences were determined in an automated sequencer using the dideoxynucleotide chain termination procedure (Sanger et al., 1977).

Phylogenetic analyses

Phylogenetic analyses were done with multiple individuals within populations. DNA sequences were aligned using the ClustalW algorithm (Thompson et al., 1994) by the ClustalW 1.7 program (BCM Search Launcher at the Human Genome Centre, Baylor College of Medicine, Houston, TX, USA). Because different tree building algorithms make different evolutionary assumptions, data were evaluated by parsimony, neighbour joining and maximum-likelihood. All analyses were performed with PAUP, version 4.0b2, for Macintosh (Swofford, 1999). For parsimony, the branch-and-bound method was used (characters unordered, equal weight). Bootstrapping was performed with the branch-and-bound option for 2000 replicates (stepwise sequence addition, tree-bisection-reconnection [TBR], MulTrees option). For neighbour joining, distances were calculated using the Kimura 2 parameter model. Maximum-likelihood trees were constructed with a transition/transversion ratio of 2.0 by heuristic search (100 replicates, random addition sequence, MulTrees, TBR) (Swofford, 1999).

Whitefly survey

The whiteflies that were identified by RAPD PCR analysis were collected and processed.[1]

RAPD PCR analysis

Total DNA was isolated from individual whiteflies using a method developed for plants (Gilbertson et al., 1991). The DNA was amplified in a PCR. The Operon primers F2 (5'GAGGATCCCT3'), F12 (5'ACGGTACCAG3'), H9 (5'TGTAGCTGGG3') and H16 (5'TCTCAGCTGG3') (De Barro and Driver, 1997) were tested for their efficacy to distinguish whiteflies in Latin America. The reaction conditions for the first cycle were 5 min at 94 °C, 2 min at 40 °C and 3 min at 72 °C. This was followed with 39 cycles of 1 min at 94 °C, 1.5 min at 40 °C, and

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[1] For further information on specific methodologies used to produce the results published in this book, please contact the information and communication assistant of the Project (www.tropicalwhiteflyipmproject.cgiar.org).

2 min at 72 °C. The PCR products were run in agarose gels stained with ethidium bromide and visualized using UV light.

Results

Analysis of selected whiteflies in Latin America

A region of the 16S mitochondrial DNA of *B. tabaci* biotypes A and biotype B, *B. tuberculata*, *Aleurotrachelus socialis* (Bondar), *Trialeurodes vaporariorum* (Westwood) and *T. variabilis* (Quaintance) were compared by parsimony and distance analysis using Phylip version 3.57 (Felsenstein, 1993). At least two independent clones from each whitefly population of the 3’ region of the mitochondrial 16S gene were prepared and sequenced. Included in the comparison were sequence data of the mitochondrial 16S gene from *B. tabaci* biotype A of Arizona (Genbank accession: AF110722), Costa Rica (Genbank accession: AF110715) and Puerto Rico (Genbank accession: AF110719) (Frohlich et al., 1999). In the analysis of various *B. tabaci*, those of biotype A were grouped together and there was at least 97% identity with a maximum mean distance of 0.02 (Table 1) (Calvert et al., 2001). The *B. tabaci* biotype A is widespread throughout the region of the whitefly survey.

In the distance analysis, the Israel *B. tabaci* isolate (Genbank accession: AF110717) was 98.8% identical with the *B. tabaci* biotype B isolates from Colombia (Table 1). Comparisons were also made with individuals from Colombia, Arizona, Israel and Yemen and all were at least 98.2% identical (data not shown). Given the rapid spread of the B biotype, the lack of diversity between populations in Arizona USA and Colombia was expected. This gives additional evidence that *B. tabaci* biotype B was recently introduced into the Americas from the region of the Middle East.

A representative from each species or biotype was used in the analysis to find the most parsimonious tree (Figure 1). *B. tabaci* biotypes A and

Table 1. Mean distances for a 3’ region of mitochondrial 16S ribosomal gene in fifteen individual whiteflies representing different species and populations.

Whitefly[a]	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
1 *B. tabaci* B Sucre	0.00	0.01	0.01	0.01	0.10	0.10	0.09	0.10	0.10	0.22	0.35	0.35	0.37	0.37	0.36
2 *B. tabaci* B CT cass		0.00	0.00	0.00	0.08	0.09	0.08	0.09	0.09	0.21	0.34	0.34	0.36	0.36	0.35
3 *B. tabaci* B CT bn			0.00	0.00	0.09	0.10	0.09	0.09	0.09	0.21	0.34	0.35	0.37	0.36	0.35
4 *B. tabaci* Israel				0.00	0.08	0.09	0.08	0.09	0.09	0.21	0.34	0.34	0.36	0.36	0.35
5 *B. tabaci* A CT 1					0.00	0.01	0.01	0.02	0.00	0.22	0.34	0.35	0.36	0.35	0.38
6 *B. tabaci* A CR						0.00	0.01	0.02	0.01	0.23	0.35	0.36	0.37	0.36	0.39
7 *B. tabaci* A AZ							0.00	0.02	0.01	0.22	0.34	0.36	0.37	0.36	0.38
8 *B. tabaci* A PR								0.00	0.02	0.22	0.35	0.36	0.36	0.36	0.38
9 *B. tabaci* CT 2									0.00	0.22	0.34	0.35	0.36	0.36	0.38
10 *B. tuberculata* CT										0.00	0.28	0.28	0.37	0.36	0.38
11 *A. socialis* Mon											0.00	0.07	0.40	0.39	0.43
12 *A. socialis* CT												0.00	0.41	0.40	0.41
13 *T. vaporariorum* CT													0.00	0.00	0.39
14 *T. vaporariorum* AZ														0.00	0.38
15 *T. variabilis* CT															0.00

a. *B, Bemisia*; *A, Aleurotrachelus*; *T, Trialeurodes*.

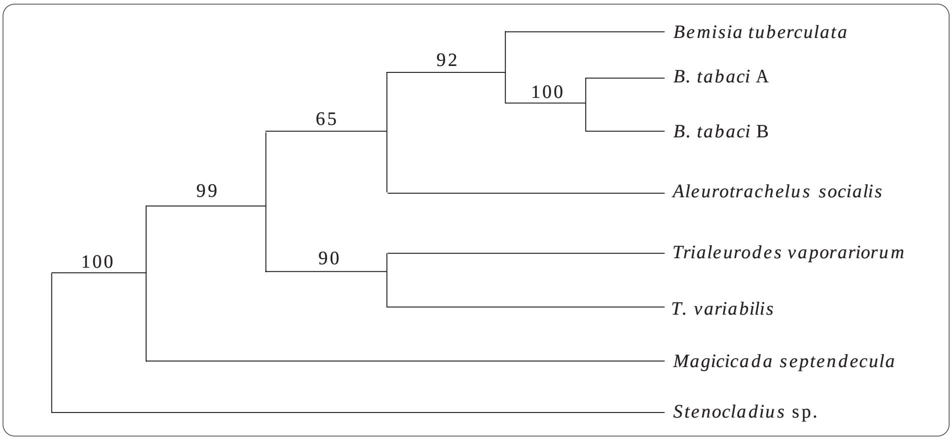

Figure 1. Cladogram showing the relationship between the whiteflies in this study. The cladogram is based on the most parsimonious tree inferred from the analysis of 485 base sites of a region of the mitochondrial 16S gene. Numbers above the branches indicate the level of statistical support for the corresponding node from 10,000 bootstrap replicates.

B were always grouped together. *B. tuberculata* was grouped with the *B. tabaci* 92% of the time and *T. vaporariorum* and *T. variabilis* were grouped together 86% of the time. Using parsimony analysis, the relationship between the three genera of *Bemisia*, *Trialeurodes* and *Aleurotrachelus* was not clear. The results of the distance analysis (Table 1) show that *Aleurotrachelus* was closer to the genus *Bemisia* than to *Trialeurodes*. An unexpected result was the relatively large mean distance between *T. vaporariorum* and *T. variabilis*.

Gene sequences and phylogenetic studies

The use of DNA sequences has become increasingly more important as a tool to study the evolution, populations and systematics of insects. Since the database for sequence information is expanding exponentially, it is certain that these methods will be even more important in the future. The several advantages to using DNA include the fact that the genotype, and not the phenotype, is examined directly. There is also an expanding base of phylogenetic information on an increasing number of gene sequences that exhibit different rates of change.

This study generated additional evidence that the *B. tabaci* biotype B is highly conserved throughout the Americas. This was expected because it is a recent introduction. The populations of *B. tabaci* biotype A are more conserved than we would have predicted from the studies that used esterase isozyme pattern as the means to detect diversity. Because the expression of esterases can be induced by insecticide applications, their phenotypes are probably not reliable for determining biotypes. Comparing the results of the molecular and esterase analyses, many esterase isozyme patterns are associated with the *B. tabaci* biotype A based on DNA markers.

Even though, in the phylogenetic analysis, *T. vaporariorum* and *T. variabilis* were in the same clade, the absolute distance was fairly high. Since the distance is equally great between *T. vaporariorum*, *T. variabilis* and the whiteflies in the other genera, there is a question of how closely related the members of *Trialeurodes* may be. Further studies should determine if these two species are members of the same genera, or whether they should be placed into separate genera.

The application of RAPD for identification of whiteflies

Whenever an increase in whiteflies occurs or they begin to affect additional crops, the introduction of the *B. tabaci* B biotype is suspected. Often, this is the case but whitefly populations are affected by many environmental factors, including the cropping system and the varieties grown. The molecular methods that generate DNA sequence data are time and labour intensive and are not suited to large scale monitoring of populations. Rapid and reliable methods are needed to distinguish between the most common whiteflies. In Latin America, these are *B. tabaci* biotypes A and B, and *T. vaporariorum* on most crops.

Using RAPDs has several advantages. Foreknowledge about any particular gene in the target organism is not needed. More than one primer can be used to increase confidence in the reliability of the method. With whiteflies and many other types of samples, the ability to store them for many months at room temperature in 70% ethanol facilitates shipping them across international borders and allows the analysis of large numbers of samples to be processed in an orderly manner.

RAPD was used (De Barro and Driver, 1997) to distinguish indigenous Australian populations of *B. tabaci* from the introduced B biotype. The authors reported on four oligonucleotide primers that they considered the most useful for identifying the native *B. tabaci* from the B biotype.

Analysis of molecular markers to identify whiteflies in Latin America

Four oligonucleotide primers (De Barro and Driver, 1997) were tested for their utility in distinguishing *B. tabaci* biotype A and biotype B, and *T. vaporariorum*. In the analysis of PCR products using the primer H9 (Operon, USA), there were differences for the range of whitefly species tested (Figure 2). For *B. tabaci* biotype B and *T. vaporariorum*, there are prominent PCR products ca. 600 and 800 bp that can sometimes make distinguishing the two species difficult. The unique PCR product in *B. tabaci* biotype B ca. 950 bp and *T. vaporariorum*'s unique PCR product at ca. 500 bp are important for distinguishing between these two species. Although confusion can occur in the interpretation between some species, the H9 primer was most useful in distinguishing between *B. tabaci* biotypes A and B. At 600 bp, some PCR products are similar in size in both biotypes but the biotype A has several unique PCR products, including doublet bands at 300-350 bp. The

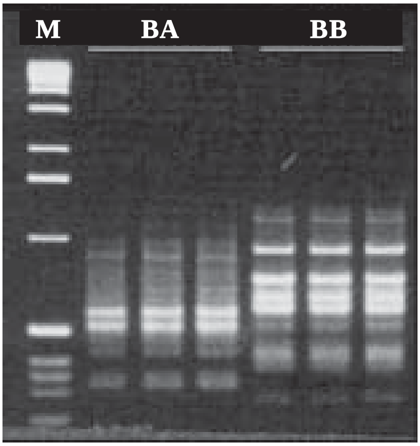

Figure 2. Using Operon primer H9, these are the RAPD-PCR DNA products from individual whiteflies. M:1kb markers (BRL), BA: *Bemisia tabaci* biotype A, BB: *Bemisia tabaci* biotype B.

biotype B has unique PCR products at ca. 600, 700 and 900 bp compared with one product of ca. 850 bp in the biotype A.

The primer H16 (Figure 2) was most useful in distinguishing between the whitefly species. While there can be some common bands in the 500 to 1000 bp range for both *B. tabaci* biotypes, three products in *B. tabaci* biotype B of ca. 350, 450 and 550 bp were consistently useful for identification of the B biotype.

When primers F2 and F12 were used, there were larger numbers of PCR products. These can be used for distinguishing the whitefly species but, because of the large numbers of bands, were generally less useful than H9 and H16. When using RAPDs for the identification of the whiteflies in this study, analysing the individual whiteflies with both the H9 and H16 primers is recommended.

Mapping the Distribution of Whitefly Species and Biotypes in Latin America

This survey was undertaken to map the distribution and range of whiteflies in various countries in Latin America. For this purpose, RAPDs were used to characterize the principal whitefly pests in many countries of Central America and the Caribbean region.

The survey using molecular markers complemented the activities where whitefly specimens were analysed by light microscopy. For the molecular analysis, at least one and often two primers were used and the results were compared to the morphological identification. The use of RAPD data was the only method to distinguish between the biotypes A and B in the *Bemisia* complex.

This survey was extensive but not exhaustive, therefore results should be interpreted as a representation of the predominate whiteflies populations in various regions of Cuba, the Dominican Republic, Guatemala, Honduras, Costa Rica, Panama, Colombia and Venezuela.

Dominican Republic

In the departments of Azua, Barahona, Peravia, Santiago, Montecristo and San Juan, the predominant whitefly was *B. tabaci* biotype B. Plants tested include eggplant (*Solanum melongena* L.), tomato (*Lycopersicon esculentum* Mill.), okra (*Abelmoschus esculentus* [L.] Moench), melon (*Cucumis melo* L.), cucumber (*Cucumis sativus* L. var. *sativus*) and hibiscus (*Hibiscus* spp. L). Only one sample of tomato in Azua was classified a biotype A. In the lowland tropics of the Dominican Republic, the biotype B was introduced nearly a decade ago and has nearly excluded the indigenous biotype A. In the department of Vega, common bean (*Phaseolus vulgaris* L.), tomato and potato (*Solanum tuberosum* L.) were tested and *T. vaporariorum* was the only whitefly found.

Guatemala

In the department of Zacapa, the predominant whitefly was *B. tabaci*. In Zapcapa, the B biotype was present on all the hosts tested, which included okra, melon, watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai) and cucumber. In Japala, both *B. tabaci* biotype B and *T. vaporariorum* were found. The B biotype was found on okra, melon, watermelon, cucumber, tomato, eggplant, tobacco (*Nicotiana tabacum* L.) and weed species. *T. vaporariorum* was the predominate whitefly in several host plants including paw paw (*Carica papaya* L.), "*tomate-manzana*" (*Lycopersicon esculentum* var. beef tomato), cherry tomato (*Lycopersicon esculentum* var.

cerasiforme [Dunal] A. Gray), common bean and some samples of tomato.

Honduras

In Honduras, the predominant whitefly was *B. tabaci* biotype A. Most of the samples were common bean, tomato and cucumber from the Comayagua Department. The paw paw in Comayagua was host to *T. vaporariorum*. On chilli peppers (*Capsicum* spp. L.) in the Francisco Morazan Department, the A biotype was also the only whitefly identified.

Costa Rica

In the lowland tropics of Costa Rica, *B. tabaci* biotype A is still the predominant whitefly. In the departments of Guanacaste, San José, Heredia and Alajuela, most of the samples were biotype A. The A biotype was found on common bean, melon, watermelon, tomato, chilli peppers, cucumber and others. In the departments of Arajuel and Puntarenas, the B biotype was present. It was the only whitefly found in the two samples from Arajuel but in Puntarenas both biotype A and B were present. *T. vaporariorum* was present in the Alajuela Department and was common in Cartago where it was found on chilli peppers and tomato.

El Salvador

Only a limited number of samples (15) have been tested and all were *B. tabaci* biotype B.

Panama

The principal whitefly in the Department of Chiriqui was *T. vaporariorum*. In the other regions of the country, *B. tabaci* biotype B was the predominant whitefly. Populations of *B. tabaci* biotype A are still found and in some areas the populations are mixed. This suggests a relatively recent introduction of *B. tabaci* biotype B in Panama.

Cuba

B. tabaci biotype B was the only whitefly found in the samples from Cuba. The B biotype has been in Cuba for nearly 10 years and clearly has become the predominant whitefly.

Colombia

Most of the samples tested were from the north coastal region of Colombia, where the principal whitefly is *B. tabaci* biotype B. The B biotype is affecting tomato, col (*Brassica oleracea* L. var. *capitata* L.), eggplant, melon and cassava (*Manihot esculenta* Crantz). The A biotype is still present but the introduction of the B biotype is relatively recent.

Venezuela

Samples were collected from the departments of Zamora and Jiménez, where the principal whitefly was the B biotype. In the Urdaneta Department, there was a mixture of the A and B biotypes. Only a few samples were analysed and more are needed before conclusions should be made.

A Regional View of Whitefly Populations

Most of the samples that were determined to be in the *B. tabaci* complex by morphological methods were classified as either biotype A or B (Table 2). In general, about 15% of the samples could not be amplified; only 23% of the samples from Venezuela could be amplified. This may demonstrate the importance of the proper sample handling. About 5% of the samples also had RAPD PCR products that could not be identified. These may well be either different

Table 2. The species of whitefly[a] by country (%) as determined using random amplified polymorphic DNA (RAPD) analysis.

Country	Number of samples	*B. tabaci* biotype A	*B. tabaci* biotype B	*T. vaporariorum*	NI[b]	NB[c]
Guatemala	185	8.1	60.5	26.0	5.4	0
Cuba	44	0	100.0	0	0	0
Dominican Rep.	106	13.2	81.1	0	0	5.7
Colombia	173	0.6	63.6	14.4	5.8	15.6
Venezuela	262	4.6	18.3	0	0	77.1
Costa Rica	160	61.3	6.9	12.5	3.1	16.2
Panama	198	4.6	33.3	40.9	9.1	12.1
El Salvador	15	0	100.0	0	0	0
Honduras	138	88.4	2.9	0	7.3	1.4

a. *Bemisia tabaci, Trialeurodes vaporariorum*.
b. Samples with amplified PCR products but unable to identify.
c. No products were amplified.

species or unique biotypes. This level of uncertainty did not detract from the survey and those unidentified samples are a source of potentially different whitefly populations that merit further study.

B. tabaci biotype A, *B. tabaci* biotype B and *T. vaporariorum* were the principal whiteflies found in the region. Even at the department level, a predominant whitefly usually could be identified. Occasionally, within the same region, mixed populations were present but these were normally separated on different hosts. *T. vaporariorum* was present in many regions but tends to be at higher elevations. Only in the mid-altitudes do *B. tabaci* and *T. vaporariorum* populations overlap. In some countries, including Cuba and the Dominican Republic, the exclusion of the *B. tabaci* biotype A is almost complete, at least on the principal crop species tested for this survey. The pattern in Central America is more complex. In Cuba, Guatemala, Panama and the north cost of Colombia, the process of domination of the B biotype appears to be well advanced. In Colombia, biotype B is a recent introduction (Quintero et al., 1998) and already is the predominant whitefly in the region. This survey documents the expanding range of *B. tabaci* biotype B (Polston and Anderson, 1997) and adds details to the distribution within the countries in the study. In Honduras and Costa Rica, the A biotype is still the major whitefly present. It is probably just a matter of time until the B biotype becomes the principal whitefly in those areas. Nevertheless, these are the zones that need to be monitored closely and where strategies to decrease the probability of introduction of the B biotype should be developed.

Developing a specific marker to identify whiteflies in the Bemisia *complex*

RAPDs are produced by using short oligonucleotides, generally of 10 base pairs. This allows the generation of many amplified PCR products, using just one primer. The lack of specificity of the short oligonucleotides can lead to results that are difficult to interpret because of similar size PCR products. Also, shorter oligonucleotides are even more sensitive to single base changes. RAPD markers can be converted in a sequence characterized amplified region (SCAR). This specific marker utilizes a pair of oligonucleotides generally 20-25 base in length and amplifies a single region of a genome (Ohmori et al., 1996). This makes interpretation of the results much simpler than with RAPDs. Instead of a group of bands with a degree of variability, SCAR produces single bands in the target organism.

The SCAR for the *B. tabaci* biotype B was developed by cloning the RAPD PCR products of the primer H9. The cDNA clones were analysed and sequenced. The primers were designed and tested. One set of primers only amplified *B. tabaci* biotype B (Figure 3). This set of primers did not amplify any tested population of *B. tabaci* biotype A or *T. vaporariorum*. Further testing is needed to confirm that these primers amplify this PCR product in geographically separated populations of *B. tabaci* biotype B. Given the lack of diversity in the evolutionary studies, this SCAR is expected to be useful for rapidly identifying all populations of biotype B.

The current research is directed to developing SCARs for *B. tabaci* biotype A and *T. vaporariorum*. Each set of primers will amplify bands of different sizes. This will allow the primers to be mixed together and will distinguish between the whiteflies in a single PCR reaction. Thus more extensive surveys possible will be possible and the technique will be more consistent in all the laboratories that need the diagnostic capability to rapidly identify whitefly pests.

Conclusions

Our detailed molecular analysis is helping define both the range and the variability of *B. tabaci* biotypes A and B in Latin America. Similar studies in other regions of the world have been made and will allow a global view of the *Bemisia* complex. The molecular method of RAPDs proved essential in distinguishing between the *B. tabaci* biotype A and B but, because of the nature of the method, comparing results between laboratories is difficult. Therefore we are developing SCARs to simplify the identification of whiteflies.

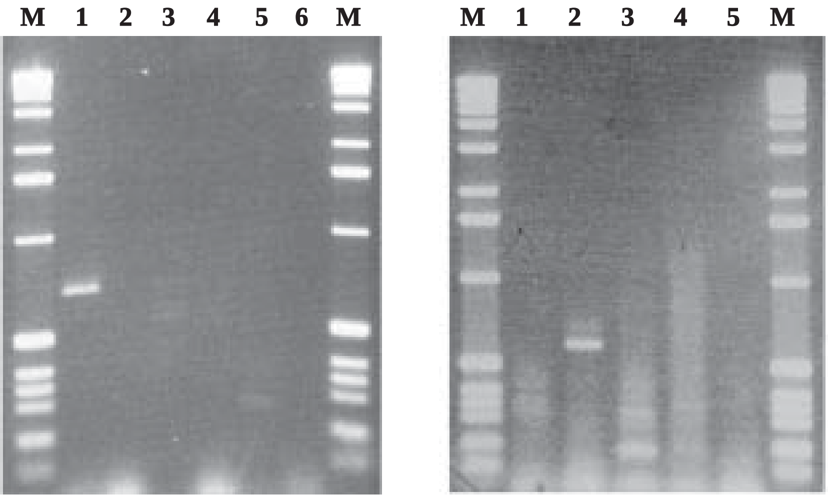

Figure 3. Two SCARs were developed to distinguish *Bemisia tabaci* biotype A (1) and *Bemisia tabaci* biotype B (2). Lane 1 *Bemisia tabaci* biotype A, lane 2 *Bemisia tabaci* biotype B, lane 3 *Bemisia tuberculata*, lane 4 *Trialeurodes vaporariorum*, lane 5 *Aleurotrachelus socialis*, 6:DNA free reaction and M:1kb markers (BRL).

Already we can identify *B. tabaci* biotype B using a SCAR, and we are making progress in the development of SCARs for *B. tabaci* biotype A and *T. vaporarorium*. Specific unambiguous results of one amplified band for each species or biotype of whitefly will simplify the interpretation of results and become a rapid and relatively inexpensive method to use in surveys of whiteflies.

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CHAPTER 3.14

Conclusions and Recommendations

*Francisco Morales**

Despite over 40 years of research on *Bemisia tabaci* (Gennadius) (Homoptera:Aleyrodidae) as a pest and vector of plant viruses, this whitefly species continues to cause considerable damage to food and industrial crops in Mexico, Central America and the Caribbean. *B. tabaci* first became a pest of cotton (*Gossypium hirsutum* L.) in Middle America, and then a major vector of plant viruses in common bean (*Phaseolus vulgaris* L.) and tomato (*Lycopersicon esculentum* Mill.) plantings throughout the region. The expansion of non-traditional crops in the 1980s, mainly vegetables for export, further aggravated the problem by providing more reproductive hosts for the whitefly pest and a higher demand for pesticides. The introduction in the early 1990s of a more aggressive and polyphagous biotype (B) of *B. tabaci* in the Americas further jeopardized past efforts to control this pest.

In this region, whitefly-borne geminiviruses (begomoviruses) are more prevalent during the dry months of the year (November through April), when *B. tabaci* populations encounter more favourable weather. The area planted to crops such as melon (*Cucumis melo* L.), common bean, tomato, eggplant (*Solanum melongena* L.), cucurbits and sweet and hot peppers (*Capsicum* spp. L.) has been reduced greatly or completely abandoned during this period of the year because of the frequent outbreaks of whiteflies and whitefly-borne viruses. The development of *B. tabaci* from a harmless insect to a major pest is closely related to the introduction and intensive use of pesticides after the Second World War. Pesticide abuse remains a major factor in *B. tabaci* and begomovirus epidemics in this region. Whiteflies are known to develop resistance to insecticides, the frequent use of which eliminates natural predators of *B. tabaci*. It is not uncommon to see farmers spraying insecticides on susceptible crops every other day and even daily, up until harvest time, in open defiance of food safety regulations. The high levels of pesticide residues in tomato, pepper, common bean, eggplant and other vegetables attacked by *B. tabaci* disqualifies these products for sale in international markets and constitutes a serious health hazard for the millions of rural and urban consumers in the region, where produce is not tested for toxic residues.

But the main reason for the persistence of these pest problems is the lack of qualified technical assistance for farmers, particularly

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those who have attempted to diversify their subsistence crops with more profitable cash crops such as tomato and pepper. This situation is a reflection of the increasingly deteriorating economic situation of the region since the mid 1970s, which has negatively impacted on the capacity of national agricultural research institutions and international agricultural research centres to conduct research and provide the necessary technical assistance to farmers affected by severe phytosanitary problems such as whiteflies and begomoviruses. Moreover, the diversion of foreign aid funds from crop improvement to natural resource management has further contributed to increased yield losses due to pest problems, abandonment of profitable crops grown by small-scale farmers, higher production costs and considerably more environmental and food contamination.

Whitefly and begomovirus control strategies in the region differ according to countries, agricultural areas and crops. In north-western Mexico, the use of virus-resistant common bean varieties and more efficient insecticides for vegetables has greatly reduced the impact of these pests. In the Yucatán Peninsula, Mexico, small-scale farmers were using physical control methods during the first 3 weeks after transplant of tomato and pepper. In the Dominican Republic, a crop-free period was legally imposed throughout the country to break the cycle of the whitefly before the main planting season. And in most Central American countries, susceptible crops were only planted during the rainy season, when whitefly populations are at their lowest level. These whitefly/virus control strategies have reduced, but not eliminated, the production losses associated with outbreaks of *B. tabaci* due to various factors. The constant flow of improved cultivars developed in the past through conventional plant breeding methods has been greatly reduced because of the radical changes that have occurred in the past decade in favour of molecular breeding methods. Second, technical assistance to small-scale farmers is practically nil, which leaves plant protection in the hands of pesticide salespeople. Susceptible crop-free periods reduce whitefly incidence but do not increase the productivity of prime agricultural land that must be devoted to less profitable crops or remain idle during several months of the year. The new insecticides in the market are effective for whitefly control but are very expensive for resource-poor farmers.

The Tropical Whitefly Integrated Pest Management (TWF-IPM) Project has successfully identified, tested and validated the most efficient and sustainable whitefly control methods used in over 10 countries in Middle America. So far, the most valuable IPM measure identified is genetic resistance to control whitefly-transmitted viruses, particularly in the case of common bean. Unfortunately, practically no crop improvement work is being done in the case of vegetables such as tomato and pepper despite their tropical American origin. Most vegetable varieties are bred in temperate countries and thus are not adapted to the tropical conditions and pests that exist in the region. The TWF-IPM Project has successfully used physical barriers (micro-tunnels) to protect sensitive crops during the first critical month of their life cycle. This strategy also protects susceptible plants against other insect-borne viruses and pests, and greatly increases their productivity per unit area with minimum inputs. The identification and use of bio-control agents of *B. tabaci* has been advanced greatly in the region, particularly in countries such as Cuba and the Dominican Republic. Finally, an enhanced understanding of the whitefly species, biotypes and plant viruses present in the region, has helped understand the ecology and epidemiology of these crop production problems and, more important, has contributed to the implementation of sustainable and efficient IPM methods.

It is imperative to re-orient agricultural research in the region by understanding that spending our limited resources on purely environmental or sociological issues, without a food production component, only perpetuates misery and increases environmental degradation and human health problems. There is considerable need to strengthen national and international research and extension services to provide timely and effective technical assistance to small-scale farmers. We must help resource-poor farmers to diversify their cropping systems with more profitable crops in order to maximize the income derived from their limited land resources. Traditional crops are the basis of food security but they are merely subsistence crops. High-value crops, particularly vegetable and fruit crops, can help small-scale farmers improve their livelihoods but this can only be achieved with proper and timely technical assistance provided by multi-disciplinary groups of agricultural specialists. Finally, there is a vacuum in the area of agricultural economics and marketing for small-scale farmers. Unless these farmers are incorporated into the market chains, they will be at the mercy of the vagaries of the open markets and intermediaries who profit from the misery and lack of market knowledge of the resource-poor farmer. The TWF-IPM Project expects to address these issues and scale up the dissemination of the IPM measures validated to date in Central America, Mexico and the Caribbean region.

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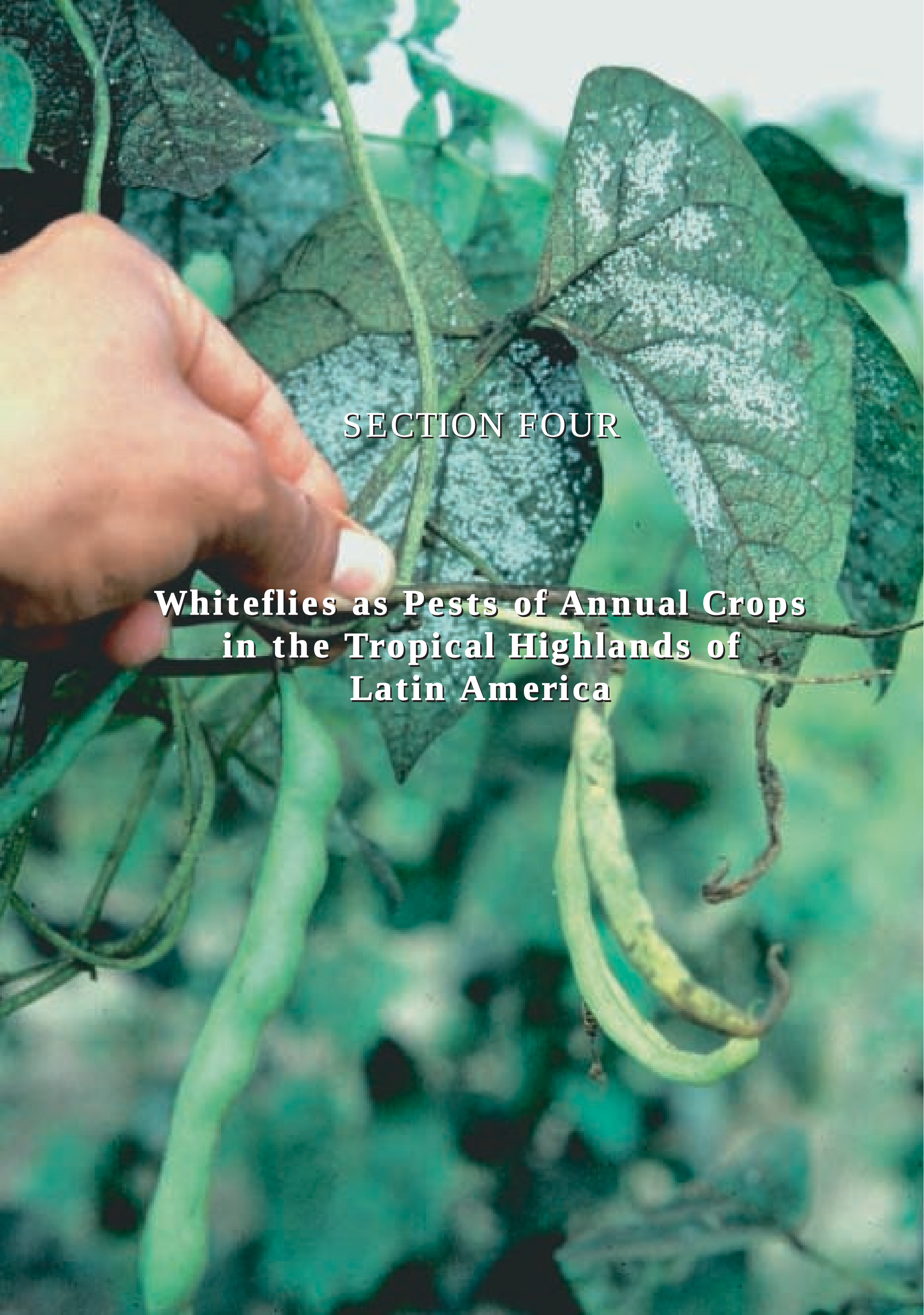
SECTION FOUR

Whiteflies as Pests of Annual Crops in the Tropical Highlands of Latin America



BLANCA 268

CHAPTER 4.1

Introduction

*César Cardona**

According to Strong et al. (1984), 724 species of whiteflies (Homoptera: Aleyrodidae) have been described from the tropics but only 420 from temperate regions. Most of these insects do not represent an economic threat to agriculture and only a handful can be described as serious pests. In a recent survey conducted in Central America and Colombia, Caballero (1992) listed the 30 commonest and economically most important whitefly species recorded from 84 host plant species. Among these, the sweetpotato whitefly *Bemisia tabaci* (Gennadius), the greenhouse whitefly *Trialeurodes vaporariorum* (Westwood) and the citrus whitefly *Aleurocanthus woglumi* Ashby were regarded as key pests. Of lesser importance were *Trialeurodes abutiloneus* (Haldeman) and three species affecting cassava: *Aleurotrachelus socialis* Bondar, *Bemisia tuberculata* Bondar and *Trialeurodes variabilis* (Quaintance).

T. vaporariorum and *B. tabaci* are by far the most important whitefly species affecting annual crops in the Latin American highlands and these two species are the focus of the present review. While reference is made to *B. tabaci* and *T. vaporariorum* as virus vectors, emphasis is on the importance of these insects as direct pests causing mechanical damage. The whiteflies affecting cassava (*Manihot esculenta* Crantz), a semi-perennial crop, are not discussed.

Legumes and Horticultural Crops in the Andean Highlands

The Andes encompass a wide range of latitudes and climates but in the tropics especially, altitude is the main determinant of temperature and biological character. Areas between 1000 and 2000 m above sea level are known as highlands. Those above 2000 m have a quasi-temperate climate and are collectively known as the "high Andes". Transitions from tropical lowlands to temperate highlands may be abrupt and the alpine zones form ecological islands, large or small. As indicated by Winograd et al. (1998), the Andean backbone and its lateral ranges contribute remarkable diversity to the ecosystems of Colombia and Ecuador. In parts of Colombia, small ridges and spurs connect the three main ranges, circumscribing a series of intermountain basins, each of which hosts a distinctive local community of natural biodiversity. Agricultural systems and their ecology are correspondingly diverse.

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The tropical highlands of Colombia and Ecuador include extensive agricultural areas with mean temperatures of 12-20 °C, variable rainfall patterns (700-1600 mm) and relative humidity ranging from 50% to 85%. Small-scale farmers grow common bean and snap bean (*Phaseolus vulgaris* L.), tomato (*Lycopersicon esculentum* Mill.), potato (*Solanum tuberosum* L.) and other horticultural crops, both for home consumption and for sale. Crops are usually grown on steep, erosion-prone slopes, with no irrigation and little or no use of fertilizers.

In contrast, the lowlands of Colombia and Ecuador experience mean temperatures of 26-28 °C, similarly variable rainfall patterns (600-1600 mm) and prolonged dry seasons that favour the buildup of whitefly populations. Cotton (*Gossypium hirsutum* L.), soybean (*Glycine max* [L.] Merr), melon (*Cucumis melo* L.) and, to a lesser extent, tomato are grown on large farms that occupy a significant proportion of the agricultural land. Many small-scale farmers also grow less than 1 hectare of legumes, vegetables or tobacco (*Nicotiana tabacum* L.).

Status of Research on *Trialeurodes* and *Bemisia*

The centre of origin for *T. vaporariorum* has not been identified. According to Vet et al. (1980), the species is most probably indigenous to tropical and subtropical America. *T. vaporariorum*, a well-known pest of greenhouse crops in Europe, has long been recorded on annual field crops in highland areas of Colombia (Posada, 1976) and Ecuador (Merino and Vásquez, 1962). It is confirmed as a competent vector of numerous plant viruses (Díaz et al., 1990; Duffus, 1987; 1996; Wisler et al., 1998; Salazar et al., 2000) but it is more important as a direct pest of several crops. This whitefly was viewed as a minor pest until the early 1980s (Cardona, 1995). Since then, *T. vaporariorum* has become a key pest on dry beans, snap bean, tomato, potato and ornamentals and is considered the most important whitefly species in the tropical highlands of South America. Damage is due to the continuous sucking of sap from the phloem by nymphs and adults and to the abundant excretion of honeydew that falls on leaves and fruits and serves as a substrate for fungi that block photosynthesis. Both the yield and quality of crops are affected. A series of trials in Colombia (Cardona et al., 1993; Prada et al., 1993) showed that high infestation levels (up to 450 nymphs/leaf) can reduce the yields of common bean by 38% and of snap bean by 54%.

Whitefly problems in snap bean production in Colombia have increased dramatically since the early 1980s and illustrate the need for integrated pest management (IPM) work on whiteflies. In an attempt to reduce pesticide use in whitefly-affected areas, a pilot project, funded by the International Development Research Centre (IDRC) of Canada, was initiated in the Sumapaz region of central Colombia. Surveys showed that all of 286 farmers interviewed were using insecticides as the sole method to manage whiteflies. Insecticide use averaged 11 applications in a 70-day cropping cycle. Most of the insecticides used were organophosphates and carbamates belonging to toxicological category I (highly toxic) as defined by Metcalf (1994): those insecticides with acute oral LD_{50} values of less than 50 mg/kg or dermal LD_{50} values of less than 200 mg/kg. Field tests indicated that seven of the 10 most widely used insecticides were ineffective for whitefly control, due to resistance (Cardona, 1995). Research showed that simple sanitation measures such as leaf roguing and destruction of crop residues reduced overall whitefly infestation levels if implemented on a community-wide basis. In addition, selective use of granular insecticides delayed the need for the first foliar insecticide application, enhancing natural enemy activity. A relatively simple IPM package based on cultural control and sanitation practices, timely application of effective insecticides and reliance on natural biological control resulted in a 66% reduction in insecticide use (Cardona, 1995). Farmers' participation in the overall process was essential for widespread adoption of the system proposed. However, it was concluded that, before continuing with IPM training and scaling up, it was necessary to verify that patterns of *T. vaporariorum* distribution, reproductive hosts plants, perceptions of the problem and insecticide use were similar throughout the Andean region.

B. tabaci also has been documented as a pest of several crops in Colombia (Posada, 1976; Bolaño, 1997) and Ecuador (Merino and Vásquez, 1962). Except for sporadic outbreaks on cotton in Colombia (Alcaraz et al., 1990), this insect was generally regarded as a secondary pest until 1993 when serious outbreaks occurred in the western provinces of Manabí, Guayas, and Los Ríos in Ecuador. Mendoza et al. (1995) estimated that vegetable growers in the Guayas region lost US$400,000 per year as a result of whitefly attacks. Up to 10,000 ha of soybean were destroyed in the Guayas and Los Ríos provinces (Mendoza, 1996). Similar outbreaks occurred in 1996 in the north-western Departments of Sucre, Córdoba and Atlántico in Colombia (Quintero et al., 2001). The change in *B. tabaci*'s pest status was attributed to the introduction into the region of the B biotype (Quijije et al., 1995), which was first detected in Colombia in 1997 (Quintero et al., 1998).

A New Foundation for Whitefly IPM in the Andes

In view of the increasing importance of whiteflies in the Andean highlands, the launching in 1997 of a new project for "sustainable integrated management of whiteflies as pests and vectors of plant viruses in the tropics" by the Systemwide Programme on Integrated Pest Management was especially timely. A sub-project was established, under the co-ordination of the Centro Internacional de Agricultura Tropical (CIAT), to look specifically at "Whiteflies as pests in the tropical highlands of Latin America".

The purpose of the diagnostic phase of this sub-project (and of others addressing different whitefly problems around the world) was to provide a sound basis for future IPM efforts by gathering and analysing baseline data and so being able to characterize properly the nature of major whitefly problems. Specifically, in the Andean highlands, there was a need to describe the patterns of distribution of *T. vaporariorum* and *B. tabaci*, especially in relation to altitude, as well as the distribution of the newly introduced B biotype of *B. tabaci*. Information on the range of reproductive host plants and identification of natural enemies was also desired. Additional socio-economic information on crop losses, farmers' perception of the problems and patterns of insecticide use, as well as biological assessment of levels of insecticide resistance, were needed as a foundation for moving forward to more widespread adoption of IPM.

Surveys were conducted between October 1997 and December 1998 in whitefly-affected areas of Colombia and Ecuador while insecticide resistance testing continued through May 1999. The research was carried out in close collaboration among staff of CIAT and partner organizations, the Instituto Nacional Autónomo de Investigaciones Agropecuarias (INIAP) in Ecuador and the Corporación Colombiana de Investigación Agropecuaria (CORPOICA) in Colombia. The methods used and the results of the work are reported in the following two chapters.

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CHAPTER 4.2

Colombia and Ecuador

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As part of the diagnostic phase of the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project (see Introduction and Chapter 4.1, this volume), partners in the sub-project concerned with “Whiteflies as pests in the tropical highlands of Latin America” conducted extensive field surveys in Colombia and Ecuador from October 1997 to December 1998. The sub-project was co-ordinated by the Centro Internacional de Agricultura Tropical (CIAT) and carried out in collaboration with the Ecuadorian national research organization: the Instituto Nacional Autónomo de Investigaciónes Agropecuarias (INIAP), and the Colombian national research organization: the Corporación Colombiana de Investigación Agropecuaria (CORPOICA). The present chapter summarizes the diagnosis and characterization of problems caused by the greenhouse whitefly *Trialeurodes vaporariorum* (Westwood) as a pest of annual field crops in Colombia and Ecuador, and its relationship to the sweetpotato whitefly *Bemisia tabaci* (Gennadius).

The surveys focused on the tropical highlands (1000-3000 m above sea level) along the Andean corridor of Colombia and Ecuador (0° 30'-11°N, 73°-78° 30'W). Within this zone, surveys covered areas representing about 97% of whitefly-affected areas (Figure 1). Representative sampling also was carried out in the lowlands and mid-altitudes in order to construct an altitudinal gradient from sea level up to 3000 m. Samples were collected in mid-altitude valleys within the highland tropics of Colombia (400-1000 m above sea level). And, in the tropical lowlands (less than 400 m above sea level), surveys were conducted along the northern (Atlantic) coast of Colombia (7° 30'-11°N, 72°-77°W) and the Pacific coast of Ecuador (0°-1° 30'S, 79°-80° 30'W), including the Galápagos Islands of Ecuador (2°-3°S, 90°-90° 30'W) in the Pacific Ocean. INIAP staff conducted field surveys and interviews with farmers along the coast of Ecuador. CIAT personnel sampled the northern highlands of Ecuador. CORPOICA and CIAT staff conducted fieldwork in Colombia.

Surveys followed the standardized methodology agreed among the project partners. Twenty survey areas were defined in Colombia and Ecuador based on the importance of annual crops and previous knowledge of

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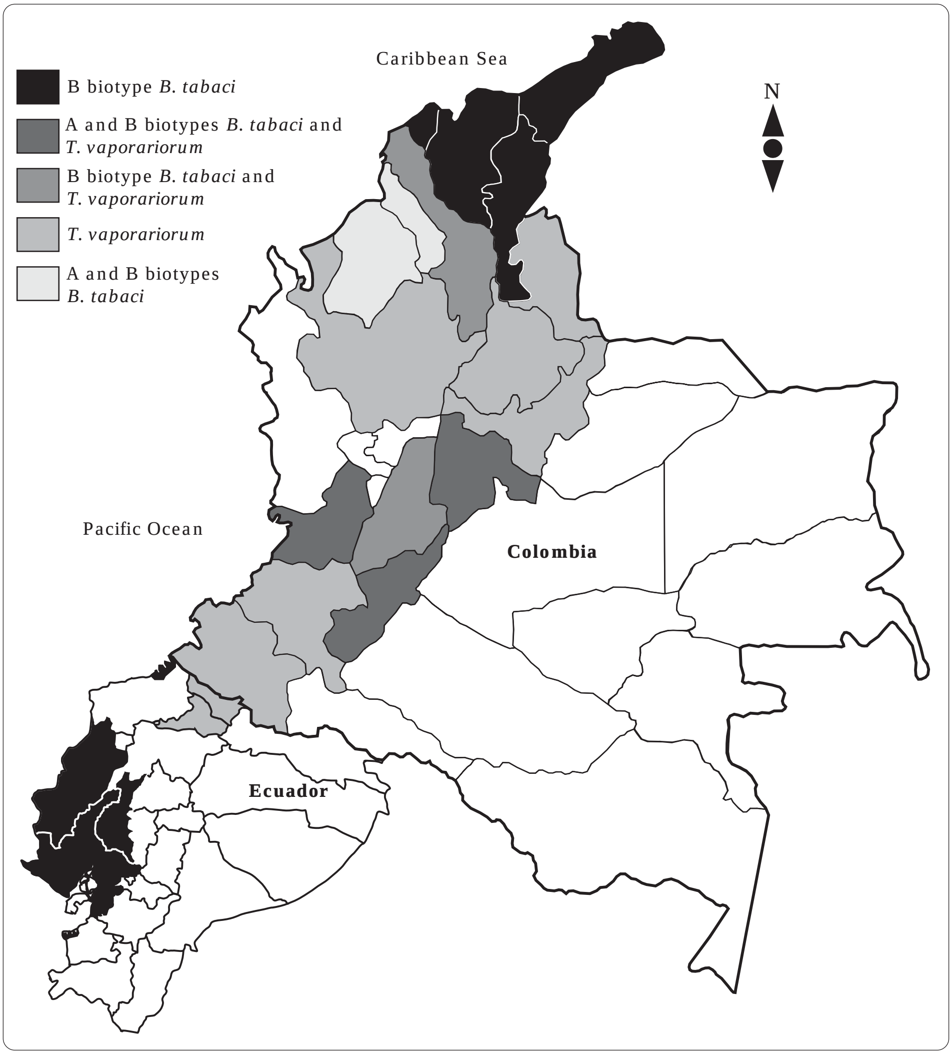

Figure 1. Survey area and distribution of whitefly species (*Bemisia tabaci* and *Trialeurodes vaporariorum*) in Colombia and Ecuador. Uncoloured areas were not sampled.

whitefly incidence. Nine areas were chosen to represent the lowlands along the Pacific coast of Ecuador and the Atlantic coast of Colombia where cotton (*Gossypium hirsutum* L.), soybean (*Glycine max* [L.] Merr.) and tomato (*Lycopersicon esculentum* Mill.) are grown on large farms that occupy a significant proportion of the agricultural land. Eleven areas were chosen to represent the highlands of northern Ecuador and the main agricultural areas along the three mountain ranges in Colombia. Highland areas in both countries are occupied by small-scale farmers who grow common and snap bean (*Phaseolus vulgaris* L.), tomato, potato (*Solanum tuberosum* L.) and other horticultural crops for consumption and sale. Between 10 and 20 data sets were collected and processed in each survey area. A complete data set consisted of a producer questionnaire and biological samples of whiteflies and natural enemies from one of that producer's fields. Three hundred and twenty-five data sets were processed, based on visits and interviews with 274 farmers and 168 extension agents, private technical assistants, insecticide salesmen and government officers in charge of rural development projects. Insecticide resistance was also assessed in whiteflies from 40 sites, as detailed in Chapter 4.3 (this volume).

Whiteflies and natural enemies were identified at CIAT and whitefly biotypes determined using esterase banding patterns in polyacrylamide gel electrophoresis (PAGE) and random amplified polymorphic DNA/ polymerase chain reaction (RAPD-PCR) analysis (Quintero et al., 1998). Biological samples for whitefly species and biotype identification were taken at 215 sites; 74 samples for identification of natural enemies were collected.

Whitefly populations were sampled in 14 crops. In the tropical highlands, emphasis was placed on common bean (73% of samples taken), tomato (18%) and potato (7%). In mid-altitude valleys, most samples were taken on tomato (58%) and common bean (18%). Cucurbits (38%), tomato (31%), pepper (*Capsicum annuum* L.) (10%) and eggplant (*Solanum melongena* L.) (5%) were the crops most intensively surveyed in the tropical lowlands. Yield losses due to the greenhouse whitefly on snap beans were measured.

Increased Biological Understanding

Species

An analysis of whitefly species composition along an altitudinal gradient suggests that *T. vaporariorum* and *B. tabaci* occupy well-defined niches and habitats (Figure 2). In the tropical lowlands, 91.5% of samples collected were identified as *B. tabaci*, whereas 100% of the whiteflies collected in the tropical highlands were *T. vaporariorum*. The species co-exist in all mid-altitude valleys where samples were taken, with *T. vaporariorum* predominating. Thus, in Colombia and Ecuador, *T. vaporariorum* occurs between 780 and 2830 m above sea level and is the only species present above 1600 m, whereas *B. tabaci* is the only species present at lower elevations (500 m or less) (Quintero et al., 2001).

This distribution of the two major whitefly species may be related to climatic conditions, especially temperature regimes, which are determined in the tropics principally by elevation. As much of the agricultural production in the Andes is concentrated in mid-altitude valleys, our findings clearly show the need for a correct identification of the whitefly species concerned. If it is assumed, for example, that *B. tabaci* is the major pest in mid-altitude valleys and highlands, IPM packages will be ill targeted. Our findings also dispel the notion, prevalent among technical

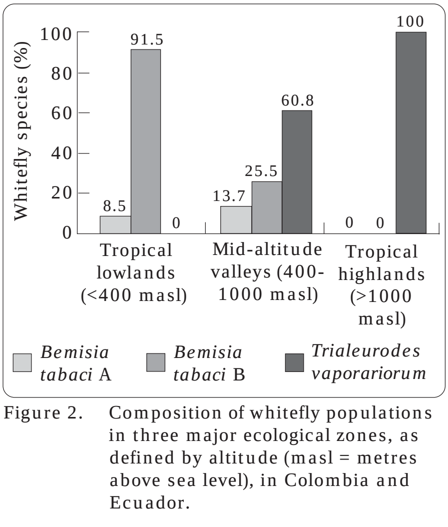

Figure 2. Composition of whitefly populations in three major ecological zones, as defined by altitude (masl = metres above sea level), in Colombia and Ecuador.

assistants and extension personnel, that *B. tabaci* is the key pest in the highlands and mid-altitude valleys of the Andean zone. *T. vaporariorum* is the key pest in this region.

Biotypes

No evidence for the existence of different biotypes of *T. vaporariorum* was found in more than 400 samples examined by RAPD-PCR. Thus it is concluded that there is no basis for continued biotype analysis of the greenhouse whitefly within the context of the TWF-IPM Project.

Both A and B biotypes of *B. tabaci* were detected among the 214 samples evaluated by RAPD-PCR (Figure 3). The B biotype is the only one of importance along the coast of Ecuador. It is present also throughout the northern (Atlantic) coast of Colombia and in the Departments of Valle, Huila, Tolima and Cundinamarca. The A biotype of *B. tabaci* was detected only in four departments in Colombia: Córdoba, Sucre, Valle and Huila (Figure 2). The prevalence of the B biotype in the coastal areas of Ecuador and Colombia represents a serious threat to agriculture in the region. Recent work (CIAT, unpublished results) has shown that the presence of the B biotype can also be a major threat to agriculture in mid-altitude valleys such as the Cauca Valley in Colombia. In a survey conducted in 2003, the B biotype was present in 63% of samples taken (as opposed to 11.5% in 1997). The increased incidence of the B biotype was also evidenced by the common occurrence of such physiological disorders as irregular ripening of tomato, and silver leaf symptoms on squash (*Cucurbita moschata* Duchesne). This underlines the need for continuous monitoring of whitefly species composition.

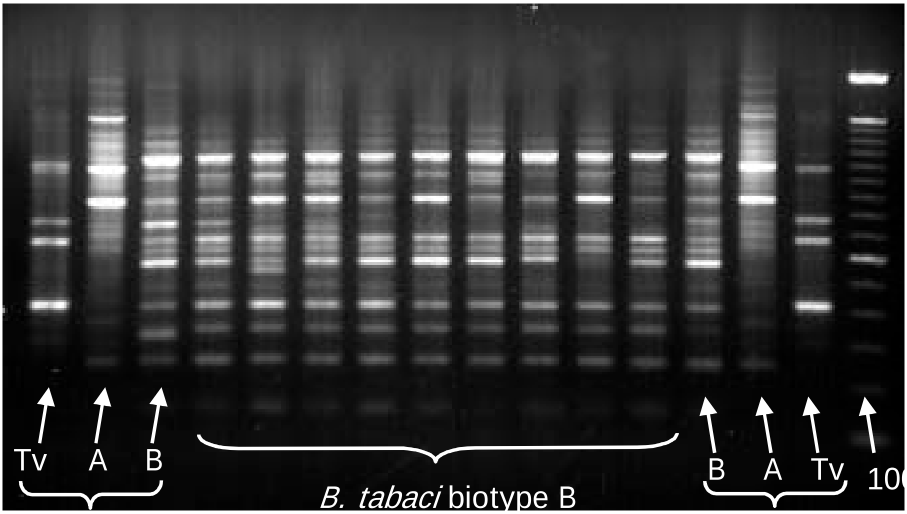

Figure 3. RAPD patterns obtained with primer OPA-04 for a sample of whiteflies collected in the northern coastal zone of Colombia. Lane 1: *Trialeurodes vaporariorum* (control); lane 2: *Bemisia tabaci* biotype A (control); lane 3: *B. tabaci* biotype B (control); lanes 4-12: *B. tabaci* biotype B (field sample); lane 13: *B. tabaci* biotype B (control); lane 14: *B. tabaci* biotype A (control); lane 15: *T. vaporariorum* (control); lane M: marker (1 kb ladder).

Reproductive hosts

In Colombia, *T. vaporariorum* was found breeding on common bean and snap bean, tomato, potato, squash, cucumber (*Cucumis sativus* L. var. *sativus*), eggplant, pepper, pea (*Pisum sativum* L.), tobacco (*Nicotiana tabacum* L.), cabbage (*Brassica oleracea* L.), coriander (*Coriandrum sativum* L.) and several species of ornamentals. In mid-altitude areas, *T. vaporariorum* also reproduces on cotton and soybean. All *T. vaporariorum* samples in Ecuador were taken on common bean and tomato. Snap bean and common bean are by far the most important reproductive hosts for *T. vaporariorum* in Colombia and Ecuador, followed in order of decreasing importance by tomato and potato (Table 1).

In Colombia, the A biotype of *B. tabaci* was recorded breeding on soybean, tobacco, tomato, cotton, poinsettia (*Euphorbia pulcherrima* Willd. ex Klotzch), broccoli (*Brassica oleracea* L. var. *italica* Plenck) and eggplant. In both Colombia and Ecuador, the B biotype was found reproducing on cotton, tomato, eggplant, squash, pepper, cucumber, common bean, snap bean, soybean, tobacco, cabbage, lettuce (*Lactuca sativa* L. var. *capitata* L.), sweetpotato (*Ipomoea batatas* [L.] Lam.), melon (*Cucumis melo* L.), watermelon (*Citrullus lanatus* [Thunb.] Matsum. & Nakai), pumpkin (*Cucurbita ficifolia* Bouché), lima bean (*Phaseolus lunatus* L. var. *lunatus*), forage Arachis (*Arachis pintoi* Krapov. & W. Gregory), grape (*Vitis vinifera* L.), poinsettia and additional *Brassica* species. Table 1 shows the main crops affected by *B. tabaci*.

In greenhouse studies on the comparative biology of biotypes A and B on 20 cultivated host plants, the B biotype bred faster than the A biotype on cauliflower (*Brassica oleracea* L. var. *botrytis* L.), cabbage, squash and tomato. Percentage emergence of the B biotype was significantly higher on 13 out of the 20 host plants studied; the A biotype did not breed on squash or beet (*Beta vulgaris* L. subsp. *vulgaris*). The B biotype did more damage to lettuce, squash, cabbage, cauliflower, cucumber, *Brassica* sp., soybean and watermelon than did the A biotype. Snap bean was affected also and suffered significant reductions in biomass as a result of damage by the B biotype. Common bean, on the other hand, was not as severely affected as other host plants.

The B biotype successfully colonized plants of four *Cucurbita* species tested. The A biotype did not survive to the adult stage on pumpkin

Table 1. Main crops serving as reproductive hosts of whiteflies in major agro-ecological zones of Colombia and Ecuador.

Whitefly species	Tropical lowlands	Mid-altitude valleys	Tropical highlands
A biotype of *Bemisia tabaci*	Eggplant	Tobacco	-
	Tomato	Soybean	-
B biotype *of B. tabaci*	Squash	Squash	-
	Cotton	Cotton	-
	Eggplant	Tobacco	-
	Brassica sp.	-	-
	Tomato	-	-
Trialeurodes vaporariorum	-	Common bean	Common bean
	-	Snap bean	Snap bean
	-	Tomato	Tomato
	-	-	Potato

and squash. On *Cucurbita mixta* Pangalo (= *Cucurbita argyrosperma* C. Huber subsp. *argyrosperma*) and *Cucurbita pepo* L., the B biotype reached the adult stage in ca. 20 days, 33% faster than the A biotype. The ability of the B biotype to successfully colonize hosts that are not attacked by the A biotype and its ability to reproduce faster on certain host plants are important diagnostic tools (Brown et al., 1995; Schuster et al., 1996). When *C. moschata*, *C. ficifolia* and *C. pepo* plants were infested with either A or B biotype adults, full expression of silverleaf symptoms, as described by Yokomi et al. (1990) and Costa and Brown (1991), were obtained 21 days after infestation with the B biotype. No silvering was observed on any plant infested with the A biotype.

Apart from silverleaf, other symptoms of B biotype attack that have been described in the literature were observed in numerous different fields: uneven ripening of tomato (Schuster et al., 1996; Polston and Anderson, 1997), chlorotic pods and petioles on snap beans (Hassan and Sayed, 1999), white stem streaking of *Brassica* spp. and phytotoxic disorders on poinsettia and lettuce (Brown et al., 1995).

Natural enemies

Survey work confirmed that *Amitus fuscipennis* MacGown and Nebeker is the most common parasitoid of *T. vaporariorum* in the highlands of Colombia and Ecuador. Other natural enemies of *T. vaporariorum* recorded were the parasitoids *Encarsia hispida* DeSantis, *Encarsia pergandiella* Howard, *Encarsia nigricephala* Dozier and *Encarsia* sp., and the predators *Delphastus pusillus* Leconte, *Hippodamia convergens* (Guérin-Ménenville), *Cycloneda sanguinea* (L.), *Coleomegilla maculata* (DeGeer) and *Chrysoperla* sp. The fungi *Verticillium lecanii* (Zimm.) and *Paecilomyces fumosoroseus* (Wize) were found in the Departments of Antioquia, Cauca Valley and Cundinamarca in Colombia (López-Avila et al., 2001).

Additional studies by Manzano et al. (2000) demonstrated that *A. fuscipennis* is abundant on common bean crops where no chemical treatments have been made, and remains active even after heavy pesticide use. Some studies have been conducted on the biology or efficacy of this species as a natural enemy of *T. vaporariorum* in the Andean zone (Márquez and Valencia, 1991; Medina et al., 1994; Manzano et al., 1999). Recent results (Manzano et al., 2000) suggest that *A. fuscipennis* could potentially be a good biological control agent of *T. vaporariorum* in Colombia, in environments that are not overly dry or warm. However, as the parasitoid seems to be negatively affected by certain combinations of climatic conditions, its limitations need to be further studied in order to ascertain its true potential as a natural enemy.

The entomopathogen *V. lecanii* has been tested under laboratory and field conditions in Colombia (González and López-Avila, 1997) with variable, usually poor, results.

The natural enemies of *B. tabaci*, found at many different sites in Colombia and Ecuador, included the aphelinid parasitoids *E. hispida*, *E. pergandiella*, *E. nigricephala*, *Encarsia* sp. and *Eretmocerus* sp., as well as the predators *D. pusillus* and *Chrysoperla* sp. The relative importance and efficiency of these natural enemies has not been assessed.

Yield losses

Three trials aimed at measuring losses due to *T. vaporariorum* on snap bean were conducted during 1998 and 1999.

The establishment of these trials coincided with the appearance in Colombia of another serious pest of common bean, the melon thrips, *Thrips palmi* Karny. As a result, the trials had to be redesigned to account for the incidence of the new pest. In large replicated plots, different insecticide regimes were established to measure yield responses and to partition losses due to thrips and whiteflies. The latter was achieved by using spinosad (Tracer®) Dow Chemical Co.) as a selective insecticide for *T. palmi* control and buprofezin (Applaud®, ICI) in mixture with monocrotophos (Azodrin®, Shell Co.) for whitefly exclusion. The mean loss due to *T. palmi* was 4.1 t/ha or 46% of the potential yield. *T. vaporariorum* caused losses of 2.1 t/ha or 23% of potential yield (Rendón et al., 2001).

Increased Socio-Economic Understanding

Farmers' perceptions of the whitefly problem

From 88% to 100% of farmers surveyed were male; 41% to 63% were owners who had been on their farms for over 5 years, planting an average of 1.2 ha with three or more crops planted in succession through the year. Table 2 shows that most farmers knew the insects; they were able to identify them as whiteflies and seemed to be aware of whitefly-related problems. The whitefly was regarded as an endemic pest by about 49% of farmers in the tropical highlands of Colombia and Ecuador, by 23% in mid-altitude valleys (400-1000 m) in Colombia and by 82% in the tropical lowlands of Colombia and Ecuador. A minority claimed to be able to predict whitefly epidemics, which they usually associated with dry weather. Whitefly problems in the survey area are distributed extensively, the insects being present in 72% of farms visited. The lowest incidence was in the Departments of Cauca and Cauca Valley, Colombia, where the insects were present in 46% of farms visited.

Whiteflies ranked as the most important pest of common bean in 50% of the sites visited, and as the key pest in 45% of the tomato farms that were surveyed. A relatively small percentage of farmers gave whiteflies the highest rating as pests when compared with other insects on the farm (for example, *T. palmi* on common bean or fruit borers on tomato) but a significant proportion thought that whiteflies could reduce yields by 50% or more. Up to 30% of melon growers and 34% of those planting tomato indicated that they have abandoned crops as a result of devastating whitefly attacks. Most farmers (76% of those growing cucurbits, 80% of those growing tomato) could identify the insects as pests but very few associated the presence of whiteflies with virus diseases (Table 2).

Technicians' perceptions of the whitefly problem

Virtually all extension agents and technical assistants interviewed considered that whiteflies were a major problem on tomato and common bean. Most (80%-93%) regarded whiteflies as pests, while 27%-39% considered that whiteflies act as both pests and vectors. Up to 70% thought that the situation in most crops had changed as a result of whitefly incidence. Very few were aware of new technologies to solve the problem, considered that the area planted to several crops had been reduced or that insecticide use had increased as a result of whitefly incidence. All acknowledged that using insecticides was the only recommendation they could make to farmers.

Table 2. Main characteristics and perceptions of farmers surveyed in the whitefly-affected areas of Colombia and Ecuador.

Percentage of farmers who:	Tropical highlands	Mid-altitude valleys	Tropical lowlands
Are male	88	100	100
Own their land	63	41	64
Have lived on the farm for more than 5 years	64	41	68
Receive technical assistance	15	18	42
Plant three or more crops simultaneously	47	68	36
"Rotate"[a] crops and/or fields	82	79	59
Plant more than twice a year	56	64	11
Know whiteflies	82	92	88
Regard whiteflies as a problem	67	64	65
Regard whiteflies as endemic	49	23	82
Relate whitefly incidence to climatic conditions	53	59	83
Consider that they can predict whitefly incidence	24	14	53
Give highest rating to whiteflies as pests	15	18	21
Consider that whiteflies can cause 50% or more yield losses	40	14	40
Identify whiteflies as pests	71	32	80
Identify whiteflies as virus vectors	0	5	0
Identify whiteflies as both pests and vectors	1	9	7

a. Rotation in this case sometimes means shifting the crop from one place on the farm to another.

Insecticide use

All farmers who attempted control of whiteflies (up to 70%) in mid-altitude and highland areas of Colombia and Ecuador used insecticides as the sole method of control. Virtually all applications against whiteflies were preventive, with little or no regard for insect populations and the phenological stage of crops. Few farmers received formal technical assistance; instead, they received frequent visits by insecticide salesmen who recommended a wide array of insecticides (up to 34 different brands) for whitefly control. The three crops most heavily sprayed were tomato, common bean and potato but, as shown in Table 3, many applications did not have whiteflies as the main target insect. Nevertheless, the heavy use of insecticides against other pests favours the development of the high levels of resistance detected in whiteflies in the Andean zone (Chapter 4.3, this volume).

Table 3. Percentage of farmers using insecticides to control whiteflies and other pests on selected crops in major ecological zones of Colombia and Ecuador.

Crop	Ecological zone	Using insecticides	Spraying against:		
			Whiteflies	Whiteflies and other insects	Other insects
Common bean	Tropical highlands	89	33	39	17
Common bean	Mid-altitude valleys	75	0	75	0
Tomato	Tropical highlands	90	11	63	16
Tomato	Mid-altitude valleys	92	8	50	34
Tomato	Tropical lowlands	86	38	48	0
Potato	Tropical highlands	86	0	0	86

Up to 57% of insecticide applications were reportedly made with organophosphate insecticides (OPs) alone. However, OPs also were reported as being used in mixtures with carbamates and pyrethroids (Table 4) to the extent that 80%-100% of the applications had OPs as ingredients. Given the high levels of insecticide resistance to OPs and pyrethroids that were detected in our studies, it is not surprising that a significant proportion of farmers (up to 60%) were beginning to use novel insecticides. This was most evident in tomato-growing areas in the tropical lowlands and in the mid-altitude valleys of Colombia.

Farmers had easy access to at least 36 different active ingredients and 40 commercial brands. Most insecticides used belong to the toxicological category I (extremely dangerous), as defined by Metcalf (1994) and are used with minimal safety precautions for handling and application. In the survey, the most widely used group of pesticides were OPs, with 40% to 50% share of the use, followed by carbamates and pyrethroids (Table 5). Novel products such as insect growth regulators, juvenoids and nicotinoids were reported as becoming popular (Rodríguez and Cardona, 2001). Also used were soaps, oils and botanicals.

Insecticides were reportedly applied to tomato and snap bean as often as three times a week (Rodríguez and Cardona, 2001). The number of applications per cropping season varied among regions but it is interesting to note that 24% of farmers surveyed made 10 or more applications (Table 6).

Most farmers used very low (0.2 cc/L) or very high (4.8 cc/L) dosages of concentrate, well below or above the 2 cc/L average dosage usually recommended by manufacturers. Up to 71% of farmers surveyed in Colombia and 55% in Ecuador took their own decisions on timing of applications as opposed to those who have access to technical assistance. More than half reported spraying on a calendar basis, regardless of infestation levels, and relatively few had received any kind of technical assistance.

Table 4. Percentage of farmers using organophosphates (OP), carbamates (CAR) and pyrethroids (PYR) to control whiteflies in major ecological zones of Colombia and Ecuador.

Insecticide group	Tropical highlands	Mid-altitude valleys	Tropical lowlands
OP	55	33	57
CAR	4	0	2
PYR	4	0	8
OP + CAR	5	45	5
OP + PYR	24	22	25
CAR + PYR	0	0	0
OP + CAR + PYR	8	0	3

Table 5. Percentage participation of organophosphate, carbamate and pyrethroid insecticides in chemical control of whiteflies in selected tropical highland areas of Ecuador and Colombia.

Insecticide category	Northern Ecuador–southern Colombia	Departments of Cauca and Cauca Valley, Colombia	Department of Antioquia, Colombia
Organophosphates	40	50	50
Carbamates	22	14	25
Pyrethroids	22	9	13
Novel products[a]	16	23	12
Others (soap, oils, etc.)	0	4	0

[a] Includes insect growth regulators, juvenoids and nicotinoids.

Table 6. Patterns of insecticide use for whitefly control in Colombia and Ecuador.

Number of insecticide applications per cropping season[a]	Percentage of farmers
1-3	34.8
4-6	25.0
7-9	8.8
10	7.4
> 10	24.0

a. The mean number of applications per season against the B biotype of *Bemisia tabaci* in the tropical lowlands of Colombia and Ecuador is 6.5.

Conclusions

In general, the results of both the biological surveys and the interviews with producers and their technical advisors underline the need for IPM research and implementation. A highly dangerous situation has developed for small-scale farmers in Colombia and Ecuador over the past 15 years: commercial pressures and an inadequate knowledge base have influenced farmers to adopt a "chemical culture", involving excessive dependence on pesticides. The situation has been aggravated by the introduction into the region of the B biotype of *B. tabaci*, a major pest of annual crops. The results, their consequences and recommendations for further action are discussed in more detail in Chapter 4.4.

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CHAPTER 4.3

Insecticide Resistance in Colombia and Ecuador

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Farmer interviews conducted as part of extensive on-farm surveys for the diagnostic phase of the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project (Rodríguez and Cardona, 2001; Chapter 4.1, this volume) showed that insecticide use against whiteflies in Colombia and Ecuador is excessive. In the tropical highlands and mid-altitude valleys, farmers spray their crops 5 to 6 times on average to control *Trialeurodes vaporariorum* (Westwood). The mean number of applications against the B biotype of *Bemisia tabaci* (Gennadius) in the tropical lowlands of Colombia and Ecuador was estimated at 6.5. Over-reliance on insecticides for whitefly control is so widespread that 30% of 325 farmers interviewed reported that they make more than 10 applications per cropping season. The frequency of applications in many cases is as high as two to three times per week. Most farmers complained about the limited control achieved by conventional insecticides and many farmers are now using novel insecticides such as buprofezin, pyriproxyfen, diafenthiuron and imidacloprid reportedly with better results. However, the 10 most widely used insecticides identified in the surveys comprised nine conventional products—dimethoate, carbofuran, chlorpyriphos, methamidophos, methomyl, profenofos, monocrotophos, cypermethrin and malathion—and only one of these novel insecticides, imidacloprid. The high toxicity of several of the conventional products in widespread use raises concerns over both human and environmental health, underlining the need for alternative approaches based on IPM.

Insecticide resistance in whiteflies is well documented as a widespread phenomenon in those countries in which monitoring of resistance has been conducted (Horowitz and Ishaaya, 1996). Recent reviews on the subject are those of Dittrich et al. (1990), Cahill et al. (1996b), Denholm et al. (1996) and Palumbo et al. (2001). In the Andean zone, Buitrago et al. (1994) detected high levels of resistance to cypermethrin and deltamethrin in populations of *T. vaporariorum* from the central highlands of Colombia. Resistance to organophosphates and carbamates was not as high, and ranged from low to moderate. Cardona et al. (1998; 2001) reported that the B biotype of *B. tabaci*, the main whitefly species in the lowlands of Colombia, was highly resistant to methamidophos and methomyl.

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A more general account of whitefly problems in the high Andes, mid-altitude valleys and lowlands of Colombia and Ecuador is presented elsewhere (Chapter 4.2, this volume), as are the results of biological surveys and farmer questionnaires, intended to characterize whitefly problems in these areas. Here, a summary is presented of findings on the resistance of whiteflies to conventional insecticides in the survey areas. Also included are baseline responses of *T. vaporariorum* adults to monocrotophos, lamda-cyhalothrin, bifenthrin, carbosulfan, carbofuran, thiamethoxam and imidacloprid. Baseline responses of *T. vaporariorum* nymphs to buprofezin, diafenthiuron and imidacloprid are included. The knowledge of whitefly problems synthesized in these studies is intended to provide a sound basis for pursuing IPM approaches in the Andean region of Latin America.

Methods and Materials

Experimental approach and locations

Field surveys and insecticide resistance testing were carried out between October 1997 and May 1999. In accordance with the common research methods agreed among the project partners, the vial technique was adopted to establish baseline data in the laboratory and for field assessment of resistance to conventional insecticides. The technique involves assessing the rate of mortality of a sample of whiteflies within a vial coated with a known concentration of insecticide (Plapp et al., 1990; Cahill and Hackett, 1992). Baseline data for imidacloprid were obtained following the methodology developed by Cahill et al. (1996a).

Laboratory work to establish baseline data and select diagnostic dosages was conducted at the headquarters of the Centro Internacional de Agricultura Tropical (CIAT) in Cali, Colombia. Average laboratory conditions were 24 °C and 75%-80% relative humidity (RH).

Field monitoring extended from northern Ecuador (Imbabura and Carchi provinces) to the north-eastern department of Guajira in Colombia. With the exception of coastal areas in Ecuador, all major areas affected by whiteflies were covered. Sites were chosen according to previous data on heavy insecticide use but care was taken not to perform tests in areas with mixed populations of whitefly species, as indicated in biological surveys carried out in parallel (Chapter 4.2, this volume). Thus, no insecticide resistance work was conducted in mid-altitude valleys where mixed populations of *B. tabaci* and *T. vaporariorum* had been recorded. In total, insecticide resistance was measured at 40 sites in Colombia and Ecuador. Between two and four insecticide resistance tests were conducted in each target area. Resistance of *T. vaporariorum* collected from common bean (*Phaseolus vulgaris* L.), tomato (*Lycopersicon esculentum* L.) and potato (*Solanum tuberosum* L.) was measured in highland areas along the Andean corridor from northern Ecuador to northern Colombia. Resistance of *B. tabaci* biotype B from tomato, eggplant (*Solanum melongena* L.), squash (*Cucurbita moschata* Duchesne) and cotton (*Gossypium hirsutum* L.) was measured in lowland areas of the region known as the “northern coast” of Colombia and in the Cauca Valley (a mid-altitude valley).

Baseline data and diagnostic doses

Adult whiteflies obtained from separate mass rearings of *T. vaporariorum* and *B. tabaci* biotype A, maintained at CIAT for over 10 years (without exposure to insecticides) were used to obtain baseline data. Technical grade insecticides were dissolved in acetone and 250 µL samples of the solution were pipetted into each 20-mL vial. The acetone was allowed to evaporate completely before introducing the test insects. Insecticides and dosages tested with *T. vaporariorum* were methamidophos (16.0, 8.0, 4.0, 2.0, 1.0 and 0.5 µg/vial), methomyl (2.5, 0.5, 0.1 and 0.02 µg/vial), cypermethrin (100.0, 50.0, 25.0, 12.5 and 6.25 µg/vial) and carbofuran (10.0, 3.3, 1.1, 0.33 and 0.11 µg/vial). Insecticides and dosages tested with the A biotype of *B. tabaci* were methomyl (25.0, 6.25, 1.56, 0.39 and 0.10 µg/vial), methamidophos (16.0, 4.0, 2.0, 1.0 and 0.5 µg/vial) and cypermethrin (450.0, 150.0, 50.0, 16.7 and 5.6 µg/vial). Five replicates of 20 insects were tested for each dosage. Acetone-coated vials were used as checks (controls). Mortality was recorded 6 hours after the adults were placed in the vials.

To generate baseline data for imidacloprid and thiamethoxam (Rodríguez et al., 2003), petioles of excised common bean leaves were immersed in aqueous solutions of commercial grade imidacloprid (10.0, 5.0, 2.5, 1.25, 0.625 and 0.312 ppm) for 48 hours to allow the leaves to take up the insecticide solution. Leaf discs were then cut and placed on agar in petri dishes, one leaf disk per petri dish (Cahill et al., 1996a) and infested with 20 *T. vaporariorum* adults per unit. Discs from leaves whose petioles had been immersed in distilled water were used as checks. Five replicates (units) per dosage were used. Mortality was recorded 6 hours after introduction of the adults into the units.

To generate baseline data with nymphs, we collected common bean leaves infested with pupae in each of the test sites. The infested material was then confined to cages. Adults emerging from these pupae were collected with an aspirator and introduced in clip cages attached to leaves of common bean seedlings. The adults (10 per clip cage) were allowed to oviposit for 24 hours. After hatching of the eggs, the area of the leaf infested with first instar nymphs was marked and the number of individuals thus obtained was recorded. Infested seedling leaves were then dipped for 5 s in 100 mL of the desired concentration of each of the insecticides tested. Formulated material was used in all tests and the desired concentrations were prepared by dilution with distilled water. Controls were dipped in distilled water. Treated plants were kept in a rearing chamber at 24 °C, 75%-80% RH and left undisturbed until adult emergence occurred. The number of surviving individuals in each treatment was recorded. Insecticides and dosages tested were buprofezin (100, 25, 6.2, 1.5, 0.3, 0.09 and 0.02 ppm), diafenthiuron (1000, 300, 100, 30, 10, 3, 1 and 0.3 ppm) and imidacloprid, (1000, 300, 100, 30, 10, 3 and 1 ppm). Four replicates were tested for each dosage.

Percentage mortality was corrected using the Abbott formula (Busvine, 1971). Tests in which check mortality was higher than 10% were not included in the analysis. Results were subjected to probit analysis (SAS Institute, 1988). Slopes, LC_{50} and LC_{90} values, as well as 95% confidence limits were tabulated. Baseline data were then used to choose diagnostic doses. Four doses were chosen empirically so as to obtain mortality ranging from 5% to 95%. After several tests, a single dose for each insect species and insecticide was chosen as the diagnostic dose (as defined by Halliday and Burnham, 1990) to be used in extensive monitoring of resistance under field conditions.

Field assessment of resistance

Resistance was assessed in the field using vials coated in the laboratory with diagnostic dosages of methomyl, methamidophos and cypermethrin, and transported to the field in ice-chests maintained at about 20 °C. These insecticides are representative of the conventional insecticides most widely used in the study area. At each site, adult whiteflies were collected with a mouth aspirator from the foliage of infested plants and transferred to the vials. Twenty adults per vial, replicated five times, were used for each test. Acetone-coated vials were used as controls. Infested vials were kept in the ice-chests for 6 hours before mortality was recorded. Any test in which mortality in control vials was higher than 10% was discarded. Percentage mortality was corrected using the Abbott formula (Busvine, 1971) and transformed to arcsine square root of proportion. Data were then analysed by a 1-way analysis of variance (ANOVA) (SAS Institute, 1988). When the *F* test was significant, comparison of means was by the least significance difference (LSD). Untransformed means are presented.

Results and Discussion

Baseline data and diagnostic doses

Table 1 presents baseline toxicology data for reference strains of *T. vaporariorum* and Table 2, the A biotype of *B. tabaci*. The LC_{50} and LC_{90} values reflect toxicities of the test

Table 1. Toxicological responses of laboratory strains of *Trialeurodes vaporariorum* to five insecticides.[a]

Insecticide	n	LC_{50} (95% FL)[b]	LC_{90} (95% FL)[b]	Slope ± SEM	X^2
Adults					
Methomyl	457	0.25 (0.15-2.6)	0.95 (0.76-14.7)	2.19 ± 0.55	4.05
Methamidophos	600	5.30 (2.5-7.6)	22.50 (15.6-48.7)	2.05 ± 0.49	0.08
Monocrotophos	710	9.70 (6.7-13.4)	175.40 (115.5-299.8)	1.00 ± 0.08	4.80
Cypermethrin	480	37.00 (22.0-55.7)	400.00 (232.7-953.5)	1.24 ± 0.18	2.95
Lambda-cyhalothrin	605	9.40 (6.1-13.5)	264.90 (170.9-455.3)	0.90 ± 0.06	0.10
Bifenthrin	380	2.40 (1.6-3.1)	6.70 (5.2-9.8)	2.90 ± 0.49	0.90
Carbofuran	504	1.97 (1.5-2.5)	6.80 (5.4-9.7)	2.37 ± 0.31	4.31
Carbosulfan	712	1.80 (1.5-2.1)	19.90 (16.3-24.9)	1.20 ± 0.05	3.30
Imidacloprid	921	5.70 (4.6-6.9)	28.40 (20.9-44.9)	1.90 ± 0.22	7.80
Thiamethoxam	670	8.60 (6.0-11.7)	101.00 (68.8-170.4)	1.20 ± 0.12	5.90
First instar nymphs					
Buprofezin	907	0.80 (0.6-1.1)	9.20 (6.9-13.0)	1.20 ± 0.09	7.30
Diafenthiuron	904	3.20 (2.3-4.4)	60.10 (41.8-92.8)	1.00 ± 0.06	10.50
Imidacloprid	483	16.50 (10.7-23.4)	171.50 (115.6-290.9)	1.21 ± 0.10	3.60

a. Conventional insecticides were tested using insecticide-coated glass vials. Imidacloprid tests were conducted using the technique developed by Cahill et al. (1996a). Nymphs were tested using a modification of the methodology described by Prabhaker et al. (1985).

b. Imidacloprid, thiamethoxam, buprofezin and diafenthiuron concentrations in ppm. All others in µg/vial. LC, lethal concentration; FL, fiducial limits.

Table 2. Toxicological responses of laboratory strains of *Bemisia tabaci* biotype A to three insecticides, using insecticide-coated glass vials.

Insecticide	n	LC_{50} (95% FL)[a]	LC_{90} (95% FL)[a]	Slope ± SEM	X^2
Methomyl	500	1.7 (1.1-2.3)	9.1 (6.7-13.7)	1.76 ± 0.21	1.73
Methamidophos	517	1.4 (0.9-1.6)	6.6 (5.3-14.7)	1.86 ± 0.47	14.89
Cypermethrin	502	14.4 (5.8-27.2)	202.9 (122.5-352.4)	1.12 ± 0.14	3.27

a. Insecticide concentration: µg/vial. LC, lethal concentration; FL, fiducial limits.

insecticides to susceptible strains of whiteflies that have not been exposed to insecticides for at least 10 years. As discussed by ffrench-Constant and Roush (1990), establishing baseline data for different insecticides is a fundamental step in resistance studies because these data will serve as a basis for future comparisons, which will allow researchers to detect any changes in insecticide resistance levels. In addition, as explained by Sanderson and Roush (1992) and Denholm et al. (1996), calculation of baseline data permits the selection of diagnostic doses, which can be used more conveniently in extensive efforts to detect and monitor resistance such as the one reported here that was carried out over numerous sites in the Andean zone of Ecuador and Colombia. Carbofuran was included because granular formulations of this insecticide are still effective against *T. vaporariorum* in Colombia (Cardona, 1995). Imidacloprid is a highly efficient insecticide that is becoming popular in the region. Careful monitoring of the efficiency of this product will be needed for future resistance management and IPM work to be undertaken by the second phase of the TWF-IPM Project. It is interesting to note that the LC_{50} value for imidacloprid for our reference strain of *T. vaporariorum* (2.4 ppm) was similar to that obtained by Cahill et al. (1996a) using susceptible strains of *B. tabaci* from Sudan and Pakistan.

The main purpose of the preliminary tests was to select diagnostic doses for methomyl, methamidophos and cypermethrin (Table 3) that were then used to compare the insecticide resistance of test samples of whiteflies from Colombia and Ecuador with that of the fully susceptible laboratory strains. We did not calculate diagnostic doses by statistical means. Instead, diagnostic doses were chosen empirically to produce >95% mortality.

Field monitoring of resistance in biotype B of B. tabaci

Responses to insecticides in our studies were arbitrarily classified as follows on the basis of mean percentage mortality:

- 0%-50% mortality: high resistance;
- 50%-80% mortality: intermediate resistance; and
- 80% mortality: low resistance.

According to this scale, the B biotype of *B. tabaci* recently introduced

Table 3. Response (corrected percentage mortality) of *Trialeurodes vaporariorum* and *Bemisia tabaci* adults to three insecticides.[a]

Whitefly species	Methomyl (2.5 µg/vial)	Methamidophos (32 µg/vial)	Cypermethrin (500 µg/vial)
T. vaporariorum	97	99	88
B. tabaci biotype A	99	100	98

a. Diagnostic dosages were tested using insecticide-coated glass vials.

into Colombia showed high levels of resistance to methomyl and methamidophos and, in some places, moderate levels of resistance to cypermethrin (Table 4). Considerable inter-population variation was detected. Variability in the response of the B biotype to insecticides has been detected also in Arizona (Sivasupramaniam et al., 1997), in Hawaii (Omer et al., 1993) and in California (Prabhaker et al., 1996). In general, insecticide resistance levels reflected insecticide use patterns. For example, the very high levels of resistance to acetylcholine esterase inhibitors (exemplified here by methomyl and methamidophos) in different sites in the departments of Atlántico, Córdoba and Sucre may reflect the extensive use of organophosphates on cotton, tomato and vegetables in the region. Lower levels of resistance to the test pyrethroid (cypermethrin) may be due to the less frequent use of this type of insecticide for whitefly control (Table 4 in Chapter 4.2, this volume). However, further interpretation of the data is hampered by the lack of knowledge of exactly where the B biotype whitefly that invaded Colombia originated and of the pattern of insecticide use in the area of origin. It is likely that the frequency of insecticide resistance was already high in the pest population at the time it was introduced.

Field monitoring of resistance in **T. vaporariorum**

The responses of *T. vaporariorum* to insecticides were arbitrarily classified on the same basis as those of *B. tabaci* (0%-50% mortality, high resistance; 50%-80% mortality, intermediate resistance; and > 80% mortality, low resistance). In the northern Ecuador-southern Colombia region (Table 5), *T. vaporariorum* populations were susceptible to methomyl in seven out of eight sites tested. Generalized resistance to methamidophos (the most widely used insecticide) was found. Extreme cases were those in northern Ecuador (Turquisal, Ibarra) and in southern Colombia (El Tambo, Gualmatán, Funes). Intermediate to low resistance to cypermethrin was detected in west-central Colombia (Table 6).

Table 4. Response (corrected percentage mortality) of *Bemisia tabaci* biotype B adults to three insecticides at sites on tropical lowland areas of the northern coast of Colombia.[a]

Site (department)[b]	Methomyl (2.5 µg/vial)	Methamidophos (32 µg/vial)	Cypermethrin (500 µg/vial)
Repelón (Atlántico)	42.8 c	2.0 d	92.0 ab
Cereté 1 (Córdoba)	54.0 b	1.5 d	87.8 abc
Cereté 2 (Córdoba)	12.4 de	0.8 d	68.8 c
Ciénaga de Oro (Córdoba)	41.8 bcd	3.0 d	94.6 a
Cotorra (Córdoba)	6.8 e	4.2 d	78.2 bc
Montería (Córdoba)	5.4 e	2.4 d	87.2 abc
Manaure (Guajira)	100.0 a	25.4 b	91.5 ab
Corozal 1 (Sucre)	9.8 e	14.0 bc	85.4 abc
Corozal 2 (Sucre)	30.0 bcde	4.8 cd	65.2 c
Sampués (Sucre)	15.4 cde	16.2 bc	88.8 abc
“CIAT” (reference strain)	100.0 a	100.0 a	100.0 a

a. Diagnostic dosages were tested under field conditions using insecticide-coated glass vials. Twenty adults per vial replicated five times were used for each test. Means within a column followed by the same letter are not significantly different at the 5% level (LSD test).

b. “CIAT” indicates a laboratory population of *B. tabaci* biotype A maintained for at least 10 years without exposure to insecticides.

Table 5. Response (corrected percentage mortality) of *Trialeurodes vaporariorum* adults to three insecticides in the southern Colombia-northern Ecuador tropical highland region.[a]

Site (country)[b]	Methomyl (2.5 µg/vial)	Methamidophos (32 µg/vial)	Cypermethrin (500 µg/vial)
El Tambo (Colombia)	100.0 a	1.8 d	95.2 a
Gualmatán (Colombia)	100.0 a	1.8 d	35.2 c
Funes (Colombia)	84.2 a	9.0 d	78.0 b
Ibarra (Ecuador)	100.0 a	4.0 d	100.0 a
Pimampiro (Ecuador)	87.4 a	46.4 bc	100.0 a
San Vicente (Ecuador)	55.6 b	59.0 b	100.0 a
Turquisal (Ecuador)	100.0 a	14.0 cd	100.0 a
“CIAT” (reference strain)	96.6 a	99.0 a	87.7 b

a. Diagnostic dosages were tested under field conditions using insecticide-coated glass vials. Twenty adults per vial replicated five times were used for each test. Means within a column followed by the same letter are not significantly different at the 5% level (LSD test).

b. “CIAT” indicates a laboratory population of whiteflies of the same species as the test populations maintained for at least 10 years without exposure to insecticides and used here as a susceptible check for toxicological studies.

Table 6. Response (corrected percentage mortality) of *Trialeurodes vaporariorum* adults to three insecticides at sites in the Antioquia-Cauca-Valle tropical highland region of west-central Colombia.[a]

Site (department)[b]	Methomyl (2.5 µg/vial)	Methamidophos (32 µg/vial)	Cypermethrin (500 µg/vial)
Carmen de Viboral (Antioquia)	96.0 a	55.0 c	69.8 d
El Peñol (Antioquia)	90.0 ab	76.4 b	80.2 cd
Rionegro (Antioquia)	100.0 a	43.4 c	84.2 bcd
La Unión (Cauca)	100.0 a	99.0 a	98.2 a
Pescador (Cauca)	62.0 c	2.6 e	67.0 d
Cerrito (Valle)	100.0 a	8.2 e	89.0 abc
La Cumbre (Valle)	100.0 a	100.0 a	98.0 a
Pradera (Valle)	79.4 bc	2.8 e	64.8 d
Tenerife (Valle)	99.6 a	3.0 e	72.4 cd
“CIAT” (reference strain)	96.6 a	99.0 a	87.8 bcd

a. Diagnostic dosages were tested under field conditions using insecticide-coated glass vials. Twenty adults per vial replicated five times were used for each test. Means within a column followed by the same letter are not significantly different at the 5% level (LSD test).

b. “CIAT” indicates a laboratory population of whiteflies of the same species as the test populations maintained for at least 10 years without exposure to insecticides and used here as a susceptible check for toxicological studies.

Whiteflies were susceptible to methomyl in all but two of the sites studied. Extreme resistance to methamidophos occurred in the Cauca Valley (Cerrito, Pradera, Tenerife) and in Cauca (Pescador). These are places where insecticide use on common bean and tomato is heaviest. Again, low to intermediate resistance to cypermethrin was detected. In the eastern mountain ranges of Colombia (Table 7), *T. vaporariorum* was susceptible to methomyl and less resistant to methamidophos than in other parts of the country, possibly because of lower insecticide use in that area.

Table 7. Response (corrected percentage mortality) of *Trialeurodes vaporariorum* adults to three insecticides at sites in the central-eastern tropical highland region of Colombia.[a]

Site (department)[b]	Methomyl (2.5 µg/vial)	Methamidophos[c] (32 µg/vial)	Cypermethrin (500 µg/vial)
Boyacá (Boyacá)	100.0 a	95.3 a	89.0 a
Tinjacá (Boyacá)	100.0 a	85.6 ab	75.2 abc
Bojacá (Cundinamarca)	100.0 a	40.0 c	57.9 cd
Pasca (Cundinamarca)	100.0 a	-	81.0 ab
Fosca (Cundinamarca)	100.0 a	71.6 bc	41.4 d
Garzón (Huila)	100.0 a	52.2 bc	88.8 a
Rivera (Huila)	100.0 a	40.0 c	88.7 a
Abrego (Norte de Santander)	98.4 a	60.3 bc	71.3 abc
Lebrija (Santander)	91.5 b	3.0 d	64.4 bc
“CIAT” (reference strain)	96.6 a	98.8 a	87.8 a

a. Diagnostic dosages were tested under field conditions using insecticide-coated glass vials. Twenty adults per vial replicated five times were used for each test. Means within a column followed by the same letter are not significantly different at the 5% level (LSD test).
b. “CIAT” indicates a laboratory population of whiteflies of the same species as the test populations maintained for at least 10 years without exposure to insecticides and used here as a susceptible check for toxicological studies.
c. Test was not conducted at Pasca location.

Conclusions

In summary, the B biotype of *B. tabaci*, recently introduced into Colombia, showed high levels of resistance to methomyl and methamidophos and, in some places, moderate levels of resistance to cypermethrin. In highland and mid-altitude areas of Colombia and Ecuador, *T. vaporariorum* showed high resistance to methamidophos, intermediate resistance to cypermethrin and low resistance to methomyl. In general, levels of insecticide resistance in whiteflies seem to be related to the intensity of use of the particular kind of insecticide. Baseline responses of *T. vaporariorum* to carbofuran and imidacloprid are presented.

These results provide a basis for future monitoring of resistance and for the development of insecticide resistance management strategies. Novel insecticides such as diafenthiuron, buprofezin, pyriproxyfen and imidacloprid are still effective for whitefly control in Colombia and Ecuador (Rodríguez et al., 2003). Because whiteflies can also develop resistance to novel insecticides (Denholm et al., 1996; Horowitz and Ishaaya, 1996; Elbert and Nauen, 2000; Palumbo et al., 2001) monitoring of resistance and development of baseline data should continue in order to facilitate management of whitefly populations. The results are further discussed and proposals for further action based on these results are presented in Chapter 4.4 (this volume).

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CHAPTER 4.4

Conclusions and Recommendations

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When the Systemwide Program on Integrated Pest Management (SP-IPM) launched its project for "Sustainable integrated management of whiteflies as pests and vectors of plant viruses in the tropics" (The Tropical Whitefly IPM Project, TWF-IPM), the nature of whitefly problems in the high Andes of Colombia and Ecuador was poorly understood. Anecdotal reports and fragmentary scientific studies, mostly published only in the "grey" literature, suggested that whitefly problems were severe and insecticide misuse widespread in this socially and economically important horticultural area (Chapter 4.1, this volume). The seriousness of the reported situation led partners in the project planning process to establish a sub-project specifically to look at "Whiteflies as pests of annual crops in the highlands of Latin America". However, at the outset of the project, the species of whiteflies responsible for the problems was uncertain and the role of insecticides and insecticide-resistance in aggravating whitefly-related problems was unclear. The first phase of this project provided, for the first time, an opportunity to survey systematically the main crop production areas of this ecologically complex region and characterize their whitefly problems, both from a biological point of view and in terms of farmers' perceptions of these problems.

Scientists and technicians of the participating national research organizations, the Instituto Nacional Autónomo de Investigaciones Agropecuarias (INIAP) in Ecuador and the Corporación Colombiana de Investigación Agropecuaria (CORPOICA) in Colombia, conducted field surveys and farmer interviews between October 1997 and December 1998, in close collaboration with their counterparts from the project's coordinating organization, the Centro Internacional de Agricultura Tropical (CIAT) (Chapter 4.2, this volume). Researchers at CIAT continued until May 1999 to complete parallel studies of insecticide resistance in whiteflies from selected sites within the study area (Chapter 4.3, this volume).

The results of these studies (Chapters 4.2 and 4.3, this volume) provide a unique description of whitefly problems in Andean agriculture, which is not only of inherent scientific value but also will now serve as a sound foundation for action to address the very considerable agricultural production, human health and

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environmental problems that the study revealed. Integrated pest management approaches are likely to play a key role in tackling these problems, both within the framework of future phases of the current project and beyond. Key lessons learned in this study and proposals for further action are discussed here and in the final chapter on Conclusions (this volume).

Conclusions

Whitefly identification and host crop importance

The greenhouse whitefly *Trialeurodes vaporariorum* (Westwood) is the most important whitefly species in highland and mid-altitude areas of Colombia and Ecuador. Although it affects numerous horticultural crops and ornamentals, *T. vaporariorum* is most important as a pest of common bean (*Phaseolus vulgaris* L.) and tomato (*Lycopersicon esculentum* Mill.). This confirms previous work (Cardona, 1995; Corredor et al., 1999) on the increasing importance of *T. vaporariorum* as a major pest in the highlands of the Andes. However, it is now recognized that whitefly problems in the area are more widespread than initially thought.

Altitudinal distribution

The results of extensive surveys showed that *T. vaporariorum* occurs in mid-altitude and highland areas ranging from 700 to 2800 m above sea level and that *Bemisia tabaci* (Gennadius), the tobacco whitefly, ranges from sea level to 900 m above sea level. Thus, these two species overlap in many of the mid-altitude valleys. This is important because proper identification of whitefly species is a critical step in the formulation of research strategies. If, for example, *B. tabaci* is erroneously assumed to be the main pest species in mid-altitude valleys or highland areas, then IPM programs will be ill targeted and may not be effective.

Farmer perceptions

Whiteflies, in this case *T. vaporariorum*, were considered to be the key pest in common bean at 50% of the sites visited and the key pest in 45% of the tomato farms that were surveyed. A significant proportion of farmers think that whiteflies can reduce yields by 50% or more. This perception may well be justified. Previous studies (Cardona et al., 1993; Cardona, 1995) and work conducted in the first phase of the TWF-IPM Project showed that *T. vaporariorum* may cause losses ranging from 23% to 54% of the potential yield in snap bean (also *Phaseolus vulgaris* L.). There is no doubt that whiteflies have a high damage potential and that farmers are correct in believing that whitefly attacks have created numerous problems and have become a major constraint to agricultural production in the region.

Insecticide use

Whiteflies in Colombia and Ecuador have become the target of intensive, in most cases excessive, use of insecticides. This in turn has resulted in the destruction of natural enemies and the creation of new secondary pest problems such as the leafminer *Liriomyza huidobrensis* (Blanchard) and pod borers such as *Epinotia aporema* (Walsingham) on common bean and the *Neoleucinodes elegantalis* (Gueneé) on tomato. As shown in the previous chapter, excessive insecticide use has resulted also in the development of high levels of resistance of whiteflies to conventional insecticides. Some farmers are now using costly novel insecticides that may become useless in a few years because of insecticide resistance. Knowledge on insecticide use patterns and insecticide resistance levels obtained in this first, diagnostic, phase of the TWF-IPM Project will be important in the formulation of insecticide resistance management strategies and in the development of IPM options aimed at reducing pesticide use.

Insecticide resistance

In highland and mid-altitude areas of Colombia and Ecuador, *T. vaporariorum* showed high resistance to methamidophos (an organophosphate), intermediate resistance to cypermethrin (a pyrethroid) and low resistance to methomyl (a carbamate), three test products chosen as exemplifying the major groups of conventional insecticides currently in use. In general, resistance levels seem to be related to intensity of insecticide use. Responses of *T. vaporariorum* to carbofuran and imidacloprid were determined, to provide a baseline for future monitoring of resistance against these products, both of which are in widespread use. The B biotype of *B. tabaci*, recently introduced into Colombia, showed high levels of resistance to methomyl and methamidophos and, in some places, moderate levels of resistance to cypermethrin.

Development of resistance to representative conventional insecticides was higher than those reported in a previous study (Buitrago et al., 1994). The diagnostic survey showed that insecticide resistance is widespread, affecting common bean and tomato growers alike. These are important findings that help explain why conventional insecticides are no longer effective and why farmers are now using excessive numbers of applications of conventional pesticides and new generation pesticides that are costly. In the long-term, the need to use more expensive insecticides may threaten the economic viability of common bean and tomato production in mid-altitude and highland areas where small-scale, usually poor, farmers predominate.

Recommendations

There is an urgent need to develop and implement IPM systems that will help re-establish the ecological equilibrium. Experience in the Sumapaz region of Colombia has shown that this is feasible: implementation of a relatively simple IPM package based on cultural control and sanitation practices, timely application of effective insecticides and reliance on natural control in certain areas resulted in a 66% reduction in insecticide use (Cardona, 1995). Although the system was economically and technically viable, adoption rates varied with geographical area within the region and were not as high as researchers expected. A main obstacle encountered was the high level of risk aversion (as defined by Tisdell, 1986) among small-scale farmers. However, overall adoption rates of 30%-35% were regarded as high by social scientists linked to the project (Nohra Ruiz de Londoño [CIAT Impact Unit], personal communication, 1996) and suggest that careful planning, active participation of farmers in decision making, and participatory research approaches may help overcome barriers to adoption.

Several promising IPM tactics have been identified (Cardona, 1995; CIAT, 2000). These include the replacement of broad-spectrum insecticides with more selective ones, and the timing of applications according to pre-established action thresholds. In addition, cultural control measures such as the incorporation of crop residues, leaf roguing and manipulation of planting dates may be effective in some circumstances. Advances have been made also in the evaluation of the parasitoid *Amitus fuscipennis* MacGown and Nebeker (Manzano et al., 2000) and the entomopathogen *Verticillium lecanii* (Zimm.) (González and López-Avila, 1997) but more work needs to be done in order to measure the effectiveness of these and other natural enemies. These preliminary efforts to develop and test IPM strategies locally, coupled with findings from the diagnostic phase of the TWF-IPM Project, can be used as a basis for developing more widely applicable IPM strategies. The overall results of these efforts should be the development of systems that will: (1) reduce levels of pesticide used by common bean farmers; (2) reduce health risks to farmers and consumers; and (3) re-establish the ecological equilibrium in areas that have been disturbed by excessive pesticide use.

The immediate objective of any future IPM initiative should be to reduce pesticide use. Farmers currently depend so heavily on chemical pesticides, and environmental disruption is of such magnitude, that for the foreseeable future it is most unlikely that any IPM system could be effective without a chemical component. In the longer term, as more farmers adopt IPM approaches and environmental equilibrium is re-established, more ambitious, pesticide-free, strategies might become feasible; however, for the moment at least, monitoring of resistance should continue and sound strategies to manage insecticide resistance will have to be developed. Testing of new chemicals that are effective but less detrimental to the environment will also be important.

Based on the diagnostic phase survey work, IPM pilot studies should be continued in the following hot spots: Sumapaz, Oriente Antioqueño, Tenerife, Pradera and Nariño in Colombia, and in the Chota valley in northern Ecuador. However, problems associated with *T. vaporariorum* extend far beyond the Colombian and Ecuadorian highlands and common bean. This whitefly also has been reported as a significant field pest affecting melon (*Cucumis melo* L.) and tomato in Mexico (Flores et al., 1995), tomato in Costa Rica (Alpizar, 1993), tomato and common bean in the Dominican Republic (FUNDESA, 1996), as well as tomato, common bean and snap bean in Venezuela (Arnal et al., 1993), Ecuador (Mendoza, 1996), Colombia (Cardona, 1995), Bolivia (CIAT, unpublished records), Brazil (Gerk et al., 1995), Peru (Núñez 1995) and Argentina (L'Argentier et al., 1996; Giganti et al., 1997). *T. vaporariorum* is also a major pest in greenhouses in Colombia (Corredor et al., 1999) and Argentina (Viscarret and Botto, 1996; Salas et al., 1999). The benefit of developing effective IPM approaches for *T. vaporariorum* is therefore likely to be felt beyond the highlands of Colombia and Ecuador.

In order to overcome existing barriers to adoption (Cardona, 1995), IPM components should be tested with local communities using participatory research methods (Ashby, 1990). A possible approach that already has shown promise in Latin America (Brown and Hocdé, 2000) is to involve Comités de Investigación Agrícola Local (CIALs) in the testing (and, in some cases, validation) of individual IPM components and of promising IPM strategies. If major changes in existing practices are found to be necessary such as the general adoption of particular cropping dates or crop rotations, then the implementation of IPM will have to be discussed with local communities and government. The success of IPM approaches will depend ultimately on establishing consensus among IPM stakeholders. Given farmers' attitudes to chemical pesticides and their current reliance on these products as the sole method of addressing whitefly problems, farmer education (Whitaker, 1993) will be most important; however, if a major shift towards IPM approaches is to be achieved, advocacy efforts directed at consumers and the various stakeholders involved in agricultural production and processing is likely to be necessary as well.

Whitefly problems in the Andean highlands are evidently serious, extensive and deep-rooted, so immediate success cannot be anticipated. However, a phased approach, as conceived within the framework of the TWF-IPM Project, promises tremendous benefits to producers, consumers and the environment. The necessary investment in research, education and policy change should therefore be undertaken without delay.

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SECTION FIVE

Special Topics on Pest and Disease Management

BLANCA 302

CHAPTER 5.1

Sustainable Integrated Management of Whiteflies through Host Plant Resistance

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Introduction

As direct feeding pests and vectors of plant viruses, whiteflies constitute a major problem in the production of cassava (*Manihot esculenta* Crantz) in Africa, the neotropics and, to a lesser degree, Asia. The largest complex in the neotropics includes *Bemisia tabaci* (Gennadius)[1], *B. tuberculata* (Bondar) and *Trialeurodes variabilis* (Quaintance). *B. tabaci* has a pantropical distribution, feeding on cassava throughout Africa, where it transmits African Cassava Mosaic Disease, and several countries in Asia, including India and Malaysia.

Whitefly feeding affects cassava in three ways. Direct damage is caused by feeding in the phloem of the leaves, inducing chlorosis and leaf fall, which results in considerable reduction in root yield if prolonged feeding occurs. Yield losses of this type are common in the neotropics. Whiteflies also produce a honeydew, which provides a medium for sooty mould growth that can reduce yields. Most importantly, *B. tabaci* is a major vector of cassava viruses.

Research on whitefly control in the neotropics has emphasized host plant resistant (HPR) and biological control. The bulk of this report will cover HPR research on whiteflies (*Aleurotrachelus socialis* Bondar) as a component of the Cassava Whitefly Integrated Pest Management (IPM) Project.

In traditional production systems, resource-limited farmers have few options available for controlling pests. The cassava germplasm bank at the Centro Internacional de Agricultura Tropical (CIAT) has nearly 6000 accessions and locally selected cultivars (land races) collected primarily in the neotropics. These traditional cultivars represent centuries of cassava cultivation in diverse habitats, having been selected by farmers over a long period in the presence of a high diversity of herbivores. These land races often possess traits that confer low to moderate levels of resistance to multiple pests. This germplasm bank is constantly being evaluated for resistance to several arthropod pests that can cause yield losses in cassava. Evaluations often are done in more than one ecosystem.

The general purpose of this project is "to reduce crop losses due to whitefly feeding damage and whitefly-transmitted viruses, and

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[1] Evidence suggests that *Bemisia tabaci* represents a species complex with numerous biotypes.

prevent further environmental degradation and food contamination due to excessive pesticide use, leading to more productive and sustainable agricultural systems”.

The project has three major objectives, which are to:

(1) Identify and access exotic or novel genes and gene combinations that can contribute to germplasm enhancement for whitefly resistance in cassava;
(2) Study the genetics of resistance and to map genes for whitefly resistance in cassava and develop molecular markers for their incorporation into improved African, Latin American and Asian germplasm; and
(3) Develop crop management options for reducing whitefly populations and the transmission of whitefly-transmitted viruses.

The research presented in this chapter is being funded by the Ministry of Foreign Affairs and Trade (MFAT) Host Plant Resistance (HPR) Project and consists of four major areas of activity:

(1) Cassava germplasm evaluation to identify sources of whitefly resistance in land race varieties in the CIAT cassava germplasm collection;
(2) Identification of genomic regions responsible for the determination of whitefly resistance in cassava;
(3) Identification of resistance mechanisms in cassava; and
(4) Tritrophic relationships to determine the effect of HPR on whitefly parasitism.

Cassava Germplasm Evaluation

Field screening of cassava germplasm can be done at several sites in Colombia. Ideally, field populations of whiteflies should be high and damage levels significant so as to distinguish susceptible cultivars. Field evaluations of cassava germplasm use a population (nymphs) scale combined with a leaf damage scale (Table 1). Evaluations are made periodically throughout the growing cycle; four to five evaluations are done, 1.5 to 2.0 months apart. During the 1998 growing cycle, cassava clones were evaluated at CIAT in the Cauca Valley and Nataima in the Tolima valley of Colombia.

From 1994 through to 1996, whitefly (*A. socialis)* populations at Nataima, Tolima, were lower than normal. Low whitefly populations provide inadequate selection pressure to ensure reliable resistance evaluation. All evaluations done at Nataima are with natural field populations of whiteflies.

During 1997-98, whitefly populations increased significantly, allowing for more accurate germplasm evaluation. During 1999, some 1651 cassava clones were planted in observational fields, and 1418 were evaluated. These clones were the F_1 progeny from crosses of cassava clones previously selected for their resistance to whiteflies, with clones with desirable agronomic characteristics. These crosses resulted in 17 families (Table 1). Four field evaluations were done over a period of several months, using a whitefly damage and populations scale, as previously described.

Results indicate that whitefly populations were high and selection

Table 1. Families of cassava clones formed from crosses of whitefly (*Aleurotrachelus socialis*)-resistant and -susceptible cultivars.

Family	Crosses[a]		Observation
	Female	Male	
CM 3317	MBra 12 (T)	MCol 1468 (S)	MCol 1468 = CMC-40
CM 5438	MBra 12 (T)	MCol 1505 (S)	
CM 7559	MNGua 2	MBra 12 (T)	
CM 8884	CG 489-4 (R)	MCol 1468 (S)	CG 489-4 = MEcu 72 X MBra 12
CM 8885	CG 489-4 (R)	MCol 1505 (S)	CG 489-4 = MEcu 72 X MBra 12
CM 8887	CG 489-4 (R)	MCol 2256	CG 489-4 = MEcu 72 X MBra 12
CM 8889	CG 489-23 (R)	MCol 1468 (S)	CG 489-23 = MEcu 72 X MBra 12
CM 8891	MCol 1468 (S)	CG 489-34 (R)	CG 489-34 = MEcu 72 X MBra 12
CM 8892	MCol 2246	CG 489-34 (R)	CG 489-34 = MEcu 72 X MBra 12
CM 8893	MCol 2256	CG 489-34 (R)	CG 489-34 = MEcu 72 X MBra 12
CM 8960	MCol 2246	MBra 12 (T)	
CM 8961	MCol 2256	MBra 12 (T)	
CM 8984	MCol 1505 (S)	CG 489-34 (R)	CG 489-34 = MEcu 72 X MBra 12
CM 8990	MCol 2026	CG 489-34 (R)	CG 489-34 = MEcu 72 X MBra 12
CM 8991	MCol 2026	MBra 12 (T)	
CM 8995	MEcu 72 (R)	MCol 1468 (S)	
CM 8996	MEcu 72 (R)	MCol 2246	

a. R, whitefly resistant cultivar; T, whitefly tolerant cultivar; and S, whitefly susceptible cultivar.

pressure was good. Of the 1418 clones evaluated, 938 (66%) had damage ratings above 3.6 and were eliminated as susceptible (Figure 1); 308 (22%) clones had an intermediate damage evaluation (2.6 to 3.5). The remaining 172 (12%) clones had damage ratings below 2.6 and are considered promising for resistance; 56 (3.9%) clones had damage ratings of 1 to 1.5, indicating possible high levels of resistance. However, one cycle (year) of field evaluation is not adequate to identify resistance confidently and these clones will continue to be evaluated for at least 3 cycles (years). A 1.0 to 1.5 rating signifies no damage symptoms and low whitefly population levels. The families that most frequently occurred in this group were CM 8984, with nine selections, and CM 8893, with six. In both these families, the resistant parent was CG 489-34, indicating that this clone may contain good heritable resistant traits.

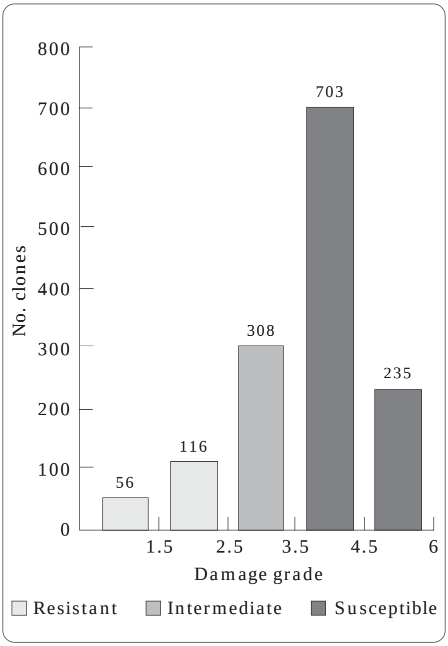


Figure 1. Evaluations of F_1 cassava clones from crosses of whitefly (*Aleurotrachelus socialis*)-resistant and -susceptible cultivars at the Corporación Colombiana de Investigación Agropecuaria (CORPOICA), Nataima, Tolima (1997-98).

Whitefly populations at CIAT were moderate to high for the fourth consecutive year. Evaluations at CIAT during 1998 concentrated on cultivars that had been selected as promising for resistance during previous years at the Instituto Colombiano Agropecuário (ICA)-Nataima.

Thirty-two clones sown in 50-plant plots were evaluated using a 1 to 6 (low to severe damage or low to high whitefly population level) scale. Whitefly population scales are based on counts of nymphs, pupae and adults. The 32 clones evaluated consisted of land race varieties, hybrids and backcrosses, and previously had shown good levels of resistance in screening trials in Tolima. Five regional or farmer varieties from the Tolima area also were included.

Results showed that nine cultivars, or 26.5%, presented very low damage levels (1.0 to 1.5) and three cultivars had damage levels between 1.6 and 2.5. The remaining 20 cultivars had damage levels between 2.6 and 5. The regional cultivars from Tolima, Azucena, Ceiba Blanca, Almidona, Llanera Precoz and Cuero de Marrano had damage and population ratings between 4 and 4.5, indicating that farmer varieties in the regions are susceptible to whiteflies and probably experiencing significant yield losses.

The nine best cultivars were MEcu 64, MPer 335, MPer 415 and MEcu 72 (all land race varieties), and CM 8424-6, CM 8424-33, CM 8424-4, CG 489-34 and CG 489-4 (all hybrids). All had damage ratings of 1.0, except MPer 415 and CG 489-34 with ratings of 1.5. MEcu 72 and CG 489-34 had maintained low damage ratings consistently, over several years. The varieties MEcu 64 and MPer 335 had excellent growth habits as well as low damage ratings and low whitefly populations. These two varieties will continue to be evaluated at CIAT and Tolima and should enter into breeding schemes for improved whitefly-resistant clones.

Identification of Genomic Regions Responsible for the Determination of Whitefly Resistance in Cassava

Previous research at CIAT determined the existence of different sources of resistance to whitefly. The most important genotypes are MBra 12 and MEcu 72, which were used as parentals in the generation of new genotypes. CG489-34 has shown the best resistance behaviour. The more susceptible genotypes, MCol 2026 and MCol 1505, also were selected and evaluated.

The goal of this project is to study and screen plants with resistance to whitefly in cassava, using molecular techniques.

Different breeding populations were obtained from the resistant and susceptible genotypes as parentals:

CG 489-34 X MCol 2026 = 131 individuals
CG 489-34 X MCol 1505 = 108 individuals
MBra 12 X MCol 2026 = 135 individuals

We selected the more contrasting individuals (resistant and susceptible) in the field for each family. DNA was extracted from the different individuals of each group and, with the parental, was mixed in a bulk. We used molecular screening (DNA restriction fragment length polymorphisms [RFLPs] and random amplified polymorphic DNA [RAPDs]) intended to find markers associated to resistance, and later it is intended to isolate the responsible genes. The parental lines and the bulk will be screened with amplified fragment length polymorphism (AFLP) markers.

Parentals from each family were evaluated with RFLP markers from the cassava map (Fregene et al., 1997). Seventy-two polymorphic probes were obtained. Sixty-two RAPD markers have been screened with the bulks of the same families; the primer OP.P3 showed a clear polymorphism between the susceptible and resistant group. This marker is being evaluated with the whole population of each cross to confirm its association with the resistance (Figure 2).

We intend to isolate and sequence the polymorphic bands and generate a sequence characterized amplified region (SCAR). This can be used to make a diagnosis of resistant materials and determine the most promising genotypes for breeding programs and farmers.

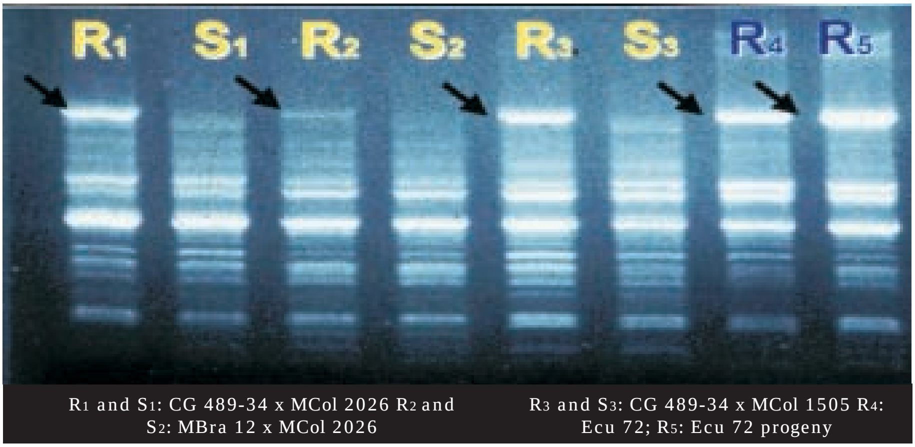
With the sequence of the genes of resistance, homologies will be established with those reported on other crops, to understand expression patterns. The RFLP, RAPD, microsatellites and AFLP markers will be used to generate a framework map and for the quantitative trait locus (QTL) analysis.

Identification of Resistance Mechanisms

Whitefly feeding behaviour

Cassava genotypes with resistance to whiteflies have been identified at CIAT. Resistant clone MEcu 72 shows high mortality for both adult and immature whiteflies, which may suggest less feeding on this genotype under natural conditions. It is necessary to identify whitefly feeding behaviour on susceptible genotypes and to make comparisons with MEcu 72 in order to better understand the mechanisms of resistance.

Electronic monitoring of insect feeding (EMIF) is a technique that permits the identification and quantification of the feeding behaviours



R_1 and S_1: CG 489-34 x MCol 2026 R_2 and S_2: MBra 12 x MCol 2026

R_3 and S_3: CG 489-34 x MCol 1505 R_4: Ecu 72; R_5: Ecu 72 progeny

Figure 2. Random amplified polymorphic DNA (RAPD) bulk analysis of whitefly (*Aleurotrachelus socialis*)-resistant and -susceptible cassava clones.

of hemipteran insects. By passing an electrical signal to a test plant, and tethering an insect with a fine gold wire, modifications caused by stylet movements and feeding behaviours can be observed as waveforms. The EMIF technique has been used extensively for the study of mechanisms of plant resistance to insects. CIAT presently owns two AC-Electronic feeding monitors, and has access to a DC version of the system (electrical penetration graph [EPG]). The AC systems have been used with leafhoppers (*Empoasca kraemeri* Ross & Moore) on common bean (*Phaseolus vulgaris* L.), and the DC systems with cassava mealybugs (*Phenacoccus manihoti* Matile-Ferrero). Preliminary observations with both systems suggested that an easy protocol could be developed for monitoring the whitefly. In addition, CIAT technicians needed to be trained in wiring techniques, operation of the system, computer display and data acquisition.

Electronic monitoring methodology

The methodology devised for electronic monitoring of whitefly feeding behaviour includes two major steps: a wiring technique to attach the thin wire to the mesonotum of an adult whitefly and the proper settings (ground voltages, signal frequency) of the electronic monitoring system.

The “normal” gold wire used with leafhoppers (12.7 µm) is too thick and stiff for the small whitefly adults. A thinner wire needed to be obtained. Since purchase of a thinner wire was difficult in the short period of time allowed for this project, thinning of the existing gold wire was necessary. To do this, a 10- to 20-cm piece of the thick wire is placed for 45 min in a solution of 3 mol nitric acid and 9 mol hydrochloric acid. This is a potent oxidizer that dissolves away part of the gold, leaving a thinner wire while conserving its electricity conducting properties. Pieces of this thin gold wire are attached with silver paint to a copper stub.

Female whiteflies from a greenhouse colony are placed by mouth aspirator on a vacuum stand and held in place by a gentle vacuum. Several individuals can be placed simultaneously. With the help of the copper stub, the thin gold wire is placed on the mesonotum of a female, and a small drop of electrically conducting silver paint is used to attach the wire to the insect. Care needs to be taken to not get silver paint on the whitefly’s eyes or wings. This affects their behaviour even more than the tethered condition. New insects need to be used should silver paint get on their wings or eyes.

Tethered whiteflies can be acclimated to the wire by placing them on a cassava leaf for 1 hour. Before the electronic monitoring session begins, whiteflies are starved for 30 minutes. The copper stub holding the tethered whitefly is attached to the alligator clip on one of the input electrodes of the electronic monitor.

A constant signal, 250 mV at 250 Hz, was established as the best setting for *A. socialis*. This signal is transmitted via an electrode inserted in the substrate of a potted plant (4-8 weeks old). Detached leaves placed on a container filled with water and sealed with the output electrode of the electronic monitor also worked and showed better conductivity than the potted plants. However, the effects of detached leaves vs. whole plants on whitefly feeding behaviour need to be evaluated further.

Feeding behaviour

During probing, stylet movements and other feeding behaviours induce changes to the constant signal. These modifications, known as waveforms, are captured in a computer using an analogue to digital converter board, displayed on the screen on a time scale, and stored for post-acquisition measurement and analysis. Once waveforms have been correlated with specific behaviours, quantification can be made on their frequency and duration.

Waveforms produced by probing *A. socialis* on CMC 40 are shown in Figure 3. Because of time limitations, waveforms cannot be correlated with specific behaviours for this whitefly species. However, it has been reported that several species of whiteflies share very stereotypical styles of feeding and that waveform patterns are very similar among them. Since waveforms shown are similar in appearance to those reported in the literature, we will describe them. All probes observed started with salivation waveforms corresponding to the intercellular path that the stylets follow in their way to the phloem. These waveforms are shown in Figures 3A and 3C as "intercellular stylet path". They have been correlated also with the deposition of a salivary sheath. In some instances, the stylets come across treachery elements in the xylem and the whitefly ingests from them. This particular waveform has not been correlated with completely, but there is evidence of ingestion. After reaching the phloem, normally whiteflies go into continued ingestion, producing a flat waveform whose beginning is illustrated toward the end of the trace (Figure 3).

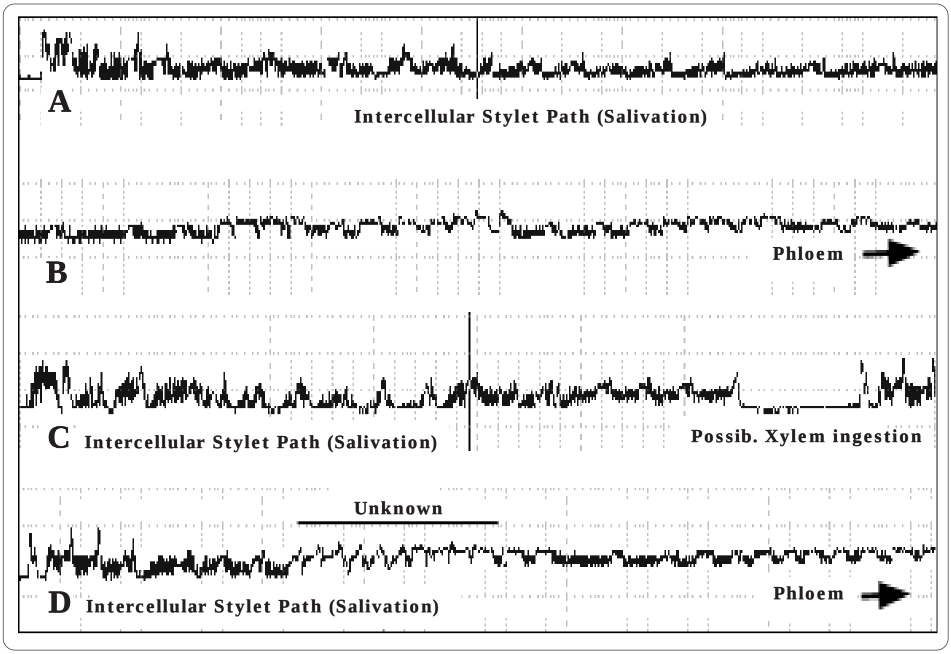


Figure 3. Waveforms produced by probing *Aleurotrachelus socialis* on cassava cv. CMC 40. A and B: Single probe (stylet penetration). B and C: Single probe forms a second whitefly. Note that waveform identified as possible xylem ingestion and an unknown waveform that has yet to be correlated with a feeding behaviour.

Figure 3D shows a new waveform that previously has not been identified or associated with any behaviour. It would be expected that, with more electronic monitoring of whitefly feeding, more patterns would be identified and more correlational work would be needed. Now that a methodology exists for the study of whitefly feeding behaviour at CIAT, comparisons can be made among resistant and susceptible genotypes.

Tritrophic Relationships: Determining Effect of HPR on Whitefly Parasitism

It has been observed that farmers will use pesticides to control high populations of cassava whiteflies. Pesticide applications will reduce the effectiveness of biological control as well as cause environmental contamination. Several years of research at CIAT has identified cultivars with varying levels of resistance to whiteflies, especially the species *A. socialis*.

Present research is investigating the compatibility between HPR and biological control. The combination of these two methods could reduce pest populations below economic injury levels or extend the usefulness of HPR. In addition, it is important to know the effect that plant resistance, especially antibiosis, might have on parasitoid populations.

The compatibility of the parasitoid *Encarsia hispida* DeSantis was evaluated on whitefly immatures feeding on whitefly resistance genotypes of cassava. The resistant varieties tested were MEcu 72, and CG 489-4; the susceptible check was CMC 40. Parasite development time, fecundity and survival were measured.

Plants of the abovementioned varieties at 1 month of age were infested with *A. socialis* eggs by exposing the leaf undersurface to ovipositing adults for 36 hours. After 15 days, when whitefly immatures were in the second instar, they were exposed to *E. hispida* parasites. *E. hispida* was obtained by collecting cassava leaves with parasitized whitefly pupae in the field at CIAT, placing the leaves in blackened plastic traps and capturing emerging parasites. The results at this point are preliminary.

Results to date indicate that the female longevity of *E. hispida* was similar on varieties CMC 40 (17.8 days) and CG 489-34 in experiments 1 and 2 (12.2 and 13.8 days, respectively) (Figure 4). *E. hispida* longevity on MEcu 72 was greatly reduced, only 7.7 days, indicating that this variety has factors or characteristics that affect the longevity of the parasite. These factors could be the number of tricomes or a chemical component that is interfering with parasitoid feeding (Table 2).

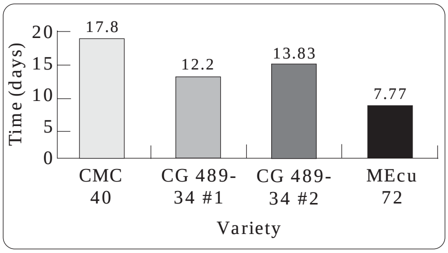


Figure 4. Longevity of the parasitoid *Encarsia hispida* on the whitefly *Aleurotrachelus socialis* on three cassava varieties, CMC 40, CG 489-34 (experiments #1 and #2) and MEcu72.

E. hispida emergence from *A. socialis* pupae was measured on the varieties CG 489-34 and MEcu 72. Results show a low rate of emergence of the parasitoid, 0.43 for CG 489-34 and 0.86 for MEcu 72. These

Table 2. Whitefly (*Aleurotrachelus socialis*) survival, development time and fecundity on four cassava varieties.

Variety	Percentage survival of immatures[a]	Development time/days[a]	Number eggs/female	Tricomes/cm^2 on second leaf
MBra 12	75.0 a	32.18 c	107	4.653
CMC 40	66.0 ab	32.07 c	64	189
CG 489-34	65.5 ab	33.13 b	46	15.290
MEcu 72	27.0 c	34.45 a	15	33.048

a. Means within a column followed by the same letter are not significantly different at the $P = 0.05$ level, using LSD test.

preliminary results indicate that host plant resistance in cassava may affect the development or emergence of parasitoids. Experiments using susceptible varieties (CMC 40) still need to be completed. Since *E. hispida* is the most frequently collected parasitoid in cassava fields, a much higher emergence is expected.

The survival of *E. hispida* when associated with *A. socialis* on the cultivar MEcu 72 decreased rapidly when compared to the other cultivars. One hundred percent mortality occurred within 19 days (Figure 5). Longest survival was on CMC 40, with individuals living for 40 days. Variety CG 489-34 was intermediate; in both experiments with this cultivar, individuals survived until 34 days. These data indicate that there may be varietal influence associated with *E. hispida* survival. Since MEcu 72 and CG 489-34 are resistant varieties, this resistance factor or mechanism may be affecting parasitoid survival.

Host plant resistance and biological control agents increasingly are accepted as complementary pest-control tactics that can reduce the use of chemical pesticide, especially with difficult-to-control pests such as whiteflies. Cassava is most often grown by resource-limited, small-scale farmers in the tropical and sub-tropical regions of the world. HPR and biological control offers a low-cost, practical, long-term solution for

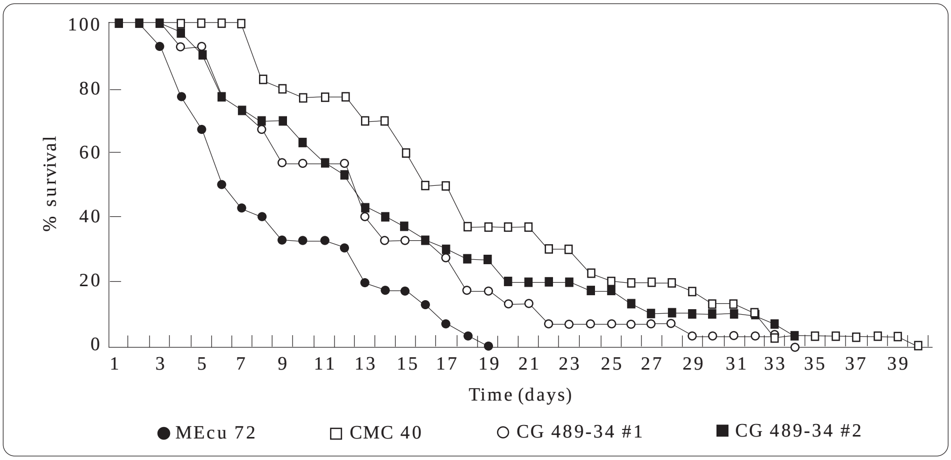


Figure 5. Survival of *Encarsia hispida*, parasitoid of *Aleurotrachelus socialis*, on three varieties of cassava, MEcu 72, CMC 40 and CG 489-34 (experiments #1 and #2).

maintaining lower whitefly populations, reducing crop losses, and limiting or eliminating the need for applications of costly insecticides. This is especially important in crops such as cassava, which has a long growing cycle (1 year or more) that would require numerous pesticide applications to achieve adequate whitefly control. Research on cassava whitefly control at CIAT initially emphasized HPR. More recently, a concentrated effort is being made to identify and use natural enemies in an IPM context (see Chapter 5.2).

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CHAPTER 5.2

Biological Control of Whiteflies by Indigenous Natural Enemies for Major Food Crops in the Neotropics

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Introduction

Whiteflies as direct feeding pests and virus vectors constitute a major problem in cassava (*Manihot esculenta* Crantz) and associated crops in Central and South America and the Caribbean region. There is a large complex in the neotropics, where 11 species are reported on cassava alone (Bellotti et al., 1999). In the northern region of South America (Colombia, Venezuela, and Ecuador) the major species is *Aleurotrachelus socialis* (Bondar) (Castillo, 1996). Two additional species, of lesser importance, but frequently found on cassava, are *Bemisia tuberculata* (Bondar) and *Trialeurodes variabilis* (Quaintance). High populations of *A. socialis*, frequently observed in the region, can cause serious yield reductions in cassava. There is a correlation between duration of attack and yield loss: infestations of 1 month resulted in a 5% yield reduction, of 6 months in a 42% reduction, and of 11 months in a 79% reduction (Vargas and Bellotti, 1981).

Until recently, the *Bemisia tabaci* (Gennadius) biotypes found in the Americas did not feed on cassava. It has been speculated that the absence of African cassava mosaic disease (ACMD) in the Americas may be related to the inability of its vector, *B. tabaci*, to colonize cassava (Costa and Russell, 1975). Since the early 1990s, a new biotype (B) of *B. tabaci*, considered by some to be a separate species (*Bemisia argentifolii* Bellows & Perring) (Perring et al., 1993) has been found feeding on cassava in the neotropics (França et al., 1996; Quintero et al., 1998). More recently, in greenhouse studies done at the Centro Internacional de Agricultura Tropical (CIAT), it has been shown that *B. tabaci* females will oviposit on cassava, and immatures will feed and pupate; however, few adults emerged (CIAT, 1999). It is considered that ACMD now poses a more serious threat to cassava production, because most traditional varieties in the neotropics are highly susceptible to the disease. In addition, the B biotype of *B. tabaci* as a virus vector causes heavy crop losses on numerous other crops in the neotropics and these often are grown in association with cassava, or in the same area. The possibility of virus diseases moving between these crops, or the appearance of previously unrecorded viruses, has become a potential threat.

Host plant resistance and biological control agents (e.g., parasitoids, predators and entomopathogens) increasingly are accepted as a means of pest control that reduces environmental

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contamination and other disadvantages arising from excessive use of chemical pesticides. Many natural enemies are found associated with species of whiteflies on cassava in the neotropics, especially in Colombia and Venezuela (Gerling, 1986; Gold et al., 1989; Castillo, 1996; Evans and Castillo, 1998). Although a large complex of parasitoids have been collected from cassava whiteflies, little is known about the levels of parasitism they impose on the whitefly population, either as individual species or collectively.

For instance, during 1994 and 1995, CIAT personnel carried out surveys for cassava whitefly natural enemies in three regions of Colombia: the Andes, the Atlantic Coast and the Eastern Plains (Castillo, 1996). One hundred sites were visited in 20 departments (states). The three regions differ in altitude, temperature and precipitation, as well as in their cassava cultivation patterns. The Andean region contains many small plantations dispersed over a wide area; in the Atlantic Coast, cassava cultivation is generally in extensive and uniform areas; while in the Eastern Plains, cassava is cultivated in small "patchy" areas near the foot of the easternmost range of the Andes.

The whitefly species collected on cassava during these surveys were *A. socialis*, *B. tuberculata*, *T. variabilis*, *Tetraleurodes* sp. and *Aleuroglandulus malangae* Russell. The latter two species were collected for the first time in very low numbers in the Atlantic Coast and Andean region. *A. socialis* represented 64.5% of the adults and 86% of the immature (eggs, nymphs, and pupae) collected across the three regions. *T. variabilis* was represented by 27.5% of the adults and 11.0% of the immatures, while *B. tuberculata* represented 8.0% of the adults and 3.0% of the immatures (Castillo, 1996).

A. socialis and *B. tuberculata* had a wide distribution, both being collected in almost all sites surveyed. *T. variabilis* was collected from the Atlantic Coast and the Andean region, but not from the Eastern Plains. *A. socialis* populations were highest in the Eastern Plains and lowest in the Andean region. *B. tuberculata* numbers were highest on the Atlantic Coast and Eastern Plains, and almost all *T. variabilis* were collected from the Andean region. The data from this study showed that *A. socialis* and *B. tuberculata* populations decreased with increasing altitude, and were most abundant below 900 m. *T. variabilis* was most abundant at higher elevations, above 1400 m. *A. socialis* populations were highest when temperatures were above 23 °C, while *T. variabilis* populations were higher at lower temperature (<19 °C). *B. tuberculata* were most abundant in those areas with higher temperatures. Annual precipitation did not have a significant effect on distribution of any one of the three species.

A species richness of whitefly parasitoids was collected during these surveys. Ten species were collected from *A. socialis*, *B. tuberculata* and *T. variabilis*. Several of these were unrecorded species (Castillo, 1996; Evans and Castillo, 1998). The parasitoids collected were represented by three genera, *Encarsia* (four species), *Eretmocerus* (four species) and *Amitus* (two species). Three of the *Encarsia* species were *E. hispida* DeSantis, *E. pergandiella* Howard and *E. bellotti*. None of the *Eretmocerus* genus was identified to species and one *Amitus* was identified to species, *A. macgowni* Evans and Castillo. *Eretmocerus* sp. "b", *E. hispida*, *A. macgowni* and *E. bellotti* were collected parasitizing *A. socialis*. *E. bellotti*, *E. hispida*, *E. pergandiella* and *Eretmocerus* sp. "a" were collected from *T. variabilis*. Only *Eretmocerus* sp. "c" was collected parasitizing *B. tuberculata* (Castillo, 1996).

The highest level of parasitism was around 15% measured on *A. socialis* in the Eastern Plains; almost 14% was measured on *B. tuberculata* and 12% on *T. variabilis*, both in the Andean region. *E. hispida* represented the greatest number of individuals collected, 1845 or about 64% of all parasitoids collected. *Eretmocerus* sp. "b" was the second most commonly collected (485 individuals), followed by *A. macgowni* with 159 individuals. *E. hispida* and *Eretmocerus* sp. "b" were collected in all three regions. The greatest complex of species was found in the Andean region, whereas only *E. hispida* and *Eretmocerus* sp. "b" were observed in the Coastal and Eastern Plains regions.

The results of this initial phase of the surveys indicated a rich complex of parasitoids associated with cassava whiteflies, many of which are unrecorded species that could prove useful in biological control programs. It was further realized that an expanded survey would prove beneficial and increase our knowledge of the natural enemy complex associated with whiteflies.

A second phase to these studies was undertaken with the objectives to:

(1) Further define the complex of natural enemies, especially parasitoids, and quantify their association with whitefly species;
(2) Expand these surveys to include other crops in addition to cassava;
(3) Determine which natural enemies regulate whitefly populations by evaluating parasitoid species under controlled conditions (laboratory studies with *E. hispida*, a parasite of *A. socialis*, were initiated); and
(4) Determine if the B. biotype of *B. tabaci* is infesting cassava fields.

Methods

Survey methodology

Three geographic areas were selected for exploration for whitefly species and their parasitoid natural enemies. These were the Atlantic Coast (the Departments of Atlántico, Córdoba, Bolívar and Magdalena), and the mid altitude Central Highlands (Departments of Cauca, Valle del Cauca, Caldas, Quindío and Risaralda) of Colombia. The surveyed Ecuadorian regions were the coastal area and the highlands (Sierra).

The Colombian departments surveyed represented two distinct ecological zones. The Atlantic Coast, which is hot and fairly dry, has temperatures ranging from 27 to 36 °C and an average relative humidity (RH) from 25% to 70%. The Andean region is much cooler, with temperatures ranging from 22 to 33 °C, and RH ranging from 7% to 100%. The Atlantic Coast is also characterized by a 4- to 6-month dry season. Dry periods in the Andean region are usually 2 to 3 months. Altitude was 0 to 200 m for the collection sites on the Atlantic Coast, and 25 to 1750 m in the Andean region.

Several crop hosts, including cassava, common bean (*Phaseolus vulgaris* L.), tomato (*Lycopersicon esculentum* Mill.), eggplant (*Solanum melongena* L.), cotton (*Gossypium hirsutum* L.), cucumber (*Cucumis sativus* L. var. *sativus*) and snap bean (*P. vulgaris*) were sampled. From each collection site, 100 leaves were collected randomly and 1 square inch leaf area was examined to determine the whitefly species present and their respective densities.

The rate of parasitism was determined by collecting 40 leaves randomly from the field and removing a 1-inch square from each leaf sampled. Only one whitefly species was allowed to remain on each leaf square and the emergence of parasitoids is recorded for each whitefly species. This methodology allowed us to accurately determine the parasitoid species associated with each whitefly species. Identifications remain pending for some whitefly and parasitoid species.

Methodology for parasitism studies with Encarsia hispida

Field surveys carried out over several years have shown that the parasitoid *E. hispida* is frequently found parasitizing cassava whiteflies. A colony of this species was established on *A. socialis* on cassava in the greenhouse, and activities were initiated to study *E. hispida* biology and behaviour.

To ensure successful rearing of whitefly parasitoids, a colony of the whitefly A. *socialis* was maintained on cassava (var. CMC-40) in the greenhouse. Potted 5-week old cassava plants were infested weekly by exposing them to *A. socialis* adults for oviposition for 48 hours. The parasitoid colony was initiated by collecting cassava leaves with parasitized *A. socialis* pupae from the field. These were placed in a black plastic box (emergence chambers) with a clear glass bottle attached to an opening in the lid where emerging parasites are drawn to the light and collected. The parasitoids were removed from the bottle with an aspirator and identified.

The desired parasitoid species (*E. hispida*) was collected in sufficient numbers and released into nylon-mesh cages (50 x 50 x 90 cm) in the greenhouse (25 to 32 °C, 60%-80% RH). Each cage contained potted cassava plants infested with second to third instar *A. socialis* nymphs; 16 to 18 days after release, adult parasitoids began to emerge. Once emergence was detected, cassava leaves (now with parasitized pupae) were collected and placed in the "emergence chamber" where parasitoids were collected and species identification confirmed. Collected parasitoids were used to maintain the parasite colony, or utilized in the various studies described in this report.

E. hispida parasitism studies were carried out in the greenhouse (25 to 32 °C; 60%-80% RH), using *A. socialis* as host species. Three methodologies were evaluated to determine parasitism efficiency:

(1) Four nylon-mesh cages (50 x 50 x 90 cm) were used, containing two potted cassava plants infested with whitefly (*A. socialis*) nymphs. Sixty parasitoids were released into each cage with four repetitions for each instar.
(2) Four whitefly-infested cassava plants were used; four infested leaves were placed in a petri dish (100 x 15 mm), and 30 parasitoids were released into each petri dish with 16 repetitions for each instar.
(3) A muslin "bag" was placed over an entire cassava leaf and sealed at the petiole. Each leaf was infested with 80 nymphs, and 20 parasitoids were released into each bag with four repetitions per instar.

Results

Surveys on the Colombian Atlantic Coast and the departments of Cauca and Valle del Cauca resulted in four species of whiteflies being confirmed as feeding on cassava: *Aleurotrachelus socialis*, *Bemisia tuberculata*, *Trialeurodes* sp. (probably *T. variabilis*) and *Tetraleurodes* sp. (Table 1). *A. socialis* was the predominant species

Table 1. Whitefly species and their associated parasitoid complex collected on cassava from three geographical regions of Colombia.

Region	Whitefly species	Parasitoid species
Atlantic Coast	*Aleurotrachelus socialis*	*Encarsia* sp. *Eretmocerus* sp.
	Bemisia tuberculata	*Encarsia* sp. *Eretmocerus* sp. *Metaphycus* sp.
	Trialeurodes sp.	*Encarsia* sp. *Eretmocerus* sp.
	Tetraleurodes sp.	
Valle del Cauca	*A. socialis*	*Encarsia* sp. *Eretmocerus* sp.
	B. tuberculata	
Cauca	*A. socialis*	*Encarsia bellotti* *Eretmocerus* sp. *Signiphora aleyrodis*
	B. tuberculata	*Encarsia pergandiella* *Eretmocerus* sp. *Euderomphale* sp. *Signiphora aleyrodis*
	Trialeurodes sp.	*Encarsia hispida* *Encarsia pergandiella* *Eretmocerus* sp.

found in all three areas; however, its population was considerably higher in Valle del Cauca and lowest in Cauca (Figure 1). All four species were collected from the Atlantic Coast, whereas *Tetraleurodes* sp. was not collected in either Cauca or Valle del Cauca.

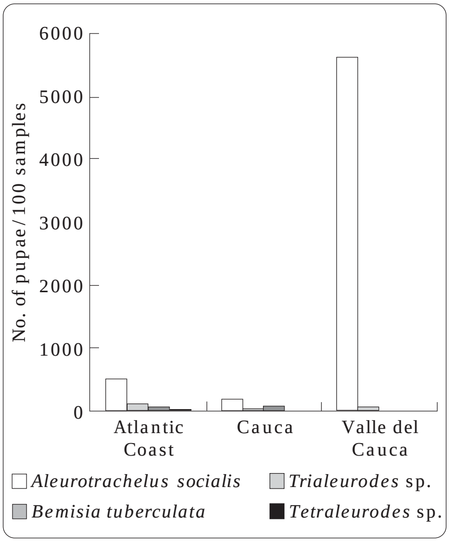

Figure 1. Whitefly population densities on cassava in three geographic zones of Colombia.

Pupal populations of *A. socialis* ranged from an average of about 55 per square inch on leaf samples from Valle del Cauca to about 5 on the Atlantic Coast and 2 in Cauca (Figure 1). The data also showed that whenever there are high whitefly densities or populations, *A. socialis* is the predominant species regardless of geographic area. However, when densities are low at sampling sites, other species may predominate (Figure 2). On the Atlantic Coast, under low population densities, *B. tuberculata* and *A. socialis* had similar populations, whereas in Valle del Cauca, *B. tuberculata* predominated. In Cauca, *Trialeurodes* had the highest population.

Numerous whitefly parasitoids were collected from the three areas (Table 1). These parasitoids belong to the genera *Encarsia*, *Eretmocerus*, *Metaphycus* and *Euderomphale*, and indications are that the collection contains several unrecorded species. The hyperparasitoid

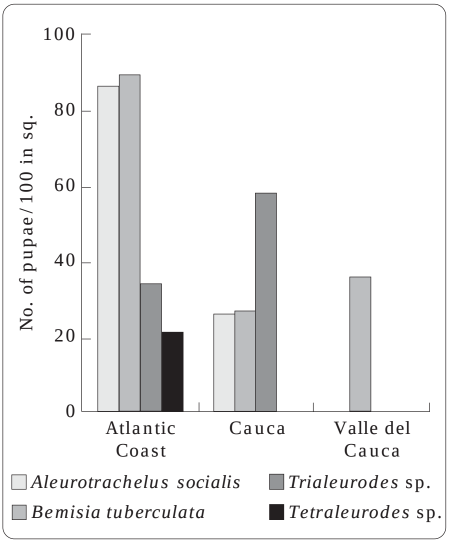

Figure 2. The whitefly species complex at collecting sites with low population densities in three geographic zones of Colombia.

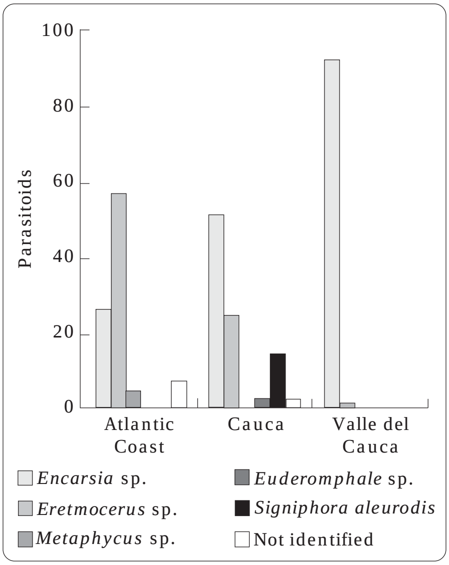

Figure 3. Parasitoid species (%) collected from whiteflies on cassava in three geographic zones of Colombia.

Signiphora aleyrodis was collected only in Cauca. *Encarsia* was the genera most frequently collected in Cauca and Valle del Cauca (Figure 3), with the highest percentage in the latter. On the Atlantic Coast, *Eretmocerus* was the most frequent genus. *Metaphycus* was only collected on the Atlantic Coast and *Euderomphale* only from Valle del Cauca.

Levels of infestations were high for *Aleurodicus* sp., *A. socialis*, *B. tuberculata* and *Tetraleurodes* sp. for the coastal region of Ecuador. *T. vaporariorum*, which is found infesting common bean, was found for the first time at high infestation levels in cassava in the coastal and highlands regions of Ecuador.

The association between whitefly species and parasitoid species was evaluated also. Results indicate a sizable parasitoid species complex associated with each whitefly species and that this can be influenced by geographic area. For example, with *A. socialis*, the predominant parasitoid genus in the Atlantic Coast was *Eretmocerus*, while in Cauca and Valle del Cauca, *Encarsia* predominated (Figure 4). In Valle del Cauca, 99.6% of the parasitism of *A. socialis* was by *Encarsia* and 0.4% by *Eretmocerus*. The most numerous complex of parasitoids was found associated with *B. tuberculata* (Table 1). *Eretmocerus* and *Metaphycus* were the predominant genera on the Atlantic Coast and *Eretmocerus* in Cauca. The hyperparasite *S. aleyrodis* was collected from both *A. socialis* and *B. tuberculata* pupae.

Four species of parasitoids were collected from Ecuador. The complex of parasitoids collected from whiteflies in cassava in Ecuador has been identified only to the generic level (Greg Evans, Mike Rose, personal communication, 1999). In the coastal region, *A. socialis* was parasitized by *Encarsia*, followed by *Amitus* and *Eretmocerus*; *Aleurodicus* was parasitized by *Euderomphale* sp. *Tetraleurodes* and *Trialeurodes* were mostly parasitized by *Eretmocerus* sp.

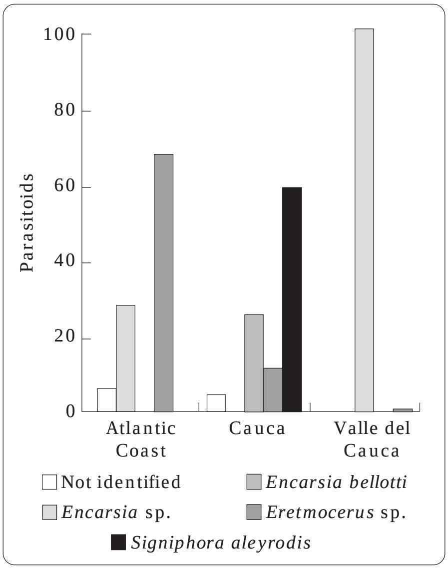

Figure 4. Parasitoid species (%) collected from the whitefly *Aleurotrachelus socialis* in three geographic zones of Colombia.

and *B. tuberculata* was parasitized by *Encarsia* sp. and *Euderomphale*. In the highland region, *T. vaporariorum* was the only whitefly species collected, with about 16 pupae per 2.54 cm^2. The dominant parasitoids were *Encarsia* spp., representing 98% of the sample.

In Colombia, *Trialeurodes* sp. was most frequently parasitized by *Encarsia*. Two species of *Encarsia* were identified, *E. hispida* and *E. pergandiella*, and a third species remains to be identified.

Encarsia sp. and *Eretmocerus* spp. were frequently collected from both high and low whitefly populations. *Metaphycus* sp. and *Euderomphale* sp. are observed only when whitefly populations are low. Parasitoids of the genera *Encarsia* and *Eretmocerus* were collected from sea level to about 2400 m. *E. pergandiella* was collected in high populations at all levels of whitefly infestation.

The whitefly species complex associated with cotton and legume and vegetable crops was distinct from that described on cassava. Two whitefly species were collected, *Bemisia tabaci* and *Trialeurodes vaporariorum*. During the period of field collections (January to June 1999), *B. tabaci* was the only species collected on the Atlantic Coast (departments of Atlántico and Córdoba), while in the Andean region (departments of Caldas, Quindío, Risaralda and Valle del Cauca), *T. vaporariorum* was the only species collected, with its highest populations observed in Valle del Cauca and Caldas.

B. tabaci was collected from cotton, eggplant and tomato, while *T. vaporariorum* was collected from common bean, snap bean, cucumber and tomato. Populations of *T. vaporariorum* in general were much higher than *B. tabaci* and this was especially the case on common bean and snap bean (Figure 5). These results indicate that *B. tabaci* may be adapted more to the warmer temperatures of the coastal region, whereas *T. vaporariorum* prefers the cooler temperatures of the Andean region.

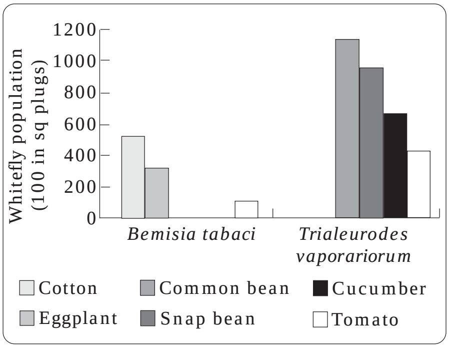

Figure 5. Whitefly nymph pupae populations on several crops in the departments of Atlántico, Córdoba, Caldas, Quindío, Risaralda and Valle del Cauca, Colombia.

Parasitoids from four genera, *Encarsia* and *Eretmocerus* (Aphelinidae), *Amitus* (Platygastridae) and *Metaphycus* (Encyrtidae) were collected. These collections have been sent to taxonomists and species identifications are pending. Therefore, in this report, they are referred to only by genera.

Encarsia was collected in all the departments except Risaralda (where only two sites were surveyed) but *Amitus* presented the highest populations, especially in Valle del Cauca and Caldas (Figure 6). This corresponded to the two departments with the highest whitefly populations. *Eretmocerus* was the predominant species in Córdoba. Parasitoid populations were highest in Valle del Cauca and lowest in Risaralda and Quindío. Only one specimen of *Metaphycus* was collected across the sites.

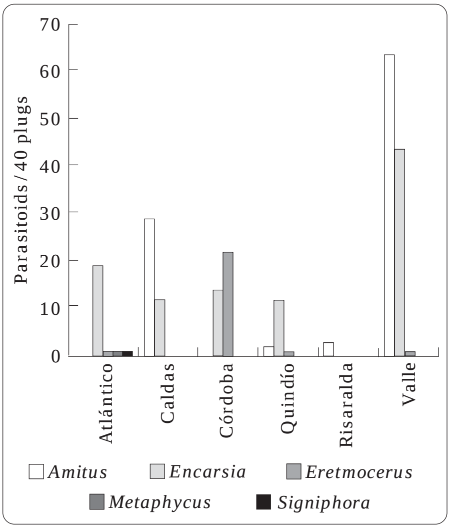

Figure 6. Whitefly parasitoids collected from several crops in the departments of Atlántico, Córdoba, Caldas, Quindío, Risaralda and Valle del Cauca, Colombia.

Encarsia parasites were collected from all the plant hosts sampled and had the overall highest densities among all collected parasitoids (Figure 7). *Amitus*, although not collected from all hosts, also presented high populations, especially on common bean and snap bean. *Eretmocerus* was associated almost exclusively with cotton (only one individual was collected from tomato).

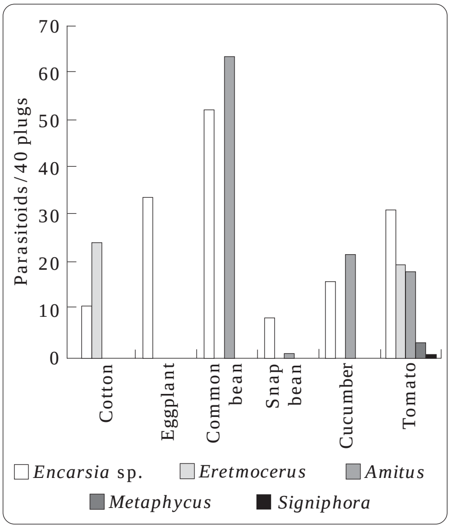

Figure 7. Whitefly parasitoid frequency collected from several crops in the departments of Atlántico, Córdoba, Caldas, Quindío, Risaralda and Valle del Cauca, Colombia.

Encarsia and *Eretmocerus* were collected from both whitefly species (Figure 8), while *Amitus* was collected only on *T. vaporariorum* and *Metaphycus* and *Signiphora* were collected only from *B. tabaci*.

During surveys, data on pesticide applications were recorded during farmer interviews. In general, parasitoid populations were higher in fields where pesticides were not applied, regardless of the whitefly species. Results show that populations of *Encarsia* are less affected by pesticide applications than populations

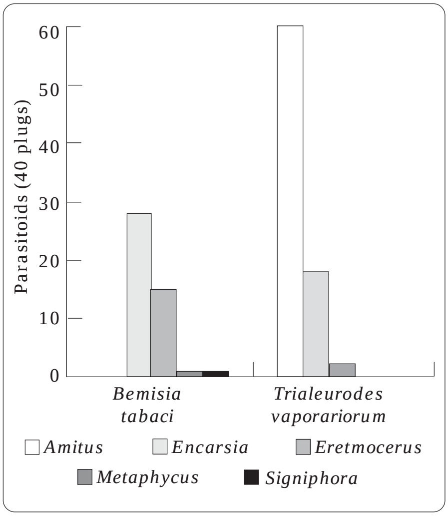

Figure 8. Relationship between parasitoid complex and whitefly species collected from several crops in the departments of Atlántico, Córdoba, Caldas, Quindío, Risaralda and Valle del Cauca, Colombia.

of *Amitus*, indicating that *Encarsia* may have acquired a degree of resistance to some of the pesticides applied.

***Parasitism studies with* E. hispida**

Of the three methodologies evaluated, the highest rate of parasitism was obtained using the third method. Parasitism levels using the first two methods were low. The highest rate of parasitism with method 1, the use of nylon-mesh cages, only reached 9% in the third instar. This improved with the second method (the use of leaves enclosed in petri dishes) to 30% in the 2nd instar and to 20% in the 3rd.

The employment of muslin bags, as described in the third methodology, gave the best results. In the first of two experiments using this methodology, parasitism rates reached about 75% in the third instar and 16% in the 1st instar, 45% in the 2nd instar and 43% in the 4th instar (Figure 9). Results of a second experiment were similar, with about 75% parasitism in the third instar and 19% in the 1st instar, 61% in the 2nd instar and 25% in the 4th instar (Figure 9). The average parasitism rate for these two experiments were about 45%, whereas average parasitism rates were only 6% using methodology 1 and were 20% using methodology 2.

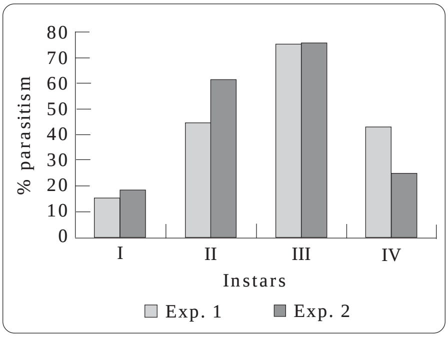

Figure 9. Cassava whitefly (*Aleurotrachelus socialis*) instar preference by the parasitoid *Encarsia hispida* in two greenhouse experiments.

These results also show that the third whitefly instar is preferred for parasitism by *E. hispida*. An average of all four experiments resulted in a parasitism rate of 21% for the 1st instar, 35% for the 2nd instar, 46% for the 3rd instar and 22% for the 4th instar. However, the average of the two experiments using methodology 3 (muslin bags) is 17% for the 1st instar, 53% for the 2nd, 75% for the 3rd and 34% for the 4th instar. The highest parasitism rate is in the 3rd instar, followed by the 2nd, 4th and 1st instar.

The time period for optimal parasitoid activity was evaluated using the third methodology described. Percent parasitism evaluations were made at 48, 72, 96 and 216 hours after parasitoid release. Peak parasitism activity occurred between 72 (35% parasitism) and 96 hours (33% parasitism) (Figure 10). However, even at 216 hours, parasitism rates still remained relatively high (nearly 29%) indicating a relatively lengthy parasitoid activity and the need to do further evaluations at time periods between 96 and 216 hours.

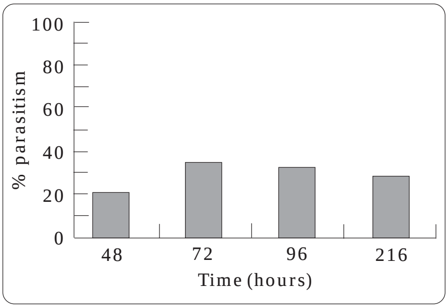

Figure 10. Parasitoid (*Encarsia hispida*) activity in relation to time of exposure to whitefly host *Aleurotrachelus socialis* on cassava in greenhouse studies.

The high rates of parasitism, especially using the muslin cage methodology, indicate that *E. hispida* could be an effective parasite in a biological control program for *A. socialis*. However, whitefly populations, especially of *A. socialis*, have been extremely high at CIAT for the past 4 or 5 years. During this period, *E. hispida* is the most frequently observed parasite. These previously described experiments were carried out under controlled conditions where the parasitoid had easy access to the whitefly prey and the parasitoid probably is not adversely influenced by environmental factors. Under natural field conditions, parasitoid activity may not be as efficient as indicated by these greenhouse studies.

Observations of pupal populations in the field at CIAT and other selected sites (i.e., Tolima) have shown that a high percentage of pupae are parasitized. However, whitefly populations remain high and cause considerable plant damage and yield loss, indicating that the actual activity of parasitoids is not sufficient to reduce whitefly populations below economic injury levels that reduce cassava yields.

Discussion

In future studies, additional parasites such as *Eretmocerus* and *Amitus* species will be evaluated in similar studies to those with *E. hispida*. Continued survey activities to identify additional natural enemies also are underway. During this phase of exploration, more emphasis will be given to regions or fields where there are low whitefly populations with the hope of identifying key parasitoids, predators or entomopathogens that are preventing eruptions of whitefly populations.

This is a collaborative project between CIAT and the University of Florida, and is funded by the United States Agency for International Development (USAID). This collaboration will provide training and improved in-country capacity for research, production, delivery and management of biological control agents. The University of Florida will provide expertise and input on parasitoid taxonomy, biology, behaviour, collecting, rearing, identification and data analysis. Since 1998, University of Florida researchers (Dr. Jorge Peña and R. Nguyen) have visited CIAT and collaborated in training CIAT personnel in some of the above areas. Whitefly parasitoids that are collected from sampling are sent to University of Florida taxonomists (G. Evans, M. Rose and A. Hammonds) for identification.

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CHAPTER 5.3

Progress of Cassava Mosaic Disease in Ugandan Cassava Varieties and in Varietal Mixtures

*William Sserubombwe**, *Michael Thresh***,
*James Legg**** and *William Otim-Nape**

A feature of the production of cassava (*Manihot esculenta* Crantz) in Uganda, as in many other countries of sub-Saharan Africa, is that many different varieties are grown and it is usual to find several varieties within individual plantings. Moreover, cassava is usually grown together with one or more intercrops, including legumes and cereals. The great diversity in the varieties grown and in the cropping systems adopted can be expected to influence the incidence and severity of cassava pests and diseases. However, this possibility has received only limited attention in relation to cassava mosaic disease (CMD), despite indications that the current pandemic in Uganda and in adjacent parts of north-west Tanzania and western Kenya is closely associated with the varieties adopted and is least damaging in areas where many varieties are grown.

The cassava component of the Natural Resources Institute (NRI) contribution to the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project has considered the role of varietal diversity in relation to CMD under epidemic conditions in Uganda. It is proposed to consider further aspects of varietal diversity and the implications of intercropping in any additional phase of the project.

Response of Local Varieties Aladu and Bao to Cassava Mosaic Disease

Ugandan farmers selected the varieties Aladu and Bao and originally grew them mainly in Apac District, where they appeared to have at least some degree of resistance to CMD. For this reason, and in the absence of more resistant varieties, they were introduced to Kumi, Soroti and other districts of Uganda in the early 1990s in attempts to rehabilitate cassava production following the devastating epidemic of CMD.

This approach was partially successful and farmers have retained both Bao and Aladu in some areas, although they are less resistant than some of the other improved varieties now available. Bao became heavily infected under the epidemic conditions encountered originally. However, as the epidemic abated and the symptoms became less severe, some farmers have attempted to return to Bao because of

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its attractive eating qualities and other desirable attributes.

Replicated plots of Bao and Aladu were established in May 1998 and recorded monthly to assess the incidence and severity of CMD. Comparable plots of the highly resistant SS4 also were established as a standard. All plants of Bao and Aladu were harvested individually in June 1999 and the data for individual plants (each of which had a full set of four neighbours) were used for analysis.

There was rapid spread of CMD to Bao and all plants were affected within 6 months after planting (MAP) (Figure 1). CMD spread less rapidly in Aladu and 15% of the plants were unaffected at the final observation 12 MAP. On average, symptoms developed in Aladu 5.1 MAP compared with 3.7 MAP for Bao. Few plants of SS4 were affected and the symptoms were less conspicuous than those of Bao and Aladu. Yields of tuberous roots were related to the stage of growth when symptoms were first expressed and Bao was more severely affected than Aladu (Figure 2). Little yield was obtained from plants of either variety that were grown from infected cuttings. Plants infected by whiteflies were affected less severely, especially those that developed symptoms at a late stage of growth.

The incidence and yield data were used to calculate the losses caused by CMD in relation to fully healthy stands and assuming that all plants were grown from healthy cuttings. The overall loss was 75% for Bao compared with 31% for Aladu. This confirms the vulnerability of Bao and the relative tolerance of Aladu, as farmers reported earlier. Moreover, sufficient symptomless plants of Aladu remained to provide cuttings for a further planting. This suggests that Aladu could be sustained even under epidemic conditions, provided that farmers select healthy cuttings at each cycle of propagation. Additional studies of this type are justified on the sustainability of a wider range of local varieties and under less extreme conditions of inoculum pressure than those encountered at Namulonge between 1998 and 1999.

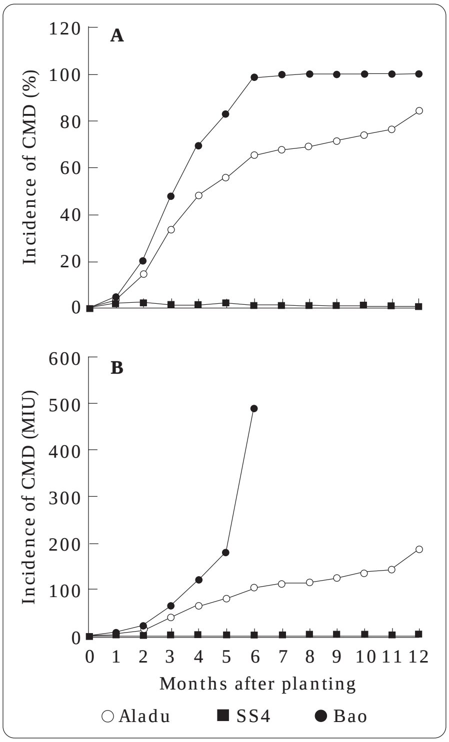

Figure 1. Mean monthly incidence of cassava mosaic disease (CMD) in local cassava varieties SS4, Aladu and Bao at Namulonge, 1998-99 experiment. Incidence data (A) as percentages and (B) transformed to multiple infection units (MIU).

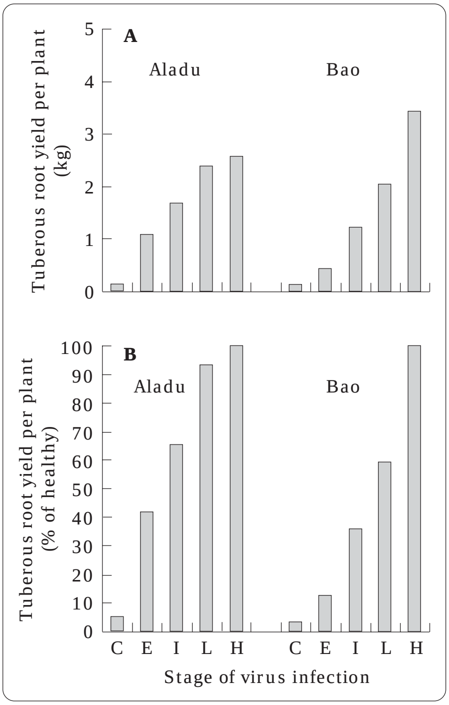

Figure 2. Relationship between time of first symptom expression of cassava mosaic disease and tuberous root yield per plant in local cassava varieties Aladu (left) and Bao (right). Data presented as (A) mean yield per plant and (B) yields expressed as percentage of healthy controls. Data for 10 plants each having a full set of four immediate neighbours. C = infected as cutting, E = early, I = intermediate, L = late infected and H = healthy (symptomless) plants. (The healthy Bao were nominal controls sampled in an adjacent plot in the absence of uninfected plants.)

Ability of Ugandan Varieties to Withstand Effects of Cassava Mosaic Disease

Experience over the last decade has shown that farmers in Uganda have overcome the effects of the CMD epidemic by adopting resistant varieties introduced from Nigeria or those selected by the National Agricultural Research Organisation (NARO) and by selecting from the local varieties already available within the country. Much information has been obtained on the performance of the resistant material that has been released to farmers through official rehabilitation schemes but little information is available on the features of the local varieties that have been adopted widely following the epidemic. Accordingly, a study has been made of the 18 local varieties listed in Table 1. They were collected from different parts of Uganda and selected for their apparent ability to withstand the most damaging effects of CMD.

Wherever possible, cuttings were collected from symptomless plants of each variety selected for study and from plants expressing mild and severe symptoms. The cuttings were planted at Namulonge in May 1998, together with the varieties Ebwanateraka, Bao, Migyera and SS4, which were included as standards. All plants were assessed monthly to record the incidence and severity of CMD. Vegetative growth and populations of the whitefly vector *Bemisia tabaci* (Gennadius) were also assessed and the plants were rated for overall vigour 12 MAP.

Populations of adult whitefly were unusually high throughout the period of the experiment and there was rapid spread of CMD both within the trial and in adjacent experiments. All the initially healthy plants of the local varieties became infected but there was relatively little spread to the resistant standards. There was wide variation in symptom severity depending on the variety and the initial health status of the planting material. Symptoms were generally highly conspicuous except in the local varieties Buhimba, Njule and Tongolo and in the resistant standards SS4 and Migyera. Nevertheless, many of the local varieties grew vigorously, especially when grown from cuttings collected from symptomless or mildly affected plants.

Table 1. Reaction of different local cassava varieties in Uganda to cassava mosaic disease (CMD) infection at Namulonge in the 1998-99 experiment. Data are for plants that were raised from cuttings collected from symptom-less (healthy) plants and from those with mild (category 2-3) and severe (category 4-5) symptoms.

Variety	Origin (District)	Incidence[a]		Category[b]	Initial CMD status[c]			Vigour index[d]		
		(%)	MIU		Healthy	Mild	Severe	Healthy	Mild	Severe
Buhimba	Hoima	34	41	MR	2.0	3.2	3.0	4.3	2.8	-
Katisa-Bakonjo	Mubende	85	190	HS	3.2	4.3	-	4.2	2.2	-
Kayumba	Mukono	68	114	S	2.7	4.5	5.0	-	2.7	1.1
Mpologoma	Mukono	-	-	-	-	4.8	4.9	-	1.9	1.3
Njule-sl	Mukono	35	43	MR	3.6	3.8	4.8	2.8	2.6	2.5
Unknown 2	Mukono	65	105	S	4.6	4.9	5.0	-	1.9	1.1
Kayinja	Mukono	65	104	S	3.7	4.1	4.9	2.8	2.6	1.7
Bao	Soroti	64	101	S	4.0	4.0	4.4	2.0	1.3	1.2
Rugogoma	Kibaale	60	92	S	4.0	4.8	4.8	3.2	1.4	2.0
SS4	Mpigi	0	-	HR	1.0	2.6	-	2.2	1.9	-
Luzira	Mpigi	72	129	-	4.0	4.9	4.5	1.8	1.2	1.0
Ebwanateraka	Mpigi	89	223	HS	4.8	5.0	5.0	1.0	1.0	1.0
Migyera	Mpigi	13	13	HR	2.6	3.6	3.2	3.8	3.4	2.6
Aladu-aladu	Soroti	55	79	I	3.8	4.6	5.0	2.1	1.2	1.1
Muwogo-omweru	Mukono	70	120	S	3.1	4.3	5.0	3.9	2.1	1.1
Matooke	Mukono	78	150	S	3.7	4.1	5.0	2.1	1.9	1.2
Tongolo	Masindi	51	72	I	3.0	3.7	5.0	2.0	1.7	1.0
Unknown 1	Mukono	68	112	S	3.7	4.8	4.9	2.7	2.0	1.3
Unknown 3	Mukono	48	64	I	3.4	4.4	4.8	2.7	2.6	2.4
Nyaraboke	Masindi	69	116	S	3.4	3.6	4.2	2.0	2.3	2.3

a. Incidence data 3 months after planting as % and as transformed to multiple infection units (MIU).
b. Variety categorization based on incidence 3 months after planting: 0%-20%, highly resistant (HR); 21%-40%, moderately resistant (MR); 41%-60%, intermediate (I); 61%-80%, susceptible (S); 80%-100%, highly susceptible (HS).
c. Symptoms rated on a 1 to 5 scale of increasing severity in relation to status of source plants from which cuttings were collected.
d. Vigour was rated on a scale of 1 (least vigorous) to 5 (most vigorous), 12 months after planting.

Samples were collected from representative plants of all varieties for polymerase chain reaction (PCR) and enzyme-linked immunosorbent assay (ELISA) analysis to determine the cassava mosaic virus(es) present and their concentration and association with the symptoms expressed. Preliminary results showed that the epidemic-associated virus (now designated *East African cassava mosaic virus*-Uganda [EACMV-Ug]) and *African cassava mosaic virus* (ACMV) occurred alone or as a mixture. The few symptomless plants that reacted positively contained mixed infections and so provided evidence of virus latency.

These initial studies confirm the ability of some local varieties to partially withstand infection. They also indicate the scope for further studies on the behaviour of local varieties and on their sustainability, especially under the less extreme conditions of inoculum pressure encountered in post-epidemic situations.

Spread of Cassava Mosaic Disease and Whitefly Vector Populations on Varieties Grown Singly and as a Mixture

Detailed analyses have been done of field data collected during an earlier M.Sc. project funded by the Rockefeller Foundation. This project assessed the spread of CMD and whitefly vector populations in four contrasting varieties grown singly and as a mixture under epidemic conditions at Namulonge in 1995-96 and 1996-97. There was much spread of CMD to the susceptible variety Ebwanateraka, little to the resistant SS4 and intermediate levels in TMS 30337 (Nase 2) and TMS 30572 (Nase 3, also known as Migyera) (Figure 3). The amount of spread to the resistant and partially resistant varieties was similar in the sole plots and in the mixture. In contrast, there was less spread to the susceptible Ebwanateraka in the mixture than when it was grown alone. The effect was significant and is likely to have been even greater under lower levels of inoculum pressure than those experienced in the two experiments. This suggests that resistant varieties may be used to provide at least some degree of protection to susceptible ones. Indeed, the effect could be of considerable practical importance because farmers may be reluctant to discard susceptible varieties that are valued because of quality or other favourable attributes.

A new varietal mixtures experiment was planted at Namulonge in September in 1998. There were four treatments each replicated four times in a complete randomized block design:

(1) Variety Bao (susceptible): alone.
(2) Variety SS4 (resistant): alone.
(3) Bao and SS4 planted alternately in 50:50 mixture.
(4) SS4 (50%) grown as an outside barrier around Bao (50%).

Monthly assessments were made of adult whitefly populations, CMD incidence and CMD severity. The assessment of whiteflies was done so that any edge or barrier effects would be detected.

The health status of the Bao planting material was unsatisfactory and there was substantial infection arising from the cuttings used. This reflects the difficulty now being experienced in Uganda in obtaining healthy material of susceptible varieties. Nevertheless, preliminary analyses show big differences between SS4 and Bao in the incidence and severity of CMD (Figure 4). There was also evidence that the incidence of CMD in Bao was less in the mixture than in the pure stand. However, there was no evidence of a barrier or edge effect using SS4 and the comparison may have been vitiated by the extent of cutting infection with Bao. A further difficulty is that Bao is a tall erect variety that outgrows SS4, which is a relatively low spreading type. This suggests that varieties of similar habit and conformation should be selected for any further evaluation of mixtures.

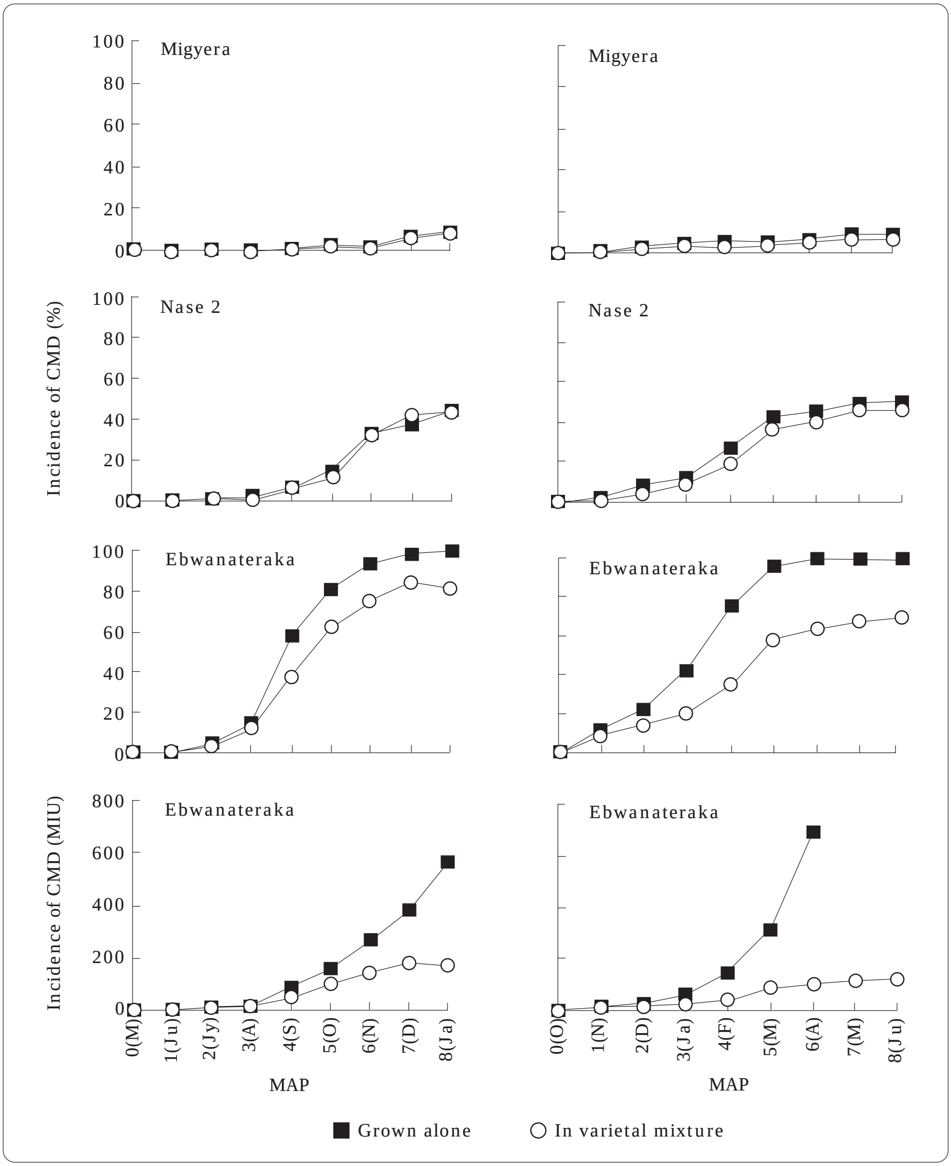

Figure 3. Mean monthly incidence of cassava mosaic disease (CMD, %) in successive months after planting in varieties Migyera, Nase 2 and Ebwanateraka when grown alone and as a mixture at Namulonge in (left) the 1995-96 and (right) the 1996-97 experiments. Incidence data for Ebwanateraka were transformed to multiple infection units (MIU) in the bottom two figures.

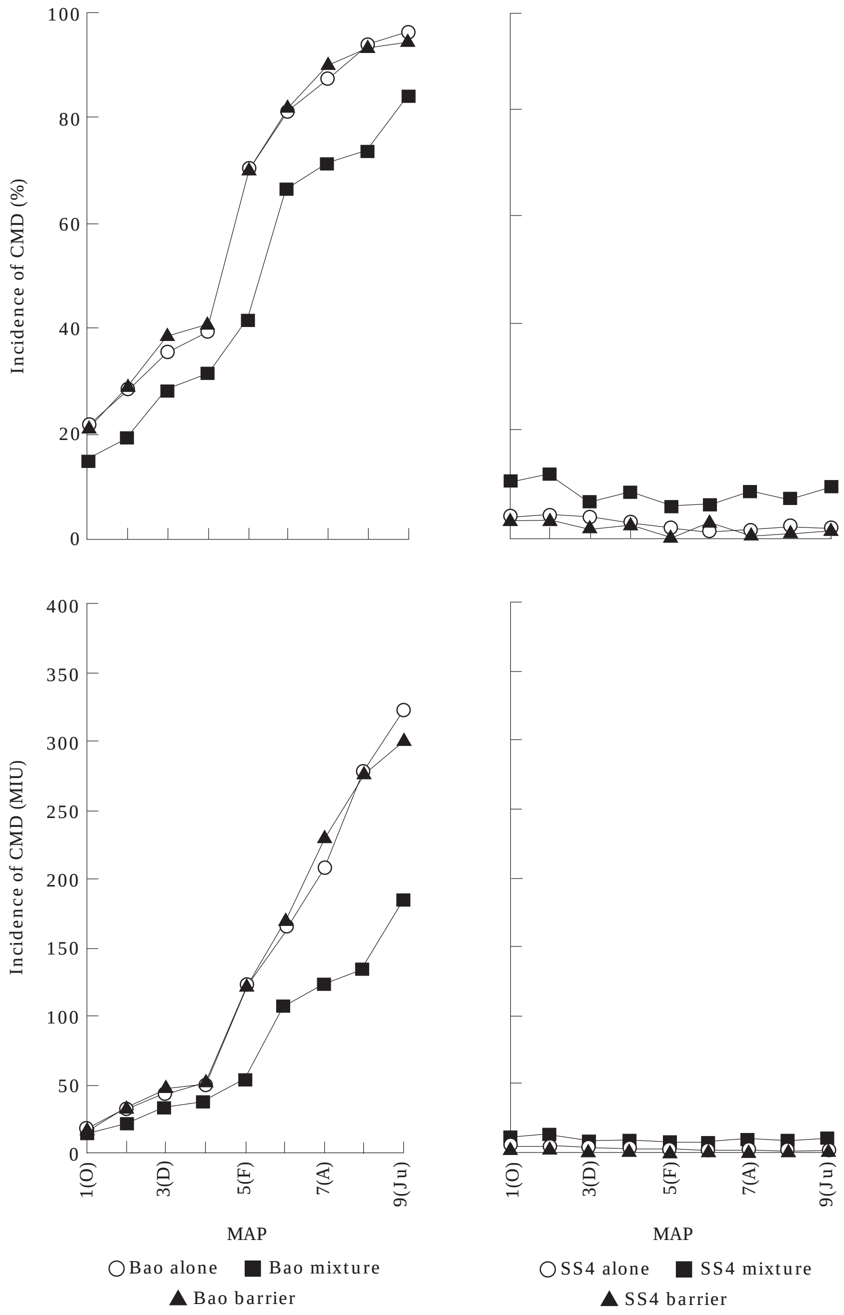

Figure 4. Mean monthly incidence of cassava mosaic disease (CMD, %) in successive months after planting (MAP) in varieties SS4 and Bao when grown alone, as a mixture and with SS4 as a barrier around Bao at Namulonge in the 1998-99 experiment. Incidence expressed as % (top) and as transformed to multiple infection units (MIU) (bottom).

The experiment was harvested 12 MAP to consider the yields of healthy and infected plants of each variety in relation to the type and healthy status of their immediate neighbours. It is particularly important to assess the ability of SS4 to compensate for the impaired growth of Bao in the mixture of the two varieties.

Conclusions

The results obtained are consistent with previous observations on the behaviour of local varieties and on the scope for adopting somewhat tolerant varieties and varietal mixtures as a means of alleviating the effects of CMD (Sserubombwe et al., 2001). Further studies on the behaviour and sustainability of local varieties, especially under the less extreme conditions of inoculum pressure encountered in post-epidemic situations, need to be made.

Acknowledgments

This study was carried out in collaboration with the NARO Cassava Programme, Uganda. The study will continue and results will be analysed and written up with short-term funds provided by the Crop Protection Programme of the UK Department for International Development (DFID).

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CHAPTER 5.4

Management of the Cassava Mosaic Disease Pandemic in East Africa

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Introduction

During the 1990s, a pandemic of an unusually severe form of cassava mosaic disease (CMD) expanded to cover a large part of East Africa, including virtually the whole of Uganda and parts of western Kenya, southern Sudan, north-western Tanzania and eastern Democratic Republic of Congo (DRC) (Otim-Nape et al., 1997; ASARECA, 1998; Legg et al., 1999b). This has been associated with the occurrence of a novel and highly virulent cassava mosaic begomovirus (Deng et al., 1997; Harrison et al., 1997; Zhou et al., 1997). Surveys to assess the prevalence and severity of CMD were conducted in Uganda, Kenya and Tanzania as part of the diagnostic phase of the Tropical Whitefly Integrated Pest Management (TWF-IPM) Project (Chapters 1.6, 1.7 and 1.8, this volume). A principal outcome of these surveys was the identification of regions that were either currently affected or threatened by pandemic expansion, based on epidemiological data (Legg et al., 1999b).

In view of the acute effects of the severe CMD associated with the pandemic, the co-ordination team of the TWF-IPM Project considered that there was an immediate need to identify sources of funding to support CMD control activities in recently affected/threatened areas. Dialogue was initiated with staff of the United States Agency for International Development (USAID) by the Project Co-ordinator and subsequently followed up in Uganda by the co-ordinator of the Africa-based Sub-Project 4. A concept note was submitted to the Office for Foreign Disaster Assistance (OFDA) of USAID in April 1998 and funding for a 1-year project was approved the following month. The project, entitled “Emergency programme to combat the cassava mosaic disease pandemic in East Africa” began in October 1998 and this first phase was completed in September 1999.

Background to Control of the CMD Pandemic

After a decade of major losses to cassava production in Uganda due to the CMD pandemic (Otim-Nape et al., 1997), substantial experience has been gained on control strategies. CMD-

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resistant cassava germplasm introduced from the International Institute of Tropical Agriculture (IITA) in the early 1980s was evaluated in multi-locational and on-farm trials during the early 1990s and, in 1993, three cultivars (TMS 60142, TMS 30337 and TMS 30572) were officially released under the names Nase 1, Nase 2 and Nase 3 (or Migyera). In addition to being resistant to infection, they were tolerant also of the effects of cassava mosaic begomoviruses (CMBs) once infected (Thresh et al., 1994). Multiplication efforts were initiated during the mid-1990s (Otim-Nape et al., 1994; GCF, 1997) and areas initially targeted included some of the first areas to be affected by the pandemic in northern and eastern Uganda (Otim-Nape et al., 1994).

In more recent years, as the pandemic has expanded southwards, the emphasis of control efforts has shifted to central and southern districts. At the end of 1997, a major project funded by the PL 480 programme of USAID and entitled “Dissemination and utilization of mosaic resistant cassava in Uganda” was initiated in the central-southern target districts of Masindi, Luwero, Mukono, Kamuli and Iganga. Principal CMD management themes within this programme were monitoring and diagnostics, plant health management within the multiplication scheme and participatory evaluation of new CMD-resistant germplasm. The project targeted districts that had been affected by the CMD pandemic for more than 1 year and therefore already were experiencing significant shortages of planting material. This project did not address districts affected more recently, in 1996 and 1997, as identified by the TWF-IPM Project diagnostic surveys.

Pandemic control in western Kenya and open quarantine

The expansion of the pandemic into western Kenya was first noted in 1995 (Gibson, 1996; Legg et al., 1999b) although it was not until 2 years later as the increasing seriousness of the problem was recognized that initiatives began to control it. The key constraint to CMD management in western Kenya was the virtual absence of any CMD resistant germplasm in the country. In order to address this problem, scientists of the East African Root Crops Research Network (EARRNET) worked with plant quarantine officials of both Uganda and Kenya to develop guidelines for the establishment of an “open quarantine” facility. This was sited at Busia, on the Kenyan side of the Kenya/Uganda border, and allowed the introduction of stem cuttings of two CMD resistant cultivars, SS4 and TMS 30572 (Nase 3), in addition to a wide range of selected clones from the EARRNET germplasm development programme, based at Serere, Uganda. It was also at this time that the pandemic control programme in western Kenya received its first major offer of financial support, from the Gatsby Charitable Foundation (GCF) in the UK. EARRNET co-ordinated the development of a project, funded by GCF, which provided for the introduction, evaluation and multiplication of CMD-resistant cultivars in western Kenya and was initially targeted to run for 3 years.

Regional pandemic control and the OFDA CMD Project

Whilst USAID and Gatsby-funded initiatives were providing substantial support for the multiplication and dissemination of CMD resistant cultivars in Uganda and Kenya, it was clear that the increasing regionalization of the CMD pandemic needed to be addressed by taking a more pro-active and co-ordinated approach. It was envisaged that such an approach would involve establishing links between control efforts in each of the East African countries affected or about to be affected by the pandemic, and targeting monitoring and control initiatives towards "threatened" zones in addition to areas already affected. This was the rationale behind the development of the OFDA-funded project, which began in October 1998.

The purpose of the OFDA CMD Project was to "boost production of cassava in Uganda, Kenya and Tanzania and enhance both short and longer term food security, through the implementation of an emergency programme to multiply and disseminate mosaic resistant cassava". Principal partners included IITA-Eastern and Southern Africa Regional Center (ESARC), the two regional root crops networks of EARRNET and the Southern African Root Crops Research Network (SARRNET) and national root crops programmes in Kenya, Tanzania and Uganda. Principal project activities were organized under five themes:

(1) Monitoring and diagnostics: conduct focused surveys in south-western Uganda, western Kenya and north-western Tanzania to provide detailed distribution maps of CMGs in the project target areas and baseline data for subsequent impact analysis.
(2) Multiplication: multiply and distribute in collaboration with project partners, elite CMD-resistant materials.
(3) Germplasm diversification: increase the range of cassava materials available to farmers in areas targeted by the project, thereby reducing future risk of production collapse.
(4) Stakeholder linkages: identify and strengthen links between key stakeholders with roles in enhancing cassava production in target areas.
(5) Farmer training: develop producer skills in identification and management of cassava pests and diseases with special focus on CMD, in addition to basic production and multiplication skills.

Implementation Highlights—The OFDA CMD Project

Significant progress was made in the attainment of project targets in each of the main themes and the project has been successful in fostering a more comprehensive regional approach to tackling the CMD problem.

Monitoring and diagnostics

An important feature of the project has been the use of regular monitoring and diagnostic surveys to assess the status of CMD in threatened zones and develop forecasts for the likely pattern of development of the pandemic. Surveys focused on the Kagera region in north-west Tanzania and Western and Nyanza Provinces in western Kenya. During the first quarter of the project, the first report was made of severe CMD in Kagera and subsequent virus diagnoses using specific polymerase chain reaction (PCR) primers confirmed the association of severe CMD in this region with the presence in diseased plants of mixed infections of *African cassava mosaic virus* (ACMV) and the Uganda variant of *East African cassava mosaic virus* (EACMV-Ug) (Legg and Okao-Okuja, 1999). The occurrence of mixed ACMV/EACMV-Ug infections is characteristic of the "front" of the CMD pandemic (Harrison et al., 1997) and the associated severe symptoms are the main reason for its acute impact on cassava cultivation in affected areas.

In western Kenya, the distribution of CMGs appears to be complex and ACMV, EACMV and EACMV-Ug have been reported (Ogbe et al., 1996; Legg and Okao-Okuja, 1999). Monitoring surveys in late 1998 demonstrated the apparent slowing of the spread of the pandemic. This was possibly as a result of the presence of major natural barriers between north and south Nyanza Province (Kisumu area), which include the Winam Gulf of Lake Victoria and the Kano Plains, an expanse of flood plain in which cassava cultivation is virtually absent (Legg et al., 1999a). Although high incidences of severe CMD have yet to be reported from south Nyanza, the pandemic-associated EACMV-Ug has been detected from Migori, near the Tanzania border.

Data obtained from monitoring and diagnostic surveys have been used to develop regional CMD maps and their presentation has been an important tool in raising awareness amongst agricultural workers based in threatened zones. It is considered also that presentation of these graphics has been a key component in persuading both national agricultural institutions and donor agencies to take seriously the threat posed by the pandemic to regional cassava production and, with it, food security.

Multiplication

The multiplication and dissemination of CMD-resistant cultivars has been the main control technique used in tackling the CMD pandemic in Uganda and it has fulfilled the same role in the OFDA CMD Project in the wider region. Approaches to multiplication differed in each of the three participating countries, however, since the availability of planting material differed substantially in each. In Uganda, stems of cultivar SS4 were obtained from the USAID-funded PL 480 Project and used to plant 15 ha in each of Rakai and Masaka Districts.

In western Kenya, by contrast, less than 4 ha of CMD-resistant cultivars were present at the outset of the project, at the open quarantine site. The OFDA CMD Project joined GCF and the Rockefeller Foundation in supporting the multiplication of this material and by June 1999 the area of CMD-resistant material had increased to more than 35 ha.

In north-western Tanzania, the last of the three countries to be affected by the pandemic, only a small number of plants of CMD-resistant varieties were present by the end of 1998 and therefore more vigorous action had to be taken to obtain resistant material. As a consequence, whilst lobbying the Tanzania Ministry of Agriculture and Co-operatives for permission to establish open quarantine in Kagera, tissue culture plantlets were requested from IITA, Ibadan, Nigeria, and an intensive programme of true irrigated rapid multiplication (IITA, 1998) of available CMD-resistant cultivars was expanded at Ukiriguru Agricultural Research Institute (ARI), Mwanza. By June 1999, cassava planting material equivalent to almost half a million cuttings had been multiplied, more than 10,000 tissue culture plantlets of CMD-resistant cultivars had been imported from IITA, Nigeria and proposals for the establishment of open quarantine had been approved.

Germplasm diversification

Broadening the genetic base of cassava is seen as a key aspect of a balanced approach to the management of cassava pests and diseases. This is particularly important with respect to the CMD pandemic, since a heavy emphasis on the development of germplasm with high levels of resistance to CMD has resulted in less attention being given to other important pest and disease constraints. A practical result of this is that the two varieties most widely multiplied for CMD control in East Africa, SS4 and TMS 30572, are relatively susceptible to cassava green mite damage. In more recent germplasm development work, greater emphasis has been placed on multiple pest and disease resistance and germplasm with these characteristics is now becoming available both through national breeding programmes and through the EARRNET regional germplasm development programme.

In Uganda, two demonstration sites have been established in Rakai and Masaka and include nine cultivars that are currently being evaluated in national on-farm trials. In Kenya, more than 500 clones were introduced to the open quarantine site from the EARRNET programme based at Serere and the best 15 of these were multiplied for “fast-track” multi-locational testing in 2000. In Tanzania, more than 20 elite cultivars from the multiplication programme are under evaluation in areas of the Kagera region most severely affected by the pandemic, and new cultivars with novel West African landrace-derived sources of CMD resistance formed part of the recently imported tissue culture consignment from IITA, Nigeria. Each of these initiatives should help ensure that a diversity of cultivars is ultimately made available to farmers that should minimize losses likely to result from future pest or disease outbreaks.

Training

Changes resulting from the CMD pandemic are dramatic, and experience obtained from Uganda shows that farmers are typically unaware of the likely impact at the outset and frequently continue with cassava cultivation for a number of years, suffering substantial yield losses, before finally abandoning the crop. Raising awareness and the provision of training in CMD management are key requirements if this type of scenario is to be avoided. Training for both extension workers and farmers therefore has been an important part of the OFDA CMD Project. Various fora have been used for training exercises including:

(1) Stakeholder workshops: held at the outset of the project in each of the three countries. Information provided on the cause, spread, impact and strategy for the management of the CMD pandemic.
(2) *In-situ* training: farmers and extension workers have been trained in cassava pest management and multiplication techniques at multiplication sites.
(3) Individual training: researchers from participating countries have been trained in virus diagnostic techniques and have been sponsored to participate in regional CMD scientific meetings.
(4) Community training: village leaders have been trained in north-western Tanzania to facilitate the transfer of information down to grassroots level.
(5) Formal course training: the project has helped sponsor courses on seed multiplication in Tanzania and on cassava production, protection and utilization in Kenya, principally targeted at local agricultural workers.

In addition to the provision of training through courses or demonstration, use has been made of the media, particularly in Tanzania, where local radio communications have provided information on the CMD pandemic to a much greater number of households than could have been achieved using other methods.

Stakeholder linkages

The key to the success the project has achieved to date is largely attributable to the active participation of many cassava stakeholders in target areas. These include non-governmental organizations, government district agriculture offices, farmer training centres and projects funded by multi-lateral donors. The inclusion of a broad range of partners has the twin advantages of fostering ownership of the project at local level and enhancing local co-ordination. The development of a local steering committee for western Kenya, which was charged with the responsibility for co-ordinating cassava development and multiplication activities in that region, was an important development in which the OFDA Project participated (ASARECA, 1999).

Conclusions

In a relatively short period of time the OFDA CMD Project, affiliated to the TWF-IPM Project, has made significant achievements in both establishing a regional collaborative mechanism for the monitoring and management of the CMD pandemic and actually implementing control measures. A key advantage of the pro-active approach the project is taking, with its emphasis on targeting threatened zones, is that lead times for the implementation of control measures may be significantly reduced compared with previous experience in Uganda. Given the magnitude of losses associated with the CMD pandemic (Legg et al., 1999b; Otim-Nape et al., 1997; Thresh et al., 1997), this is an important development. The multi-faceted approach being pursued by the Tanzania Root and Tuber Crops Programme with its partners, and supported by the Project, also is considered to be a useful model for other countries threatened by the pandemic in the near future. Given the likely continued expansion of the pandemic into other neighbouring countries, similar such vigorous responses will be essential if major losses are not to continue across the cassava-producing areas of East and Central Africa.

Acknowledgements

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Conclusions

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The Tropical Whitefly Integrated Pest Management (TWF-IPM) Project is one of the most ambitious projects ever undertaken in the IPM area. The magnitude of the project responds to the global distribution of the pest and the severe damage caused by whiteflies to major food and industrial crops in tropical and subtropical agricultural regions of the world (Jones, 2003). The emergence of a more prolific and polyphagous biotype (B) of *Bemisia tabaci* (Gennadius) has further complicated the control of this pest on alfalfa (*Medicago sativa* [L.] subsp. *sativa*), cotton (*Gossypium hirsutum* L.), common bean (*Phaseolus vulgaris* L.), tomato (*Lycopersicon esculentum* Mill.), soybean (*Glycine max* [L.] Merr.), lettuce (*Lactuca sativa* L. var. *capitata* L.), peas (*Pisum sativum* L.), broccoli (*Brassica oleracea* L. var. *italica* Plenck), collard (*Brassica oleracea* L. var. *viridis* L.), cabbage (*Brassica oleracea* L.) and okra (*Abelmoschus esculentus* [L.] Moench) (Gruenhagen et al., 1993; Godfrey et al., 1995; Morales and Anderson, 2001). In developing countries of Latin America, sub-Saharan Africa, India and south-east Asia, *B. tabaci* is an efficient vector of plant viruses affecting important food crops, particularly common bean (Morales and Anderson, 2001), cassava (*Manihot esculenta* Crantz) (Fargette et al., 1990; Gibson et al., 1996), tomato (Polston and Anderson, 1997) and peppers (*Capsicum* spp. L.) (Morales and Anderson, 2001). In subtropical and mountainous regions, the whitefly species *Trialeurodes vaporariorum* (Westwood) attacks crops such as potato (*Solanum tuberosum* L.), tomato, common bean, several cucurbits and different vegetables (Boiteau and Singh, 1988; Cardona, 1995). Until recently, cultivated grasses (Gramineae) were among the few plant species spared by whitefly pests. Within the last decade, devastating whitefly attacks have taken place in important food and industrial crops such as rice (*Oryza sativa* L.), sorghum (*Sorghum bicolor* [L.] Moench), sugarcane (*Saccharum officinarum* L.) and pastures (author's observation). Not surprisingly, whiteflies were considered by popular news media (e.g., CNN and Newsweek) as the "pest of the 20th century" and their importance as a major agricultural pest remains high in the new millennium.

Whiteflies harm plants in different ways by extracting sap, covering plants with a sticky sugary substance (honeydew) that affects the quality (e.g., sticky cotton) or promotes the growth of fungi (sooty mould) on plant surfaces blocking photosynthesis (Henneberry et al., 1996) and by transmitting viruses (Markham et al., 1994; Jones, 2003). In heavily infested plants, hundreds of immature and adult

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whiteflies can be found feeding on a single leaf, resulting in rapid plant death from the massive loss of nutrients. Sooty mould can also provoke plant death. And, finally, whitefly-transmitted viruses are among the most damaging viral pathogens known, often causing total yield losses. Genetic immunity to these viruses is rare in most of the plant species attacked by begomoviruses.

Although whiteflies have been associated with agriculture for centuries, these insects were only recognized as pests in the 1950s (Ernst, 1994), coinciding with the development and intensive use of agricultural pesticides following World War II. Half a century later, most farmers are still not aware of the taxonomic or biological differences that characterize different species and their physiological variants (biotypes). Nor are they informed about the role of these species as pests and/or vectors of plant viruses. Therefore, farmers apply insecticides whenever they see whiteflies on their crops. In rare instances, farmers who have received some technical assistance apply insecticides when the population of the pests has reached a pre-determined level, known as the "damage threshold". Whereas these "target" or "threshold" applications may be effective in controlling whiteflies as pests (Chu et al., 1995; Riley and Palumbo, 1995), they are totally ineffective in the case of whitefly-transmitted viruses. Basically, few adult individuals of a whitefly vector can transmit a virus long before its population is noticed in the field or reaches a particular density on susceptible plants.

The introduction of the "B biotype" of *Bemisia tabaci* in the Americas (Brown and Bird, 1995) has drastically increased the capacity of this whitefly species to cause damage to a larger number of different cultivated plant species and adapt to new environments. Farmers that try to escape *B. tabaci* in the tropical lowlands and mid-altitude (500-900 m) regions by growing their crops at higher altitudes are often disappointed to find yet another whitefly pest, *Trialeurodes vaporariorum*, in the highlands. Unlike *B. tabaci*, the main whitefly pest and vector of plant viruses in the tropics, *T. vaporariorum* was considered a mere nuisance in crops grown under controlled conditions (e.g., glasshouse, screen-house conditions). Currently, *T. vaporariorum* is also a major pest of field crops in the highlands and temperate regions of the world and the number of plant viruses transmitted by this species is steadily growing (Jones, 2003).

Pesticide abuse is a common factor in the case of the main whitefly pests, regardless of the crops and ecosystems affected. The excessive application of insecticides, often using inadequate chemicals, alters the delicate balance between insect pests and their biological control agents (i.e., predators, parasitoids and entomopathogens). In the absence of natural enemies, whitefly populations increase freely on susceptible crops and eventually develop resistance to the most frequently used insecticides (Omer et al., 1993; Dennehy and Antilla, 1996). Modern agricultural practices, such as the intensive cropping of diverse plant species that act either as suitable feeding or reproductive hosts, further contribute to the exponential increase of whitefly pests in disturbed environments (Godfrey et al., 1995). Last but not least, climate change has played a major role in the increasing outbreaks of whitefly pests throughout the world. As more forest and wild lands are cleared for agricultural purposes, the climate becomes drier and warmer, conditions that shorten the life cycle of whitefly pests and thus increase their populations (Chu et al., 1995).

Small-scale farmers are trying to grow more profitable crops in order to maximize the output of their limited landholdings. In the absence of proper technical assistance, farmers face several problems: first, they are not familiar with new crops, let alone with their phytosanitary problems. Exotic crops can and will encounter different pests in their new environments. Often, exotic pathogens and pests are introduced with imported plant germplasm. In the case of horticultural crops, and particularly vegetables, the new cultivars have been bred in temperate countries, even though the origin of some of these plant species is the tropics (e.g., tomato and peppers). This means that new crops may not be well adapted to tropical conditions, including their lack of resistance to local pests. Although technical assistance to small- and medium-scale farmers is not forthcoming, pesticide salesmen manage to reach every corner of the rural world. This explains the reliance of farmers on pesticides to protect their crops.

The main problem of most IPM practices is their failure to control pests before they cause economic damage. IPM is often wrongly associated with “organic agriculture”, in which chemical insecticides do not have a place. Unfortunately, once an agricultural region has been significantly modified by the introduction of new crops, and its biological equilibrium broken by the intensive use of pesticides, whitefly populations are usually too large to be controlled by non-chemical IPM practices, such as biological control agents or yellow traps. Thus, the TWF-IPM Project has been following a different approach based on the rationalization of chemical control, using the most effective yet specific and safe systemic insecticides at the proper time. This strategy rapidly reduces the number of insecticide applications to a minimum, allowing the recovery of the beneficial fauna over time. Once this condition is met, other IPM strategies can be incorporated, such as cultural practices, use of entomopathogens, physical barriers and legal measures, among others. A significant reduction in the use of pesticides also contributes to lower production costs and higher profits for farmers, which will become critical factors for emerging economies in the age of free trade agreements. Last but not least, men, women and children in developing countries are inadvertently consuming vegetables contaminated with highly toxic pesticides. The medium- and long-term effects of these toxic residues are difficult to determine, but they may range from chronic illness to life-threatening ailments, including cancer. Reducing pesticide use in vegetable crops to safe levels should greatly contribute to vegetables free of toxic pesticide residues, and improved health standards in rural and urban populations in developing countries.

The history of cassava mosaic disease (CMD) is over a century old (Fauquet and Fargette, 1990) and it has been almost half a century since whiteflies emerged as pests and vectors of plant viruses of economic importance. New viruses have been appearing throughout the tropics and subtropical regions of the world with a high frequency (Polston and Anderson, 1997). With the advent of molecular techniques, the list of whitefly-borne viruses has increased exponentially in the last 3 decades. Altogether, considerable research has been conducted on these pests, and yet, their impact seems to increase every year in developing countries. We have already discussed some of the factors that have contributed to the unexpected dissemination of whiteflies and whitefly-borne viruses. Undoubtedly, the advent of chemical insecticides and their intensive use in crops, such as cotton, elevated whiteflies to the category of pests. Another important epidemiological factor, the diversification of crops in developing countries, both as export commodities and cash crops, has also played a major role in the increase of these pests.

The importance of some of the new crops lies in their role as reproductive hosts to whiteflies (Tsai et al., 1996). These crops need not support very large populations of whiteflies, but rather occupy large areas or regions. The best example is perhaps soybean, which was rediscovered in Latin America in the 1970s as a potential export crop, increasing the total area planted to over 10 million hectares. Because individual soybean plants do not support large populations of whiteflies, primarily *B. tabaci*, soybean growers do not control this insect in the soybean crop. When the soybean crops mature at the end of the year, common beans are planted, provoking the migration of whiteflies from the soybean fields to the young common bean plantings. The search for more profitable crops has led small- and medium-scale farmers to diversify and intensify their cropping systems, providing an opportunity for whiteflies to dispose of different food sources throughout the year.

Crop improvement has played an important role in both the onset and control of whitefly-related problems. In the case of cassava, the lack of CMD-resistant cultivars and/or the limited adoption of mosaic-resistant germplasm has perpetuated the cultivation of CMD-susceptible cassava clones (Morales, 2001). Consequently, the causal viruses find hosts that act as virus reservoirs and permanent sources of inoculum for the whitefly vector. In sub-Saharan Africa, this situation has made possible the occurrence of multiple infections and subsequent recombination of different CMD-inducing virus species (Pita et al., 2001). The recombinant whitefly-transmitted begomovirus has caused severe yield losses in Uganda and some neighbouring countries, necessitating the implementation of a famine relief project. In the case of common bean, the downsizing of the national and international programs that worked on the development of begomovirus-resistant bean germplasm in Latin America has resulted in the abandonment of prime agricultural regions where common beans used to be produced. This happened because of the emergence of more pathogenic begomoviruses and the breakdown of the resistance available in the earlier generations of improved common bean cultivars (Morales, 2001).

The economic crisis that affects most national agricultural research programs in developing nations has greatly affected the flow of technical information to farmers and, consequently, their capacity to manage complex and severe crop production constraints such as the whitefly problem. Industrialized nations are currently controlling whiteflies with a new generation of systemic insecticides that are more efficient and selective (Chao et al., 1997). They are available in developing countries, but their price is very high for most small-scale farmers, who continue to use ineffective and highly toxic contact insecticides, often on a daily basis. These contact insecticides have practically annihilated the beneficial fauna and induced resistance in whitefly pests, giving rise to very large populations of whiteflies that can overcome most control methods, including systemic insecticides. Fortunately, some of the new systemic insecticides are being produced as “generic” compounds in developing countries, at lower prices. These problems have forced many farmers to abandon the cultivation of susceptible crops in prime agricultural regions.

The TWF-IPM Project has gathered enough evidence to suggest that the whitefly problem can be managed with the collaboration of farmers, by providing proper and timely technical assistance. The challenge for the project is to find the most appropriate communication means to disseminate the IPM technology that has been found to be effective and economically viable for small-scale farmers. To this end, the TWF-IPM Project is about to initiate its third and final phase to disseminate and scale out suitable IPM technology using farmer participatory research and different communication channels to reach affected farmers throughout the tropics, wherever whiteflies are a food production constraint. We are confident that well-informed farmers will be able to reduce the number of application of insecticides, lower production costs, increase profits, and reduce environmental contamination and human health hazards. Ultimately, farmers will be able to recover the biological equilibrium of their agricultural ecosystems, and thus prevent future whitefly outbreaks.

The information presented in this book represents a major achievement in terms of compiling the international and national (grey) literature and knowledge available on the extent and socio-economic importance of whiteflies as pests and vectors of plant viruses. This information has been essential in selecting the most promising and viable IPM strategies to be tested in Phase II of the TWF-IPM Project. The information contained in this book also reflects the interest and hard work of many colleagues who now form one of the most extensive research networks in the tropics. This joint effort has already yielded visible results in terms of raising awareness about the nature of the whitefly problem, particularly in relation to the key issue of pesticide abuse, and the use of appropriate methods to characterize whitefly pests and diagnose whitefly-borne viruses. The design, validation, implementation and success of IPM methods recommended to control whiteflies and whitefly-transmitted viruses largely depends upon a solid knowledge base.

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Acronyms and Abbreviations

Acronyms

ACIAR	Australian Centre for International Agricultural Research
AID	Agency for International Development
ANPP	Association Nationale pour la Protection des Plantes (*National Association for Plant Protection*), France
ARI	Agricultural Research Institute
ARP	African Regional Program of AVRD
ARS	Agricultural Research Service of USDA, USA
ASARECA	Association for the Strengthening of Agricultural Research in Eastern and Central Africa
ASDI	Agencia Sueca para el Desarrollo Internacional (*Swedish Agency for International Development*)
ATC	Agriculture Trading Company, Malawi
AVRDC	Asian Vegetable Research and Development Center
BBA	Biologische Bundesanstalt für Land und Fortwirtschaft (*Federal Biological Research Centre for Agriculture and Forestry*), Germany
BMNH	British Museum of Natural History
BNN	Banco Nacional de Nicaragua (*National Bank of Nicaragua*)
C&F	New Zealand Crop and Food Research
CATIE	Centro Agronómico Tropical de Investigación y Enseñanza (*Tropical Agricultural Research and Higher Education Centre*), Costa Rica
CENSA	Centro Nacional de Sanidad Agropecuaria (*National Centre for Plant and Animal Health*), Cuba
CENTA	Centro Nacional de Tecnificación Agrícola (*National Centre of Agricultural Technification*), El Salvador
CGIAR	Consultative Group on International Agricultural Research
CIAL	Comité de Investigación Agrícola Local (*Local Agricultural Research Committee*)
CIAS	Centro de Investigaciones Agrícolas del Sureste (*South-eastern Agricultural Research Centre*), Mexico
CIAT	Centro Internacional de Agricultura Tropical (*International Center for Tropical Agriculture*), Cali, Colombia

CIAZA	Centro de Investigaciones Aplicadas a Zonas Áridas (*Research Centre for Arid Zones*), Dominican Republic
CINVESTAV	Centro de Investigaciones y Estudios Avanzados (*Centre for Research and Advanced Studies*), Irapuato, Mexico
CIP	Centro Internacional de la Papa (*International Potato Center*), Peru
CNIA	Centro Nacional de Investigación Agropecuaria (*National Centre for Agricultural Research*), Nicaragua
CNRE	Centre National de Recherche sur l'Environnement (*National Centre for Environmental Research*), Madagascar
CONAL	Comisión Nacional del Algodón (*National Cotton Commission*), Nicaragua
CONVERDS	Collaborative Network for Vegetable Research and Development in Southern Africa
CORPOICA	Corporación Colombiana de Investigación Agropecuaria (*Colombian Corporation of Agricultural Research*)
COSCA	Collaborative Study of Cassava in Africa
CRDA	Centre de recherche et de documentation agricoles (*Centre for Agricultural Research and Documentation*), Haiti
CRS	Chitedze Research Station, Malawi
CSIRO	Commonwealth Scientific and Industrial Research Organisation, Australia
DANIDA	Danish International Development Agency
DARNDR	Fondo Salvadoreño de Estudios de l'Agriculture, des Ressources Naturelles et du Développement Rural (*Salvadoran Fund for studies in Agriculture, Natural Resources and Rural Development*)
DDR	German Democratic Republic
DFID	Department for International Development, UK
DICTA	Departamento de Investigación y Capacitación Agropecuaria (*Agricultural Research and Training Department*), Honduras
DIGESA	Dirección General de Salud Ambiental (*State Environmental Health Office*), El Salvador
DRC	Democratic Republic of Congo
EAP	Escuela Agrícola Panamericana (*Pan-American Agricultural School*), Honduras
EARRNET	East Africa Root Crops Research Network
ESARC	Eastern and Southern Africa Regional Center of IITA
ESCaPP	Ecologically Sustainable Cassava Plant Protection, regional project, Ghana
EXTOXNET	Extension Toxicology Network
FAO	Food and Agriculture Organization of the United Nations, Italy
FCA	Florida Collection of Arthropods

FIRA	Fideicomisos Instituidos en Relación con la Agricultura (*Agriculturally Related Trusts*), Mexico
FOFIFA	Centre National de Recherche Appliquée au Développement Rural (*National Centre for Applied Research in Rural Development*), Madagascar
FSCA	Florida State Collection of Arthropods
FUNDESA	Fundación para el Desarrollo Agropecuario (*Foundation for Agricultural Development*), Santo Domingo, Dominican Republic
GCF	Gatsby Charitable Foundation, UK
GCRIO	Global Change Research Information Office, USA
GIIT	Grupo Interinstitucional e Interdisciplinario de Tomate (*Interinstitutional and Interdisciplinary Tomato Group*), Nicaragua
GTZ	Deutsche Gesellschaft für Technische Zusammenarbeit (*German Agency for Technical Cooperation*)
HORTI	Horticultural Research and Training Institute, Tanzania
HPR	Host Plant Resistance Project of MFAT
IARCs	International Agricultural Research Centres
ICA	Instituto Colombiano Agropecuario (*Colombian Institute of Agriculture and Livestock*)
ICIPE	International Centre of Insect Physiology and Ecology, Kenya
ICTA	Instituto de Ciencia y Tecnología Agrícolas (*Institute of Agricultural Science and Technology*), Guatemala City, Guatemala
IDIAP	Instituto de Investigación Agropecuaria de Panamá (*Panamanian Agricultural Research Institute*)
IDRC	International Development Research Centre, Canada
IICA	Instituto Interamericano de Cooperación para la Agricultura (*Inter-American Institute for Cooperation on Agriculture*)
IIE	Institution of International Education, UK
IITA	International Institute of Tropical Agriculture, Nigeria
IITA-ESARC	International Institute of Tropical Agriculture-Eastern and Southern Regional Center, Nigeria
INIA	Instituto Nacional de Investigación Agraria (*National Institute of Agricultural Research*), Mexico
INIAP	Instituto Nacional Autónomo de Investigaciones Agropecuarias (*National Institute for Agricultural Research*), Ecuador
INIFAP	Instituto Nacional de Investigaciones Forestales y Agropecuarias (*National Institute for Agricultural and Forestry Research*), Sinaloa, Mexico
INISAV	Instituto de Investigaciones de Sanidad Vegetal (*National Institute for Research on Plant Health*), Havana, Cuba
INRAB	Institut National de Recherche Agronomique du Bénin (*Benin National Institute for Agricultural Research*)

IOBC	International Organisation for Biological and Integrated Control for Noxious Animals and Plants
IRA	Institut de Recherche Agronomique (*Agricultural Research Institute*), Cameroon
ISA	Instituto Superior de Agricultura (*Higher Institute for Agriculture*), Dominican Republic
ISTRC-AB	International Society for Tropical Root Crops-Africa Branch
JIC	John Innes Centre, UK
KARI	Kenya Agricultural Research Institute
LILIANA	Instituto de Investigaciones de Hortícolas “Liliana Dimitroya” (*Horticultural Research Institute “Liliana Dimitroya”*), Cuba
LZARDI	Lake Zone Agricultural Research and Development Institute, Tanzania
MAG	Ministerio de Agricultura y Ganadería (*Ministry of Agriculture*), Nicaragua
MARNDR	Ministère de l’agriculture, des ressources naturelles et du développement rural (*Ministry of Agriculture, Natural Resources and Rural Development*), Haiti
MFAT	Ministry of Foreign Affairs and Trade, New Zealand
MIP	Manejo Integrado de Plagas (*Integrated Pest Management*)
MSU	Montana State University, USA
NAARI	Namulonge Agricultural and Animal Production Research Institute, Uganda
NARES	National Agricultural Research and Extension Systems
NARO	National Agricultural Research Organisation, Uganda
NORAD	Norwegian Agency for Co-operation for Development
NRCRI	National Root Crops Research Institute, Nigeria
NRI	Natural Resources Institute, UK
NRTCIP	National Root and Tuber Crops Improvement Project, Ghana
OFDA	Office for Foreign Disaster Assistance of USAID
OIRSA	Organismo Internacional Regional de Sanidad Agropecuaria (*Regional International Organization for Plant Protection and Animal Health*), Nicaragua
PAUP	Phylogenetic Analysis Using Parsimony
PCCMCA	Programa Cooperativo Centroamericano para el Mejoramiento de Cultivos Alimenticios (*Centroamerican Cooperative Programme for the Improvement of Food Crops*), Guatemala
PHYLIP	Phylogeny Inference Package
PNUMA	Programa de las Naciones Unidas para el Medio Ambiente (*United Nations Environment Programme*), Geneva
PPRI	Plant Protection Research Institute, South Africa
PPRSD	Plant Protection and Regulatory Services Directorate, Ghana

PROFRIJOL	Programa Cooperativo Regional de Frijol para Centro América, México y el Caribe (*Regional Co-operative Bean Programme for Central America, Mexico and the Caribbean*)
PROMIPAC	Programa Regional de Manejo Integrado de Plagas en América Central (*Regional Integrated Pest Management Program for Central America*)
REDCAHOR	Red Colaborativa de Investigación y Desarrollo de Hortalizas para América Central, Panamá y República Dominicana (*Collaborative Network for Vegetable Research and Development for Central America, Panama and Dominican Republic*)
SADC	Southern Africa Development Community
SARH	Secretaría de Agricultura y Recursos Hidráulicos (*Secretariat of Agriculture and Hydraulic Resources*), Mexico
SARI	Selian Agricultural Research Institute, Tanzania
SARRNET	Southern African Root Crops Research Network
SDC	Swiss Agency for Development and Cooperation
SEA	Secretaría del Estado de Agricultura (*State Agricultural Secretariat*), Dominican Republic
SENASA	Servicio Nacional de Salud Animal (*National Service for Animal Health*), El Salvador
SP-IPM	Systemwide Programme on Integrated Pest Management
TRTCP	Tanzania Root and Tuber Crops Programme
TWF-IPM	Tropical Whitefly Integrated Pest Management Project
UA	University of Arizona-Tucson, USA
UFL	University of Florida-Gainesville, USA
UNA	Universidad Nacional Agraria (*National Agricultural University*), Nicaragua
UNDP	United Nations Development Programme
USAID	United States Agency for International Development
USDA	United States Department of Agriculture
USNM	United States National Museum
UTLA	Universidad Técnica Latinoamericana (*Latin American Technical University*)
UW	University of Wisconsin-Madison, USA

Abbreviations

AC1	replicase
ACMD	African cassava mosaic disease
ACMV	*African cassava mosaic virus*
A.f.	*Aleurothrixus floccosus*
AFLP	amplified fragment length polymorphism
ANOVA	analysis of variance
A.p.	*Aleyrodes proletella*
AV1	capsid protein
B.a.	*Bemisia afer*
BCaMV	*Bean calico mosaic virus*
BDMV	*Bean dwarf mosaic virus*
BG	begomovirus
BGMV	*Bean golden mosaic virus*
BGYMV	*Bean golden yellow mosaic virus*
B.h.	*Bemisia hirta*
BMMV	*Bean mild mosaic virus*

BR	Brazil
B.t.	*Bemisia tabaci*
B.t.-A	*Bemisia tabaci* A biotype
B.t.-B	*Bemisia tabaci* B biotype
C	plant infected as cutting
CAR	carbamates
CLCuV	*Cotton leaf curl virus*
CMD	cassava mosaic disease
CMG	cassava mosaic geminivirus
CMV	cucumoviruses, *Cucumber mosaic virus*
CO	Colombia
COI	cytochrome oxidase I
CPMMV	*Cowpea mild mottle virus*
CVYV	*Cucumber vein yellowing virus*
DAS	double antibody sandwich
DNA	deoxyribonucleic acid
DR	Dominican Republic
DSM	Dar es Salaam
E	early infected plant
EACMV	*East African cassava mosaic virus*
Eg	Egypt
EIL	economic injury level
ELISA	enzyme-linked immunosorbent assay
EM	electron microscopy
EMIF	electronic monitoring of insect feeding
EPG	electrical penetration graph
ES	El Salvador
FFS	farmer field school
FL	fiducial limits
GA	Guatemala
GIS	geographic information systems
GV	geminiviruses
H	healthy, symptomless plant
HD	Honduras
HPR	host plant resistant
HR	highly resistant
HS	highly susceptible
I	intermediate infected plant
I	intermediate resistance
ICMV	*Indian cassava mosaic virus*
IPM	integrated pest management
Is	Israel
ITS	internal transcribed spacers
L	late infected plant
LC	lethal concentration
LSD	least significant difference
MABs	monoclonal antibodies
MAB-BS	a broad spectrum monoclonal antibody used to detect bi-partite begomoviruses
MAB-GA	a monoclonal antibody used to detect the original Middle American isolates of *Bean golden yellow mosaic virus*-Guatemala
MAP	months after planting
MIU	multiple infection units
Mo	Mochis
MR	moderately resistant
MYMV	*Mungbean yellow mosaic virus*
NARS	national agricultural research systems
NCM	nitrocellulose membrane
nt	not tested
O.c.	*Orchamoplatus citri*
OLCV	*Okra leaf curl virus*
OPs	organophosphate insecticides
ORF	open reading frame
PAGE	polyacrylamide gel electrophoresis
PCR	polymerase chain reaction
PepGMV	*Pepper golden mosaic virus*
PepLCV	*Pepper leaf curl virus*
PHYVV	*Pepper Huasteco yellow vein virus*
PR	Puerto Rico
PTY1	monoclonal antibody to detect potyviruses
PVY	*Potato virus Y*

PYR	pyrethroids
QTL	quantitative trait locus
R	whitefly resistant cultivar
RAPD	random amplified polymorphic DNA
RF	resistance factor
RFLPs	restriction fragment length polymorphisms
RH	relative humidity
S	susceptible
S	whitefly susceptible cultivar
SACMV	*South African cassava mosaic virus*
SCAR	sequence characterized amplified region
S_{EA}	East African strain
SiGMV	*Sida golden mosaic virus*
SLCV	*Squash leaf curl virus*
S.p.	*Siphoninus phillyreae*
SPCFV	*Sweetpotato chlorotic fleck virus*
SPCSV	*Sweetpotato chlorotic stunt virus*
SPFMV	*Sweetpotato feathery mottle virus*
SPLV	*Sweetpotato latent virus*
SPMMV	*Sweetpotato mild mottle virus*
SPSV-SEA	East African strain of SPCSV
SPVD	sweetpotato virus disease
T	whitefly tolerant cultivar
T.a.	*Tetraleurodes andropogon*
Tan	Tanzania
TAS	triple antibody sandwich
TBR	tree-bisection reconnection
TEV	*Tobacco etch virus* (Potyviridae: Potyvirus)
TLC	tomato leaf curl disease
TLCV	*Tomato leaf curl virus,* now ToLCV
TMV	*Tobacco mosaic virus* (*Tobamovirus*)
ToLCV	*Tomato leaf curl virus*
ToMoTV	*Tomato mottle Taino virus*
ToMoV	*Tomato mottle virus*
ToYMoV	*Tomato yellow mottle virus*
ToYMV	*Tomato yellow mosaic virus*
TPV	Texas pepper virus (= *Pepper golden mosaic virus*)
T.r.	*Trialeurodes ricini*
T.v.	*Trialeurodes vaporariorum*
TVTV	*Tomato vein thickening virus*
TYLCV	*Tomato yellow leaf curl virus*
Ug	Uganda
WmCSV	*Watermelon chlorotic stunt virus*
WTVs	whitefly-transmitted viruses
Xam	*Xanthomonas axonopodis* pv. *manihotis*