

Figure 16. SCAR'S for the identification of whiteflies from Yucatán. Lanel 1 = molecular marker (1Kb). Lanes 2 and 3 = Whieflies from mint (*B. Tabaci*). Lanes 4 and 5 = whiteflies from Tamaulipas. Lanes 6–10 whiteflies from Yucatan, Lanes 7 and 8 correspond to biotype B of *B. tabaci*, Lane 9 corresponds to *T. vaporariorum*. 11 = *B. tabaci* control. Lane 12 = Biotype B of *B. tabaci*. Lane 13 = *T. vaporariorum*. Lane 14 = Molecular marker (1Kb).

The last sequences obtained for tomato begomoviruses found in Yucatan, indicate that *Tomato mottle virus* was the predominant virus in the pilot sites selected. Aphid-transmitted viruses, particularly potyviruses, have also been detected in pepper tomato and cucurbit samples (**Table 13**). Yucatan is also the only region in Continental Latin America, where an Old World virus, *Tomato yellow leaf curl virus* (TYLCV) has been reported in tomato. Specific PCR assays with tomato samples from Yucatan confirmed the presence of this monopartite begomovirus, mixed with bipartite begomoviruses as well (**Figure 17**).

Table 13. Serological assay of selected plant samples from Yucatan, Mexico.

Plant Sample	Locality	GEMINIVIRUS		POTYVIRUS		CMV	
		Atc. M. 4C1-3f7 Absorbance*		AGDIA Absorbance**		Atc. M. 5 ⁹ -1F9-1D5 Absorbance*	
Ya-ax-ak	Hunucmá	0.08	-	0.93	+	0.13	+
Chilli habanero c/s	Hunucmá	0.08	-	0.02	-	0.002	-
Chilli habanero línea 11	Mocochá	0.09	-	0.08	-	0.01	-
Tomato	Dzán	0.25	+	0.41	+	0.06	-
Negative Control		0.01	-	0.06	-	0.008	-
Positive Control		1.22	+	2.00	+	0.39	+

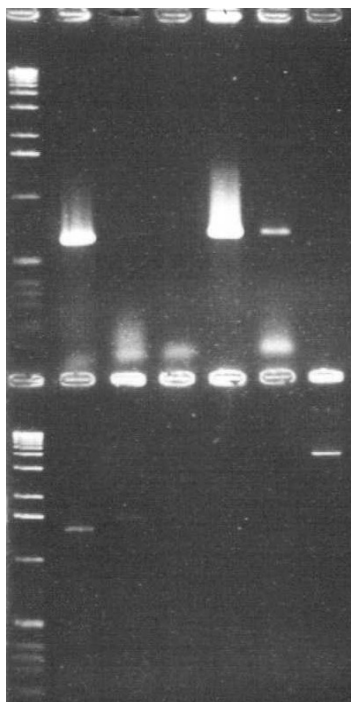


Figure 17. PCR assay for TYLCV. Lane 1 = 1 Kb molecular marker. Lane 2 = Positive control for TYLCV. Top gel: Lanes 3-7 Tomato samples from Yucatan. Bottom gel: Lanes 3 – 7: same samples assayed for bi-partite begomoviruses (sample 7 was positive).

The presence of both Old World (TYLCV) and New World begomoviruses in Yucatán, complicate the breeding for resistance to these different viruses. Fortunately, the potential sources of resistance to begomoviruses sent by AVRDC (**Table 8**), had been originally evaluated and selected for Old World begomoviruses, including TYLCV. These materials were evaluated in three different localities of the Yucatan Peninsula, Mocochoá, Yaxhoom and Uxmal. The first locality had very low virus incidence (after Hurricane Isidore) and was used to evaluate the agronomic characteristics of the imported germplasm. The second and third localities had medium and high virus pressure, respectively, and provided good data on the potential of these entries as sources of resistance. In Yaxhoom, FLA 456-4 was the best entry (no symptoms of viral infection), followed by FLA 478-6-1-0, FLA 653-3-1-0 and FLA 505. In Uxmal, FLA 456-4 was again the most resistant entry, but all of the FLA lines behaved well. The remaining materials did not produce.

Based on these results, FLA 456-4 was selected as parental material to improve susceptible local tomato cultivars, such as ‘Maya’. The crosses were made at AVRDC, Taiwan, by Dr. Peter Hanson, and segregating (F2-F4) lines (**Table 14**) were sent to Yucatan. **Table 15** shows the results of this evaluation.

As it can be concluded from **Table 15**, the resistance to begomoviruses present in FLA 456-4, was not transferred to the segregating materials evaluated in Yucatan, and the lines were susceptible to early blight. A new set of crosses is being prepared now at AVRDC to exploit the high levels of resistance found in the FLA tomato lines.

Table 14. Tomato materials at different levels of inbreeding and range from F2 to F4, derived from crosses involving FLA 456-4 as a source of begomovirus Resistance.

Entry	Generation	Previous	Quantity
CLN2714-7	F2	21606-7	50 semillas
CLN2714-117	F2	21606-117	50 semillas
21602-21	F3		30 semillas
21602-40	F3		30 semillas
21602-76	F3		30 semillas
21602-94	F3		30 semillas
21602-105	F3		30 semillas
21602-175	F3		30 semillas
21602-264	F3		30 semillas
21602-3	F3		30 semillas
21602-56	F3		30 semillas
21602-90	F3		30 semillas
CLN2674-129-27-11	F4	22995-2	50 semillas
CLN2674-129-27-11	F4	22989-11	50 semillas
CLN2674-138-9-30	F4	22995-30	50 semillas
CLN2679-199-9-14	F4	23027-14	50 semillas
CLN2679-199-9-26	F4	23027-26*	50 semillas
CLN2679-199-12-8	F4	23028-8	50 semillas
CLN2679-199-16-29	F4	23030-29*	50 semillas
FLA456-4	check		50 semillas

Table 15. Results of the field evaluation of segregating tomato materials derived from crosses with FLA 456-4 in Yucatán.

Material	No. plants	%/virus	Severity	% E. blight
CLN2714-7	29	96	4	75
CLN2714-117	18	97	5	80
21602-21	25	99	5	66
21602-40	22	92	5	78
21602-76	16	89	5	90
21602-94	24	100	5	92
21602-21	6	100	5	67
21602-105	7	100	5	77
21602-175	24	100	5	86
21602-3	17	98	5	66
21602-56	8	100	5	87
CLN2679-199-12-8	18	97	5	98
CLN2679-199-16-29	11	92	5	49
CLN2674-129-27-11	2	100	5	76
21602-264	2	100	5	88
CLN2674-138-9-2	5	100	5	67
FLA-456-4	60	25	2	30

A survey of cultivated and wild hosts of *Bemisia tabaci* in horticultural farms of northern Yucatan, revealed the existence of 58 wild and 14 cultivated plant species. The wild species belong to 22 different botanical families, and the cultivated species to three major families: *Leguminosae* (*Vigna unguiculata*), *Cucurbitaceae* (*Cucurbita*, *Citrullus* spp.) and *Solanaceae* (*Lycopersicon*, *Capsicum*, *Solanum*, *Nicotiana* spp.). **Table 16** shows the distribution of the whitefly-transmitted viruses found in Yucatan by the coordinator of this project in Mexico, Dr. Raul Diaz-Plaza (INIFAP) in the various host plants identified.

Table 16. Ecology of begomoviruses* in Yucatan, Mexico.

Host	PHYVV	PepGMV	TYLCV	ToMoV
Malvaceae	-	+	+	+
Leguminosae	+	-	+	-
Euphorbiaceae	+	-	-	-
Cucurbitaceae	+	-	+	-
Convolvulaceae	-	-	+	-
Amaranthaceae	+	-	+	-
Solanaceae				
Tomato	+	+	+	+
Sweet pepper	+	+	+	+
Habanero pepper	+	+	+	+

* PHV = *Pepper huasteco yellow vein virus*; PepGMV = *Pepper goldem mosaic virus*; TYLCV = *Tomato yellow leaf curl virus*; and ToMoV = *Tomato mottle virus*.

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IPM measures implemented in Yucatán

The INIFAP group in Yucatán has been working on whitefly control since the Peninsula was severely affected by this pest and the viruses it transmits in 1989. Yucatan was the first place where the microtunnel technology was adopted by small-scale farmers in Latin America (**Figure 18**).



Figure 18. Chilli production under microtunnels in the state of Yucatán (1999).

This strategy worked efficiently until new chemical products appeared in the market for whitefly control (mainly the new neonicotinoids; e.g. imidacloprid). The salespeople from the agrochemical companies that market these products, convinced farmers to switch to chemical control instead of dealing with nets, which require more intensive labour and higher production costs. Thus, at the onset of the TWFP-Phase II in Yucatán, most producers of horticultural crops had abandoned the use of anti-whitefly nets and reverted to chemical control using imidacloprid. A preliminary survey of the area affected by whiteflies in the state of Yucatán, clearly showed that imidacloprid was not controlling all the viruses transmitted by insects in either tomato or peppers (**Figure 19**).

The national program scientists of INIFAP-Yucatan were already conducting tests on IPM practices to control the whitefly *B. tabaci* when the project started. Their main strategy was the use of natural barriers (e.g. maize, cucumber, eggplant) and intercropping chillies with other plant species (mostly horticultural species, such as mint, leeks, basil and coriander) of economic value, previously shown to possess some whitefly-repellent properties.

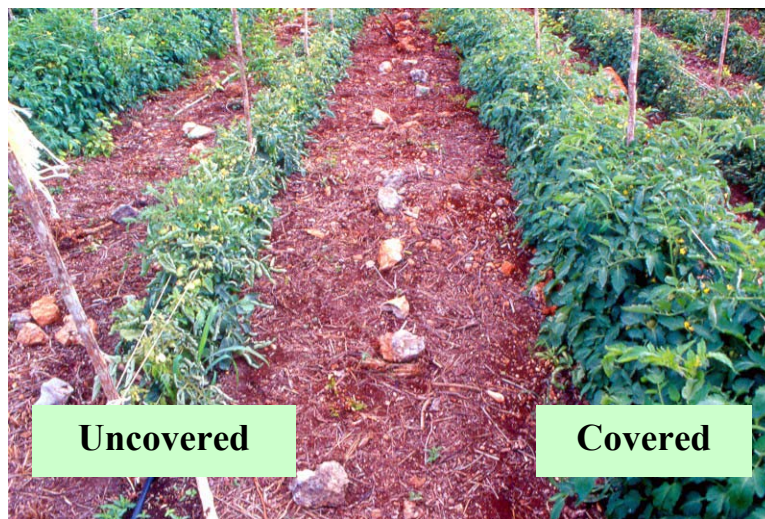


Figure 19. Effect of microtunnels on tomato production in Yucatán, Mexico.

Whereas the main objective of the project was to show small-scale farmers that although the use of nets implies higher production costs, these are offset by the higher yields and quality obtained, the TWFP decided to merge the two strategies (natural barriers/repellent plants and microtunnels). These experiments were evaluated during the first year of the project in three different localities (Mocochá, Hunucma and Yaxhoom), and clearly showed that these strategies do not work. Basically, even under moderate whitefly pressure, virus incidence was approximately 55% in both the control (traditional planting) and plots protected by live barriers and inter-planted with whitefly-repellent crops (**Table 17**).

Table 17. Effect of the association of chilli and whitefly-repellent plants on whitefly-borne virus incidence in Hunucmá, Yucatán.

Treatment	Virus Incidente (%)				F _{0.05}	F _{0.01}
	Rep. 1	Rep. 2	Rep. 3	Rep. 4		
Coriander	33	45	43	55		
Mint	44	45	53	58		
Basil	54	60	62	56		
Leek	56	55	72	43		
F.V.	G.L.	S.C.	C.M.	FOBS.		
Treatment	4	498	124.5	2.07 N.S.	3.06	4.89
Error	15	899.75	59.98			
TOTAL	19	1,397.75				

The microtunnel work was initiated in November 2002, three months after hurricane Isidore had caused considerable damage in the Yucatan Peninsula. One of the effects of hurricanes, is the destruction of small wildlife, mainly insects. Consequently, the whitefly pressure during the first months of 2003, when the effect of the microtunnels had to be evaluated, was extremely low, obliterating the differences between the uncovered and covered controls (less than 5% begomovirus incidence).

The microtunnel trial was established again in the last quarter of 2003, in the locality of Dzán. The trial was conducted in a commercial rather than experimental field of 1.5 has, because nine farmers decided to pool their resources to finance the trial. Having had disappointing results with some farmers that did not take good care of the experimental trials recommended by the project, we decided to let them conduct the trial, as long as they followed the instructions for the correct use of the technological package. This IPM package consists of: 1) tomato seedlings produced under screen to avoid the early infection of seedlings grown outdoors; 2) one application of a systemic insecticide to seedlings two days before transplanting; 3) cover the seedlings immediately after transplant with the screening material (Agribon, Agryl, Tricot, etc); 4) a second and last systemic insecticide application two days before removing the net cover (approximately a month after transplanting). It must be taken into account that tomato and pepper growers in whitefly-infested areas usually make over 30 insecticide applications until harvest time.

The collaborating farmers covered approximately one hectare, alternating covered and uncovered rows in hopes of using the covered row as a barrier for the whitefly. **Table 18** shows the results obtained.

These results were obtained at a time of moderate virus pressure, and, moreover, the uncovered control had been properly protected in the nursery and field using the chemical protection scheme recommended by the TWFP. The intercropping of covered and uncovered rows probably helped reduce disease severity, but it did not prevent virus infection and plant damage (**Figure 19**). Had these farmers followed their own agronomic practices in a year of high whitefly/virus pressure, total crop failure may have occurred in the uncovered treatment.

Table 18. Effect of microtunnels (Agribon) on tomato production in Dzán, Yucatan.

Variable	Uncoverd	Covered
Plant height	55 cm	90 cm
Harvest time	80 dat*	87 dat
Virus Incidence	85%	4%
Disease severity (0-4)	1 (28%), 2 (23%), 3 (34%)	1 (3%), 2 (1%)
Yield	35 MT/Ha	45 MT/Ha
Fruit quality (1-3)**	1 (40%), 2 (30%), 3 (30%)	1 (60%), 2 (25%), 3 (15%)
Net Profit	US \$ 14,200	US \$ 19,163

* dat = days after transplant; ** 1 = best quality/higher price

An increase of 25-26% in yield and net profit (ca. US \$ 5,000) under these conditions is significant, because this additional income pays for the cost of the microtunnels, including materials and labour (Total cost/Ha = US \$ 2,000). Moreover, microtunnels increase the quality of the produce and control aphid-borne viruses, which are not controlled by any insecticide. Last, microtunnels protect the investment of resource-poor farmers in times of high whitefly/virus pressure (crop insurance), when insecticides do not prevent significant losses and even total crop failure.

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Outputs

The case studies and socio-economic analyses conducted in El Salvador, Yucatán and previously in other Central American countries by the TWFP, clearly show that small-scale farmers are trying to diversify their traditional cropping systems with high-value crops to maximise the income derived from their limited land resources.

These surveys also show that the whitefly *Bemisia tabaci* and the viruses it transmits (begomoviruses) are the main factors responsible for the significant yield losses suffered by traditional and non-traditional susceptible crops alike. These problems have occurred at a time when most Latin American governments had to reduce public spending (including agricultural research and extension), in order to pay their ever-increasing external debts. Consequently, small-scale farmers who had started to grow non-traditional (e.g. vegetables) crops without any technical assistance, had to protect their investment according to the recommendations of the only technical personnel reaching them: the salespeople working for agrochemical companies. At the same time, international agricultural research centres were forced to change the focus of their research from food production to natural resource management. Some of the many negative consequences of the lack of technical assistance to small-scale farmers has been: crop failure, need to import food, higher unemployment rates, increased poverty, and extreme levels of pesticide abuse. The latter has caused severe environmental contamination and widespread health

problems in rural and urban populations. Thus, it must become clear to policy makers that shifting resources from food production-oriented research to research in natural resources and sociology work in rural areas, is extremely counterproductive in the absence of sustainable crop production components. Farmers may be organised, but unless their current crop production problems are solved, they cannot either feed themselves or improve their livelihoods.

Common bean has been one of the two main food staples of the Mesoamerican diet (together with maize) since pre-Columbian times. Without the active common bean breeding program that the Bean Programme of CIAT initiated in the 1980s, Mesoamerica would not be eating beans during almost half of the year, due to the whitefly/virus problems that affect this region every year during the dry season. Unfortunately, the shifting of research priorities at CIAT has considerably reduced the capacity of its Bean Project (not a Programme any more due to its reduced size) to produce more begomovirus-resistant common bean lines for this region. Fortunately, the breeder of the Pan American School in Honduras, Dr. Juan Carlos Rosas, has continued to improve CIAT's materials to create new common bean cultivars. The TWFP was lucky to find in El Salvador a highly promising breeding line (EAP 9510-77) that was commercially acceptable, and highly virus-resistant, to help evaluate it and promote its adoption in order to incorporate this variety into an IPM package that allowed small-scale farmers to cultivate common beans once more during the dry months of the year. This is the first time that an IPM package has been delivered together with the virus-resistant variety. Failure to do so in the past, has resulted in the breakdown of improved, virus-resistant lines, such as CENTA-Cuzclateco (DOR 364), which has broken down due to the constant pressure of viruliferous whiteflies and misuse of pesticides.

During the duration of the project, a total of 35 plots of EAP 9510-77 (later released officially as 'CENTA San Andrés') were established nation-wide in El Salvador, with yields ranging between 1,085 and 1,200 kg/ha. The higher yields can be explained by the planting of these bean plots during the end of the rainy season (August-November), when whitefly populations are at its lowest level. Planting at this time is required to register a new cultivar in El Salvador. This new variety yielded over 700 kg/ha under the very severe whitefly (>200 adult whiteflies/plant) and begomovirus incidence that occurred in the 2001-2002 dry season, with only one application of insecticide (as compared to more than 10 applications made by local farmers in the valley of Zapotitán). The local landrace 'Rojo de Seda' and one of the old improved cultivars, 'CENTA Cuzcatleco' yielded under 100 kg/ha under these conditions. The insecticide recommendation was later increased to two applications in seasons of very high whitefly incidence, in order to increase yields to the yielding capacity of this improved variety (1-1.4 MT/ha).

The new bean line was also subjected to cooking and tasting tests with both male and female farmers. All farmers accepted the new line because of its shorter cooking time (60 min vs. 70 min for 'Rojo de Seda'); thick broth and good taste. From the economic point of view, the net benefit for the EAP line was US \$ 908.15/ha vs. US \$ 786.81/ha for the latest commercial cultivar released in El Salvador (CENTA 2000), during the August-November period. During the peak whitefly season, CENTA 2000 cannot be grown without multiple insecticide applications, whereas the EAP line would yield a net profit in excess of US \$ 500/ha, with only two applications of insecticide, and production costs ranging between US \$ 80-100/ha. The statistical analysis was done using 'paired plots' and their entire area (500 m²) as the sampling unit. Yields are expressed as kg/ha, and their

significance was calculated by using the Student 't' test. The potential impact of this new bean variety for El Salvador amounts to over 33 million dollars, assuming a total bean area of 64,000 has, a single planting, and the current minimum price of US \$ 520/MT. CENTA San Andrés is expected to be adopted in other Central American countries that consume red-seeded common beans, such as Honduras, Nicaragua and Costa Rica.

The objective of bringing common bean back into cultivation in the Valley of Zapotitan, during the December-April dry season, is already a reality. This past summer season (November 2003-March 2004) approximately 35 has of CENTA San Andrés were planted in Zapotitán. CENTA is actively multiplying seed of this new cultivar for further distribution in Zapotitán and El Salvador. CENTA San Andrés has the best commercial and virus-resistance characteristics of any red-seeded common bean cultivar ever produced in Latin America.

Food security is an important concern of this project and most developing countries, but traditional crops, such as common bean, are not going to take any small-scale farmer or country out of poverty because of their relatively low market value. Fruit and horticultural crops, on the contrary, make possible the generation of substantial income in small land areas. For instance, a hectare of maize or beans does not produce more than US \$ 200-300 per growth cycle, whereas a tenth (1/10th) of a hectare planted to tomato or chilli may produce over US \$ 1,000 per planting. The Mesoamerican subproject of the TWFP has chosen two major horticultural crops: tomato and peppers (sweet peppers and chillies) to develop IPM packages that can be used by small-scale farmers to diversify their traditional cropping systems (not to replace them) and, thus, increase the profitability of their limited landholdings.

As in the case of common bean, the most viable strategy would be to use virus-resistant tomato and pepper varieties, but despite their Latin American origin, these crops have not been improved genetically to withstand all of the production problems that affect these crops in this region. So far, production of tomatoes and peppers in Latin America has only been possible due to the excessive use of pesticides, that make these products unsuitable for the European or North American markets, but not for the local market where the extremely toxic pesticide residues these products contain, are not detected.

In 2002-2003, a new project was implemented in El Salvador by the Financial Transactions Report Analysis Centre (FINTRAC) and the Centre of Investment, Development and Export of Agribusiness (IDEA). Fintrac's primary mission is to "increase the productivity and sales of our clients in a sustainable fashion. This involves incorporating small-scale producers to local, regional and global supply chains through innovative technical interventions in the field, as well as market analysis and linkages with commercial buyers". One of the "innovative technical interventions in the field" was the adoption of the microtunnel technology promoted in this area by the Tropical Whitefly Project financed by DFID. The site chosen by FINTRAC in El Salvador, was San Juan Opico, a few kilometers away from the main pilot site (Zapotitan/Ciudad Arce) of the TWFP in that country. The project started with 16 farmers in San Juan de Opico and approximately 33 has of land planted to different horticultural crops, mainly tomato and peppers protected inside microtunnels (**Figures 20 and 21**). The goal of this project was to cover 280 has last year, and it has been operating successfully for two years in El Salvador and Honduras. Another project that has shown interest in the TWFP's work in El Salvador, and particularly in the micro-tunnel technology, is the Swisscontact-Helvetas Consortium in Honduras. Their

mission is to “help the development of small- and medium-sized enterprises in the processing and marketing of agricultural products”.



Figure 20. Adoption of the micro-tunnel technology by independent small-scale farmers in San Juan Opico, El Salvador.



Figure 21. Tomato grower in San Juan Opico, El Salvador, showing results obtained with the micro-tunnels recommended by the TWFP.

This project plans to expand soon into Nicaragua. More recently, in the Cauca Valley of Colombia, where whitefly-transmitted viruses are affecting snap bean and tomato production, a

group of horticultural farmers called ‘mesa agriculture’ has also contacted the TWFP for technical assistance with the micro-tunnel technology. This information is already available to interested users through our Web Page, and a hard copy of this publication is being prepared as well.

The microtunnel technology is also being adopted by independent farmers in the pilot sites of Zapotitán (**Figure 22**) El Salvador, and Yucatán, Mexico (**Figures 23 and 24**). The situation in Yucatán is being carefully monitored, as small-scale farmers begin to see the advantages of using the micro-tunnels again. This project can be very influential in counteracting the biased ‘technical assistance’ provided by the agrochemical companies/distributors in this and other regions of Middle America.



Figure 22. Adoption of micro-tunnel technology in Zapotitán, El Salvador.



Figure 23. Adoption of micro-tunnels by independent farmers in Yucatán, Mexico.



Figure 24. Vegetable production in Yucatán using micro-tunnels.

The project is also interested in following up the socio-economic impact of implementing simple IPM measures for pest management in loroco in El Salvador, particularly within the concept of ‘backyard’ or ‘peri-urban’ agriculture with a gender focus. From the biological point of view, that is, pest control, the work conducted so far has shown that the IPM measures implemented are effective and sustainable. The collection of data continues in the pilot site in order to let the crop reach the most productive stage at the end of this year.

The TWFP is very interested in making the best use of the considerable amount of information gathered on the identification of whitefly species and biotypes, viruses transmitted by *Bemisia* and *Trialeurodes* species, crops affected, and temporal and spatial patterns of whitefly/virus spread, to develop reliable disease forecasting methods. The purpose of developing this pest/disease prognostic capacity would be to alert farmers to environmental conditions suitable for the development of high whitefly populations. The following is an abstract of a paper published in *Virus Research* by Francisco J. Morales and Peter Jones of CIAT. This is the most complete analysis on the ecology of *Bemisia tabaci* in tropical America. "Whitefly-transmitted geminiviruses are the most important constraint to common bean and horticultural crop production in the lowland tropics, particularly in Latin America. Currently, over 30 distinct species of geminiviruses transmitted by the whitefly *Bemisia tabaci* attack common bean, tomato, pepper, cucurbits and other horticultural crops in the lowlands and mid-altitude valleys of the American tropics and subtropics. A climate probability model (FloraMap) was obtained using 304 geo-referenced locations where *B. tabaci* and geminiviruses cause significant damage. *Clustering of the 304 points produced a simple model with two climatic variables: a dry season of at least 4 months with less than 80 mm of rain, and a mean temperature of the hottest month above 21° C.* A modified Koeppen climate classification showed that 55% of the geminivirus-affected localities are in the Tropical Wet/Dry region; 21.6 % in the Tropical and Subtropical Dry/Humid climates, and the remaining locations belonged to the wet Equatorial and Trade Wind Litoral climates. These findings are expected to help implement sustainable Integrated Pest Management practices in mixed cropping systems and different environments throughout the tropics". **Figure 25** shows the distribution of the points and the areas of Latin America where *B.*

tabaci and the begomoviruses that this whitefly species transmits, may cause significant yield losses.

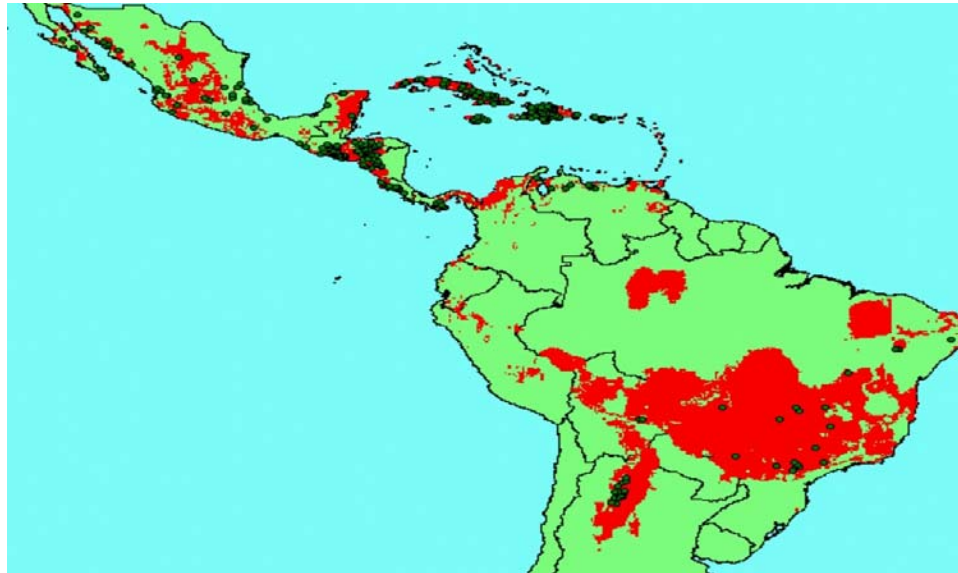


Figure 25. Distribution of hot spots (green points) and risk areas (in red) for *Bemisia tabaci* and whitefly transmitted begomoviruses.

Achievements and Obstacles

This subproject has demonstrated beyond any doubt that it is possible to manage pests and diseases as difficult and severe as those associated with the whitefly *Bemisia tabaci*, in agricultural regions where farmers have had to abandon the cultivation of susceptible basic food and cash crops. It has taken us three years to bring common bean, tomato, pepper and chilli production back to the Valley of Zapotitán (our pilot site) and other horticultural areas of El Salvador and Yucatán, Mexico, but it has been done and the small-scale farmers who have adopted the IPM methods recommended and validated by the project, have greatly profited from this work. The challenge of this subproject was not to increase productivity by 10 or 30%; whitefly transmitted viruses have the potential to cause total or significant (>50%) yield losses on all crops attacked almost every year during the prolonged dry season. Therefore, farmers prefer not to plant at all, forgoing the opportunity to gain badly needed income from their limited land resources for almost half of the year, and take their products to the market when the demand and prices are higher because of the winter season up north and the lack of internal production to supply the demand for food at that time of the year.

These achievements have not come easy for different reasons. First, most national agricultural research institutions (NARIs) in Latin America have been so badly affected by the chronic economic situation of the region, since the 1980s, that there are very few national scientists to work with or, worse yet, trained or motivated enough to conduct field experiments and work with small-scale farmers. Often, there are not enough vehicles to supervise the work, and the few vehicles available spend more time on the repair shop than on the road. The best and most experienced scientific personnel has retired or left their institutions, and the young replacements

are not well trained, Extension personnel are used to delivering ready-made products that farmers know how to use without much additional information (e.g. a new variety and a list of pesticides available in the market). They do not know how to promote more complex IPM packages that require farmer training and education on the principles of IPM and direct and indirect economic benefits derived from the adoption of IPM practices. There is also a constant rotation of personnel at NARIs from task to task, often in response to the pressure of large-scale growers on the Ministry of Agriculture. And, finally, the timely utilisation of funds is hindered by the bureaucracy that characterises official institutions in developing countries.

Despite these obstacles, the outputs proposed by the project have been achieved as mentioned earlier. The subproject has shown that it is possible to grow common bean during the dry season of the year, without the need to apply insecticides every day or every other day as it is the belief among farmers due to the whitefly problem. We have shown that the virus-resistant new cultivar only requires a couple of insecticide applications to produce above the national average, but farmers get nervous when they are not applying pesticides every day. Obviously, the constant pressure of the pesticide companies on farmers has a lot to do with pesticide abuse in the region. This is an aspect to be dealt with in the next phase of the project, particularly in Yucatán, where small-scale farmers are just beginning to realise that they should not have given up the use of micro-tunnels in order to rely exclusively on new, costly pesticides against whiteflies.

The promotion of physical barriers has been very successful to bring tomato and pepper production back during the dry season because they have seen that plants are not affected by vectors or viruses during their critical growth period, and the yield and quality of the produce results in higher prices and income. This technology has been practically 'stolen' by other projects and private organizations and has already been successfully used outside the pilot sites on a larger scale. Unfortunately, they have adopted only the physical materials (micro-tunnels) but not the IPM package that complements these physical barriers. The subproject needs to work on these aspects with the small- and medium-scale farmers who have already adopted this technology without all the complementary information.

All the previous sociological and biological work has provided enough material to understand the whitefly problem. This subproject and on-going CPP projects that have benefited from these data, are beginning to analyse all of this information in order to understand the ecology and epidemiology of whiteflies and whitefly-borne viruses with a view to implementing more rational and sustainable IPM measures. The current CPP Project ("Adaptive evolution within *Bemisia tabaci* and associated *Begomoviruses*: A strategic modelling approach to minimising threats to sustainable production systems in developing countries" by Frank van den Bosch and M.J. Jeger, is currently analysing the effect of the IPM measures implemented in the Mesoamerican subproject on whitefly/begomovirus control, particularly regarding their long-term consequences. This exercise has been possible thanks to the biological data provided by this subproject to the main CPP investigators.

Contribution of Outputs to Developmental Impact

The Tropical Whitefly project is a rare example of an agricultural research project that addresses a concrete food production problem and, at the same time, demonstrates the potential to make a substantial contribution to food security (recovery of abandoned areas for food production),

poverty alleviation (increased income from limited land resources), improved health standards (minimum pesticide use), and environmental sustainability (discourage migrant agriculture).

The Mesoamerican subproject of the TWFP has filled a large vacuum created in the past two decades by the drastic reduction in technical assistance to small-scale farmers, caused by a chronic regional economic crisis (external debt) that has affected both national and international agricultural research institutes.

The focus of the Mesoamerican subproject on mixed cropping systems, including both staple and high-value crops, responds to small-scale farmers' attempts to diversify their cropping systems in order to maximise the productivity and profitability of their scarce land resources.

The Mesoamerican subproject has inserted its research activities in the research agenda of the national agricultural research programs it has worked with, thus making sure that our respective research agendas coincide on the basic need to produce food in a sustainable manner.

The Mesoamerican subproject of the TWFP has implemented sustainable IPM technology that helps small-scale farmers produce more food and improve their livelihoods, but with minimum use of pesticides. Thus, the project has the potential to benefit the environment by reducing pesticide use. In fact, pesticide abuse has been an increasingly important problem ever since the focus on crop improvement was changed through pressure from environmentalist groups to natural resource management, unfortunately independently of any food production component. In the absence of technical assistance or concrete food production technology, small-scale farmers can only protect their investment through the intensive use of pesticides, thus, poisoning themselves, their families, their agricultural products, the environment and, finally, all the unaware consumers of heavily contaminated produce in developing countries that do not have food safety standards. Natural resource management projects without a food/crop production component, alleviate neither poverty nor hunger in developing countries.

A recent study by DFID claims that it takes US \$ 11,000 to take a single person out of poverty in Latin America. This figure probably represents the radical change that has taken place in agricultural research priorities in Latin America since the 1990s, from crop improvement to natural resource management. With proper technology and a viable crop production component, the TWFP has shown that a resource-poor farmer can overcome poverty with an additional investment of less than US \$ 1,000. The economic feasibility of this strategy has been demonstrated in the Central American sub-project, where both the private sector and the Government of El Salvador have invested successfully for the past two years in agricultural projects aimed at increasing horticultural production, by facilitating credit to small-scale farmers using the technology promoted by the TWFP. These outputs, namely virus-resistant varieties and sustainable IPM technologies, are already in the hands of farmers in our current pilot sites.

Now, we have to scale up these IPM packages to reach all the regions affected by whiteflies and whitefly-transmitted viruses in the tropics. However, much more crop improvement efforts are necessary to improve horticultural crops, mainly tomato, sweet and hot peppers and cucurbits, for resistance to the whitefly/virus pests specific to each region, particularly in Latin America. Most of this work could be carried out by the International Agricultural Research Centres (IARCs), if the supporting industrialised nations realised that, without a long-term, solid crop improvement

program at these centres, poverty alleviation will continue to be an impossible goal. Also, over-reliance in biotechnology at the expense of the traditional genetic improvement field work that IARCs used to do in their most productive years, has greatly slowed down the process of crop improvement rather than accelerate it, as was expected.

Follow-up Action

Phase III of the TWFP-Mesoamerica will focus on technology dissemination, impact assessment and policy issues.

In the case of common bean, the subproject needs to assess the socio-economic impact of the work done so far in the pilot sites. We need to document how fast is the new cultivar, CENTA San Andrés, going to be adopted in the valley of Zapotitán, in El Salvador and neighbouring countries. How much are small-scale farmers going to benefit from having this new variety, and the opportunity to grow it throughout the year. How much are we going to reduce production costs because of the possibility of reducing pesticide applications to 10-20% of the current application volumes. This point, however, requires more participatory work with farmers, which is contemplated in Phase III. Finally, how much is the Government saving in food imports and how is the urban consumer benefiting from increased bean production in the region.

In the case of horticultural crops, the technology needs to be disseminated following a true farmer participatory approach, and through farmer field schools. The main objective of this approach is to explain to small-scale farmers the benefits of the physical control methods and the biological principles behind the IPM measures recommended, rather than to teach the method itself. In those areas where farmers have been using the method for three years, we will conduct an impact assessment exercise. Linked to this particular activity, we have the possibility of study the impact of policy (e.g. intervention of the Salvadoran and Mexican Governments to reduce the cost of the nets used for physical control, and/or provide loans for small-scale farmers who want to adopt this technology).

The loroco case in El Salvador will be followed up into the third year of production before an impact assessment study is conducted. The dissemination of technology for this and previous crops will be channelled through different communication media: publications, radio, television, and electronic media in order to reach as many potential users as possible. Fortunately, the TWFP has already worked in all of the countries affected by whiteflies and viruses in this region, which facilitates the dissemination of information through known channels.

One of the past research activities with the most potential impact is the development of whitefly-transmitted virus-resistant tomato germplasm. Segregating populations are already in the hands of two national programs (CENTA-El Salvador and INIFAP-Mexico). This work is proceeding with the collaboration of AVRDC. This is the first time that we have identified sources of resistance to whitefly-borne viruses of tomato in Latin America.

A Concept Note has been drafted, which describes the proposed approaches to deliver these technological breakthroughs to small-scale farmers. Two previous meetings organised by the System-wide IPM Programme have served as the channel to link up with other SP-IPM groups,

particularly with the specialists on Farmer Participatory Research (lead by CABI), in order to agree on the best approach to execute Phase III.

Publications and Trainings

CIAT. 2003. "A United Effort against a Global Pest: Helping Poor Farmers Reduce Crop Losses and Grow more Food in a Sustainable Way" A5 Colour brochure. 10pp. [English] (D)

CIAT "Tropical Whitefly IPM Project: Collaborative Research to solve a Global Problem" Colour brochure 20pp.

Morales, F.J. 2003. The Whitefly *Trialeurodes vaporariorum* as a potential Constraint to the Development of Sustainable Cropping Systems in the Mesothermic Valleys of the Bolivian Highlands, August 2003, CIAT, Colombia, 11pp. (C)

Morales, F.J. 2003. Promoting economic growth in small farm communities of El Salvador through suitable pest management, p.8, Agricultural Research and Extension Network, No.47, January 2003, ISSN 0951-1865. (D)

Morales, F.J.; Martinez, A.K.; Velasco, A.C. 2003. New outbreaks of begomoviruses in Colombia. *Fitopatol. Col.* 26: 75-79. (A)

In Press

Morales, F.; Jones, P. The ecology and epidemiology of whitefly-transmitted Viruses in Latin America. *Virus Research.* (A)REPORTED AS IN PRESS IN PPR1 SEP 03.

Morales, F.J. Integrating IPM and Sustainable Livelihoods in Central America. *In:* Pachico, D. (ed.). *Scaling up and out: Achieving widespread impact through agricultural research.* Centro Internacional de Agricultura Tropical (CIAT), Cali, Colombia. NB. On 20.1.04 PL forwarded an internal CIAT email dated 5 Sep 03 saying: "Still a way to go and annual reports intervening - so 2004 the likely publication time".

Internal Reports

Morales, F. 2003. The Systemwide Tropical Whitefly IPM Program. *In:* CIAT (Centro Internacional de Agricultura Tropical). *Integrated Pest and Disease Management in Major Ecosystems: Annual Report, Project PE-1.* Cali, CO. p. 224.

CIAT in Perspective 2001–2002: From Risk to Resilience. Rebuilding El Salvador's granary through integrated management of whiteflies. Cali, CO. p. 32.

Morales, F. 2002. The Systemwide Tropical Whitefly IPM Program: From Phase I to Phase II. *In:* CIAT (Centro Internacional de Agricultura Tropical). *Integrated Pest and Disease Management in Major Ecosystems: Annual Report, Project PE-1.* Cali, CO. p. 238.

Anderson, P. 2001. Systemwide Project on Integrated Sustainable Management of Whiteflies as Pests and Vectors of Plant Viruses in the Tropics. *In:* CIAT (Centro Internacional de Agricultura Tropical). *Integrated Pest and Disease Management in Major Ecosystems: Annual Report, Project PE-1.* Cali, CO. p. 208.

Other Dissemination of Results: Training

La Mosca Blanca como transmisora de enfermedades virales. (Field guide for Farmers and Extension personnel). Tropical Whitefly IPM Project.

Morales, F. 2004. Whiteflies (Homoptera:Aleyrodidae) As Virus Vectors. Presentation 15th IPPC, Beijing, China (Abstract).

Control Físico de Mosca Blanca (*Bemisia tabaci*) para el manejo de enfermedades virales en cultivos hortícolas (Field guide for Farmers and Extension personnel) Tropical Whitefly IPM Project.

Perez, Juana E. 2004. Manejo y control de mosca blanca utilizando microtuneles en los cultivos de tomate y chile dulce. CENTA-CIAT.

Escamilla, E. 2004. Uso de coberturas para el manejo de afidos en el cultivo de Loroco. CENTA-CIAT.

Perez, Juana E. (2004) Evaluación de líneas autofecundadas e híbridas de tomate por su resistencia a virus transmitidos por mosca blanca. CENTA – CIAT.

Betancourt, M. De J.; Pérez Cabrera, C.A. 2004. Parcelas Demostrativas de Producción de Frijol con la Variedad CENTA San Andrés en el valle de Zapotitán. CENTA – CIAT.

The Central American sub-project supervised on M.Sc. Thesis, University of El Salvador, and trained three scientists from the NARIs of El Salvador, México, and one from the Univ. of El Salvador, at CIAT.

Listing and reference to key datasets generated:

Morales, F. 2004. Socioeconomic Survey. El Salvador. Access. Tropical Whitefly IPM Project.

Morales, F. 2004. Begomovirus Characterization DB. El Salvador/Mexico. Word Document. Tropical Whitefly IPM Project.

Morales, F. 2004. *Bemisia tabaci* biotypes. El Salvador and Mexico DB. Word Document. Tropical Whitefly IPM Project.

Activity 3. Developing integrated pest management components.

Monitoring of the changing situation with whitefly populations in the Andean zone

Rationale: Continuous monitoring of changes in whitefly populations and species composition in target areas is one of the most important objectives of the DFID-funded project on Sustainable Management of Whiteflies. This is needed to develop appropriate management systems and, if necessary, to modify existing systems so as to be able to cope with new situations.

Materials and Methods: In 2004 we processed a total of 105 whitefly samples (adults and pupae) collected in the Cauca Valley and northern coast regions of Colombia. Samples were taken from beans, snap beans, cucurbits, tomatoes and several other annual crops. We used RAPD techniques (primer OPA-04) to identify pupae and adults. Identification was based on morphological characteristics of pupae and comparison between RAPD patterns in samples brought from the field with those of existing mass rearings of different whiteflies maintained at CIAT (Figure 1).

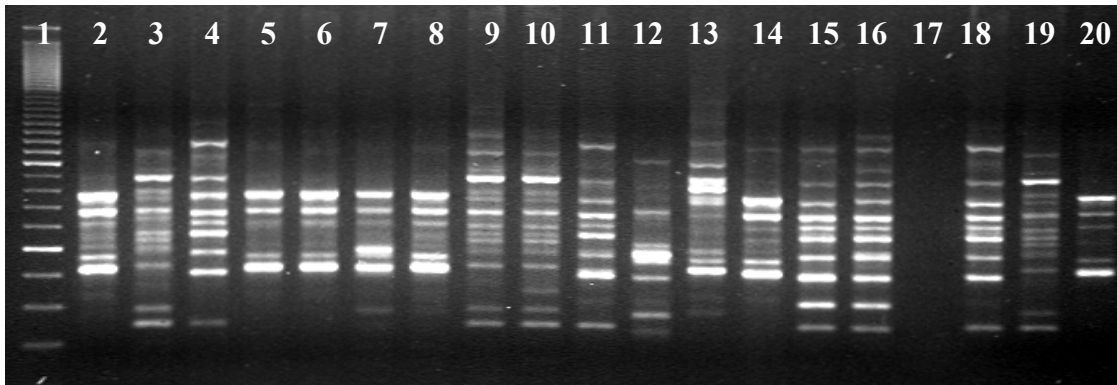


Figure 1. RAPD's for whitefly adults and pupae collected in the Cauca Valley. Amplifications using the OPA-04 primer. 1, DNA molecular marker (100pb); 2, *T. vaporariorum* from reference rearing maintained at CIAT; 3, *B. tabaci* biotype A from reference rearing; 4, *B. tabaci* biotype B from reference rearing; 5-8, adults (5-6) and pupae (7-8) of *T. vaporariorum* collected in Darién on beans; 9-10, *B. tabaci* A adults collected on soybeans in Jamundí, 11, *B. tabaci* biotype B collected on soybeans in Jamundí; 12, parasitized pupa of *B. tabaci* collected on soybeans in Jamundí; 13-14, *T. vaporariorum* adults on beans in Jamundí; 15-16, *B. tabaci* biotype B pupae on beans in Jamundí; 17, free; 18, *B. tabaci* biotype B from reference rearing; 19, *B. tabaci* biotype A from reference rearing); 20, *T. vaporariorum* from reference rearing.

Results and Discussion: Analysis of 105 samples taken in 24 locations in the Cauca Valley (Colombia) showed that 42% of the whiteflies collected belonged to the B biotype of *Bemisia tabaci*, the most aggressive form of whitefly known to date. This biotype was found affecting snap beans, tomatoes, cucumber, melon, soybeans, pepper, tobacco, and grapes. As in 2003, we found that the B biotype is now occupying niches previously reserved to the A biotype or to *T. vaporariorum*. As shown in Figure 2, species composition in the Cauca Valley has changed

drastically in the past seven years. In 1997, *T. vaporariorum* was by far the most important species, representing 73% of the samples taken while the A biotype represented 15% of samples analyzed. At present, the A biotype is difficult to find (1.6% of samples), *T. vaporariorum* represents 11% of the samples and the B biotype is the predominant species with 42% of the samples. Up to 39% of crop samples examined were affected by a combination of *T. vaporariorum* and the B biotype of *B. tabaci*.

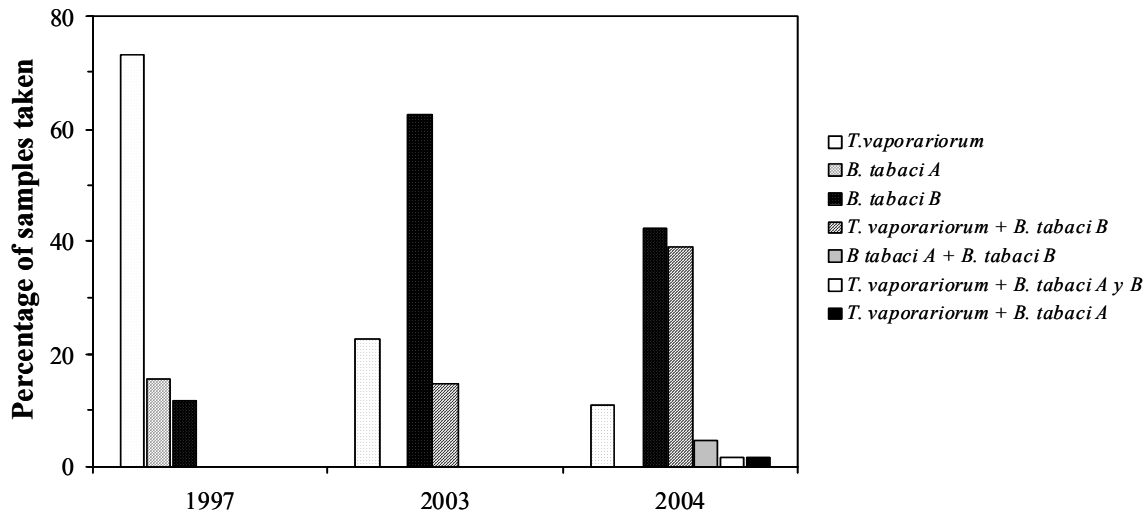


Figure 2. Changes in whitefly species composition in the Cauca Valley of Colombia (1997-2004).

Detailed monitoring of species composition on snap beans in the Pradera reference site revealed that at higher altitudes (1270-1840 m) *T. vaporariorum* is still the dominant species (**Figure 3**). At altitudes ranging from 975 to 1120 m, most individuals collected in the Pradera region belong to the B biotype of *B. tabaci* attacking different crops either alone (33.3% of samples taken) or in combination with *T. vaporariorum* (53.4% of samples). The B biotype is an aggressive form of whitefly that is causing all the serious problems described in our 2003 Report. In snap bean growing areas, it has become the causal agent of a physiological disorder known as pod chlorosis, which renders the produce useless. Most serious, it has become a very effective vector of a geminivirus that is devastating snap beans in the region.

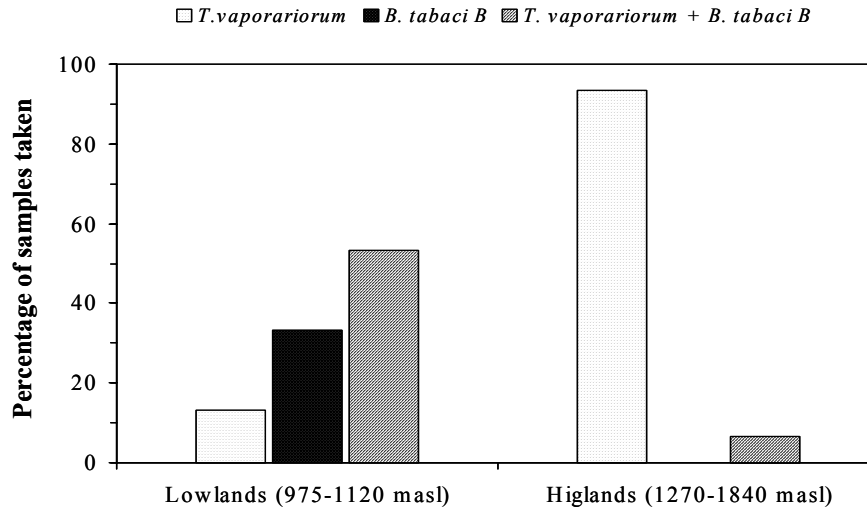


Figure 3. Whitefly species composition in the Pradera (Cauca Valley) reference site; 2004 survey. masl, meters above sea level.

Contributors: I. Rodríguez, H. Morales, C. Cardona.

Monitoring of insecticide resistance in whitefly populations

Rationale: Monitoring of insecticide resistance is another major objective of the DFID-funded project on Management of Whiteflies in the Tropics. Both major whitefly species and their biotypes in the Andean zone are the targets of excessive use of insecticides. This is reflected in ever increasing levels of resistance to insecticides and difficulties in control. The main purpose of a continuous monitoring of insecticide resistance is to develop alternative management strategies that will help to overcome resistance or delay the onset of this phenomenon.

Materials and Methods: In 2004 we established base-line data for five insecticides commonly used to control adults of the B biotype of *B. tabaci*: monocrotophos, carbofuran, carbosulfan, bifenthrin, and imidacloprid. These data will serve as the basis to establish diagnostic dosages for the species. These in turn will be used for periodic monitoring of resistance levels.

Using previously established diagnostic dosages for nymphs, we tested populations of whiteflies in the Cauca Valley in Colombia. Adult resistance levels were monitored under field conditions by means of the insecticide-coated glass vial technique. Resistance of first instar nymphs was measured using the foliage dipping technique. Systemic novel insecticides (mostly neonicotinoids) were tested using the petri dish technique (see 2003 Annual Report).

Results and Discussion: In general, it can be said that nymphal populations of both *T. vaporariorum* and *B. tabaci* biotype B are still susceptible to the insect growth regulators buprofezin and diafenthiuron and to imidacloprid, a novel neonicotinoid (**Table 1**). However, reduced responses to buprofezin in the Pradera site deserve further monitoring.

Table 1. Response (percentage corrected mortality) of nymphs of *Trialeurodes vaporariorum* and *Bemisia tabaci* biotype B to three insecticides in three consecutive growing seasons. Cauca Valley (Colombia). Diagnostic dosages in ppm.

Race	Percentage corrected mortality ^a		
	2001 B	2002 B	2003 B
<i>Trialeurodes vaporariorum</i>			
buprofezin (16 ppm)			
‘CIAT’ ^b	98.4 a A ^b	100.0 a A	97.6 a. A
La Cumbre	100.0 a A	100.0 a A	100.0 a A
Pradera	87.0 b A	77.4 a A	81.4 b A
diafenthiuron (300 ppm)			
‘CIAT’	98.2 a A	100.0 a A	96.2 b A
La Cumbre	92.6 a B	97.8 a A	100.0 a A
Pradera	88.6 a A	93.9 a A	90.5 c A
imidacloprid (300 ppm)			
‘CIAT’	100.0 a A	98.3 a A	93.7 a A
La Cumbre	92.8 b A	93.2 a A	99.0 a A
Pradera	84.9 b A	92.6 a A	93.2 a A
<i>Bemisia tabaci</i> biotype B			
buprofezin (16 ppm)			
‘CIAT’	---	98.4 a A	96.9 a A
Rozo	---	80.6 b A	87.8 b A
La Unión	---	100.0 a A	87.7 b B
Santa Helena	---	100.0 a A	92.2 b B
diafenthiuron (300 ppm)			
‘CIAT’	---	100.0 a A	91.7 a B
Rozo	---	100.0 a A	91.5 a B
La Unión	---	98.2 a A	91.7 a B
Santa Helena	---	100.0 a A	95.1 a B
imidacloprid (300 ppm)			
‘CIAT’	---	91.1 b A	98.3 a A
Rozo	---	89.3 b A	90.1 b A
La Unión	---	100.0 a A	89.1 b B
Santa Helena	---	100.0 a A	98.6 a A

^a For each species and product, means within a column followed by the same lowercase letter and means within a row followed by the same uppercase letter are not significantly different at the 5% level by LSD. Each species and product were analyzed separately.

^b A susceptible strain of *B. tabaci* biotype B maintained at CIAT.

Future work on integrated pest management of whiteflies as pests of beans and snap beans in the Andean zone should include studies on the relative efficiency of the two most important parasitoids affecting whitefly populations in the region: *Encarsia nigricephala* and *Amitus fuscipennis*. Given the excessive use of insecticides, it is important to know what is the present response of these natural enemies to some of the most commonly used insecticides. The base-line data in **Table 2** should in the future serve as the basis for possible development of insecticide tolerance in populations of these natural enemies, an optional strategy for management of the whitefly problem.

Table 2. Response^a of adults of *Encarsia nigricephala* and *Amitus fuscipennis* to different insecticides.

Insecticide	No. of Individuals tested	CL ₅₀ (CL 95%) ^b	CL ₉₀ (CL 95%)	χ^2	b ± EEM	P > χ^2
<i>E. nigricephala</i>						
methamidophos	400	0.67 (0.440 – 0.900)	4.04 (2.910–6.800)	1.72	1.64 ± 0.24	0.19 ns ^c
methomyl	400	0.00915 (0.004 – 0.015)	0.062 (0.044 – 0.110)	0.56	1.54 ± 0.30	0.45 ns
carbosulfan	400	0.09 (0.060 – 0.120)	0.38 (0.280 – 0.670)	0.22	2.04 ± 0.38	0.64 ns
cypermethrin	400	0.65 (0.040 – 1.880)	11.09 (5.680 – 21.65)	0.57	1.04 ± 0.26	0.45 ns
<i>A. fuscipennis</i>						
bifenthrin	400	0.023 (0.005 – 0.427)	0.171 (0.118 – 0.276)	1.00	1.47 ± 0.33	0.31 ns
carbofuran	400	0.074 (0.050 – 0.097)	0.380 (0.286 – 0.576)	0.70	1.80 ± 0.24	0.40 ns

^a Values of CL₅₀ y CL₉₀ in µg of active ingredient/ vial.

^b Confidence limits at 95%.

^c ns, not significant at the 5% level.

Comparison of toxicological responses of the whitefly and their parasitoids indicate that all of the insecticides tested are much more toxic to the parasitoids than to the whitefly (**Table 3**) with up to 100-fold higher tolerance in the herbivore. Nevertheless, the data show that both natural enemies studied do possess innate mechanisms of defense against toxic substances, which may be exploited by continuous mass rearing and selection for higher levels of tolerance followed by mass releases in the field. As such, resistant strains of one or both parasitoids would become management components in an integrated pest management system.

Table 3. Comparative responses of the whitefly *Trialeurodes vaporariorum* and its parasitoids *Encarsia nigricephala* and *Amitus fuscipennis* to different insecticides.

Insecticide	CL ₅₀ <i>T. vaporariorum</i>	CL ₅₀ Parasitoid	Response Ratio
<i>E. nigricephala</i>			
methamidophos	5.30 ^a	0.670	7.91
methomyl	0.25 ^a	0.009	27.77
carbosulfan	1.80 ^b	0.090	20.00
cypermethrin	37.0 ^a	0.650	56.92
<i>A. fuscipennis</i>			
bifenthrin	2.40 ^b	0.023	104.35
carbofuran	1.97 ^a	0.074	26.62

^a As determined by Cardona et al. (2001).

^b As determined by Rodríguez et al. (2003).

Contributors: I. Rodríguez, H. Morales, M. F. Montenegro, and C. Cardona.

Management strategies for whiteflies

Rationale: Whiteflies have become the target of excessive pesticide use by snap bean and dry bean farmers in the Andean zone. A management system for whiteflies that contribute to reduce pesticide use has been developed and tested with farmers in Colombia and Ecuador (see 2002 and 2003 Annual Reports). In 2004 we tested other alternatives to further reduce the need for toxic insecticides and initiated diffusion of technology activities at both sites in Colombia and Ecuador.

Materials and Methods: Two large-scale trials were conducted in areas of the Pradera reference sites where *T. vaporariorum* is still the predominant species. We compared different approaches for whitefly control based upon judicious and less detrimental use of chemicals. Seed treatments and drench applications of novel systemic insecticides were compared with the timing of foliar applications of conventional (less costly) products, in some cases with applications based upon pre-established action thresholds developed in previous experiments (see 2002 and 2003 Annual Reports). These treatments were compared with farmers' practices. These trials were used as demonstration plots for farmers in the area.

Results and Discussion: As in previous trials, and as compared with farmers' practices, alternative management strategies based on judicious timing of applications and use of action thresholds resulted in yields that did not differ from those obtained by farmers with their traditional management approaches (**Table 4**). Crop appearance, damage (sooty mold) levels, and final produce quality (as judged by farmers attending field days) did not differ either. Use of systemic insecticides as seed dressing and proper timing of foliar applications resulted in higher benefit/cost ratios with a 60-70% in the amount of applications made per cropping cycle.

Table 4. Yields (tons/ha) and economic returns obtained with different approaches for control of the greenhouse whitefly *Trialeurodes vaporariorum* in Pradera, the reference site.

Treatment	Yield (Tons/ha)		Benefit/Cost Ratios	
	Trial 1	Trial 2 ^a	Trial 1	Trial 2 ^a
Seed treatment with imidacloprid followed by two foliar applications of conventional insecticides at pre-established action thresholds.	11.1a ^b	11.9	1.43	1.77
Seed treatment with imidacloprid followed by three foliar applications of conventional insecticides at pre-established crop growth stages.	10.7a	11.5	1.38	1.62
Farmers' practices (6-7 foliar applications of conventional insecticides).	9.3a	11.9	1.14	1.65

^a Un-replicated demonstrative trial. No statistical analysis performed.

^b Means followed by the same letter are not significantly different at the 5% level by LSD.

These trials were used to initiate diffusion of technology activities in the area. A field day was organized in collaboration with ICA and the Municipal Technical Assistance Unit. Attendance was good (76 people, 15 of them women). Farmers were informed on the purposes of the demonstration plots and received training on whitefly biology and safe management of insecticides. Farmer's schools activities were initiated with 12 farmers who received training on whitefly sampling, safe management of pesticides and use of action thresholds for rational whitefly control. Diffusion activities will be strengthened if the second phase of the special project on whiteflies is approved.

Screening for virus resistance in snap beans

Rationale: In some areas, the B biotype of *Bemisia tabaci* has become a vector of a new viral disease on snap beans. There is urgent need to develop virus-resistant snap bean varieties in order to replace 'Blue Lake', the highly preferred but extremely susceptible commercial variety.

Materials and Methods: In collaboration with the Virology Unit and the Bean Breeding section, we screened 238 genotypes (snap beans and dry beans) for resistance to the new virus. We used three replications per genotype. The nursery was established in a hot spot area (La Tupia, Pradera) with high incidence of the disease. Materials were rated 43 days after planting for virus symptoms using a 1 - 5 visual scale (1, no apparent damage; 5, severe damage).

Results and Discussion: Twenty-three genotypes (9.7%) were rated resistant (damage scores 1-2); 39 (16.4%) were intermediate (2.1-3 scores), and 176 (73.9%) were susceptible (>3 in damage scores). Best materials in two consecutive trials are shown in **Table 5**. Some of these materials are well-known sources of resistance to BGMV.

Table 5. Response of selected snap beans and dry beans genotypes to a new virus disease transmitted by *Bemisia tabaci* in the Cauca Valley of Colombia.

Identification	Pedigree and Genealogy	Damage Scores ^a	
		2003	2004
EMP 496	EMP 250[A 769 x {(A 429 x XAN 252)F1 x (V 8025 x Pinto UI 114)F1}F1]F1	1.3	1.3
DICTA 113		1.0	1.3
MN 13942-22	(TLP 35xG 21212)F1 x ICTA LIGERO/-MC-1P-MQ-MC-3C-MC-MC	1.0	1.3
DOR 476		1.0	1.7
Tio Canela 75		1.3	1.7
DOR 390		1.3	1.7
BAT 304		1.0	1.7
EAP 9020-14		1.0	1.7
MR 14143-28	(RAB 651x Tio Canela 75)F1 x (RAB 608 x SEA 15)F1/-MC-2P-MQ-MC-6C-MC-MC	1.7	1.7
MN 13942-22	(TLP 35 x G 21212)F1 x ICTA Ligero/-MC-1P-MQ-MC-11C-MC-MC	1.0	1.7
MEJ 1-197	ICA Pijao x (ICA Pijao x G35877)F1/-(NN)P-(NN)Q-(NN)P	1.3	1.7
EAP 9504-30B		1.0	2.0
9653-16B-3		1.0	2.0
A 429		1.0	2.0
G 35172		1.7	2.0
MR 14143-28	(RAB 651 x Tio Canela 75)F1 x (RAB 608 x SEA 15)F1/-MC-2P-MQ-MC-3C-MC-MC	1.0	2.0
MR 14143-28	(RAB 651 x Tio Canela 75)F1 x (RAB 608 x SEA 15)F1/-MC-2P-MQ-MC-4C-MC-MC	1.3	2.0
MN 13942-22	(TLP 35 x G 21212)F1 x ICTA Ligero/-MC-1P-MQ-MC-1C-MC-MC	1.0	2.0
MN 13942-22	(TLP 35 x G 21212)F1 x ICTA Ligero/-MC-1P-MQ-MC-5C-MC-MC	1.0	2.0
URG 401	(G 35649 x G 3807) x G35023	1.7	2.0
URG 215	BAT 338 x G 35252	1.3	2.0
MEJ1-121	((ICA Pijao x G 35172) x ICA Pijao)F1/-19P-(NN)P(F8)-(NN)P	1.3	2.0
MEJ1-253	ICA Pijao x (ICA Pijao x G 35877)F1/-(NN)P-(NN)Q-(NN)P	1.3	2.0
G 5746		3.0	5.0
Blue Lake ^b		5.0	5.0

^a On a 1-5 visual scale (1, no damage; 5, severe damage).

^b Susceptible commercial check.

Contributors: J. M. Bueno, M. Castaño, F. Morales, S. Beebe, C. Cardona.

Publications

Book Chapters-In Press

- Anderson, P.; Bellotti, A.; Cardona, C.; Hanson, P.; Legg, J.; Morales, F.; Riis, L. 2004. Sharing Methods, Comparing Results. *In:* Whiteflies and whitefly-borne viruses in the tropics: Building a knowledge base for global action.
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Activity 4. Identification of genomic regions responsible for conferring resistance to whitefly in cassava.

Introduction

Whiteflies are one of the most serious pest and disease vectors that affect agricultural production around the world. In cassava (*Manihot esculenta* Crantz), whitefly can cause between 70 to 80 percent of yield loss. The most important source of resistance genes was a genotype MEcu-72 (Arias, 1995). Due to the importance of whiteflies as a pest, it is necessary to know about the nature of genes that confer resistance in genotypes like MEcu-72. For this purpose we are using F1 segregation and the genetic expression of the cross MEcu-72 (resistant genotype) x a very susceptible genotype (MCol-2246) and molecular markers. This would help to accelerate selection of resistant materials to the whitefly and also to isolate resistant genes. It is hypothesized that these resistant genes may also be effective against other whitefly species, especially *Bemisia tabaci*, the species that is a vector of ACMD, a virus that causes severe crop losses in Africa and Asia. Whitefly resistant genotypes (such as MEcu 72) from the neotropics are displaying resistance to *B. tabaci* in greenhouse trials being carried out by NRI in the UK (CIAT Progress report 2003).

The application of molecular genetic analysis for cassava breeding has been limited compared to other crops. Recently progress has been made in the development of genomic and bioinformatics tools to increase our knowledge of cassava genome structure and cassava gene function. Expressed Sequence Tag (EST) provides an immediate and productive method of gene discovery. In cassava a total of 14168 ESTs were obtained in CIAT and Perpignan University (Lopez, et al, submitted), of these 105 have SSRs, for which we designed primers.

Materials and Methods: For the present work we have used the F1 cross (family CM 8996, 276 individuals) between MEcu-72 (as the resistance parent) and MCol-2246 (as the susceptible parent) elite cassava cultivars from Ecuador and Colombia, respectively. The parents and their offspring were evaluated in the field in two places: Nataima (Tolima) and Santander de Quilichao (Cauca). With this evaluation we intend to identify the gene segregation in the offspring and select the resistant and susceptible materials. Both parents were evaluated with 343 cassava SSRs (Simple Sequences Repeat) (Mba et al, 2001) including 156 cDNA SSRs developed (Mba et al, submitted). We are using AFLPs (Vos, et al, 1995) and to find markers associated with resistance for mapping and ultimately cloning the resistant genes. We are using silver staining to visualize the allelic segregation of the markers. Cassava RGAs primers were done in the parents and the polymorphisms were mapped in the F1.

We designed primers SSRs from ESTs sequences using the software Primer 3 and these SSR were amplified in the parents and the polymorphisms were mapped in the F1.

Results

Field evaluation

The field evaluation showed high pressure being exerted by the *A. socialis* in Nataima (2003 and 2004), and Santander de Quilichao (2004) where test materials had high damage ratings;

however, some materials had lower levels of damage in the evaluations. We can conclude that these genotypes show a resistance level similar to parental MEcu 72. These evaluations were analyzed with the molecular markers to find putative associations.

SSRs from ESTs

We designed 51 pairs of SSRs primers (**Table 1**) of which 29 were polymorphics for the cross (**Figure 1**).

Table 1. SSRs from ESTs primers designed.

No.	EST Name	Motif	No. Repeat	No.	EST Name	Motif	Repeat	No.
1	cn1375-1	atgg	5	27	cn1304-1	atg	9	
2	cn1004-1	tatt	6	28	cn1351-1	aga	10	
3	cn1098-1	aga	6	29	si.03.G1.5-1	cca	10	
4	cn1388-1	tct	6	30	gi17923193gbBM260153.	taa	11	
5	cn1457-1	agc	6	31	rni.06.I21.5-1	tct	11	
6	cn255-1	tcc	6	32	si.02.O10.5-1	aat	11	
7	cn416-2	gat	6	33	cn1635-1	aag	12	
8	cn44-1	tta	6	34	m.01.H14.5-1	tc	12	
9	cn700-3	ttc	6	35	si.01.E12.5-1	tc	12	
10	c.04.C18.5-3	atg	7	36	cn1460-1	ag	13	
11	c.05.I1.5-1	tct	7	37	cn1498-1	at	13	
12	cn1186-1	ttc	7	38	cn1587-1	ata	13	
13	cn2269-1	tgg	7	39	cn2418-1	ag	13	
14	cn393-1	cat	7	40	m.04.K18.5-1	ct	13	
15	cn732-1	aag	7	41	aflp_28-2	ga	15	
16	cn764-2	tca	7	42	m.06.H4.5-1	ct	15	
	gi17922797gbBM259765.1B							
17	M259765-2	aag	7	43	rni.06.N9.5-1	at	15	
18	m.04.K21.5-1	ctt	7	44	cn1009-1	ct	16	
19	m.05.I2.5-1	ttc	7	45	cn1722-1	tct	17	
20	rni.06.N10.5-1	tta	7	46	m.05.L3.5-1	ag	17	
21	rni.09.D10.5-1	ctg	7	47	cn47-1	ct	18	
22	si.03.B22.5-1	tct	7	48	m.09.N13.5-1	ct	18	
23	cn1131-1	tcc	8	49	gi17922797gbBM259765.1BM259765-1	agc	19	
24	m.10.J19.5-1	gat	8	50	m.08.G23.5-1	at	20	
25	m.11.K5.5-1	gat	8	51	cn1880-1	at	29	
26	rni.05.L17.5-1	tga	8					

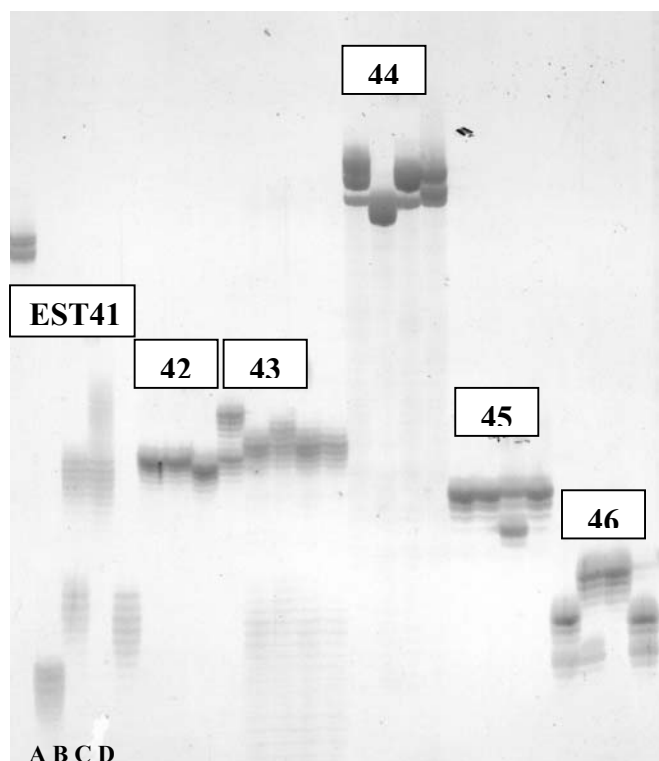


Figure 1. SSRs from ESTs in parentals, A is MNig2, B is CM21772 (both parentals used in ESTs), C is MEcu 72 and D is MCol 2246.

Mapping

For the construction of a linkage map 246 markers were analyzed, 103 SSRs, 2 RGAs, 121 AFLPs and 20 SSRs from ESTs. A genetic linkage map of cassava was constructed with 111 markers segregating from the heterozygous female parent (MEcu-72) of an intraspecific cross (**Figure 2A and 2B**). The map consists of 20 linkage groups, which represent the haploid genome of cassava. These linkage groups span is 879.8 cM and the average marker density is 1 per 7,9 cM. The position of 111 markers, shown in the **Figure 2**, on the framework (LOD = 25 and tetha (θ) = 25) molecular genetic map of cassava. Map distances are shown in Kosambi map units and analyzed by Q gene. So far, 41 SSRs markers were mapped on the cassava framework map (Fregene et al, 1997), the other 70 markers are new. The molecular data are being analyzed using QTL packages (Q gene) to determine linkages between the SSR, RGAs and AFLPs markers and the phenotypic characterization.

Association between Molecular Markers and Resistance

The molecular data are being analyzed using QTL packages (Qgene) and Simple Linear Regression at the 5% level was done using SAS. Putative associations were found between molecular markers and the field phenotypic characterization (65 markers SSRs, RGA and AFLPs, shown by *bold* in the **Figure 2A-2B**). We observed that SSRY39 marker anchored in the linkage group K are associated with the resistance.

Figure 2A. Location of putative QTL's (identified by colors) for partial resistance to whitefly in three localities on the female (MEcu-72) derived framework map. These results are based on Qgene analysis. Distances in centimorgans (cM) and significance levels are indicated on the right. The most significant markers are indicated in bold and blue square show the Linkage Group K which is localized a putative QTL in the marker *SSRY39* (identified by *bold*).

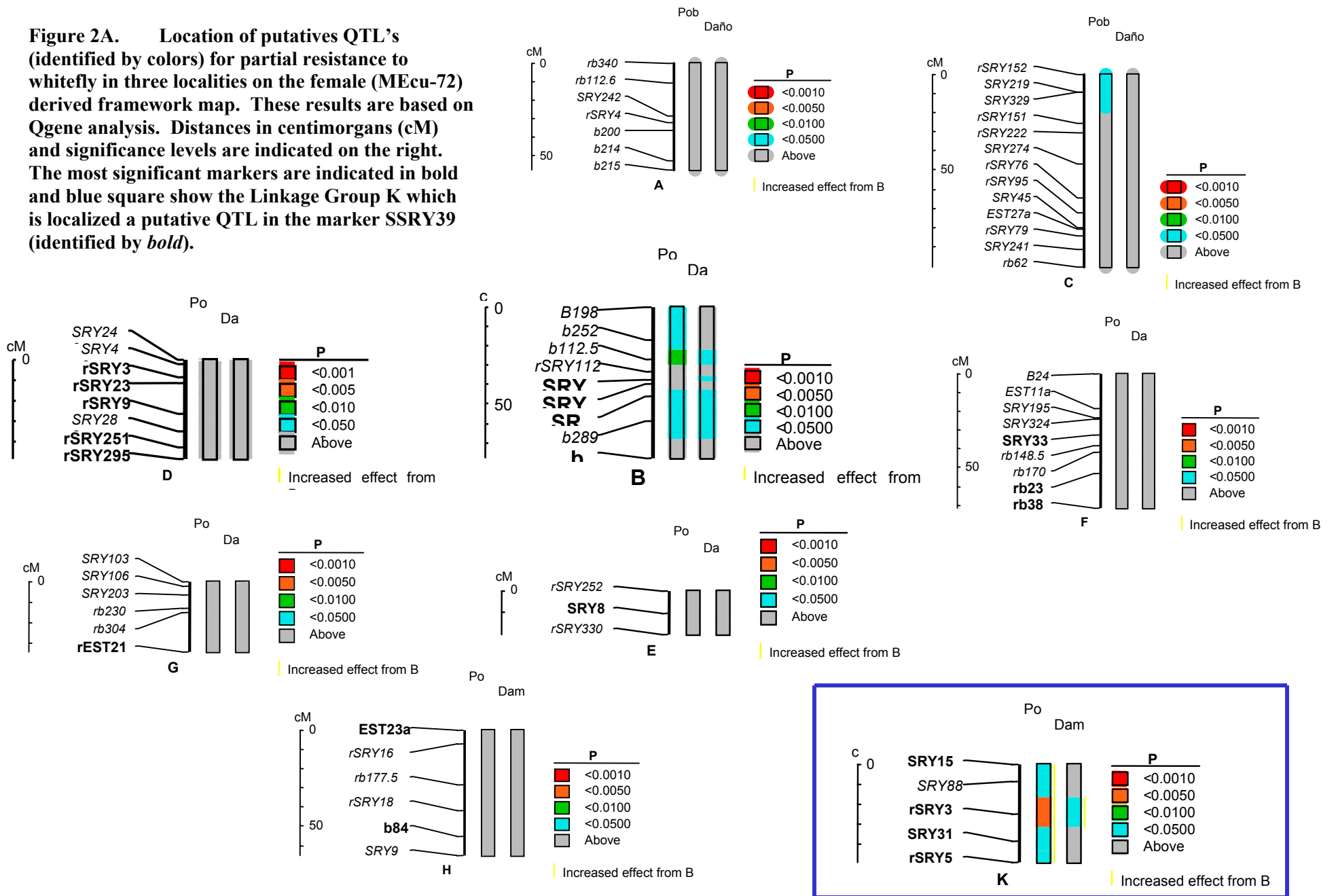
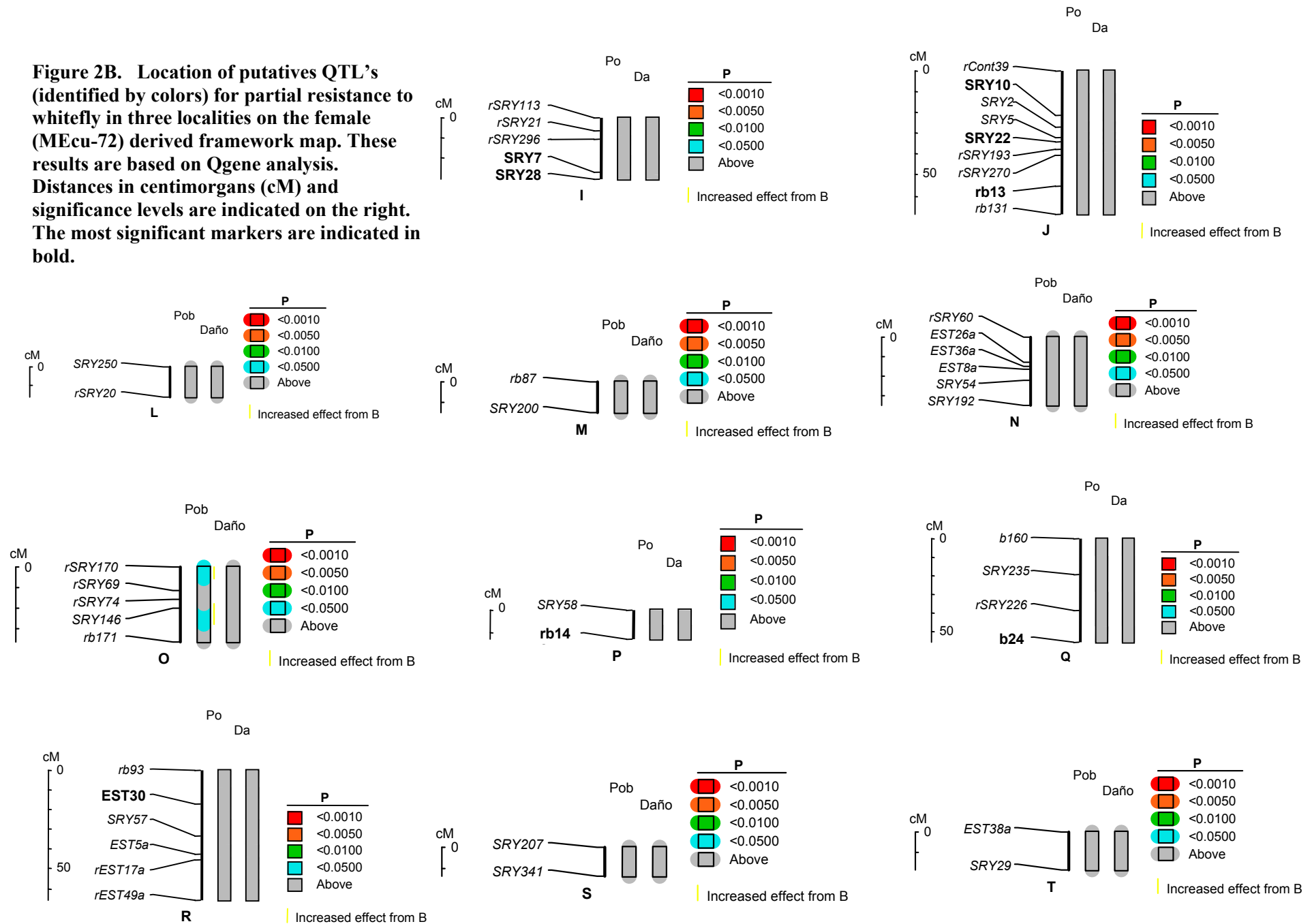


Figure 2B. Location of putatives QTL's (identified by colors) for partial resistance to whitefly in three localities on the female (MEcu-72) derived framework map. These results are based on Qgene analysis. Distances in centimorgans (cM) and significance levels are indicated on the right. The most significant markers are indicated in bold.



Ongoing Activities

- Saturation of linkage map of MEcu 72, using SNPs.
- Design of SCARs for marker-assisted selection.
- QTL analysis for whitefly resistance.
- Subtractive hybridization of the amplicon MEcu 72 (tester) and MCol 2246 (driver), during which amplified portions of differentially expressed genes are enriched and common sequences are depleted.
- Cloning and screening of the resulting products of expressed sequences during the defense response of MEcu 72 to whitefly attack.
- Microarray of clones in order to identify differentially expressed sequences.

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