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Ecologically Sustainable Cassava Plant Protection in South America and Africa: An Environmentally Sound Approach

1996 Annual report of Activities in South America



Prepared by project personnel in Colombia and Brazil

South American component of a global project involving:

Centro Internacional de Agricultura tropical (CIAT) at Cali, Colombia;
The International Institute for Tropical Agriculture (IITA) in Nigeria,
and the Empresa Brasileira de Pesquisa Agropecuária, Centro Nacional
de Pesquisa de Mandioca e Frutas Tropicais (EMBRAPA/CNPMPF) at
Cruz das Almas, Bahia, Brazil.

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“Ecologically Sustainable Cassava Plant Protection in South America and Africa: An Environmentally Sound Approach”

Technical Report on Activities conducted during 1996 by PROFISMA (Proteção Fitossanitária Sustentável da Mandioca): the South American component of a global UNDP project

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Table of Contents

1 EXECUTIVE SUMMARY	1
2 PROJECT COORDINATION	13
2.1 COORDINATION AT CIAT, BY STEVEN LAPOINTE	13
2.2 COORDINATION AT EMBRAPA/CNPMP, BY ARISTOTELES PIRES DE MATOS	17
2.3 PHASE TWO PROPOSALS, BY LINCOLN SMITH	19
3 TRAINING AND PARTICIPATORY RESEARCH	22
3.1 TRAINING, BY BERNARDO OSPINA PATIÑO	22
3.2 PARTICIPATORY EVALUATION OF TECHNOLOGY TESTING TRIALS	27
4 STRATEGIC RESEARCH AT CIAT	52
4.1 BIOLOGICAL CONTROL OF CASSAVA GREEN MITE, BY LINCOLN SMITH	52
4.2 ENTOMOLOGY, BY ANTHONY C. BELLOTTI	80
4.3 PLANT PATHOLOGY, BY ELIZABETH ALVAREZ AND JOHN B. LOKE	84
4.4 VIROLOGY, BY LEE A. CALVERT	95
4.5 CROP MANAGEMENT, BY MABROUK EL-SHARKAWY, LUIS FERNANDO CADAVID, SARA MEJÍA DE TAFUR, AND MANUEL GERARDO CAYÓN	100

5 STRATEGIC RESEARCH AT CNPMF	108
5.1 BIOLOGICAL CONTROL OF THE CASSAVA GREEN MITE, BY ALOYSEIA CRISTINA S. NORONHA.....	108
5.2 BIOLOGICAL CONTROL OF THE CASSAVA MEALYBUG (CM), BY JOSÉ MAURICIO SIMÕES BENTO.....	118
5.3 CASSAVA CROP MANAGEMENT, BY JOSÉ EDUARDO BORGES DE CARVALHO.....	121
5.4 PLANT PATHOLOGY, BY CHIGERU FUKUDA.....	127
5.5 CASSAVA INTENSIVE DIAGNOSTIC SURVEY IN NORTHEAST BRAZIL, BY JOSE HUMBERTO ALMEIDA DE CERQUEIRA AND CARLOS ESTEVÃO LEITE CARDOSO	138
6 PARTICIPANTS AND INSTITUTIONS	146
7 ACRONYMS	148
8 PUBLICATIONS	149

PROFISMA (Proteção Fitossanitária Sustentável da Mandioca): the South American component of a global UNDP project, "Ecologically Sustainable Cassava Plant Protection in South America and Africa: An Environmentally Sound Approach".

Technical Report on Activities conducted during 1996.

1 EXECUTIVE SUMMARY

PROFISMA completed its fourth project year at the close of 1996, thus terminating GLO/91/013. The project has made notable progress in basic and applied research in biological control, integrated pest management and integrated crop management in cassava, emphasizing participation of farmers in Northeast Brazil in the identification of high priority problems and the development and evaluation of technological solutions.

Support of activities in Africa

Three predatory phytoseiid mites (*Neoseiulus idaeus*, *Typhlodromalus aripo* and *T. manihoti*) imported from Brazil, via quarantine in Amsterdam have become established in Africa. Two of these appear to be reducing cassava green mite (CGM) populations by 30 to 90% in the lowland humid tropics. *T. aripo* is dispersing rapidly, and has moved up to 200 km from an initial release site in southern Benin. Estimated benefits for 4 countries in West Africa are on the order of US \$60 million per crop cycle.

These predators do not appear to be effective in dry or subtropical regions, such as the East African plateau. This region has been targeted by IITA for finding new natural enemies in Latin America adapted to this climate. CIAT identified regions with climates homologous to target zones in Zambia in central Brazil and southern Mexico using GIS (geographic information system) techniques. A graduate student from Zambia has been working with a Brazilian scientist to explore the Brazilian region to find predatory mites that would be suitable to release in the East African plateau.

Nine Strains of the predatory mite *Typhlodromalus manihoti* from several high altitude sites in Colombia were sent to IITA, Benin via quarantine at the University of Amsterdam (Mitox) for eventual release in Africa.

IITA is eager to release the fungal pathogen *Neozygites c.f. floridana* in Africa to help provide additional biological control of cassava green mite, especially in dry regions. This pathogen naturally causes epizootics in Northeast Brazil, that periodically kill virtually all of the cassava green mites in a field in one or two weeks. Characterization of the biology and taxonomy of *Neozygites c.f. floridana* strains using molecular genetic techniques have continued. The strain isolated from cassava green mite is highly specific to this genus, indicating that it would be safe to release in Africa. Preliminary experiments with RAPDs indicated that this would probably not be an efficient technology to use to identify *Neozygites* strains. Experiments with AFLP (amplified fragment length polymorphism) are showing more promising results.

PROFISMA's methodology for farmer participatory research was presented at IITA with the aim of facilitating the transfer and adaptation of these methods and experiences to the African countries with which ESCaPP is working. Although ESCaPP has been implementing its own strategy for training and farmer research (based on farmer field schools), they became very interested in organizing a training event for African scientists based on PROFISMA's participatory research strategy. A proposal was jointly developed and submitted by IITA to IDRC-Canada seeking financial support for its implementation.

Basic research at CIAT

Acarology. In addition to the research mentioned above, the CIAT acarology program collected phytoseiid predatory mites in high altitude regions in Colombia, for possible release in high altitude zones in Africa. Predatory mites (*Typhlodromalus tenuiscutus*) from a dry region in coastal Ecuador, with climate similar to the target release zone in Northeast Brazil, were sent to CNPMF via quarantine at CNPMA/EMBRAPA.

The suitability of different prey and natural food sources for different phytoseiids was evaluated to learn more about the specificity of candidate species. The exploration database was analyzed to learn more about the geographic and climatic distribution of phytoseiid species to help select those most likely to succeed in various target release zones. The database was also analyzed to learn about the degree of host plant specificity of candidate phytoseiid mites.

A computer simulation model was developed to help plan mass-rearing of predatory phytoseiid mites. Model predictions were compared to experimental results to help "validate" the model.

A taxonomic key to 30 phytoseiid species commonly found on cassava in northern South America was developed in collaboration with a Brazilian taxonomist.

The microscope slide collection of tetranychid mites was reviewed to determine the geographic and mite host species range of *Neozygites* spp. In vitro culturing techniques were improved thanks to collaborative work by Brazilian and U.S. scientists working at the Boyce Thompson Institute (Ithaca, New York).

Entomology. Field experiments were conducted in the llanos of Colombia to evaluate the efficiency and possible interactions between three parasitoids (*Aenasius vexans*, *Apoanagyrus diversicornis* and *Acerophagus coccois*) of the cassava mealybug (*Phenacoccus herreni*). Mealybug populations were low, and parasitism rates of 15.5% and encapsulation of 37.6% were observed. The parasitoid *A. vexans* had the highest percent parasitism, and total parasitism rates were highest in the field where all three parasitoid species were released.

Experiments with a Y-tube olfactometer indicate that the parasitoids *A. vexans* and *A. diversicornis* are attracted to volatiles from mealybug-damaged cassava leaves, suggesting that the plant produces synomones (odors that attract parasitoids to help protect the plant from mealybugs).

A survey of 100 field sites in Colombia helped determine the geographic and climatic distribution of 3 common species of whiteflies attacking cassava (*Aleurotrachelus socialis*, *Bemisia tuberculata* and *Trialeurodes variabilis*). Ten species of parasitoids were recovered from the three whitefly species, suggesting that some of these may be potential biological control agents. Seven of these were recorded for the first time in Colombia, and four of these are new undescribed species.

Plant pathology. Farms at 34 sites in Colombia were visited, revealing that root and stem rots caused by *Phytophthora* spp. are a major problem, causing root losses of up to 80%. Cassava in fields at CIAT is severely affected by root rots despite chemical treatment of stem cuttings (planting material), planting on ridges, crop rotation, and the relative long dry seasons. Average root yield reduction caused by root rots was calculated to be 7.1 T/ha per

growing cycle for the period 1981-1995 by analysis of 60 field experiments. From the survey, 75 *Phytophthora* isolates were isolated, and these are currently being analyzed for biological and genetic differences. New *in vitro* and greenhouse methods of inoculation were developed to test varietal resistance, and 22 cultivars were inoculated with 4 *Phytophthora* spp. isolates. We developed a PCR method to characterize cassava root rot pathogens, which has been adopted by CNPMF/EMBRAPA scientists in Brazil.

In vitro methods were developed to culture of *Phytophthora* species isolated from cassava, free of bacterial contamination. We developed bioassays to study pathogen variability and to test cassava germplasm for resistance. *Trichoderma* spp., isolated from the rhizosphere of cassava plants were screened *in vitro* for antifungal activity against a range of fungal pathogens: *Phytophthora drechsleri*, *P. nicotianae*, *Diplodia manihotis* and *Fusarium oxysporum*. Filtrates from liquid cultures of *Trichoderma* isolate 11TSM-4 caused marked reduction in growth of the pathogen *D. manihotis*, and filtrates of 14PDA-4 were most effective in reducing the growth of *F. oxysporum*, suggesting promise for biological control.

An effective thermotherapy method was developed to eliminate *Phytophthora* infections in cassava stem cuttings (planting material). This would permit stopping a major source of infection of newly-planted cassava fields.

Fusarium solani and *F. oxysporum* strains were obtained from 86 cassava samples obtained from different ecological zones in Colombia. Four methods of classification were tested for differentiation of the strains, including morphology, growth, pathogenicity and site-directed PCR (rDNA). Screening of resistance has been conducted on 51 cassava cultivars by inoculating them with 28 *Fusarium* strains.

In a survey of cassava growing areas in Colombia, 55 strains of *Diplodia manihotis* were isolated, 8 of which were highly pathogenic to cassava plants. Three inoculation methods were used for pathogenicity studies, including stem injection, stem wounding and root cylinder inoculation. In a search for genetic resistance, 20 cassava cultivars have been inoculated with 40 strains of *D. manihotis*.

Virology. An epidemic of geminivirus is causing extremely severe losses (50-100%) of cassava production in Uganda. We discovered that the geminivirus causing the East African epidemic is distinct from either African cassava mosaic virus (ACMV) or East Africa cassava

mosaic virus (EACMV), and it may actually be a hybrid of these viruses. The discovery of the true cause of the epidemic (i.e., not whiteflies) has made it possible to begin solving the problem by focusing on the evaluation and distribution of cassava varieties resistant to this new virus.

In surveys conducted in Northeast Brazil, the disease caused by cassava vein mosaic virus (CVMV) was found to be very prevalent, and often the majority of plants in a field are infected with the virus. While there are indications that yield losses of 10-15% occur, further studies are needed to quantify losses due to CVMV. Such studies were not previously possible because of the difficulty of detection of the presence of the virus in symptom-less plants.

A rapid method to detect the presence of CVMV was developed at CIAT using polymerase chain reaction (PCR) methods. This test is capable of detecting all isolates tested to date, even when visible symptoms are not present. This method has been transferred to laboratories in Brazil. Isolates of CVMV from six states in Brazil were characterized, and they differ significantly at the molecular level; however, this needs to be correlated with biological characteristics.

Cassava frogskin virus has been expanding its geographic range in Latin America. It is now endemic in parts of the state of Bahia, Brazil. This probably occurred because of the major drought in 1992, that caused farmers to import cassava stem cuttings from neighboring states that border the Amazon region. Cassava frogskin disease (CFD) can cause the complete failure of a crop and is an important new pathogen that needs to be controlled. A microscopic analysis of the roots of CFD-affected cassava revealed very different structural features that are being evaluated as method to select CFD-tolerant cassava varieties, and the CIAT core germplasm collection is currently being screened.

Whitefly transmitted geminiviruses are major problems in Africa and India. With the "B" biotype of *Bemisia tabaci* colonizing cassava in the Americas, increased efforts are being made to characterize the threat that geminiviruses pose to cassava.

Agronomy. Long-term response of cassava to NPK (nitrogen, phosphorous, potassium) levels in acid infertile soils were measured at Santander de Quilichao, Colombia. After 13 consecutive cropping cycles without application of NPK fertilizer, dry root yield was only

about 4 t/ha. With application of 100 kg/ha each of NPK, root yields were dramatically increased (about 10 t/ha dry root). The largest observed response was for K, indicating that K is the most limiting nutrient in these soils, which have high organic matter content (ca. 7%). To sustain reasonable productivity, K fertilizer must be applied annually since most K uptake (> 60%) is removed in the harvested roots.

Long-term effects of surface mulch (grass), fertilizer and tillage on cassava productivity in poor sandy soil were measured at Pivijay, in northern Colombia. Dry root yield was significantly increased by the application of either mulch or NPK fertilizer at a moderate levels. Conventional tillage increased yield only in the presence of NPK fertilization without mulch. The combination of surface mulch and no tillage gave the highest root yield with and without fertilizer. These findings indicate that consecutive cultivation of cassava in these poor soils would lead to very low yields unless appropriate measures are followed to maintain soil fertility.

Information management. We scanned and formatted 11 major documents (2,800 pages) including recent CIAT Cassava Program annual reports, CIAT Working Documents, scientific meeting proceedings and Cassava Newsletters to be included in a CD-ROM being published by IITA and the University of Florida, Gainesville.

Basic and applied research in Brazil

Acarology. Five shipments of the phytoseiid predatory mite *Typhlodromalus tenuiscutus*, originally sent from CIAT, were made from the EMBRAPA/CNPMA quarantine facility to EMBRAPA/CNPMPF (2,150 individuals). A colony was established and mass-rearing. Mass rearing of *Neoseiulus californicus* was carried out in a screenhouse on *Tetranychus urticae* on jack bean plants, enabling the production of 33,500 individuals in a one month period. 10,800 *T. tenuiscutus* were released at 2 sites in Bahia, and 34,020 *N. californicus* were released at 4 sites in Bahia and Pernambuco. 28 specimens of *N. californicus* were recovered during monthly monitoring of the release sites, suggesting that it may be establishing.

In experiments using jack bean leaf discs that had previously been immersed in neem (*Azadirachta indica*) extract, we found that neem extract caused a decrease in survivorship of

the two-spotted spider mite (*Tetranychus urticae*) but not its phytoseiid predator *N. californicus*. Studies on neem's effect on cassava green mite are under way.

CNPMF has 21 isolates of the mite pathogen *Neozygites* cf. *floridana* from several regions, in Brazil; however nine of them have lost their viability. Some isolates have been stored for up to 20 months as "mummies" and still remain viable, whereas others have lost viability in as little as 6 months. Variation in viability appears to be partly caused by the quality of the mummy and the amount of time that it spent in the field before being collected. Evaluation of different conditions for storing mummies indicates that freezing (-10°C) is generally better than 4°C or 25°C. Isolates from Cruz das Almas and Piritiba do not sporulate at relative humidity lower than 95% (at 24°C).

Neozygites isolates from *T. urticae*, collected in Cali, Colombia, and in Jaguariuna, State of São Paulo, Brazil, were pathogenic to both *T. urticae* and *M. tanajoa*; on the other hand, the isolates from *M. tanajoa*, collected in Brazil, were pathogenic only to *M. tanajoa*, thus showing a very high specificity.

The efficiency of field inoculation of *Neozygites* sp. to control CGM was studied by making ten inoculations (May 22 to June 24) using 40 mummified mites as source of inoculum. Although the first appearance of infected mites control and experimental fields occurred almost simultaneously, by August 2, the pathogen had spread all over the release field, whereas it was not found in the control field. The delay in initiation of an epizootic may be due to the low mite population at the time (25 individuals/leaf), low release rate or to climatic conditions.

Entomology. A total of 111,610 parasitoids (*Aenasius vexans*, *Apoanagyrus diversicornis* and *Acerophagus coccois*) were produced, and 35,950 of them were released in six cassava fields in Bahia and Pernambuco. *A. diversicornis* dispersed the most, reaching up to 304 km by April 1996. *A. coccois* dispersed more slowly, reaching 180 km from the releasing site by September/1995, while *A. vexans* has not dispersed from its release site as of November, 1996, despite the large number of releases. A negative correlation between number of parasitoids and mealybugs has been observed for both *A. diversicornis* and *A. coccois* in the State of Bahia, although *A. coccois* has a stronger effect on mealybug

populations. Although mealybug populations fluctuate seasonally, the peaks are now about 20% the size of those observed before establishment of the parasitoids.

Plant pathology. Cassava witches' broom disease (CWBD), caused by a phytoplasma, is a very serious cassava problem in the "Serra da Ibiapaba", State of Ceará. An important achievement on the research of CWBD was the observation that elimination of infected planting material of susceptible varieties increased root yield. On farm trials to evaluate resistant germplasm were established in 60 rural communities, resulted in the selection of 2 improved clones that were preferred by 60% of the farmers to the local variety. Projected impact of adoption of these varieties, assuming adoption by 60% of the growers, implies replacement of about 3,000 ha of cassava and would represent 60,000 t increase in yield or about US\$2.5 million increase in profits.

A survey for the cassava root rot incidence was carried in three cassava growing regions of the State of Bahia. Isolations carried out under laboratory conditions showed that the pathogens involved in the cassava root rot development in the rural communities evaluated in the Bahia were *Fusarium* sp., *Phytophthora* sp. and *Scytalidium lignicola*. A total of 179 varieties from the EMBRAPA/CNPMP Cassava Breeding Program were evaluated in on-farm experiments in Sergipe, Paraíba and Alagoas, permitting the selection of several varieties in each region that showed resistance to root rot. A farmer participatory trial showed that disease incidence is lower and root yield is higher when cassava is planted in ridges rather than on level ground.

Field evaluation of 30 cassava hybrids, from the EMBRAPA/CNPMP Cassava Breeding Program, in the municipality of Umbauba, State of Sergipe, enabled the selection of five of them that were evaluated again in a on farm trial in a randomized block design, with three replicates. Eighteen cassava varieties/hybrids were evaluated for root rot incidence in a on farm trial carried out at the COPAL Quitéria, Municipality of Alagoa Grande, State of Paraíba, in a root rot naturally infested area. One hundred and seventy nine hybrids, from the EMBRAPA/CNPMP Cassava Breeding Program, were evaluated in on farm trials in the municipality of Viçosa, State of Alagoas, where root rot incidence was relatively high.

Due to erratic geographic and temporal incidence of cassava root rot, CNPMPF decided to develop laboratory evaluation techniques to help complement field trials to identify cassava

genotypes resistant to root rot pathogens. Several inoculation techniques were tested under laboratory and greenhouse conditions which generally showed that isolates of *Fusarium* was not very virulent, but that isolates of *P. drechsleri* and *Scytalidium lignicola* expressed high virulence.

Simultaneous inoculations of the antagonistic fungi *Trichoderma polysporum*, *T. harzianum* or *T. pseudokoningii* with the pathogen *Phytophthora drechsleri* were able to prevent it from producing normal sized lesions in some varieties of cassava, suggesting some possibilities for biological control of root rots.

Virology. The main objective in 1996 was to determine root yield losses due to the cassava vein mosaic virus (CVMV). Such an experiment requires a large amount of virus-free planting material, but the PCR diagnostic method was inadequate for testing this many plants. Consequently, the production of CVMV-specific antiserum became the main objective since this is easier to perform. Several modifications on the CVMV purification technique, such as viral separation through polyacrylamide gel electrophoresis; however, all have failed to eliminate the presence of host protein in the purified virus.

We evaluated 20 host plants as possible alternate hosts of CVMV, but the virus multiplied only in cassava.

Agronomy. Experiments on duration of manual weed control at 2 sites in Bahia indicate that it has a significant effect on yield only during the first 30 to 60 d after planting. Other experiments combining various forms of weed control and intercropping showed that weeding within double rows of cassava and planting pigeon pea, cowpea or jack bean between the double rows produced higher yields than leaving weeds between the double rows. Although weeding between the double rows tended to increase yield (though not significantly), this was at a 30% higher cost due to labor.

A field trial on the effect of intercropping with jack bean or cowpea between cassava double rows to reduce erosion showed no difference because of a drought in which no soil loss by surface run-off was recorded.

On farm trials in Acarau showed that adding bagana (palm leaf waste) mulch at 180 m³/ha, or more, increased cassava root yield (25.1 t/ha) in comparison to fields without bagana (8.1

t/ha). Another trial in Russas showed no effect, possibly due to rainfall that occurred during the period of initial growth of the crop.

Farmer participatory research. The final training course "Participatory Methods for Evaluation of Technology Trials with Farmers" held at CNPMF/EMBRAPA in April was attended by 30 researchers and extension workers, representing 6 collaborating research and technology transfer institutions in the states of Bahia, Ceará, Pernambuco and Paraíba, Brazil. The course improved their skills, abilities and attitudes for using two participatory methods in the evaluation of technology testing trials with farmers.

The first series of farmer participatory research experiments at 18 COPALs in 4 states of Northeast Brazil were completed and evaluated and the second series was initiated. The various experiments included evaluation of regional varieties for yield and resistance to root rot, cassava green mite and whitefly, planting on ridges to control root rot, use of chemical fertilizers, organic compost or legume companion crops, to improve soil fertility, and row spacing. A few experiments failed because of drought or because of inadequate follow-up by the technicians. However, most experiments produced results that increased yields by 10 to 67%. In several cases the EMBRAPA scientists learned of local varieties that performed better than their "improved" varieties, thus providing them with new germplasm to incorporate in their breeding programs. They also began to learn more about the traits other than yield that farmers value, which will contribute significantly to the relevance of future crop breeding efforts.

Formation of a second generation of COPALs (farmer research committees) was initiated during 1996 in the State of Ceará through a project presented by local institutions to the Banco do Nordeste do Brasil (BNB). This project financed the establishment of COPALs in six communities of cassava growers that had been identified during the participatory survey carried out by PROFISMA in 1994. Financial support received from BNB included seed money to start the first COPAL FPR experiments, funds for farmer training events and for fuel and per diems for technicians assisting the COPAL. The COPALs were autonomously organized by local institutions followed the same methodology taught by PROFISMA.

Field days were held in Ceará and Pernambuco in 1996 with the aim of stimulating more support for COPAL activities at local and state level. The COPAL methodology for

enhancing farmer participation in research and technology transfer activities and the initial FPR results obtained by PROFISMA were presented to 160 farmers, technicians and decision-makers.

PROSERTÃO is a rural development project executed by the Government of the State of Sergipe, Northeast Brazil, with financial support from IFAD through a 25 million dollars loan during a seven years period. PROSERTÃO staff approached CNPMF looking for technical assistance to solve the problem of root rots which plagues 95% of the municipalities in the project area. A joint proposal was developed in which PROSERTÃO funds activities by CNPMF that include: 1) the implementation of a training program in FPR & COPAL methodology for PROSERTÃO personnel, 2) establishment of COPALs in some selected cassava growing areas, and 3) execution of farmer participatory activities to develop and transfer technologies to control cassava root rot.

Socioeconomics. Intensive Diagnostic Survey was conducted in 18 communities, involving 21 cassava growers in each community (9 COPAL members, 6 non-COPAL members, and 6 cassava growers from the neighborhood) in Northeast Brazil. Data were collected on land possession and usage, family composition, educational level, cassava production technology, and commercialization of cassava. Data were also collected on incidence of pests and diseases and the methods being used to control them. Over 50% of cassava growers had less than 5 ha. surveyed do not own the land they cultivate. About 29.9% of the land used as pasture, 29.9% is left under fallow, and 13.2% is natural vegetation, all of them generally used for grazing.

About 89% of the daughters, 81% of the sons, and 57% of the parents do not have any school education. The head of the family is predominantly a man older than 18 years; 52.5% of them are 50 years old or less. Approximately one third of the family members does not work in the rural community: they are either school age children or young people whose activities are other than rural labor. About 29%, besides working on their land, also sell their labor in order to make extra money.

Most of the growers (64%) prepare the soil for planting only by manual hoeing. About 73% of the cassava growers use their own cassava stakes to start a new planting, and only

42% of them reported carrying out any kind of selection of stake quality prior to harvesting. Only 64% said that the stake must be 10 to 20 cm long have a diameter greater than 2 cm.

Farmers considered leafcutting ants, root rots, hornworm, cassava green mite, and whiteflies to be the most important pest and disease problems. Although witches' broom disease is fairly uncommon, it can cause extremely high yield losses (72%). Farmers used pesticides only to control leafcutting ants, hornworms and anthracnose, and lime or ash to help control root rots.

The majority of the cassava production in the Northeast (77%) is sold as cassava flour, 19% as fresh roots, 1.6% as animal feed, and only 0.7% as dry chips. About a third of the aerial plant parts are used for feeding animals.

Project management

Due to anticipated reduction of funding in 1997, the CIAT coordinator in Brazil decided to eliminate his own position, as well as those of some junior staff in Brazil and at CIAT. Project coordination by CIAT during 1997 was devolved to other staff in Cali, Colombia.

A concept note entitled, "Ecologically Sustainable Cassava Production Systems for Latin America" was prepared by CIAT and sent to 25 donors in May with the goal of finding additional donors to support a subsequent project based on the successes of this project. A second concept note entitled, "Ecologically Sustainable Cassava Plant Protection: A Global Strategy" was written jointly by CIAT and IITA in July to unify the Latin American and African proposal concepts. The training component of PROFISMA Phase 2 was prepared by CNPMF in collaboration with IPRA (CIAT program on farmer participatory research) personnel and was sent to the Chairman of the EMBRAPA International Cooperation Department.

2 PROJECT COORDINATION

2.1 Coordination at CIAT, by Steven Lapointe

The uncertainty of continued funding of this very successful project caused major disruption during 1996 and resulted in considerable effort directed to a search for continued support. The one-year extension provided by UNDP for 1997, while considerable and generous, given the current financial status of that agency, was insufficient to maintain all activities. After careful weighing of the alternatives, the CIAT coordinator in Brazil decided to eliminate his own position, as well as some junior staff positions in Brazil and at CIAT. This resulted in enough savings to allow retention of most project junior staff and the continuation of essential activities through 1997.

Project coordination by CIAT during 1997 was devolved to staff at Cali, Colombia. The Training Officer in Cruz das Almas assumed even greater responsibilities for coordination and execution of activities in Brazil. The refractory nature of the Brazilian national program staff at CNPMF to such large projects requiring close collaboration and sharing of responsibilities and executive authority with another organization (i.e., CIAT), was a major impediment that was only partially overcome through daily contact and the project's ability to work directly with state agencies and farmer communities. The placement of a CIAT scientist at CNPMF should be carefully considered in the future and should be contingent on careful delineation of responsibilities and authority for execution of such large projects. CIAT and CNPMF should review recent experiences in an effort to improve institutional collaboration.

External Advisory Committee (EAC) Review

The second and final external review of the African activities (ESCaPP project; Ecologically Sustainable Cassava Plant Protection) took place during June, 1996 in Benin and Nigeria. The three External Advisory Committee (EAC) members that reviewed PROFISMA in Cali in August 1995 (John Borden, Don Roberts, Asuncion Raymundo) were joined by Dr. Mohamed Dahniya. Bernardo Ospina (farmer participatory research trainer) and Steve Lapointe (Project Coordinator) represented the Latin American activities (PROFISMA; Portuguese acronym for Ecologically Sustainable Cassava Plant Protection). The EAC team

was split into 2 groups, one visited Nigeria (IITA headquarters and ESCaPP/Nigeria field sites) and the other toured ESCaPP/Benin field sites during the first 4 days of the review. The groups reunited in Cotonou for 2 days of presentations.

ESCaPP presented impressive achievements. A large number of national program scientists and extensionists have been trained in production methods, and these, using local extensionists as multipliers, have established on-farm trials with grower participation. We visited several of these in Benin. The National Coordinator for ESCaPP in Benin, Norbert Maroya, was particularly impressive - organized, articulate, in touch with field activities and obviously motivated and committed to the project. Overall, we were impressed with the enthusiasm and commitment of national program staff to the project - a marked contrast with PROFISMA, perhaps due to the fact that IITA maintains control to a greater extent than PROFISMA over finances and activities.

By far the most impressive accomplishment of the project to date is the establishment, spread and measured impact of *Typhlodromalus aripo* (a phytoseiid mite predator of cassava green mite [CGM]). Field trials have demonstrated reductions in CGM numbers of up to 90%, and economic impact regarding increased root yields of 30%, corresponding to profits of about US\$ 60/ha per crop cycle. Area-wide benefit will undoubtedly be a significant (in the \$10-100 millions per year), and benefit will continue year-after-year because of the self-sustaining nature of classical biological control. This will no doubt represent an extremely high return on the initial donor investment, causing widespread impact on food production and economic development in the lowland humid tropics of sub-Saharan Africa, with no adverse environmental impact.

Major accomplishments of PROFISMA

Below are the major accomplishments of PROFISMA as the first phase of the UNDP project draws to a close:

I. Biological Control

1. Successful introduction and establishment of 3 species of parasitoid natural enemies for control of the cassava mealybug *Phenacoccus herreni* in Northeast Brazil. Two species

(*Acerophagus coccois* and *Apoanagyrus* [= *Epidinocarsis*] *diversicornis*) are established and spreading rapidly in Bahia and Pernambuco causing reductions of mealybug populations.

2. Establishment of a phytoseiid predator mite (*Neoseiulus californicus*) from northern S. America in Northeast Brazil for control of the cassava green mite (CGM). Establishment is tentative and spread is not yet documented. Protocols for exploration, collection, shipping, quarantine, rearing and release were developed for rapid, multiple introductions of additional species of natural enemies in 1996 and 1997.
3. Developed *in vivo* production methods for and methods for genetic characterization of the fungal pathogen (*Neozygites* c.f. *floridana*) for control of CGM. Methods for isolating and growing *Neozygites* in artificial media are being developed to facilitate further study and exploitation of this fungus as an agent for biological control of CGM in Latin America and Africa. Genetic characterization allows taxonomic differentiation between fungus isolates to help select the best strains and to identify them for quarantine purposes and to monitor it after being released in Africa and elsewhere.
4. Contributed to biological control of CGM in Africa in collaboration with ESCaPP through climate homologue mapping, and collection and characterization (biological and genetic) of pest and predator mites. Regions to explore in Minas Gerais, Brazil and southern Mexico were identified as ecological homologs for Zambian target release zone.

II. Farmer Participatory Research in NE Brazil

1. Completion of extensive and intensive participatory diagnostic surveys of cassava growers in NE Brazil. Major biotic and abiotic constraints to production in small-scale cassava production systems were identified through participatory surveys that emphasized farmers' perceptions and priorities. Approximately 1,600 farmers were contacted.
2. Trained a core of state extension agents and national program scientists in Farmer Participatory Research (FPR) methods.

3. Established 30 Local Agricultural Research Committees (COPALs) in 4 states of Northeast Brazil.
4. These COPALs, with support from extensionists and national program researchers, will have planted and evaluated two cycles of experiments in their communities addressing pest and disease constraints identified by farmers.

III. Areas of global collaboration between IITA and CIAT

1. Exploration and introduction of natural enemies (phytoseiid mites and *Neozygites*) from Brazil and northern South America into Africa for control of CGM.
2. Mapping of climate homologies between regions of South America and Africa for targeting natural enemy explorations.
3. Biological and genetic characterization of natural enemies (phytoseiid mites and *Neozygites*).
4. Scientific exchange visits.
5. Training of African and South American technicians in mite taxonomy.
6. Sharing and compilation of information resources; production of CD-ROM including databases, bibliographies, and "gray literature" technical documents.
7. Global project coordination and exchange.

2.2 Coordination at EMBRAPA/CNPMPF, by Aristoteles Pires de Matos

PROFISMA computer database

A computer database called Participatory Research System (SPPAR), is currently in the final elaboration phase. Data from one rural community has already been entered to test it. The database runs in FOX-PROT for Windows versions 3.1 and 95, and it has image storage capability.

Workshop on the cassava root rot disease

On April 8 to 11, 1996, a workshop on cassava root rot diseases was held at EMBRAPA/CNPMPF. Plant pathologists from Research Institutions and Universities of Northeast and West Central Brazil, EMBRAPA/CNPMPF, and CIAT discussed research approaches and problems related to this group of diseases. As a result of the workshop, the following suggestions were elaborated:

- 1) Carry out pathogenicity tests in order to know what pathogen occurs in each cassava growing area;
- 2) Identify and characterize cassava root rot-causing pathogens;
- 3) Study root rot pathogen interaction regarding disease development;
- 4) Establish methods to evaluate cassava genotypes for resistance to root rot pathogens;
- 5) Evaluate accessions of the Cassava Active Germplasm Bank (CAGB) of EMBRAPA/CNPMPF for resistance to root rot pathogens;
- 6) Continue running field evaluations of cassava genotypes for resistance to root rot;
- 7) Continue studies on biological control and effect of cultural control practices on root rot development.

Weed management consultant

During 1996 Dr. Robinson Antonio Pitelli, Professor of Weed Management, Universidade Estadual Paulista, Jaboticabal, State of São Paulo, was a consultant for the activities related to: i) effects of weeds on cassava yield, ii) effects of cover crops, including weeds, on cassava yield and arthropod populations. After one week evaluating our activities, Dr. Pitelli found

them well-planned and well-conducted. He stressed the interdisciplinary characteristics of our activities since a Weed Scientist, Entomologists, Plant Pathologists, Agronomists, Soil Scientists and a Plant Taxonomist are working together. His main suggestions for future actions were: i) survey of pathogens on weeds; ii) take also into consideration, besides the relative importance, the index of importance value, when discussing recorded data; iii) develop basic studies on nutrient uptake by weeds, weed response to mineral and organic fertilizers, and weed response to drought and nutritional stress.

Farmer participatory research (FPR) consultant

The last component of the FPR training program, "Participatory Methods for Evaluation of Technology Testing Activities with Farmers", was taught during April, 1996, by Mrs. Teresa Gracia and Mr. Luiz Alfredo Hernández, both from CIAT. This was a ten-day training session, that included theoretical classes and practical field work. Thirty researchers and extension workers, from six state institutions of four States, attended the course.

Visiting COPALs

During 1996, two World Bank's missions, together with EMBRAPA's representatives, visited the COPAL Cadete in Bahia. The first visit to the COPAL Cadete was a suggestion of the CNPMF Director of Research and Development since the World Bank representatives were interested in visiting small growers communities. The second visit was a consequence of the first one. In both occasions the visitors were very impressed with the level of grower participation on the research activities carried out by the COPAL.

Following the first visit of the World Bank mission, an EMBRAPA consultant for technology transfer was recommended to visit EMBRAPA/CNPMF in order to better know the COPAL methodology implemented by PROFISMA. After discussing with PROFISMA staff the theoretical bases of the farmer participatory research methodology adopted for the COPAL model, he visited the COPAL Colônia Agrícola Roberto Santos, located in the municipality of Inhambupe, and the COPAL Chapada, located in the municipality of Aporá, both in the State of Bahia.

By early December, 1996, the Coordinator of Agricultural Development of PROSERTÃO (an IFAD-funded development project in Sergipe), together with 13 extension workers from that project, visited EMBRAPA/CNPMPF in order to maintain contact with the coordinators of PROFISMA and the IFAD-funded project conducted by EMBRAPA/CNPMPF. He expressed an interest in visiting a COPAL in order to observe *in situ* how the COPAL methodology has been implemented among cassava growers. The PROSERTÃO team visited the COPAL Colônia, located in Inhambupe, State of Bahia and was favorably impressed.

Graduate students (Institutional Relationship)

Three graduate students from the Universidade Federal da Bahia, have been conducting their research at EMBRAPA/CNPMPF, working on PROFISMA activities as dissertation subjects, including: 1) effects of cover crops on physical and chemical properties of soil in cassava, 2) weed management in cassava growing areas, and 3) control of cassava root rot disease. PROFISMA-CNPMPF Scientists are the major professors of these students.

2.3 Phase Two Proposals, by Lincoln Smith

A concept note entitled, "Ecologically Sustainable Cassava Production Systems for Latin America" was prepared by CIAT and sent to 25 donors in May with the goal of finding additional donors to support a subsequent project based on the successes of this project. The donors contacted included: United Nations Development Programme (UNDP), Carnegie Corporation of New York, The Ford Foundation, International Development Research Centre (IDRC), World Bank, International Fund for Agricultural Development (IFAD), Henry P. Kendall Foundation, W. K. Kellogg Foundation, John D & Catherine T. MacArthur Foundation, The Pew Charitable Trusts, The International Foundation, Inc., Agency for International Development (USAID), W. Alton Jones Foundation, Inc., Jessie Smith Noyes Foundation, Weeden Foundation, The Charles Stewart Mott Foundation, Andrew W. Mellon Foundation, The Rockefeller Foundation, Inter-American Development Bank, Ciba-Geigy Foundation for Cooperation with Developing Countries, Swiss Agency for Development and

Cooperation (SDC), Swiss Centre for International Agriculture (ZIL), The McKnight Foundation.

A second concept note entitled, "Ecologically Sustainable Cassava Plant Protection: A Global Strategy" was written jointly by CIAT and IITA in July to unify the Latin American and African proposal concepts. This concept note was sent to UNDP for consideration of possible funding of a second phase of the PROFISMA/ESCaPP project beginning in 1997.

The training component of PROFISMA Phase 2 was prepared by PROFISMA's Training Coordinator in collaboration with EMBRAPA personnel and was sent to the Chairman of the EMBRAPA International Cooperation Department. The Plant Pathology and Entomology components of PROFISMA Phase 2 were prepared by PROFISMA/CNPMF staff, based on cassava growers' demands as identified by the participatory diagnostic surveys. These components will be discussed with the CNPMF cassava research team for further adjustments, before sending to donors.

Visiting UNDP Headquarters

The EMBRAPA/CNPMF Director of Research and Development, together with CIAT's representatives, visited UNDP headquarters in order to discuss financial support for the extension of the Project, starting in 1997.

Arrangements for continuing PROFISMA activities during 1997

The project coordinator for PROFISMA is terminating his position at the end of December 1996. This will substantially reduce project costs, helping us to retain other essential technical personnel during 1997. However, it will reduce the amount of supervision and coordination of activities between CIAT and CNPMF. It also greatly decreases our capability to communicate with donors in the search for funding to support subsequent projects that will extend and solidify the progress that this project has obtained to date.

A money-saving policy was implemented at EMBRAPA/CNPMF during 1996, based on two main actions: 1) EMBRAPA's financial aid for operational costs, and administrative and scientific personnel; and 2) rationally reducing project personnel. Two field workers, the

project secretary, one lab technician, the Virologist and the Environmentalist were dismissed. In addition, EMBRAPA did not charge PROFISMA overhead costs to UNDP during 1996. Such procedures made it possible to maintain PROFISMA activities during 1997, though at reduced levels. The operational budget for 1997 will be provided by EMBRAPA, which is also expected to supply additional personnel when necessary. PROFISMA personnel will be paid using carryover from the 1996 budget plus bridging money provided by UNDP, as previously discussed with UNDP representatives.

3 TRAINING AND PARTICIPATORY RESEARCH

3.1 Training, by Bernardo Ospina Patiño

Courses

During 1996, the last component of the training model proposed by PROFISMA was implemented through a course on “Participatory Methods for Evaluation of Technology Trials with Farmers” held at CNPMF/EMBRAPA in April, 1996. This last training stage was attended by 30 researchers and extension workers and was aimed at improving their skills, abilities and attitudes for using two participatory methods in the evaluation of technology testing trials with farmers. These trainees, representing 6 collaborating research and technology transfer institutions in the states of Bahia, Ceará, Pernambuco and Paraíba, were responsible, upon finishing the course, for assisting the COPALs and their communities in the harvest and evaluation of the trials installed during the 1995-96 growing season.

Establishment of new COPALs

Formation of a second generation of COPALs was initiated during 1996 in the State of Ceará through a project presented by local institutions to the Banco do Nordeste do Brasil (BNB). This project financed the establishment of COPALs in six communities of cassava growers that had been identified during the participatory survey carried out by PROFISMA in 1994. Financial support received from BNB included seed money for the first COPAL experiment, funds for training events for farmers, and fuel and per diems for technicians assisting the COPAL. Organization of the COPALs although autonomously conducted by local institutions followed the same methodology proposed by PROFISMA. One interesting aspect of this effort at the state level is that the six new COPALs are working on the same line of research: testing the use of organic compost and earthworms.

Field days

Two field days were organized during this period with the aim of getting more support for COPAL activities at local- and state-levels. During these events, the COPAL methodology for

enhancing farmer participation in research and technology transfer activities and the initial results obtained by PROFISMA were presented to larger audiences of farmers, technicians and decision-makers. One event was held in the State of Ceará, organized by EMATERCE and hosted by the COPALs located in the communities Nova Veneza and Valparaíso in Ubajara. Total attendance to this event was near 100 persons including farmers from the six new COPALs established this year. The second field day was organized in the State of Pernambuco under the coordination of IPA and EMATER-Pe, the two local collaborating agencies. This event was hosted by the COPAL in the community São Bento do Una and was attended by 60 persons.

Joint training activities PROFISMA/ESCaPP

Profisma's training coordinator participated in 1996 annual evaluation of ESCaPP activities. During this meeting held at Cotonou, Benin, PROFISMA's methodology for farmer participatory research was presented, and despite the fact that ESCaPP is implementing its own strategy for training and farmer research, they became very interested in organizing a training event for African scientists based on PROFISMA's participatory research strategy. The aim of this activity would be to facilitate transference and adaptation of these methods and experiences to the African countries with which ESCaPP is operating. A proposal was jointly elaborated and has already been submitted by IITA to IDRC-Canada seeking financial support for its implementation. The proposal is based on a three to four weeks course on FPR and COPAL methodologies to be held both at CIAT (Colombia) and EMBRAPA/CNPMPF (Brazil) with a target audience of 13 selected scientists from Benin, Cameroon, Ghana and Nigeria. These trainees are currently collaborating with ESCaPP. If the proposal is approved, the proposed period for this event is the first quarter of 1997. The importance of this activity lies not only in the fact that it was recommended by the UNDP-assembled External Advisory Committee, but also for its characteristics of being an inter-projects (ESCaPP and PROFISMA), inter-centers (CIAT, IITA, EMBRAPA), and inter-continental training activity (Africa, South America).

Joint training activities PROFISMA/PROSERTÃO

PROSERTÃO is a rural development project executed by the Government of the State of Sergipe, Northeast Brazil, with financial support from IFAD through a 25 million dollar loan during a seven year period. This project includes an agricultural development component, and initial information collected by a survey of the target area identified cassava root rot as the main constraint for agricultural production. This disease was found in 95% of the municipalities included in the area of influence of the project. Given the magnitude of the problem and considering the fact that cassava is the main source of income for small-scale farmers in the region, PROSERTÃO staff approached CNPMF looking for technical assistance to solve the problem. After initial discussions and exchange visits, a proposal was formulated that includes a) the implementation of a training program in FPR and COPAL methodology for PROSERTÃO personnel, b) establishment of COPALs in some selected cassava growing areas and c) execution of participatory technology generation and transfer activities for control of cassava root rot. These last activities would be based on a combination of improved cultural practices and the use of resistant/tolerant cassava varieties and hybrids. This genetic material is being developed by CNPMF through the project “Development of Improved Cassava Germplasm for the Semi-arid Zone of Northeast Brazil”, financed also by IFAD and executed jointly by CIAT and CNPMF since 1992. The successful results obtained with this project coupled with PROFISMA’s experiences adapting participatory methods for farmer participation in research and technology generation and transfer activities to Northeast Brazil, could be fundamental factors in the execution of these activities. Additionally, the existence of training material already in Portuguese and the possibility of using some COPALs as training sites will facilitate the implementation of this activity and the dissemination of the COPAL methodology in other regions of Northeast Brazil, a spin-off effect of the PROFISMA project.

COPAL visits to EMBRAPA/CNPMF headquarters

Visits to CNPMF research station were organized this year for the COPALs located in the State of Bahia. The objective of this training activity was to allow farmer groups to become more familiarized with PROFISMA’s activities, to increase their knowledge about specific

topics selected by them and to discuss alternative technological options that are currently available or under development at CNPMF and that could be incorporated in future technology testing activities to be implemented by the COPALs. For each visit, which lasted two days, PROFISMA staff prepared presentations, practical demonstrations and visits to laboratories and cassava fields, following agendas designed with collaboration of EBDA personnel who are responsible for technical assistance to the COPALs. These agendas were designed to meet farmers' demands and priorities. A total of six COPALS visited CNPMF during this year with participation of 160 cassava growers from the municipalities of Piritiba, Alagoinhas, Inhambupe, Anguera, Aporá and Cruz das Almas.

Participatory evaluation of trials established by the COPALs in the 1995-96 growing season

During 1996, the group of researchers and extension workers collaborating with PROFISMA in the States of Bahia, Ceará, Pernambuco and Paraíba, who are responsible for giving year-round technical assistance to the COPALs, faced a great challenge with the harvest of the first technology testing trials established by the COPALs. After covering the three training stages (diagnoses, planning and evaluation), this group of technicians finally was able to use the participatory methods for evaluation of technology trials with farmers. PROFISMA's strategy for this activity was defined as a combination of two methods: **a) agronomic evaluation** (the one commonly realized by researchers to determine yields, dry matter content, and other parameters) and **b) technology evaluation by farmers** (evaluation of technology trials using complementary techniques). The technique utilized was the so called Preference Ranking. For each evaluation the procedure was as follows:

- The COPAL coordinated the activities and requested assistance from other interested farmers in the community.
- A short meeting was held to discuss the activities, the organization and to assign responsibilities.
- Farmers participated in the harvest of each treatment, each replication and each block.

- Cassava roots and aerial plant parts harvested for each replication and each experimental block were laid on the floor grouped by treatments, and farmers were asked to rate them (the best, the second, the last, etc.) using small pieces of paper with printed numbers. Each farmer represented one vote, and he voted on all the treatments. During their ranking of the different treatments, farmers were allowed to discuss among themselves about the criteria they were using. Technicians were asked to avoid any participation in this step.
- After all the treatments have been ranked by the farmers, a final counting was made assigning points to each classification. For example, in an experiment with four treatments, the best treatment was assigned number 1 and was worth four points; the second, was assigned number two and was worth three points; the third, number three and was worth two points and the last one, was assigned number fourth and was worth one point. By adding the points assigned to each treatment, a final Preference Ranking, according to farmers criteria was obtained.
- Farmers were invited to comment on this ranking (especially regarding the criteria they used).
- Agronomic evaluation was conducted jointly by technicians and farmers. Each treatment was weighed (cassava roots and aerial plant parts). For dry matter determinations a sample was taken for each treatment, and the dry cassava content was determined in front of the farmers using the hydrostatic balance method.
- A rapid analysis of the results was made on the spot and was used to stimulate the final discussion with the farmers. Raw, semi-processed data of the results of the experiment was left with the COPAL the same day.
- A more complete report on each experiment was sent later to each researcher and extension worker assisting the COPAL in the interpretation of the results, its presentation to the larger community, and more important, the drawing of conclusions and proposals for the second experiment..

First experiences and results obtained by PROFISMA about the use of participatory methods for evaluation of technology trials with farmers are very encouraging. The technicians involved, despite the fact that they are in most of the cases new practitioners, have

been able to adapt the methods to their own conditions and have stimulated farmers to make their own evaluations. Conversely, the farmer members of the COPALs and the other farmers that collaborated with them managed to produce their own evaluation results and made a very important step forward in the process of building local, community level capacity to participate actively in technology generation and transfer activities. Another very important result of these activities was the strengthening of the collaborative relation between the technicians and the COPALs.

In the majority of the trials evaluated, results obtained with the agronomic evaluation were consistent with those obtained in the subjective evaluations made by the farmers. Their choices were supported by yield and dry matter content data. However, in some cases, it was clear that farmers' choices were not necessarily those options with the best agronomic results. *This fact helped farmers to gain awareness about the importance of diversifying the technological options available for them and increasing the number and the frequency of technologies tested in their communities.* In these cases, farmers reacted very enthusiastically and showed great interest to continue the testing and adaptation process with the technological option that showed good potential to be incorporated in their farming systems. Appendix I presents more detailed information on the participatory technology testing trials evaluated by the COPALs during the 1995-96 growing cycle.

3.2 Participatory Evaluation of Technology Testing Trials

3.2.1 State of Bahia

COPAL: Colonia Agricola Roberto Santos, Inhambupe

The objective of the experiment was to evaluate six cassava varieties for resistance/tolerance to cassava green mites (CGM). This had been prioritized by farmers as the main constraint for cassava production in the region. The six varieties were selected locally by the farmers from genetic materials that they have planted over the years. This experiment did not include any variety from EMBRAPA/CNPMF. *Farmers in this community represent an atypical example of cassava growers since they are used to put heavy amounts of fertilizers in their crops (cassava, citrus and passion fruit) and have been doing it for many*

years. The excellent results obtained in this experiment confirm this. Figure 3.2.1.1 presents the results obtained for yield (ton/ha), dry matter content (ton/ha) and the preference ranking that farmers made of the treatments included in the experiment. Two of the varieties gave yields higher than 35 tons, four of them gave yields higher than 30 tons, figures that are well over state and national average yields for cassava. The local variety, used as check in this experiment (Platina) gave the lowest yield (27 ton/ha) although farmers ranked it as 2nd in their preferences. The two varieties that gave higher yield were ranked by farmers in 3rd and 4th place. Finding out about the excellent performance of other varieties different from the local one was one of the main conclusions and lessons learnt by the members of this COPAL and their collaborating farmers.

To increase farmers knowledge about CGM and to create local capacity for making sound decisions about cassava crop management, farmers from this COPAL were trained to identify the CGM, and to make field evaluations (damage score) of the level of incidence of this pest. Farmers were then able to perform their own weekly evaluations on the pest attack and each variety was ranked according to the incidence of the pest. The information obtained by farmers was sent to the CNPMF entomologist assisting them who prepared a final evaluation of the CGM incidence during the cropping cycle, the cumulative damage score (Figure 3.2.1.2). It can be observed from this data the good tolerance of the local varieties to mite damage, the main reason for its popularity among farmers. However, this local variety did not give the best yield, and some of the introduced varieties that showed medium tolerance to mites were more productive. These results were enthusiastically discussed by the farmers who realized the possibility of introducing and adapting new, more productive cassava varieties into their farming systems.

Results obtained in the experiment were discussed among COPAL members and other farmers and a second experiment was planned and planted. This new experiment include the top three varieties of the first trial as well as three new varieties recommended by EMBRAPA/CNPMF from its germplasm bank.

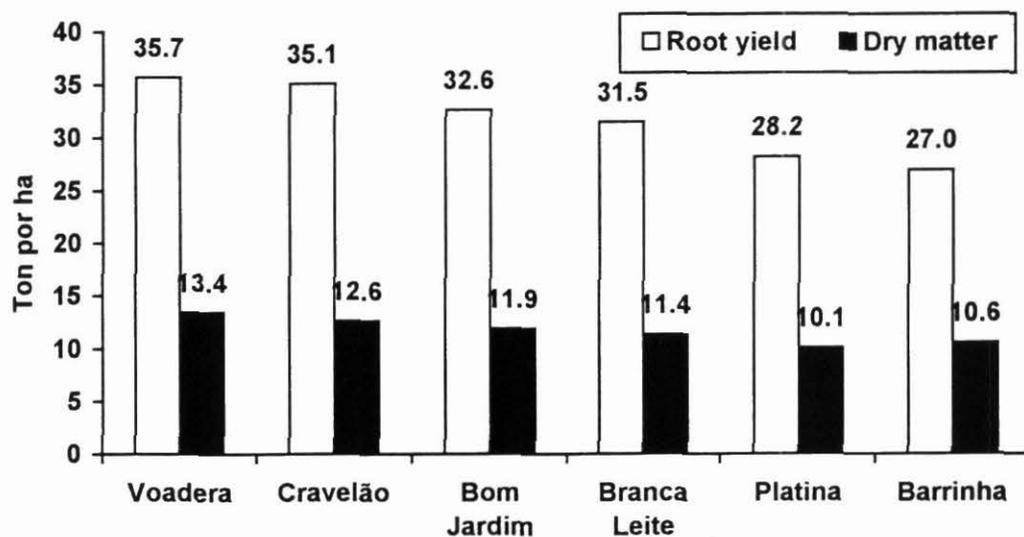
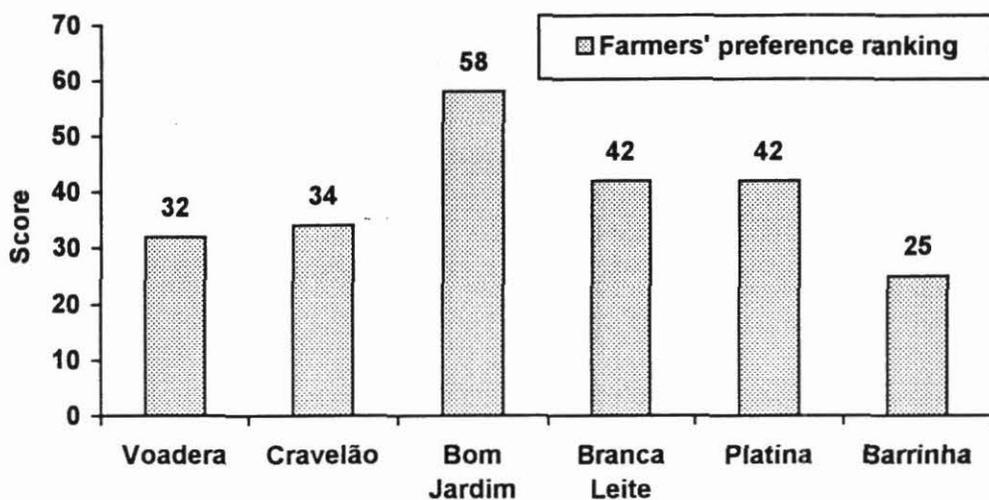


Figure 3.2.1.1. Evaluation of six cassava varieties for resistance/tolerance to cassava green mite; Agronomic and Preference Ranking Evaluation at Colonia Agricola R. Santos, Inhambupe, Bahia.

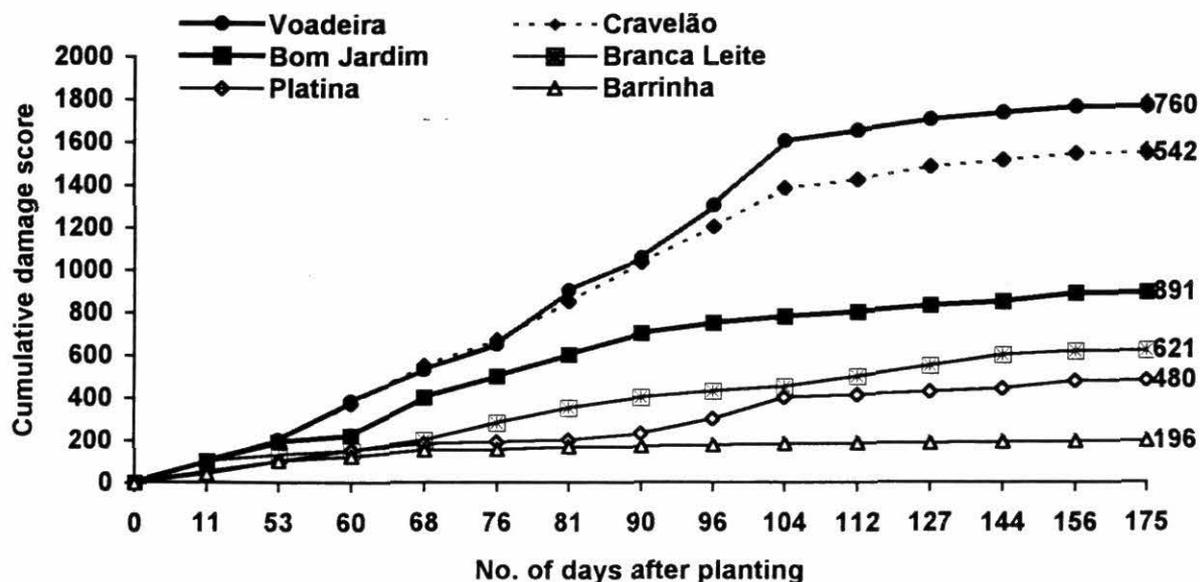


Figure 3.2.1.2. Evaluation of cassava green mite damage on six cassava varieties at COPAL: Colonia Agricola Rogerto Santos, Inhambupe, Bahia.

COPAL: Chapada, Aporá

The objective of the experiment in this COPAL was to evaluate the effect of two cassava varieties and two “cultural” practices (planting method) on the control of cassava root rot, a disease that has become a major constraint for cassava production in various states of Northeast Brazil. Farmers had prioritized this problem as their principal production constraint. One of the varieties (Osso Duro) was recommended by CNPMF researchers as a good source of resistance based on experimental results obtained in past years. The other cultivar (Cemitério) was selected by farmers as one the best genetic materials they have in the region. One of the cultural practices was recommended by researchers (planting stakes in ridges of soil) and the other is commonly used by farmers (planting on flat soil). The experiment was

planted in complete randomized blocks with four repetitions, and fertilizers were not applied. Yields obtained in this experiment were extremely low indicating the need for more intense long-term work in this community aimed at improving their cassava-based production system. The variety recommended by researchers showed a very low performance, giving the lowest yields. Low yields by this variety were also obtained in experiments in other regions during 1996 and has been attributed to a failure in the resistance that it had shown in the past. This material is being discarded by CNPMF as a source of cassava resistance to root rot. The cultural practices results were more encouraging since the recommended practice, planting in ridges, gave the best results for both varieties. The local variety planted in ridges gave 44.6% more yield than with flat planting, and the introduced variety produced 25.5% more when planted on ridges. This recommendation, which is not currently used by farmers in the region, could be the basis of future technology adaptation work. Farmer evaluation of the experiment (preference ranking), was consistent with the agronomic results, selecting as their best choices those treatments that gave the best agronomic results (Figure 3.2.1.3).

Future work in this community would be a great challenge for PROFISMA, collaborating institutions and for the COPAL itself. The community is one of the poorer farmer groups, and they are using cassava varieties that have extremely low productivity. It is surprising to notice that a few kilometers away there are other communities with varieties that are three or four times more productive. The future role of the COPAL could be crucial in the strategy of introducing and adapting better cassava varieties and improved cultural practices with the overall objective of improving cassava production, productivity and farmer incomes. Some cassava germplasm developed recently by CNPMF that is showing good resistance/tolerance to cassava root rot could be very important in future participatory research and technology transfer activities implemented through the COPAL in this region.

Results obtained in the experiment were discussed among COPAL members and other farmers, and a second experiment was planned including improved varieties introduced from EMBRAPA/CNPMF germplasm bank.

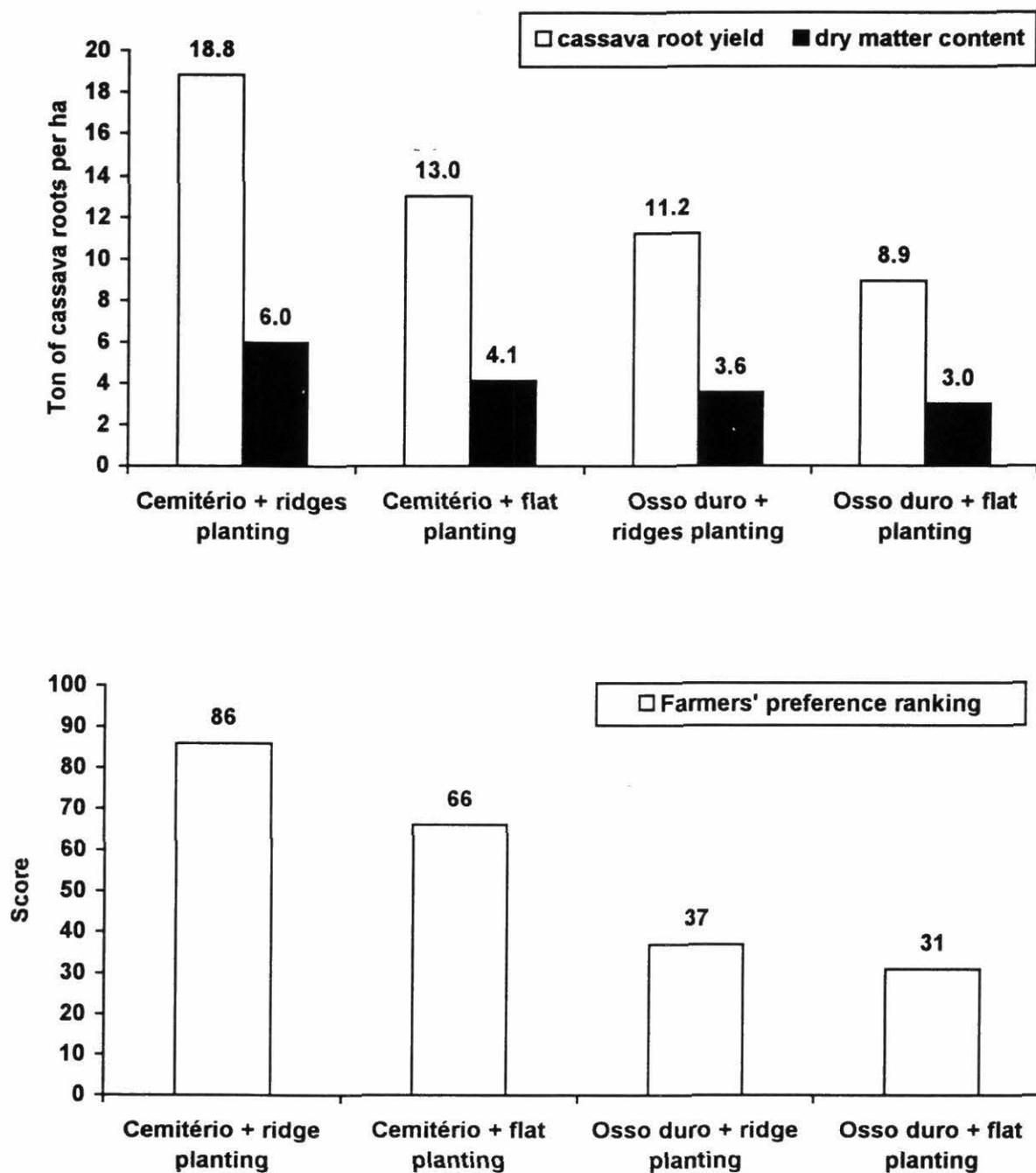


Figure 3.2.1.3. Evaluation of varietal resistance/tolerance and cultural practices for cassava root rot control. Agronomic and preference ranking evaluation at Chapada, Aporá, Bahia.

COPAL: Buril, Crisópolis

The prioritized problem in this community was also the incidence of cassava green mite (CGM). The objective of the experiment was to evaluate six cassava varieties for a resistance to CGM. Four of the cultivars were directly introduced from the CNPMF germplasm bank and had previously been identified through farmer participatory selection conducted in a germplasm development project financed by IFAD. The other two were selected by farmers from among the best locally produced material. The experiment was planted in randomized blocks with three repetitions. Planting distances were 1.0 by 1.0 m. One of the introduced varieties performed better than the local ones (Figure 3.2.1.4). The best introduced material produced 19% more roots yield than the best local variety. Two of the introduced materials showed lower dry matter (poorer quality) content than the local varieties, a factor to be considered in future breeding work. Farmers' preference ranking evaluation was not consistent with the agronomic data. They selected as the best two treatments the two local varieties (Platina and Jalé) that gave lower yields than the two best introduced materials (Maria Pau 118 and 128/08).

Farmers ranked three of the introduced materials as worse options although one of them, Maria Pau 118, was the most productive (Figure 3.2.1.4). The results were discussed with the COPAL and with other farmers of the community, and the planning of the second experiment resulted in the inclusion of these two promising clones, the introduction from CNPMF of two new ones and the inclusion of the two best local varieties as the check treatment.

Members of this COPAL also participated in training activities for evaluation of cassava green mite damage. The results of their evaluations are presented in Figure 3.2.1.5. It can be observed that Jalé, the preferred local variety did show the best resistance to cassava mites, justifying its local popularity. Three of the four varieties introduced from CNPMF showed medium tolerance to mite damage, but two of them (Osso Duro and 47/19) gave the lowest yields of the experiment. The other variety, Maria Pau 118, was the most productive, and although it was ranked very poorly by farmers, it showed them the possibility of adapting it to their cassava systems since it produced 19% more than their local best variety.

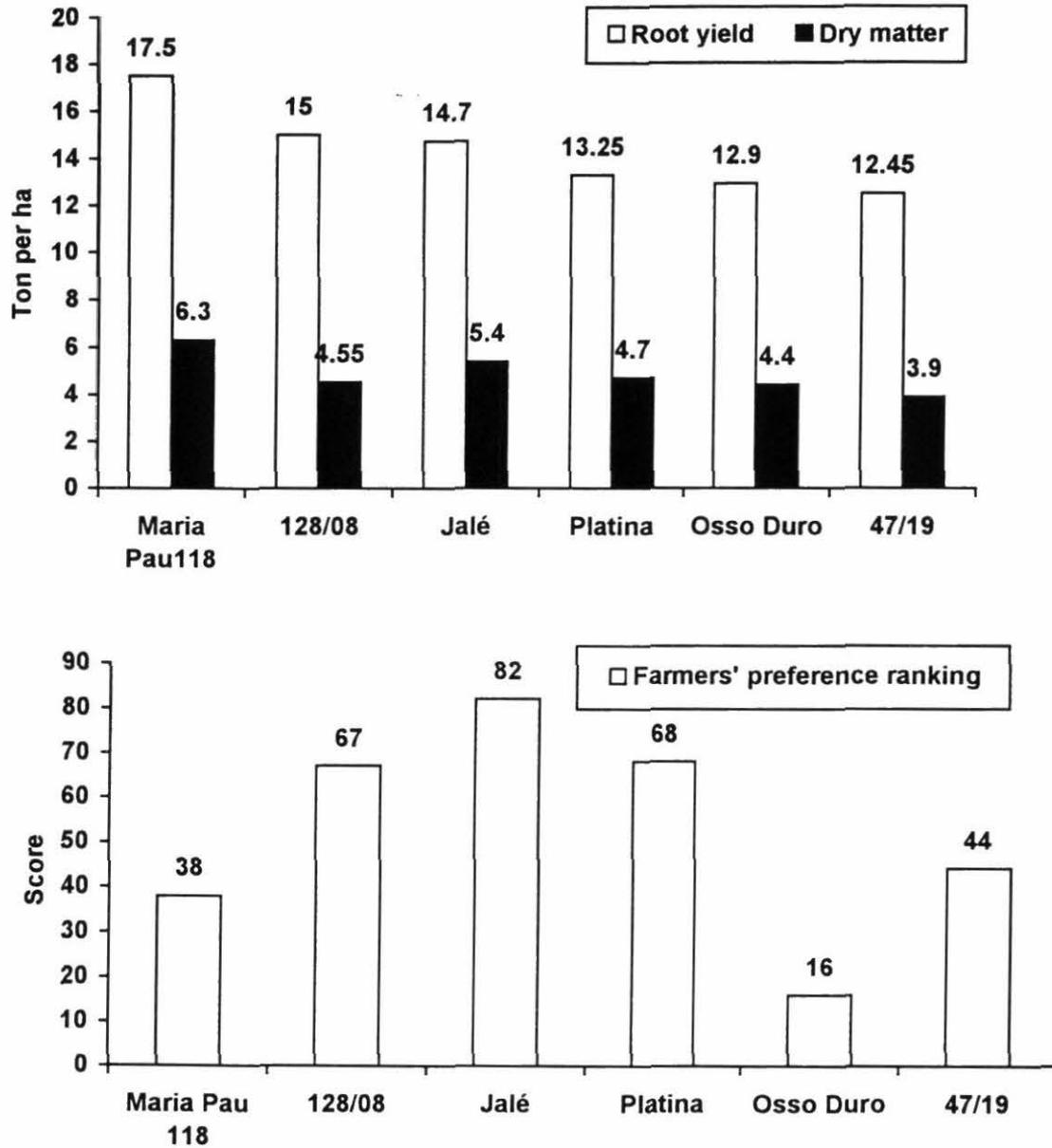


Figure 3.2.1.4. Evaluation of six cassava varieties for resistance/tolerance to CGM Agronomic and Preference Ranking Evaluation, COPAL: Buril, Crisópolis, Bahia.

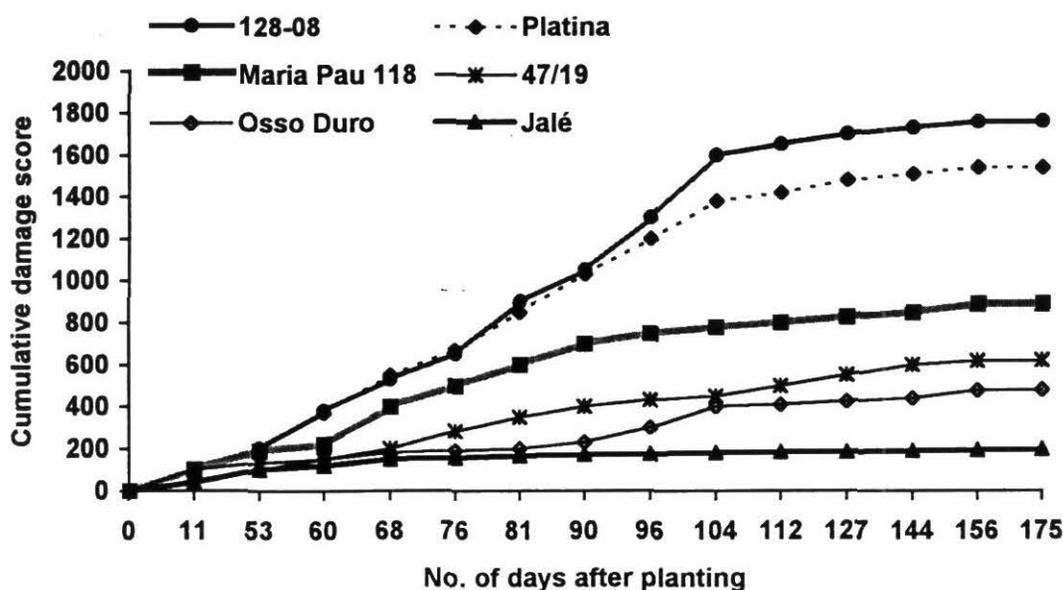


Figure 3.2.1.5. Evaluation of cassava green mite damage at Buril, Crisópolis, Bahia.

COPAL : Cadete, Cruz das Almas

Located 3 km from EMBRAPA/CNPMF headquarters, this COPAL has a comparative advantage over the rest because its proximity allows closer contact between farmers and technicians (researchers and extension workers). It also has become an excellent training and demonstration ground for PROFISMA and in general for CNPMF. The farmer group decided to plant an experiment to compare two methods of fertilization: organic compost and chemical fertilizer. Both are commonly used in the region, and there are some farmers that complain about the inefficiency and the high costs of the chemical fertilizers. The two more popular cassava varieties were used, and the experiment was planted in two different fields. Data discussed in this report refers to only one of them because soil hardness has prevented harvesting the other one. The results obtained were very interesting. First, there seems to be a varietal difference in the response to fertilization (Figure 3.2.1.6). One of the varieties (Cidade Rica) presented yield increases of 64% due to the use of organic fertilizer and of only 11% with the mixture of organic plus chemical. Chemical fertilizer alone with this variety gave the lower yield of the whole experiment. Results with the other variety (Cigana Preta) were even more interesting since the best yields were obtained without fertilizers. However, the other

variety with organic compost yielded 47% more. In relation to dry matter yield, the same tendency was observed, with the organic fertilization of the variety Cidade Rica showing the best yield (about 67% more dry matter yield per ha than the rest of the treatments). Farmer evaluation of the experiment (preference ranking), was consistent with the agronomic results, selecting as their three best choices these treatments with the best the best agronomic results (Figure 3.2.1.6).

Based on these results, the main point of discussion among COPAL members, other farmers and technicians was about the convenience of using fertilizers, especially chemical in cassava production. Farmers argued that this fertilizer is easy to get and sometimes cheaper. They are now starting to question the economic viability of this practice. Building up in this initial experience, the COPAL has already planted a second experiment in which some improved varieties recommended by CNPMF were introduced.

COPAL: Barra, São Miguel das Matas, Bahia

Cassava farmers in this community diagnosed whiteflies (*Aleurothrixus aepim*) as the main constraint to cassava cultivation in the region. The objective of the experiment designed with this COPAL was to evaluate five cassava varieties for whitefly resistance/tolerance. Three of the varieties planted were introduced from CNPMF germplasm bank and were recommended for testing by the COPAL based on excellent results obtained in other experiments in similar semi-arid ecosystems. These three clones were also selected as promising varieties by farmer groups involved in another farmer participatory research project being executed by CNPMF and CIAT in Northeast Brazil. The other two varieties were selected by the farmers as their best locally available options. The experimental design was complete randomized blocks with five repetitions (five different farmer fields located within a distance of about 1 km). Although the main objective of the experiment could not be addressed to a great extent due to the absence of a severe attack of whiteflies during the experiment, the results obtained allowed farmer members of the COPAL, other farmers of the community and technicians some important initial conclusions.

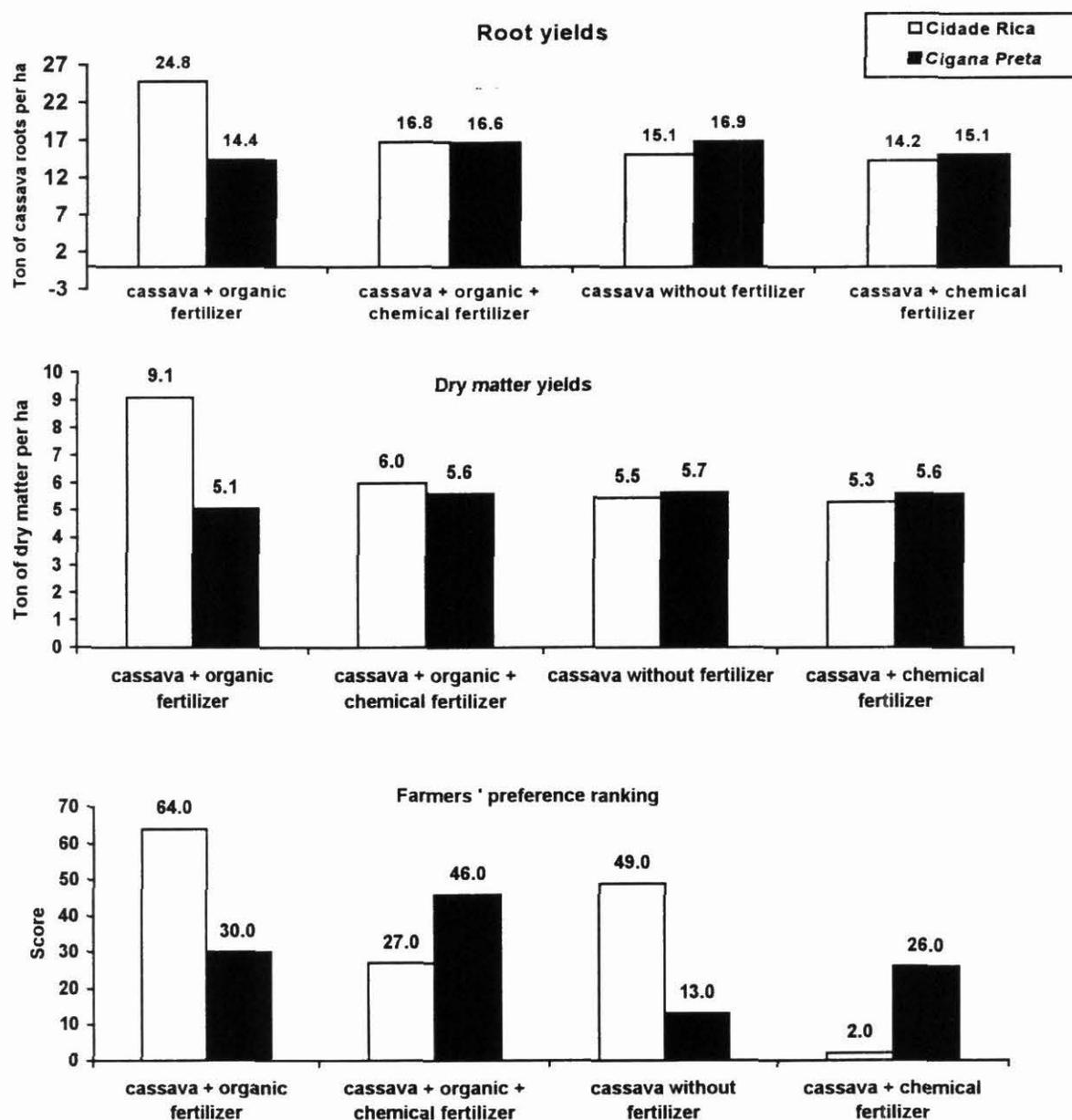


Figure 3.2.1.6. Evaluation of fertilization methods on yields of two cassava varieties at COPAL Cadete, Cruz das Almas, Bahia.

First, two of the varieties introduced from CNPMF gave yields (roots and dry matter) 50% higher than the two best local materials; a fact that caught the interest of the farmers for further testing and adaptation of these materials to the region (Figure 3.2.1.7). Second, cassava yields with local varieties is extremely low in this region considering the fact that they commonly use chemical fertilizers. The farmers' main cash crop is tobacco, and they obtain the fertilizers directly from the tobacco companies in exchange for the product, at prices fixed unilaterally by these companies. This represents a very interesting topic for future research by the COPAL, which, besides the interest for new, improved varieties, expressed interest in testing organic and chemical fertilizers and their effect in the production of tobacco and cassava. They argued that this could help them becoming less dependent from the tobacco companies. Last but not least, this experiment served to confirm the importance of allowing farmers opinions to be incorporated in the development and dissemination of cassava germplasm. As shown with the results obtained, when the cassava varieties pass through a selection process during which farmers have the chance to evaluate them using their own criteria, the possibility of good performance and good acceptance by other farmers when these materials are taken into similar ecosystems is greatly enhanced.

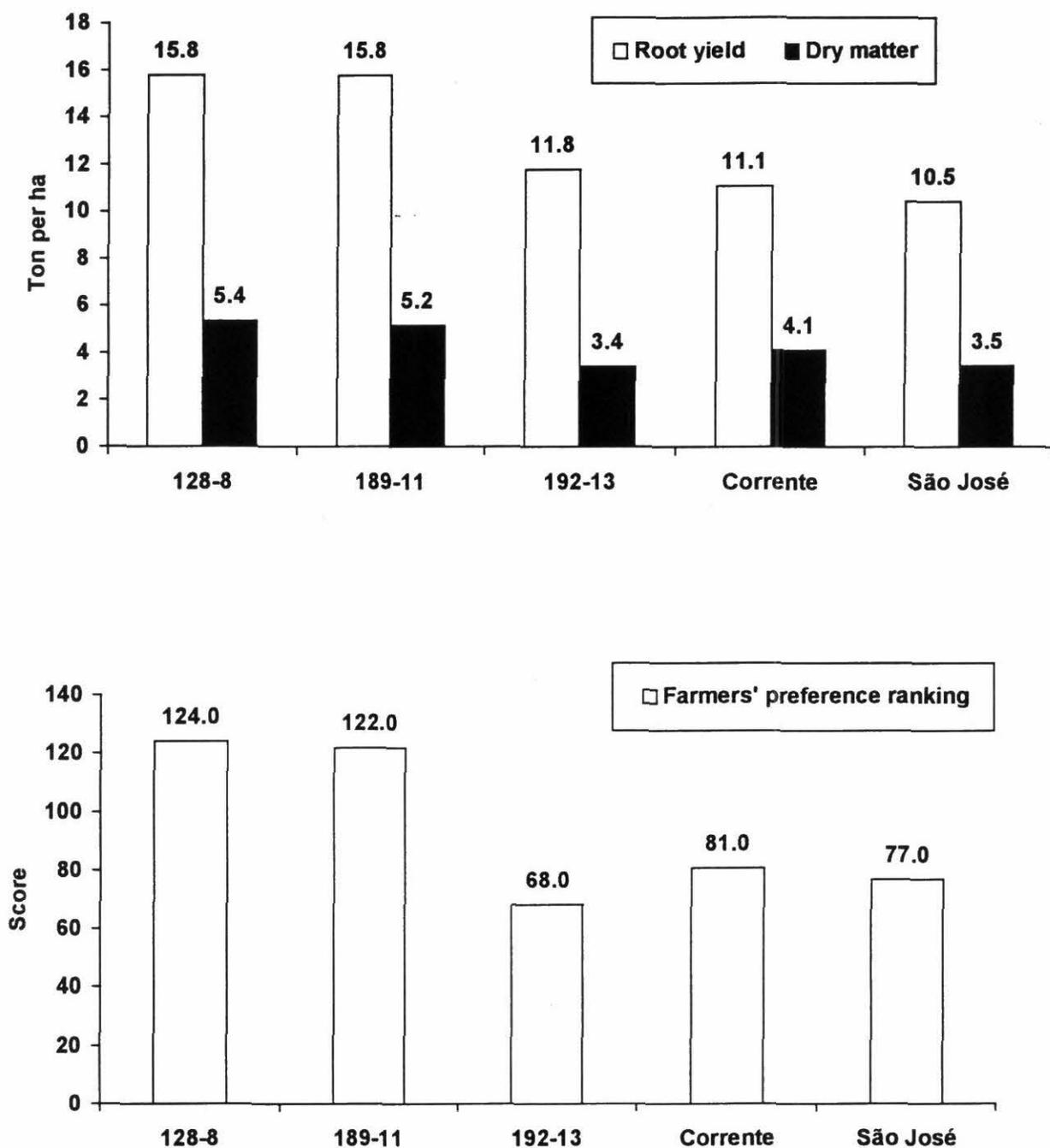


Figure 3.2.1.7. Evaluation of five cassava varieties for resistance/tolerance to whiteflies: Agronomic and Preference Ranking Evaluation at Barra, São Miguel das Matas, Bahia.

COPAL: Umbuzeiro, Anguera, Bahia

This COPAL is the only one of those established with PROFISMA's support in which the majority of the farmers are women. Cassava producing and processing in this community is done mainly by women who work on small pieces of land, usually rented. Farmers prioritized low productivity and poor fertility of their soils as the constraints on which they would like to work. Considering the lack of economic resources of the group, it was decided during the planning stage that the main objective of the experiment would be to test the effect of planting cassava inter-cropped with legumes which could help to improve soil fertility. The two legumes chosen were cowpea (*Vigna unguiculata*) and jack bean (*Canavalia ensiformis*). To facilitate crop management the double-row planting system was introduced. Farmers decided to use their best local variety, and the experiment was planted in three different farmer fields. Results obtained indicated the high degree of complexity when an attempt is made to modify a local, traditional farming system. In this case, the use of the two legumes associated with the cassava crop demanded certain specific management, especially the timing of the pruning, which was not clearly understood by the farmers, and even by the technicians giving technical assistance to them. Due to different problems, the technicians were not able to maintain a systematic, periodic presence in the community to advise farmers' decision-making on management of the experiment. As a consequence, these two legumes were allowed to grow beyond flowering stage. Competition with cassava was very severe, especially during a drought period that affected the region in the early stages of the experiment, and cassava yields were very low. The two treatments without legumes resulted in higher yields, and the introduced double row planting system was slightly better than the traditional, single row system. Farmers' preference ranking evaluation system was consistent with the agronomic results (Figure 3.2.1.8).

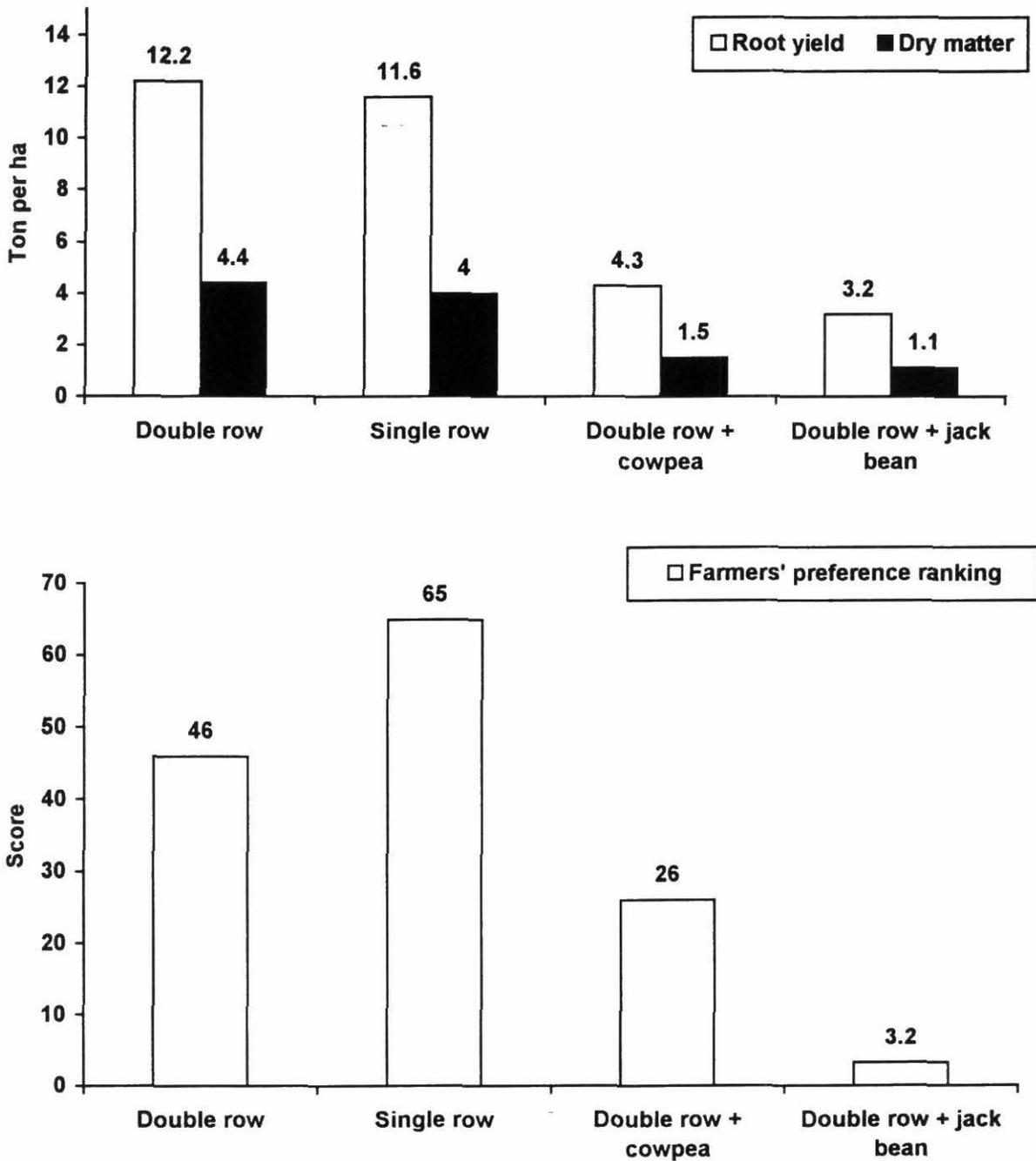


Figure 3.2.1.8. Evaluation of cassava planting systems (double row, single row and legumes intercropping) at Umbuzeiro, Anguera, Bahia.

The lesson learnt by the researchers and extension workers giving technical assistance and advice to the farmers was very important. It became clear that there is a need to maintain a systematic, periodic presence in the community to advise the COPAL farmers, especially in those moments in which they are forced to make decisions about crop management in farming systems and practices that are new to them. Another important conclusion was that during the initial experiences of farmer groups as researchers it does not seem appropriate to mix too many treatments (for example, planting distances, inter-cropping, etc.). It is better to initiate the work as simply as possible allowing farmers to gain experience little by little as the work becomes more complex.

Farmers discussions after the harvest and evaluation of their experiment indicated their desire to introduce improved cassava varieties in their future activities. It is expected that genetic material which is currently available at CNPMF's germplasm bank and that has been especially developed for these semiarid, low soil fertility ecosystems will be fundamental in the continuation of the work by this COPAL.

3.2.2 State of Ceará

COPAL: Nova Veneza, Ubajara, Ceará

The community of Nova Veneza is composed by 120 families and is located in the Serra de Ibiapaba, a region characterized by a relatively good annual rainfall and moderate climate which allows year-round production of various crops such as sugar cane, fruits, corn, beans and cassava. The importance of cassava in the region is mostly for farmers' own consumption of cassava flour (*farinha de mandioca*) since the other crops are more important as a cash and income source. Nonetheless, cassava is widely grown in the region, and the COPAL in this community identified as the main production constraint the lack of improved, more productive varieties and alternative, more efficient cropping practices, especially fertilization. Additionally, cassava production in the region has been affected drastically during the last five years by the disease known as mycoplasma witches' broom (WB) which caused a severe reduction in production, productivity and area planted.

Farmers included two local varieties in their experiment: one, Cruvela, is a local cultivar widely grown in the region and the other, Buja de Olho Roxo, was introduced as one of the varieties that shows some tolerance to WB. The experiment was planted with a randomized block design with six treatments and three repetitions. The two main characteristics of the experiment were the introduction of two legumes: *Crotalaria* and pigeon pea, as sources of green manure, and the use of Compost, a practice that the Extension Service (EMATERCE) is promoting intensively in the region. The results of the experiment are presented in Figure 3.2.2.1. It can be observed that the introduced cultivar, Olho Roxo gave higher yields for all treatments (an average increment of 48% for the whole experiment), and all the treatments in which the compost was utilized also gave higher yields (an average increase of 128% for the variety Cruvela and of 41% for the variety Olho Roxo. The overall best result was obtained by intercropping cassava with pigeon pea and using compost. This practice compared with the traditional system of planting cassava alone gave a yield increase of 225% for the variety Cruvela and of 69% for variety Olho Roxo). Results obtained with dry matter content showed the same trend suggesting a beneficial effect of the legume and the compost on the dry matter content of the cassava roots. It has been reported in the literature that pigeon pea is one of the few legumes that has a unique mechanism that allows them to access phosphate that is bound to calcium and iron soil particles. These legumes release acids from their roots which react with calcium-bound and iron-bound phosphate to release phosphate for plant uptake. Additionally, their deep rooting characteristic has a positive effect helping water infiltration.

Preference ranking evaluation conducted by farmers was somehow different from the agronomic results. Farmers gave selected as their best choice, for both varieties, a treatment which was the third place in yields and dry matter content. Their choices for the worse treatment did coincide with the agronomic evaluation (Figure 3.2.2.1).

An interesting aspect of the work conducted by this COPAL is that they are now utilizing their experiences and skills as “farmer researchers” and have installed some simple experiments with other crops such as pepper and tomato. Additionally, this COPAL is being used by EMATERCE as a demonstration, training ground in which other communities are invited for fields days on COPAL methodology, compost fertilization and other topics. In

August 1996, this COPAL hosted with great competence the First State Day for COPALs promoted by local collaborating institutions.

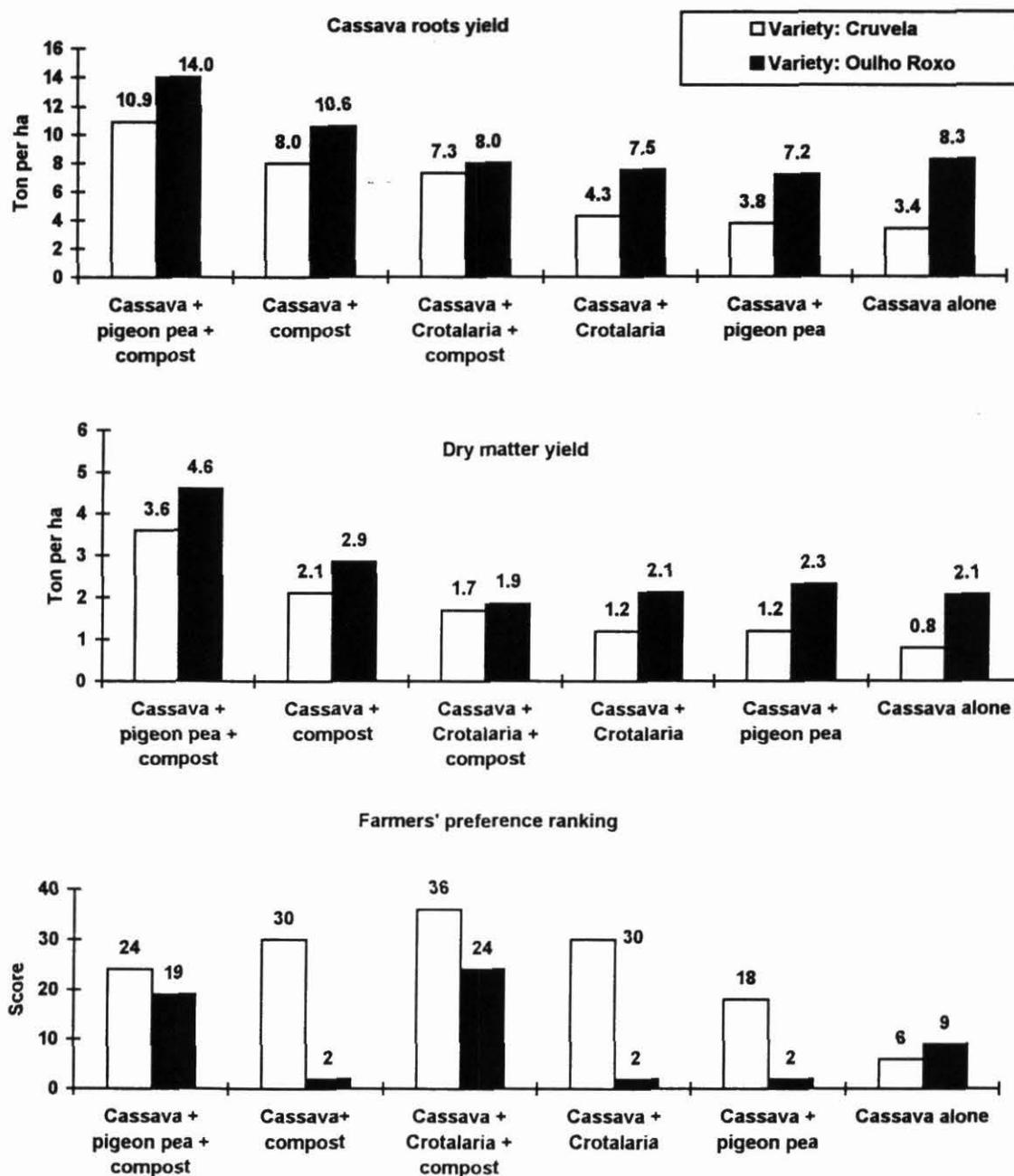


Figure 3.2.2.1. Evaluation of cassava intercropping with pigeon pea and *crotalaria* in double row planting with compost fertilization at Nova Veneza Ubajara, Ceara.

COPAL: Valparaiso, Ubajara, Ceará

This COPAL is the only farmers research group supported by PROFISMA that is located in a land reform community. The group is composed by 65 families and practices collective production and processing of various crops such as cassava, corn, beans, and banana. Cassava is a very important crop for the group, which is attempting to become self sufficient and also uses surplus cassava flour for commercialization. The crop has been affected severely during the last few years by the mycoplasma witches' broom disease (WB) and the group has been actively involved in research work conducted by CIAT, CNPMF and local institutions aimed at finding sources of genetic resistance/tolerance to this disease.

The objective defined by the COPAL for its first experiment was then related to this problem. They were getting very enthusiastic about some hybrids that have started to come out from these activities and decided to test the one they liked best, the hybrid 8709-02 (local name is Salamanta), and a local clone named Cabelo de Velha. The experimental design also included the use of the legumes pigeon pea and *Crotalaria* and compost as fertilizer. The total number of management treatments was six, with three repetitions for each.

The results obtained in the experiment were completely different for the two varieties. The hybrid Salamanta confirmed that it has indeed a great potential in the region giving higher yields than the local clone in five of the six treatments. The average yield for this hybrid for the whole experiment was 79% higher than average yield with the local variety. Considering the effect of the compost on yields, for the hybrid Salamanta, it can be observed that average yield for the three treatments that have compost was 20% higher than these respective treatments without compost. The trend for the local variety was totally opposite, with the treatments without compost giving an average yield 55% higher than these respective treatments that were fertilized with compost. This data seems to suggest that there is a genetic difference in the response to fertilizer because in both cases, the compost was prepared at the same time and applied at the same rates. The intercropping with pigeon pea was the best treatment for the local variety and was the second best one for the introduced hybrid, confirming the beneficial effects of this practice on cassava yields. The dry matter content results showed the same patterns as the yield data for both varieties. Comparing the local, traditional planting system with the best treatments for each variety, it can be observed that

for the introduced hybrid the maximum increase in yield was 26.5% whereas for the local variety this was 46.8% (Figure 3.2.2.2).

Preference ranking evaluation conducted by farmers was contrasting for both varieties. In the case of the introduced hybrid, although the local planting system had the lowest yields, farmers ranked it as their second choice, only exceeded by the treatment that included compost only. With the local variety, farmers's opinions generally matched the agronomic results, and their best two options were exactly those with highest yields (Figure 3.2.2.2). Although the introduced hybrid was ranked equally with the local variety when each was planted alone, it was rated lower than the local variety when cultivated with compost (with or without pigeon pea) – despite the fact that yields were higher by 67 to 77%. The discrepancy between farmer ratings and yield indicates the importance of using additional measures of crop characteristics to evaluate and select germplasm. The main conclusion of this experiment is the ratification of the great potential that this hybrid seems to have for the cassava systems in the region. CNPMF is intensifying its work on further testing, multiplication and release among farmers, as well as maintaining germplasm development activities to find similar or better genetic materials. In early 1997, a Field Day is planned jointly between CNPMF, local institutions and the COPAL to officially deliver this hybrid to cassava growers in the region.

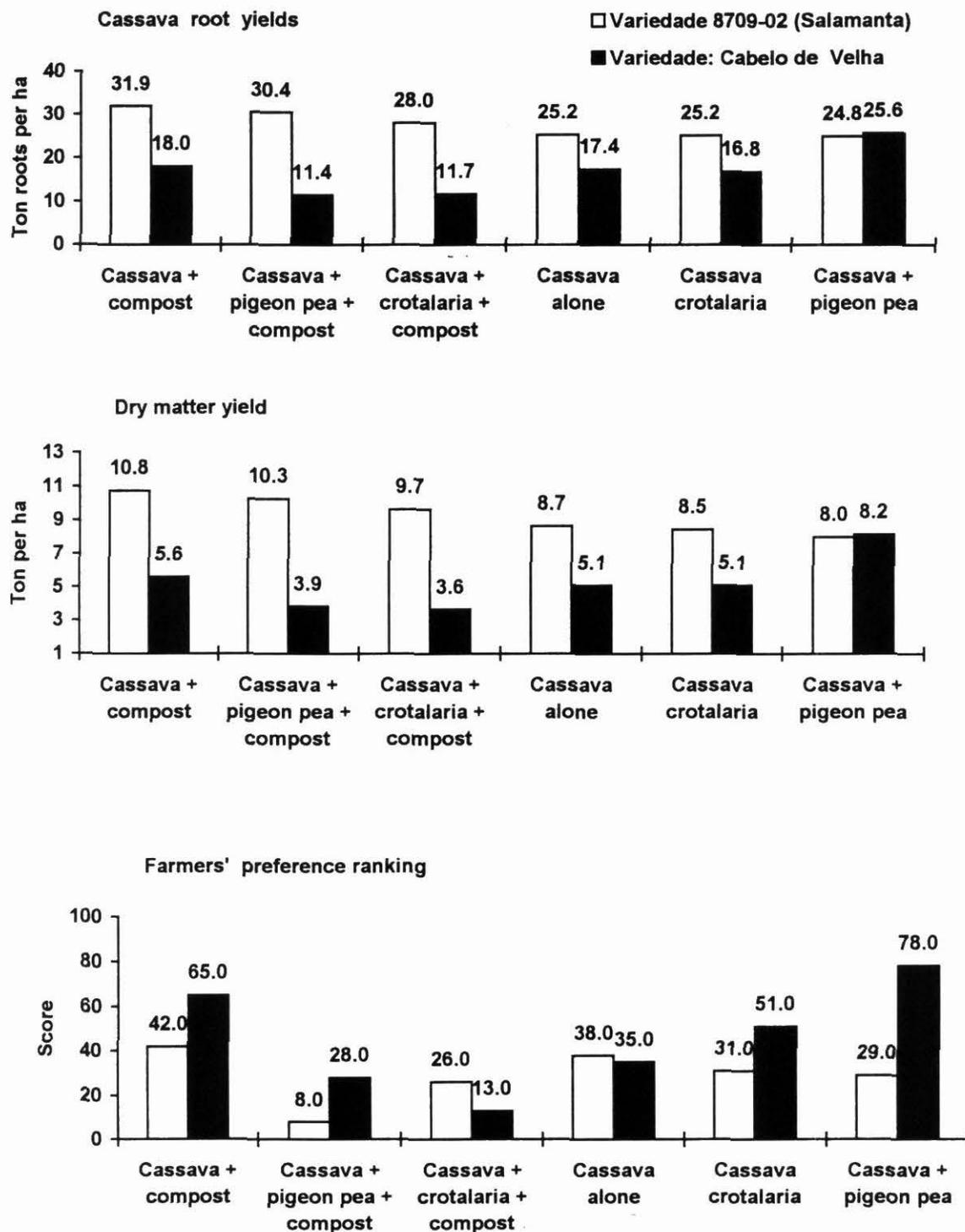


Figure 3.2.2.2. Evaluation of cassava intercropping with pigeon pea and crotalaria in double row planting with compost fertilization at VALPARAISO, UBAJARA, CEARA.

COPAL: Vila Moura, Acaraú, Ceará

This community is located in the semi-arid region of Ceará, a drought-prone area in which limited rainfall gives farmers very few agricultural options, among which cassava is on top of the list. Cassava production in the region is practiced intensively with a growing period that in average exceeds 20 months. Farmers argue that cassava production with just one “winter” is not profitable (rainy season) and plants are usually allowed to grow during the second year. The COPAL defined as its experimental objective the testing of organic compost as fertilizer in cassava production mixed with two legumes (jack bean and velvet bean). The two legumes were intended to be used as mulch and the experiment included the use of compost and a planting arrangement in double rows to facilitate farmers’ management of the experiment. Since the main objective was to increase farmers’ knowledge about the management of this new system, and especially of the two legumes, the farmers decided to harvest the experiment after one year. The best two local varieties were used. The experiment was planted in randomized blocks with six treatments and three repetitions. A similar experiment was planted in the second year switching positions between the cassava and the legumes in each plot. Evaluation of both experiments did not include determinations of dry matter and farmers’ preference ranking evaluation.

The results of the experiment are presented in Figure 3.2.2.3. It can be concluded from the data obtained that cassava production in the region with just one growing cycle gives indeed very low yields. The best results with both varieties were obtained using compost without legumes. The use of velvet bean as mulch was practically null due to severe attacks of leaf-cutting ants that destroyed it. The adaptation of jack bean to the dry climate was very good, but it became evident that some management changes have to be introduced because it competes severely with the cassava crop. Probably the best option is to use just two rows in between the cassava plants and increase the spacing. Nonetheless, farmers were very impressed with the growth of this legume and especially with the concept of growing a plant to be, later on, cut and incorporated into the soil.

The best yield in both experiments was obtained with the variety Frágoso, in the treatment cassava alone + compost (13.45 ton/ha), a yield which is around the average in the region for

two cycles. Some conclusions were evident for the members of the COPAL based on the results of this experiment:

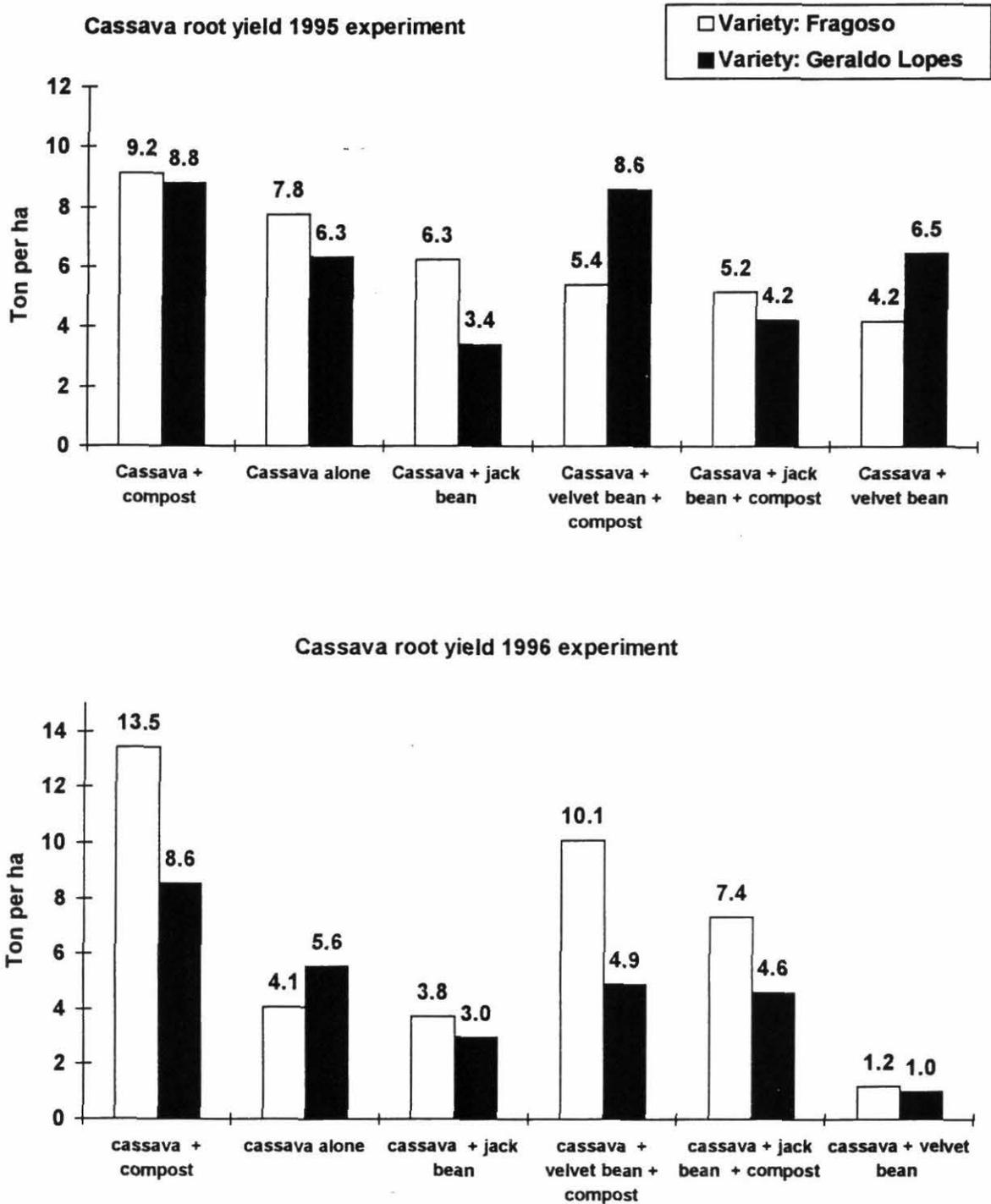


Figure 3.2.2.3. Evaluation of cassava intercropping with jack bean and velvet bean in double row planting with compost fertilization at Vila Moura, Acaraú, Ceará.

- In the region farmers are used to plant corn and beans intercropped with cassava and this makes the use of legumes for soil fertility recuperation more difficult to be adopted by farmers;
- Compost use proved to be an attractive, cheaper option for farmers;
- There is a need to study the economic viability of planting varieties with short growth period (harvest in one cycle), using organic compost. This system gave higher yields in this experiment.

COPAL: Lagoa Grande , Acaraú, Ceará

The Farmers Association of Lagoa Grande has been collaborating with local research and technology transfer institutions during the last decade. Cassava research and development projects (processing, germplasm development, crop protection) executed in the State by EMBRAPA, CIAT and local agencies had all included this group as one of the principal collaborators and beneficiaries. The Ceará Integrated Project for example, executed in Ceará during the period 1989-92, allowed the group to install a dry cassava chip agroindustry. During the last seven years, this farmer group has been the only one in Ceará that has been able to maintain every year a reasonable volume of processed dry cassava chips. The reasons behind this success are various, but probably the principal one is the excellent organizational and administrative level that the group has achieved. For example, in 1996 the profits obtained by the group with cassava processing activities were used to buy one ton of cow manure for each member. Farmers use organic fertilizer systematically in their fields and cassava production and productivity levels in the community are very good. The COPAL decided to work on cassava varieties for human consumption (sweet varieties). The main argument to justify this experimental objective was that sweet cassava varieties are almost disappearing in the region although in past years they were very popular among farmers. The COPAL feels that there is great demand and good market opportunities for these varieties and decided to start an identification/observation with those materials available in the State with the aim of adapting and selecting the most appropriate ones for their ecosystem.

The observation field was installed in an area closed to the community that belongs to a local University (Universidade do Vale do Acaraú). The integration with this University could be a very important support for the COPAL future technology adaptation efforts. The experiment included 21 varieties collected in different regions of the State. The planting system was double rows intercropped with six different legumes. These two practices were new to the group. After 15 months the experiment was harvested, and farmers performed an evaluation of the best cultivars based on criteria such as plant development, root production and especially palatability (cooking time and softness). After this evaluation, only 10 cultivars were selected for future research work. Some legumes used in intercropping with the cassava were harvested and the seeds were sold to farmers in the region. This activity could be an *additional source of income for the farmers.*

4 STRATEGIC RESEARCH AT CIAT

4.1 Biological Control of Cassava Green Mite, by Lincoln Smith

The cassava green mite (CGM, *Mononychellus tanajoa*) is one of the principal pests of cassava in Africa and Northeast Brazil. The principal objectives of the CIAT acarology unit are to find natural enemies (predators or pathogens) of CGM, evaluate them for safety and suitability as biological control agents, and send them to EMBRAPA in Brazil and IITA in Benin for release. Corollary activities include conducting research to further develop methods to evaluate natural enemies, mass rear them and release them; to publish findings and disseminate information to national programs; and to assist Brazilian scientists and extension agents working on integrated pest management of CGM.

Exploration for natural enemies

Explorations in 1996 were conducted to find phytoseiid predatory mites in two target ecological zones: 1) hot semi-arid (corresponding to NE Brazil) and 2) high altitude/subtropical (corresponding to the East African plateau). For the hot semi-arid target, we collected 5 species of phytoseiids in Manabí, Ecuador from 11 sites with 425-1500 mm annual precipitation, 6-9 dry months (<60 mm rain), 24-26°C average temperature, and 16-120 m elevation. For the high altitude/subtropical target, we collected 6 species of phytoseiids from 20 sites in Antioquia, Boyaca, Caldas, Cundinamarca, Quindio, Risaralda, Santander, and Tolima, Colombia from sites with 425-1500 mm annual precipitation, 0-4 dry months, 14-26°C average temperature, and 495-2216 m elevation. We established 7 new colonies of phytoseiids from high elevations for evaluation.

A culture of *Typhlodromalus aripo* from Palmira was established for biological investigations. This appears to be the most effective of the three mite predators established in Africa, yet little is known about why it is so effective. Also there appear to be two kinds of "*T. aripo*" which differ in their biology and may actually be different species.

Cultures of the fungal pathogen *Neozygites* sp. were collected from cassava green mite and two-spotted spider mite (*Tetranychus urticae*) at CIAT, Palmira and from CGM at Media Luna, Colombia.

Fig. 4.1.1 shows the distribution of 685 sites explored for mite predators of cassava green mite in Latin America by CIAT and EMBRAPA.



Figure 4.1.1. Sites in Latin America sampled for predators of cassava green mite.

Mite colonies maintained

Table 4.1.1 lists the colonies of phytoseiid predatory mites maintained at CIAT for experimental evaluation. Three species of Tetranychid pest mites (*Mononychellus tanajoa*, *M. caribbeanae*, *Tetranychus urticae*) were maintained to feed the predators and to use in experiments. Several species of insects (thrips *Corynothrips stenopterus*, mealybug *Phenacoccus herreni*, whitefly *Aleurotrachelus socialis*) and the foliar fungal plant pathogen, *Oidium manihoti*, were also maintained for use in feeding experiments.

Table 4.1.1 Phytoseiid strains maintained in laboratory colonies at CIAT, 1996.

Species	Origin				Date
	Country	Dept./State	Municipality	Location	Collected
<i>Typhlodromalus manihoti</i>	VENEZUELA	Yaracuy	Marín	San Felipe	Mar-95
"	BRASIL	Bahia	Cruz das Almas		Feb-93
"	COLOMBIA	Cauca	Cajibío		Jun-95
"	"	Antioquia	Barbosa	El Hoyo	Jan-96
"	"	Antioquia	Copacabana	Montañita	Jan-96
"	"	Magdalena	Medialuna		Feb-96
"	"	Guajira	Villanueva		Jan-96
"	"	Caldas	Chinchiná	San Gregorio	Aug-96
"	"	Santander	B/manga	Bijagual	Aug-96
"	"	Santander	B/manga	Los Colorados	Aug-96
"	"	Risaralda	Sta. Rosa de Cabal	UNISARC	Aug-96
"	"	Quindío	Armenia	Armenia	Aug-96
<i>T. aripo</i>	COLOMBIA	Valle	Palmira	CIAT	Sep-96
<i>T. tenuiscutus</i>	ECUADOR	Manabí	Portoviejo		Nov-94
"	"	Manabí	Puerto Cayo	Cantagallo	Dec-95
<i>T. limonicus</i>	BRASIL	Sao Paulo	Jaguariuna		Jun-90
<i>N. idaeus</i>	ECUADOR	Manabí	Rocafuerte	Entrada a Danzarín	Dec-95
<i>N. californicus</i>	ECUADOR	Manabí	Portoviejo		Nov-94
<i>G. annectens</i>	ECUADOR	Manabí	Portoviejo	Crucita/La sequita	Dec-95
<i>G. helveolus</i>	ECUADOR	Manabí	Puerto Cayo	Cantagallo	Dec-95
<i>Euseius ho</i>	ECUADOR	Manabí	Rocafuerte	Entrada a Danzarín	Dec-95

Climatic matching with Zambia

So far, none of the imported predatory mites have become established in the East African plateau. We suspect that this is because this target region is too hostile to the strains that were imported to IITA in recent years. Locations in Zambia where IITA had collected CGM were used as the basis for defining the climate of the target region (Fig. 4.1.2). Long term climatic data was associated with each of these sample sites using the average of the four nearest climatic stations, weighted by $(1/\text{distance}^2)$. Principal component analysis was then used to develop a statistical model of the climate in the target region (Fig. 4.1.3). This model was used in the GENSTAT program to develop a map of the Americas showing the degree of climatic similarity to the target release region (Fig. 4.1.4).

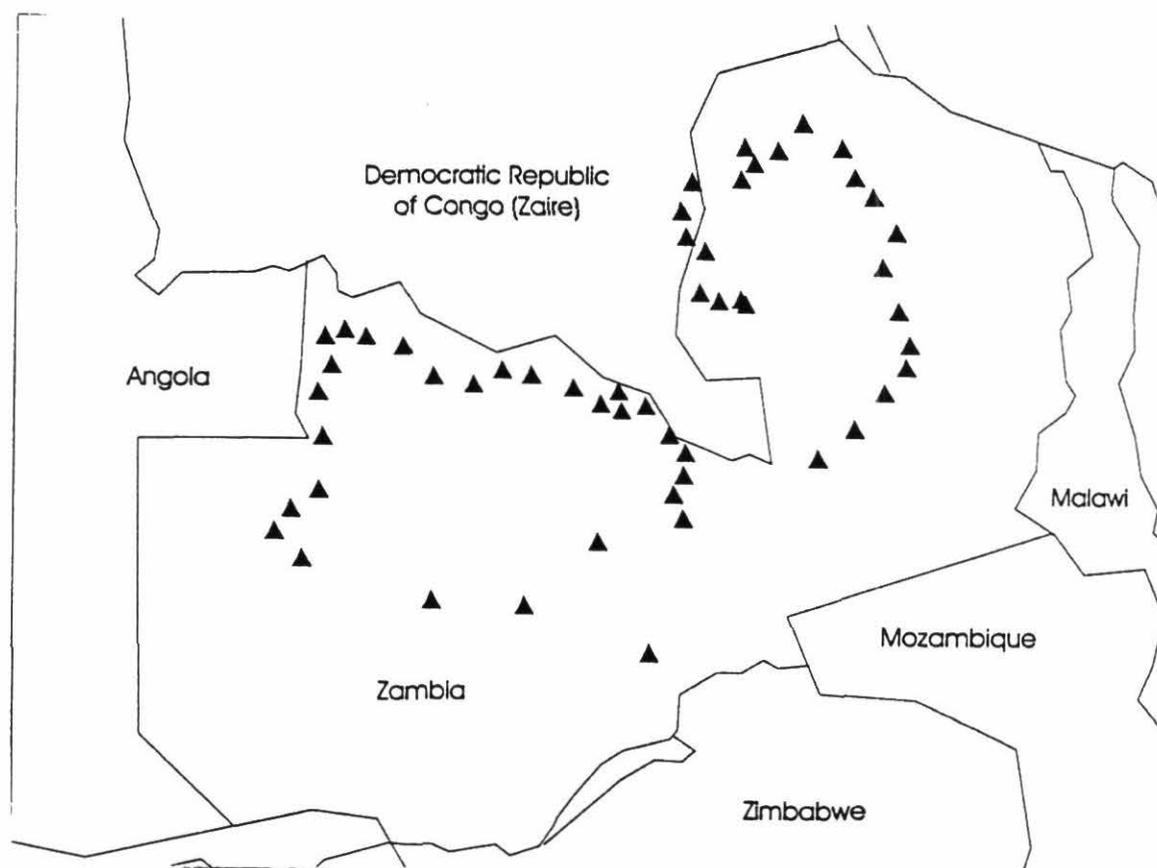


Figure 4.1.2. Sample sites in Zambia that served as the target for generating principal component analysis of climate (N=52).

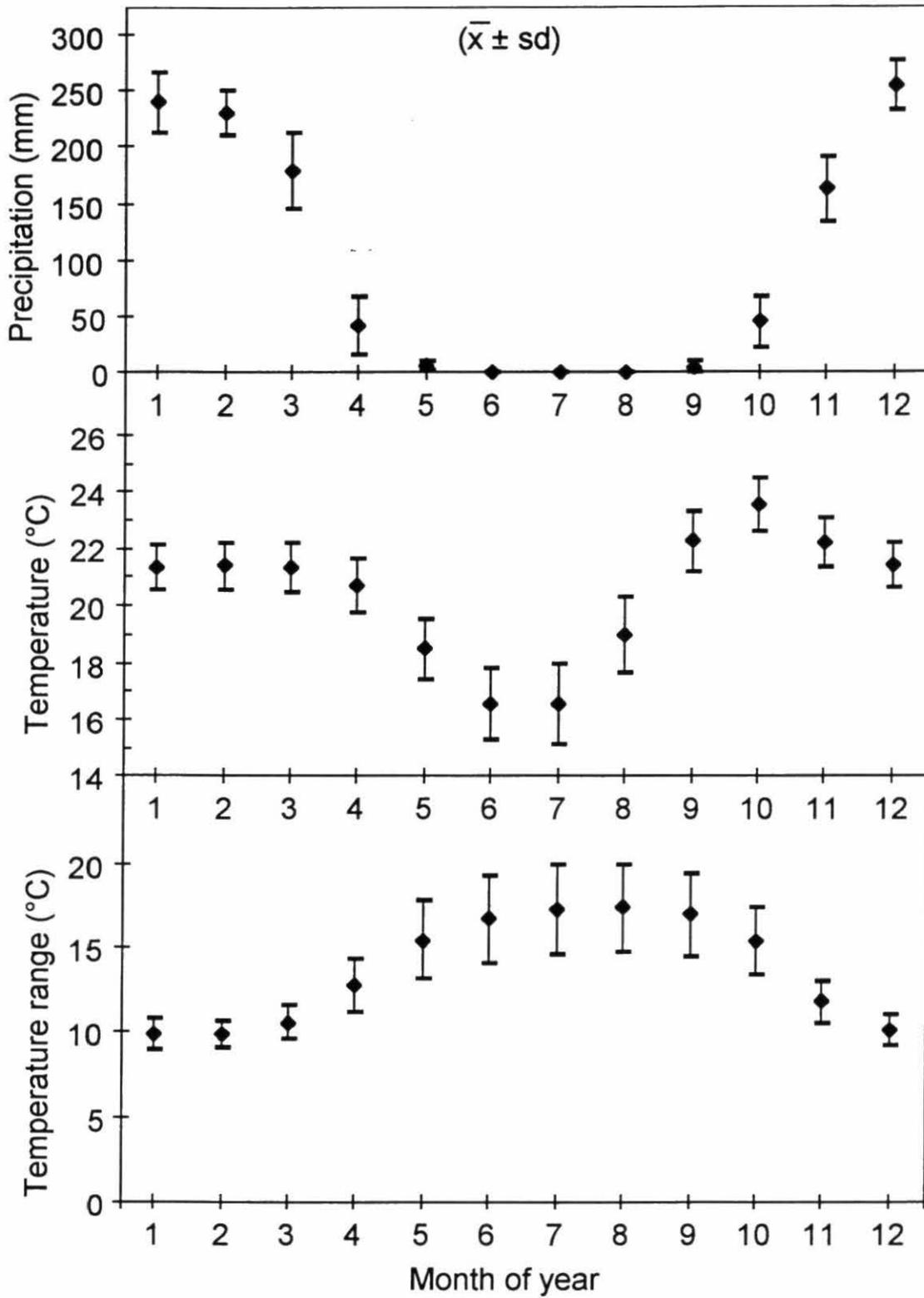


Figure 4.1.3. Climate of Zambian target region, based on 52 sample sites (precipitation, temperature, diurnal temperature range).

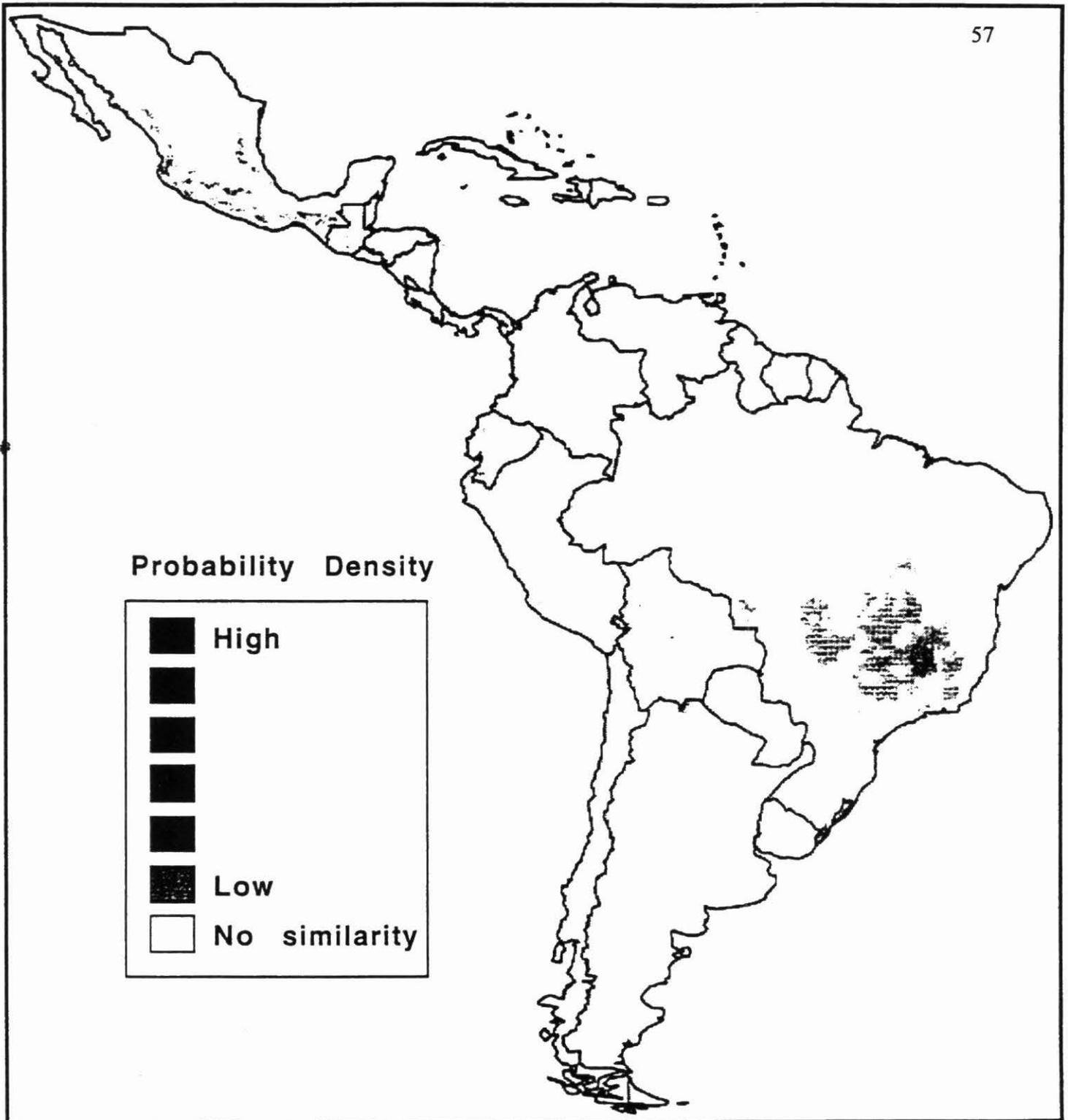


Figure 4.1.4. Map of climatic similarity to target sites in Zambia used to identify high priority regions in the Brazilian central highlands and Mexico for predator exploration.

Suitability of different prey for phytoseiids

Phytoseiid species that are candidates for exportation as biological control agents were evaluated for the specificity of prey and food sources that they can consume. This is to assure that we select species that will not harm non-target hosts after being imported to Africa or Northeast Brazil. The ability to use pollen or sugary exudates of cassava leaves, mealybugs or whiteflies is an advantage in helping to maintain predator populations when cassava green mites are scarce. *Galendromus annectens* and *G. helveolus* showed much higher longevity in the presence of tetranychid prey (*Mononychellus caribbeanae* or *Tetranychus urticae*) than for the other food sources tested (Fig. 4.1.5). *Neoseiulus idaeus* was similar except that it could also utilize pollen from castor bean (*Ricinus communis*). *Typhlodromalus tenuiscutus*, *Euseius ho* and *T. aripo* could use a greater variety of food sources, including cassava exudate (a sugary liquid found on leaves), immature whiteflies (*Aleurotrachelus socialis*) and mealybugs (*Phenacoccus herreni*); both of which produce sugary exudate.

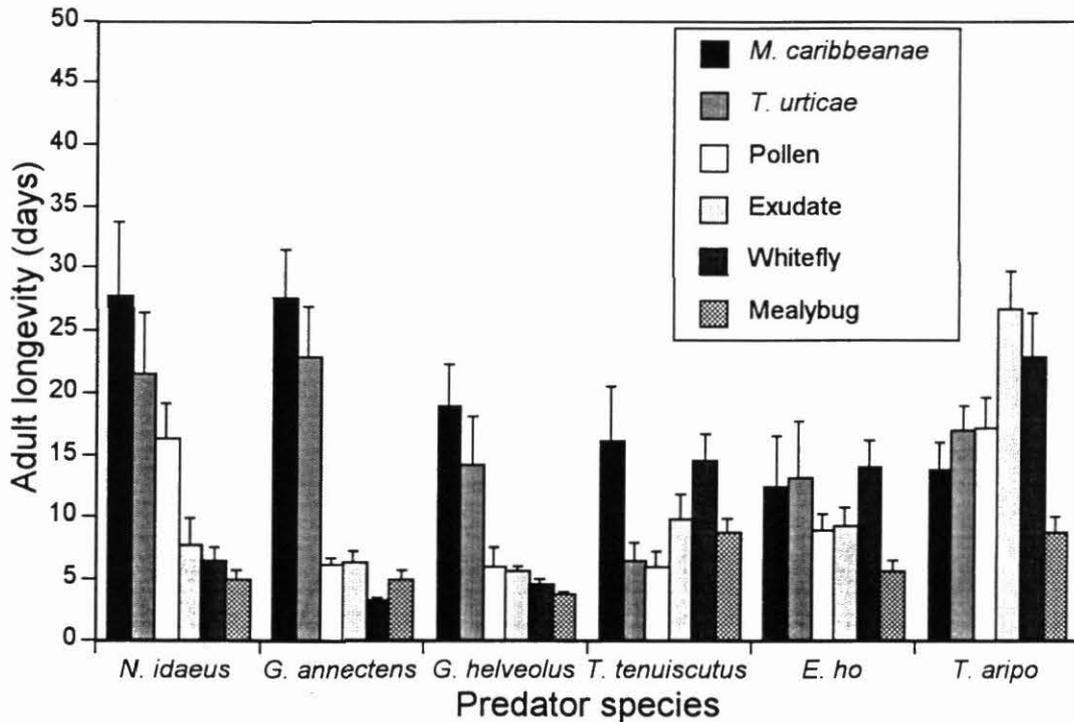


Figure 4.1.5. Effect of different food sources on female adult longevity of several species of predatory phytoseiid mites.

Predatory mites must consume protein in order to produce eggs, so fecundity is an important measure of the suitability of different food sources to increase predator populations.

Typhlodromalus tenuiscutus showed the highest specificity, laying substantial numbers of eggs only when fed on cassava mites, *Mononychellus caribbeanae* (which has previously been shown to be equal to *M. tanajoa* in preference and suitability). *Neoseiulus idaeus*, *Galendromus annectens* and *G. helveolus* also had high fecundity when provided with the two-spotted spider mite, *Tetranychus urticae* (Fig. 4.1.6). *Euseius ho* was the only species of this group that could produce many eggs when fed only castor bean pollen. *Typhlodromalus tenuiscutus* and *Euseius ho* could also reproduce at low rates when held on cassava leaves infested with various developmental stages of immature whiteflies (*Aleurotrachelus socialis*).

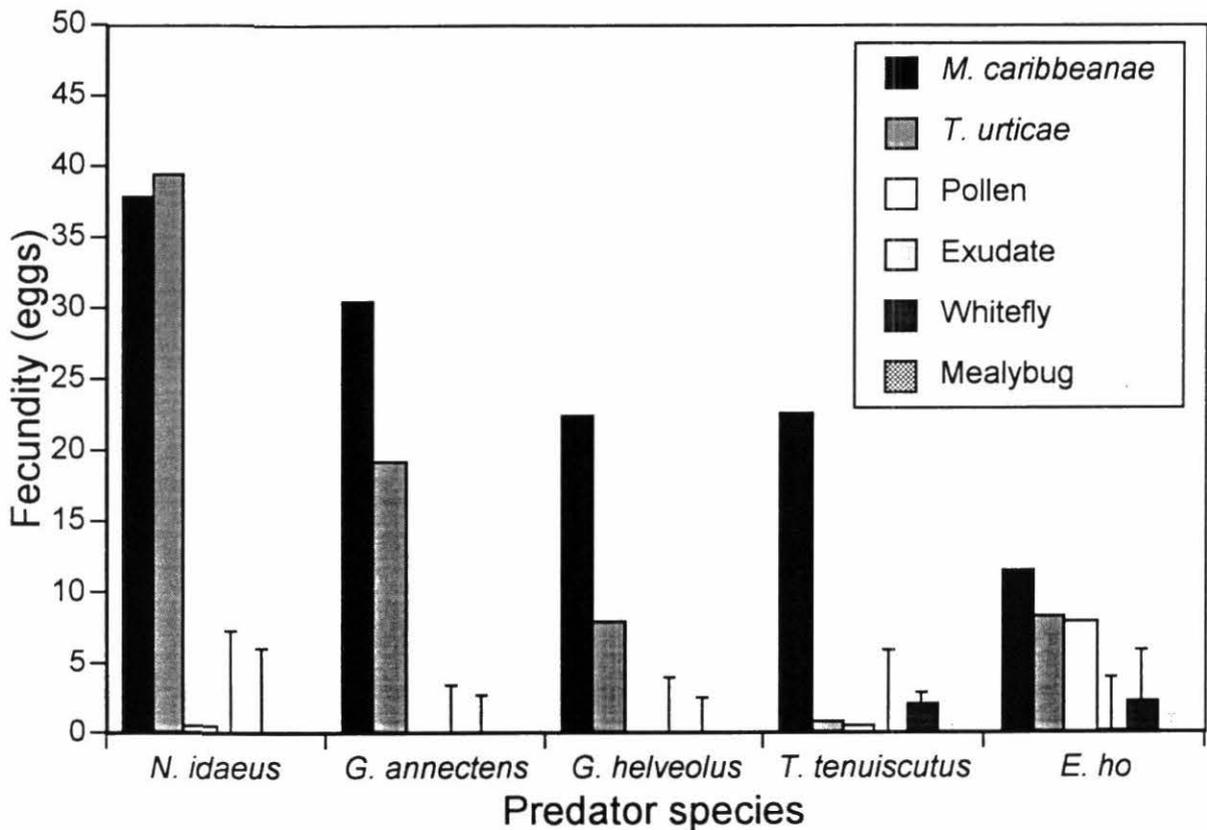


Figure 4.1.6. Effect of different food sources on fecundity of several species of predatory phytoseiid mites.

Exportation of natural enemies

Exportations of natural enemies during 1996 included *Typhlodromalus tenuiscutus* from a dry location in coastal Ecuador (Portoviejo, Manabí) which was sent to CNPMF/EMBRAPA, Cruz das Almas, Bahia via quarantine at CNPMA, Jaguariúna, São Paulo Brazil (Table 4.1.2). Strains of *Typhlodromalus manihoti* from several high altitude sites in Colombia (Barbosa, Cajibío and Copacabana, Bucaramanga, Armenia, Chinchina and Santa Rosa de Cabal) were sent to IITA, Benin via quarantine at the University of Amsterdam (Mitox). Climatic parameters associated with the sites from which the predators were collected were estimated from the average long-term data for the four nearest climate stations, weighted by the inverse of the distance squared (Table 4.1.3).

To date, three species of phytoseiids have been established in Africa. Of these, *T. aripo* and *T. manihoti* are spreading rapidly and are reducing cassava green mite populations significantly. *Typhlodromalus aripo* is estimated to be increasing root yield by 30%, which is equal to about US \$70 per season. Estimated benefits for 4 countries in West Africa are on the order of US \$60 million per year. Two species released in Northeast Brazil (*T. tenuiscutus* & *N. californicus*) have been recovered in small numbers, the latter up to 4 months after release.

Typhlodromalus manihoti and cassava green mite (CGM) were also sent to the University of Amsterdam, and CGM was sent to Boyce Thompson Institute, Ithaca, New York to establish colonies in support of collaborative basic research.

Table 4.1.2. Cultures of predatory phytoseiid mites exported from CIAT in 1996.

Date Sent	Species	Source	Colony Established	Number Sent	Number Received	Destination	% Survival
3/11/96	<i>T. tenuiscutus</i>	Portoviejo, Ecuador	12/95	400 + imms.	184 + 42 eggs	CNPMA, Brazil ¹	46%
4/23/96	<i>T. manihoti</i>	Barbosa, Colombia	1/96	910	432	Amsterdam ²	47%
"	<i>T. manihoti</i>	Cajibio, Colombia	8/95	240	219	Amsterdam	91%
5/14/96	<i>T. manihoti</i>	Guajira, Colombia	3/96	580	375	Amsterdam	65%
"	<i>T. manihoti</i>	Copacabana, Colombia	1/96	1,025	793	Amsterdam	77%
12/6/96	<i>T. manihoti</i>	Bucaramanga, Colombia (Los Colorados)	8/96	480	—	Amsterdam	ca. 60%
"	"	Armenia, Colombia	8/96	300	—	Amsterdam	ca. 60%
"	"	Chinchina, Colombia	8/96	120	—	Amsterdam	ca. 60%
"	"	Sta Rosa de Cabal, Colombia	8/96	180	—	Amsterdam	ca. 60%
"	"	Bucaramanga, Colombia (Bijagual)	8/96	180	—	Amsterdam	ca. 60%

¹ Sent to CNPMA quarantine laboratory in Jaguariuna, São Paulo, for further shipment to CNPMF, Bahia, Brazil.

² Sent to quarantine laboratory in Amsterdam, Holland, for further shipment to IITA, Benin.

Table 4.1.3. Climate associated with cultures of predatory phytoseiid mites exported from CIAT in 1996.

Species	Source	Altitude (m)	Precipitation (mm)	Temperature (°C)	Dry Months Per Year ¹	RH (%)
<i>T. tenuiscutus</i>	Portoviejo, Manabí, Ecuador	44	575	25	8	79
<i>T. manihoti</i>	Barbosa, Antioquia, Colombia	1399	2362	21	0	82
<i>T. manihoti</i>	Cajibío, Cauca, Colombia	1763	2152	18	1	80
<i>T. manihoti</i>	Villanueva, Guajira, Colombia	340	1043	27	5	74
<i>T. manihoti</i>	Copacabana, Antioquia, Colombia	1844	1649	19	1	74
<i>T. manihoti</i>	Bucaramanga, Santander, Colombia, (Los Colorados)	983	1363	22	0	85
"	Armenia, Quindío, Colombia	1500	2430	19	0	79
"	Chinchina, Caldas, Colombia	1360	2514	21	0	76
"	Sta Rosa de Cabal, Risaralda, Colombia	1580	2558	19	0	84
"	Bucaramanga, Santander, Colombia (Bijagual)	977	1363	22	1	84

¹ Months with less than 60 mm precipitation.

Climatic distribution of phytoseiids

Explorations for phytoseiids have been conducted in 14 countries in Latin America and the Caribbean with samples from over 1,097 sites. Over 87 species of phytoseiids have been encountered and over 25 new species have been described. About 67% of the samples came from cassava and 76% of the 2,943 phytoseiid records are also from cassava. Colombia, Ecuador and Venezuela account for 89% of these records, comprising at least 77 species of phytoseiids, with at least 44 in association with cassava. Because of the high number of samples and the geographic

diversity of this region we chose to use it for ecological analysis. The climate of each sample site was estimated using long-term climatic data from the CIAT GIS (geographic information system) database (average of 4 nearest climate stations weighted by distance⁻²). Duration of dry season (no. of months with <60 mm precipitation) and elevation (m above mean sea level) were considered the most useful climatic parameters available for classifying the sites. This choice was based on availability of information and usefulness for evaluating species to release in target climatic zones. Data are reported as incidence (number of samples of a climatic classification in which the species was present divided by the total of such samples in that classification) to adjust for sampling effort (Table 4.1.3).

A higher number of species were encountered in Colombia than expected based on sample effort, which probably reflects the high variation in topography and climate found within the country. In general, we have few samples from high altitude dry sites, which limited the climatic analysis. The phytoseiid species found in most environments (wet and dry, high and low) were *Typhlodromalus manihoti* and *T. aripo* (Table 4.1.4). Species found more commonly in dry regions were *Euseius ho*, *Galendromus annectens*, *Typhlodromalus rapax*, *T. tenuiscutus*, and *Neoseiulus idaeus*. Species found at intermediate elevations were *Galendromus annectens*, *G. helveolus*, and *Euseius concordis*, while *Typhlodromalus neotunus* and *Euseius naindaimeii* occurred at high elevations. Two species appeared geographically isolated: *Typhlodromalus tenuiscutus* and *Neoseiulus californicus*, and they were strongly recommended for release in Northeast Brazil, where they do not occur. These are the species that were released there in 1995 and 1996.

Table 4.1.4. Distribution of phytoseiid species with respect to elevation and number of dry months based on long-term climatic data associated with collection sites in Colombia, Venezuela and Ecuador.

No. of Dry Months	Elevation above sea level (meters)				
	< 800	800-1200	1200-1600	>1600	
<i>Typhlodromalus manihoti</i>					
< 3	0.54	0.25	0.25	0.29	
3-5	0.35	0.25	0.25	0.23	
> 5	0.24				
<i>Neoseiulus anonymus</i>					
< 3	0.05	0.15	0.15	0.07	
3-5	0.06	0.13	0.13	0.06	
> 5	0.07				
<i>Galendromus annectens</i>					
< 3	0.02	0.10	0.10	0.03	
3-5	0.04	0.05	0.05		
> 5	0.09				
<i>Typhlodromalus aripo</i>					
< 3	0.03	0.06	0.06	0.08	
3-5	0.05	0.05	0.05	0.06	
> 5	0.05				
<i>Galendromus helveolus</i>					
< 3		0.10	0.10	0.08	
3-5	0.03	0.08	0.08	0.03	
> 5	0.05				
<i>Neoseiulus idaeus</i>					
< 3					
3-5	0.04				
> 5	0.01				
<i>Euseius ho</i>					
< 3	0.04	0.01	0.01	0.05	
3-5	0.01	0.03	0.03	0.03	
> 5	0.16				
TOTAL SAMPLE					
	Number of sites in each classification cell				
	< 800	800-1200	1200-1600	>1600	Total
< 3	111	410	86	32	639
3-5	728	75	35	4	842
> 5	245	0	1	1	247
Total	1084	485	122	37	1728

Phytoseiid associations with cassava

The previously described database of exploration records for phytoseiid mites was analyzed to learn more about the host plant associations of phytoseiids. About 76% of the 2,943 phytoseiid records are from cassava, so relatively little can be said about associations with plant species other than cassava. Colombia, Ecuador and Venezuela account for 89% of these records, and were selected to restrict the geographic range for this analysis. In this subset, 81.5% of the records are from cassava. The 2,084 records were classified as coming from either cassava or "other" host plant and analyzed by chi-square. Most of the host plants had only 1-3 records per phytoseiid species, so it is not worth looking at more than the aggregated "other" plant category. The 17 most frequently encountered phytoseiid species are listed in Table 4.1.5. If we want to use phytoseiid species that have a close association with cassava, we should focus on the species in the upper part of the table. *Typhlodromalus tenuiscutus*, *N. idaeus*, *Typhlodromips dentilis*, *T. manihoti*, and *G. helveolus* had high associations with cassava. With respect to phytoseiids targeted for release in NE Brazil, of the recently collected species from the dry coast of Ecuador, this includes: *T. tenuiscutus*, *N. idaeus*, *G. helveolus* and *G. annectens*. This strong association with the target plant favors the choice of these predators to introduce for biological control of cassava green mite. *Euseius concordis*, *Amblyseius chiapensis*, *E. ho*, and *Iphiseiodes zuluagai* were negatively associated with cassava, which suggests that they may not be good candidates. *Iphiseiodes zuluagai* had a high association with citrus, and this species has been reported on citrus in other countries such as Brazil.

Table 4.1.5. Association of the 17 most common phytoseiid mites with cassava or other plants based on explorations in Colombia, Venezuela and Ecuador.

Phytoseiid species	Proportion of field samples (%)		Number of samples
	Cassava	Other plants	
<i>Typhlodromalus tenuiscutus</i>	97	3*	64
<i>Neoseiulus idaeus</i>	96	4*	53
<i>Typhlodromips dentilis</i>	96	4*	75
<i>Typhlodromalus manihoti</i>	94*	6*	650
<i>Galendromus helveolus</i>	91	9*	102
<i>Galendromus annectens</i>	88	12	116
<i>Neoseiulus anonymus</i>	86	14	181
<i>Typhlodromalus aripo</i>	83	17	124
<i>Proprioseiopsis cannaensis</i>	82	18	57
<i>Typhlodromalus peregrinus</i>	82	18	49
<i>Amblyseius aerialis</i>	79	21	106
<i>Typhlodromalus rapax</i>	76	24	78
<i>Phytoseiulus macropilis</i>	70	30	43
<i>Euseius concordis</i>	59*	41*	132
<i>Amblyseius chiapensis</i>	51*	49*	47
<i>Euseius ho</i>	50*	50*	129
<i>Iphiseiodes zuluagai</i>	31*	69*	78

* Values significantly different than expected (based on marginal values, i.e. 81.5% in cassava), indicated by the chi-square standardized deviation for the cell being greater than 2 or less than -2.

Evaluation of *Neozygites*

Natural outbreaks of the fungal pathogen *Neozygites c.f. floridana* in Northeast Brazil have been observed to rapidly kill off populations of cassava green mite (CGM, *M. tanajoa*). Although subsequent investigations by IITA scientists have shown that apparently the same species occurs in Africa. The lack of virulent epizootics suggests that it would be worthwhile to introduce a more virulent strain from South America.

Pathogenicity of *Neozygites* strains

Cultures of the fungal pathogen *Neozygites c.f. floridana* were collected from the cassava green mite and two-spotted spider mite (*Tetranychus urticae*) at CIAT and from CGM at Media Luna, Colombia. Other cultures obtained from Brazil and Africa were included in a laboratory evaluation of host specificity on three mite species: *M. tanajoa*, *M. caribbeanae* and *T. urticae*. Single mite mummies were placed in the center of a cassava leaf disk under conditions of high humidity (95%) and 16 hours of darkness to produce conidia. Twenty-five recently emerged

female mites were placed on the disk to be exposed to the conidia for 2 d at 65% RH, 12 h photoperiod. Afterwards they were transferred to fresh leaf disks and were examined daily for signs of infection and mortality. None of the 3 strains from *M. tanajoa* infected *T. urticae*, and one of the strains from *T. urticae* failed to infect *M. tanajoa* (Fig. 4.1.7). All 5 strains infected *M. caribbeanae*. This confirms that there is substantial difference among the strains and that those isolated from *Mononychellus* appear to be more specific than those from *T. urticae*.

Geographical distribution of *Neozygites*

This pathogen is currently being studied for use as a biological control agent of CGM, but little is known about its geographic distribution or host range. We reviewed 10,782 specimens of tetranychid mites on microscope slides from our museum collection, representing 907 sample sites, to determine the host range and geographic range of this pathogen. These specimens were previously collected during foreign exploration for natural enemies of *M. tanajoa*. Both conidia and hyphal bodies can be detected in the slides. Signs of *Neozygites* infection (presence of hyphal bodies) was observed in 14 species of tetranychid mites from 11 neotropical, 2 African and 2 Asian countries, indicating that this pathogen may be much more important than was previously thought (Tables 4.1.6 and 4.1.7). *Neozygites* infection was also observed in *Tetranychus kanzawai* specimens from China and Indonesia. The highest frequency of infection, as indicated by the presence of hyphal bodies, was found in *M. caribbeanae* and *M. tanajoa* (Fig. 4.1.8). Significantly low frequencies occurred in *Oligonychus peruvianus*, which is common on cassava but which is generally protected beneath silk webbing, and *Tetranychus* sp. (which is probably mainly *T. urticae*, which cannot be identified to species without males). Although it is not certain that only one species of *Neozygites* is represented by these results, they are in agreement with the previously described laboratory pathogenicity tests for *N. c.f. floridana* indicating that *Mononychellus* is very susceptible compared to other tetranychids.

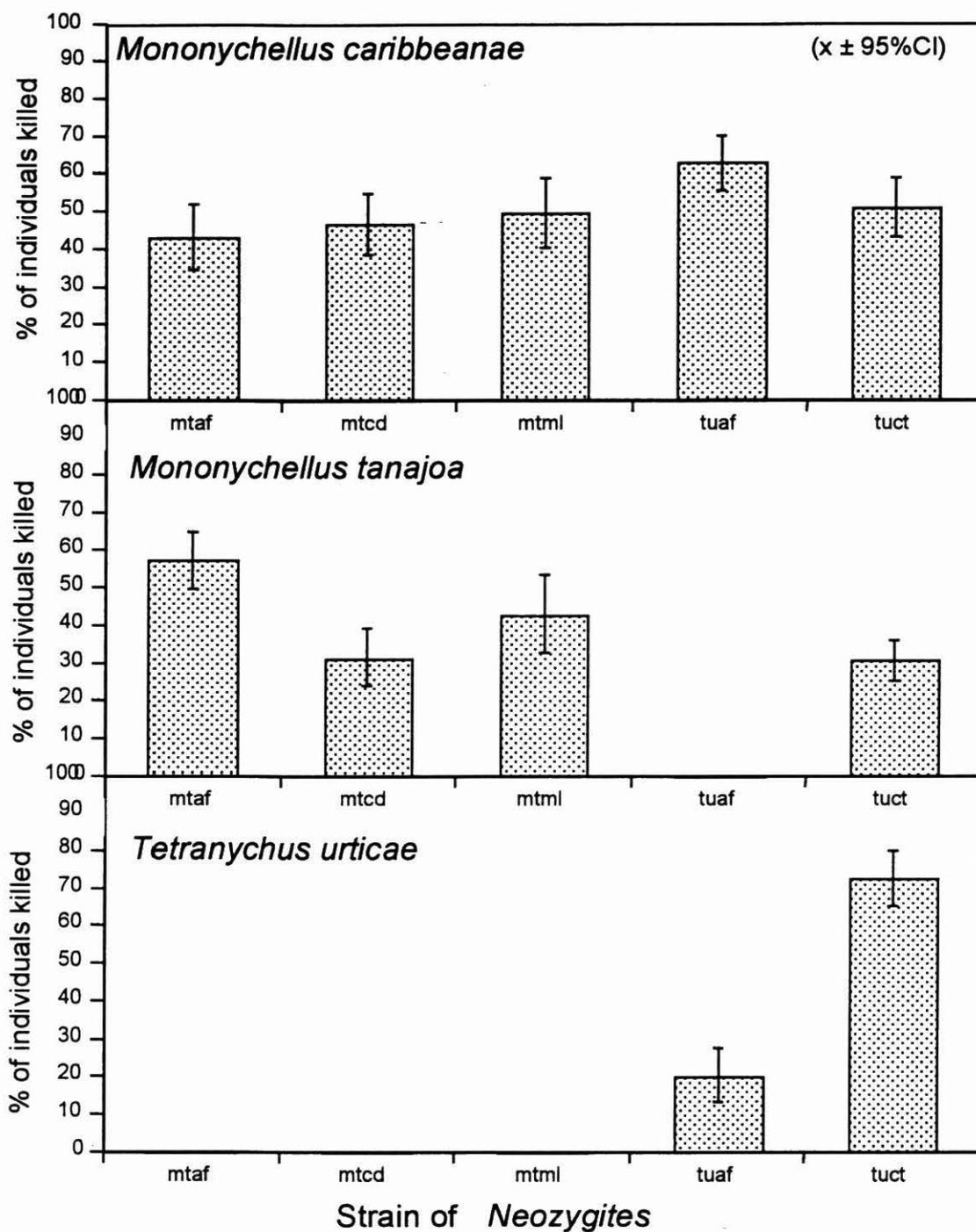


Figure 4.1.7. Pathogenicity of five strains of *Neozygites* c.f. *floridana* on 3 species of tetranychid mites associated with cassava (mt - from *M. tanajoa*; tu - from *T. urticae*; af - from Benin, Africa; cd - from Cruz das Almas, Bahia, Brazil; ml - from Media Luna, Magdalena, Colombia; ct - from CIAT, Valle de Cauca, Colombia).

Table 4.1.6. Species of Tetranychid mites held in the CIAT collection that show signs of infection by *Neozygites* sp. as indicated by the presence of hyphal bodies.

Species	Samples	Species	Samples
<i>Eutetranychus banksi</i>	1	<i>Tetranychus canadensis</i>	1
<i>Mononychellus caribbeanae</i>	38	<i>Tetranychus cinnabarinus</i>	1
<i>Mononychellus mcgregori</i>	5	<i>Tetranychus desertorum</i>	1
<i>Mononychellus planki</i>	1	<i>Tetranychus marianae</i>	3
<i>Mononychellus tanajoa</i>	43	<i>Tetranychus mexicanus</i>	1
<i>Oligonychus gossypii</i>	5	<i>Tetranychus tumidus</i>	4
<i>Oligonychus peruvianus</i>	4	<i>Tetranychus urticae</i>	1
		<i>Tetranychus</i> sp.	6
Total			115

Table 4.1.7. Countries from which specimens of Tetranychid mites held in the CIAT collection show signs of infection by *Neozygites* sp. as indicated by the presence of hyphal bodies.

Country	Samples	Country	Samples
Neotropics		Africa	
Brazil	19	Benin	3
Colombia	38	Mozambique	1
Cuba	12		
Ecuador	11	Asia	
Mexico	10	China	3
Nicaragua	1	Indonesia	1
Panama	3		
Paraguay	1		
Peru	3		
Trinidad	4		
Venezuela	14		
Total			124

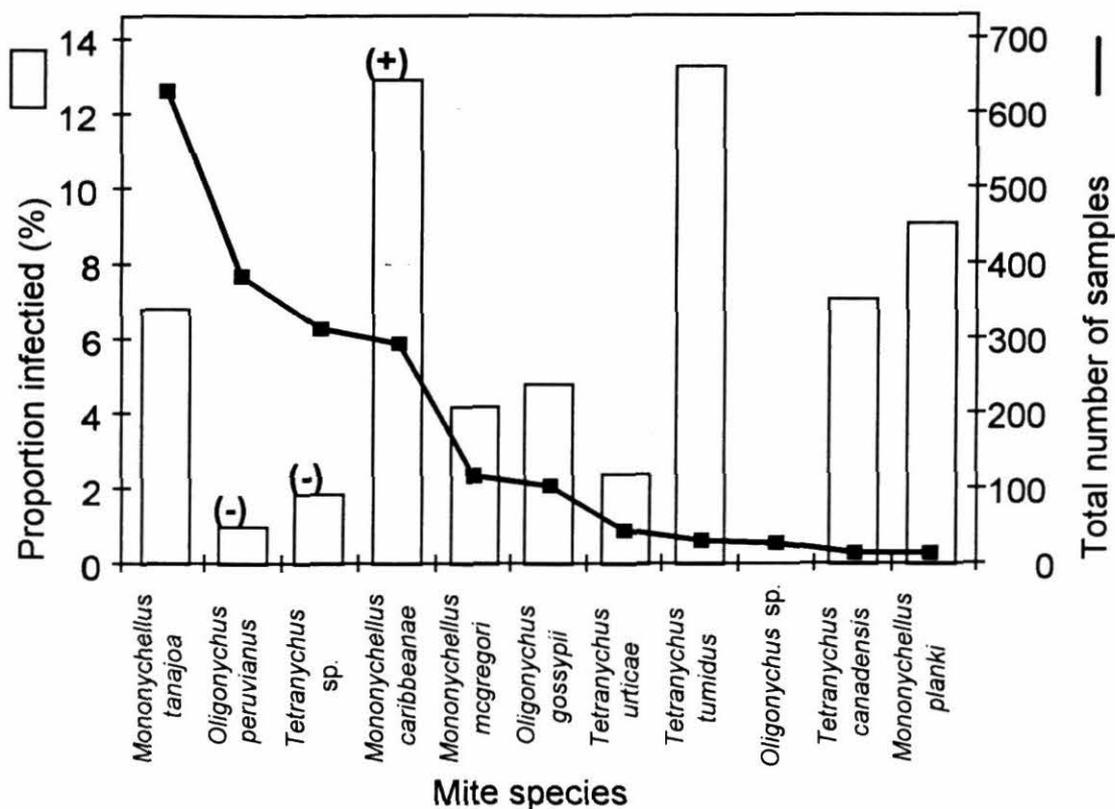


Figure 4.1.8. Proportion of samples containing individuals infected by *Neozygites* sp. (as indicated by the presence of hyphal bodies) in the 11 most common species of tetranychid mites in the CIAT collection (+, higher than expected; -, lower than expected; chi-square test with standardized deviations greater than 2).

DNA characterization of *Neozygites*

Strains of *Neozygites* c.f. *floridana* have been shown to differ substantially in their pathogenicity (see above); however, they can not be distinguished by morphology. Field research conducted by IITA in Benin and CNPMF in Brazil indicate that strains from Northeast Brazil are more virulent than those in Africa. However, it is critical to develop methods to identify strains of *Neozygites* before neotropical strains of the fungus can be released in Africa.

Characterization of DNA using PCR-based techniques should be able to permit the identification of individual strains; however, this has never been done with this genus of fungus. Despite the obstacles that are normally encountered in such pioneering research, substantial progress has been made. *In vitro* culturing methods developed by Don Roberts (Boyce Thompson Institute, USA) and Luis Leite (Instituto Biologico, Brazil) have been successfully

adopted and modified. We can now grow strains of the fungus from both *T. urticae* and *M. tanajoa* (Roberts & Leite could only grow a *T. urticae* strain) on artificial media (Grace's Insect medium + yeast extract + lactoalbumin). Problems with bacterial contamination have been resolved by adding antibiotics to the media (Fig. 4.1.9). This has removed a major obstacle to our work, now permitting us to produce sufficient quantities of uncontaminated DNA. Nevertheless, the growth rate of strains isolated from *M. tanajoa* still grow very slowly (Fig. 4.1.10). Because the *Neozygites* strains isolated from *M. tanajoa* appear to be more specific in host range than those from *T. urticae*, this suggests that growth rates may be improved by further refinement of the growth media, perhaps by including compounds found in cassava leaves. Further work will be done in this area.

Five strains of *Neozygites* c. f. *floridana* were multiplied *in vitro* for DNA extraction. Preliminary evaluations using RAPD (random amplified polymorphic DNA) and AFLP (amplified fragment length polymorphism) techniques have been performed. The initial RAPD trials using 5 primers (A2, A3, B2, B9 & D2; Operon Technologist, La Jolla, CA) were able to distinguish *Neozygites* c.f. *floridana* from a distantly related fungus (*Phaeoisariopsis griseola*, bean angular spot), but not among *T. urticae* strains of *Neozygites*. The AFLP method generally provides many more bands (DNA fragments), thus increasing the power to detect differences. An unreplicated AFLP trial showed strong differences among 5 *Neozygites* strains, while the patterns for different samples from a single batch of one strain were clearly identical. Confirmation of these results will mean that we can readily identify individual strains of this pathogen, thus opening the way to make releases in Africa.

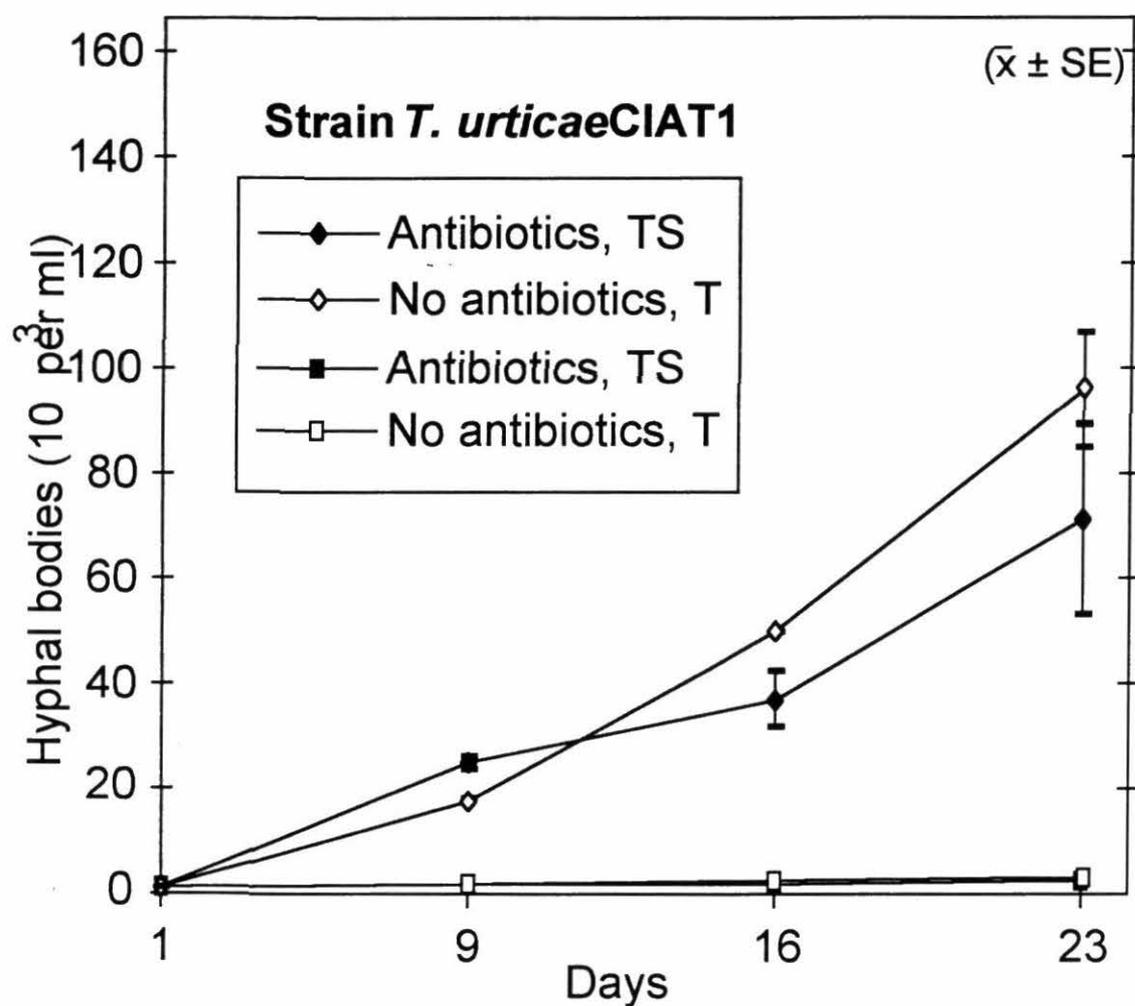


Figure 4.1.9. *In vitro* multiplication of a *Neozygites cf. floridana* strain isolated from *T. urticae* (TuCIAT1) with and without antibiotics (streptomycine + tetracycline) in two different tissue culture media (T, TNMFH + lactalbumin + yeastolate; TS, TNMFH + lactalbumine + yeastolate + fetal bovine serum).

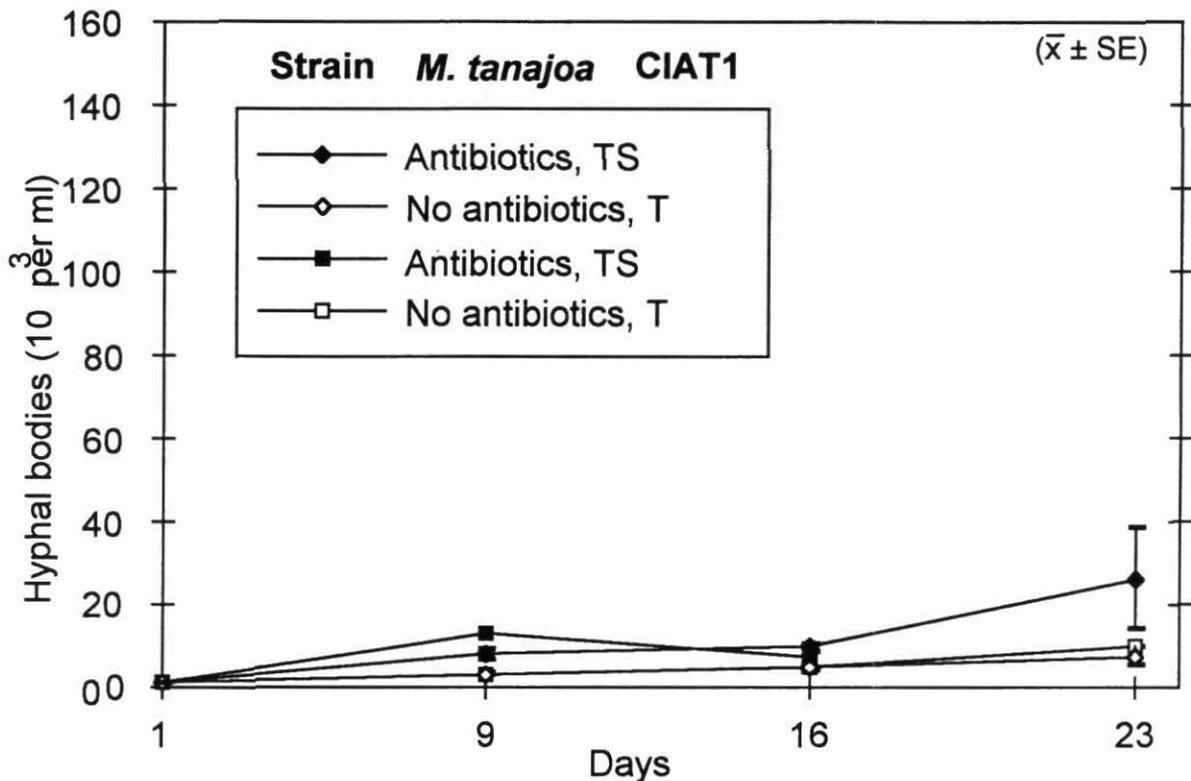


Figure 4.1.10. *In vitro* multiplication of a *Neozygites cf. floridana* strain isolated from *M. tanajoa* (MtCIAT1) with and without antibiotics (streptomycine + tetracycline) in two different tissue culture media (T, TNMFH + lactalbumin + yeastolate; TS, TNMFH + lactalbumine + yeastolate + fetal bovine serum).

Collaborative research on *Neozygites*

Strains of *Neozygites c.f. floridana* (Zygomycetes: Entomophthorales) and cultures of cassava green mites were sent to Boyce Thompson Institute (BTI), Ithaca, New York several times to support collaborative strategic research. Don Roberts and Luis Leite (a Brazilian graduate student) successfully developed an *in vitro* method of rearing a *Tetranychus urticae* strain of *Neozygites*. This was critical to help CIAT scientists obtain sufficient quantities of uncontaminated DNA to allow them to start using molecular methods to characterize strains of the fungus. The BTI scientists also investigated the process of invasion of host mites by the

fungus. This strategic research should help us understand the differences in host specificity of different strains of this pathogen.

Samples of *Neozygites c. f. floridana* were sent to Siegfried Keller at the Swiss Federal Res. Station for Agronomy, Zurich for taxonomic identification. So far, none of the strains from Africa, Brazil or Colombia have shown any morphological characters that distinguish them. This emphasizes the need to develop molecular methods to identify strains.

A strain of *Neozygites sp.* from *T. urticae* was released to Biocontrol, SA and Coinbiol, SA, in Palmira, Colombia for possible development as a commercial biological control agent.

Phytoseiid mass rearing techniques

Mass rearing of *Typhlodromalus tenuiscutus*

Experiments were conducted to help optimize the use of the Mesa-Bellotti method of rearing phytoseiid predators to assist mass-rearing of predators in NE Brazil. This system consists of using small plastic containers with a screen shelf. Fresh mite-infested leaves are placed in the bottom of the container to feed the phytoseiid predators. After 2-3 days additional fresh, mite-infested leaves are added on top of the shelf. The predators then move from the old leaves to the new leaves, which are removed in 2-3 days, when the next batch of fresh mite-infested leaves are added. This system was designed specifically for predators that require *Mononychellus* prey, which must be grown on cassava leaves. This particularly applies to *Typhlodromalus tenuiscutus* which we are now releasing in NE Brazil. The experiment on colony production was initiated with 50, 100 and 200 females in Mesa-Bellotti trays, and adult females were harvested weekly for 8 weeks, starting at week 3.

An experiment was conducted to measure the effect of initial number of *T. tenuiscutus* females used to establish Mesa-Bellotti mass-rearing cultures on the number of adult female progeny harvested. After the third week of rearing, all adult females were counted and removed from the rearing system. It can be seen that this method results in a continued decrease in production of the culture, which suggests that not enough females are retained by this harvesting method to sustain levels of oviposition (Fig. 4.1.11.). The highest production of females from Mesa-Bellotti cultures was from those initiated with 100 females, the least from those started with 50 females

(Fig. 4.1.12a). Those initiated with 200 females fluctuated from week to week the most, suggesting possible crowding effects. Production of adult female *T. tenuiscutus* in the Mesa-Bellotti mass-rearing system per number of females used to initiate the culture was highest at the lowest density tested (50 females; Fig. 4.1.12b) suggesting the effect of competitive interactions at the higher densities.

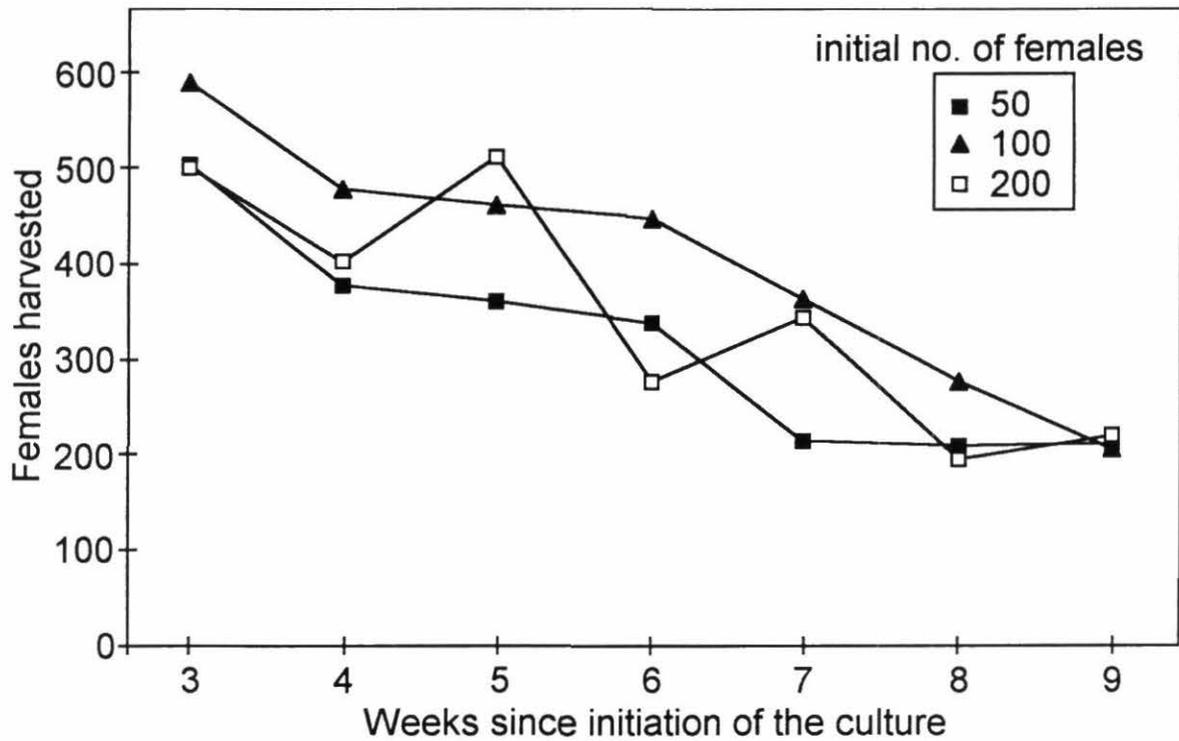


Figure 4.1.11. Effect of initial number of *T. tenuiscutus* females used to establish Mesa-Bellotti mass-rearing cultures on the number of adult female progeny harvested.

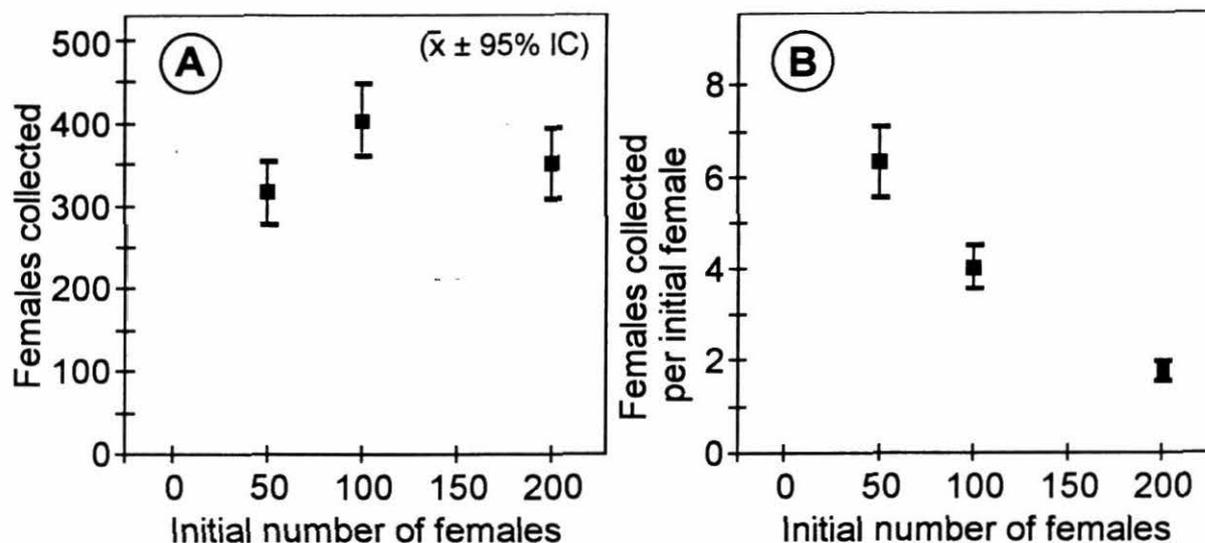


Figure 4.1.12. Production of female *T. tenuiscutus* in the Mesa-Bellotti mass-rearing system: A) number of adult females harvested per container, B) number of adult females harvested per initial female per container.

Computer model of mass rearing

Investigations were conducted to develop a stage-structured computer model to help optimize and facilitate the management of the mass-production of phytoseiids (Fig. 4.1.13). Data on development time, fecundity, survivorship, rate of stage-specific prey consumption, and intraspecific competition were collected using *T. tenuiscutus* to provide inputs for the model. Development time from egg to adult was 6.0 days at 25°C, 67% of the eggs reached adulthood, the sex ratio was 67% females, and the average longevity of adult females was 30.8 days. The periods of preoviposition, oviposition and postoviposition were 1.8, 16.4 and 4.6 days, respectively, and average fecundity was 29.3 eggs. The population parameters were: intrinsic rate of natural increase $r_m = 0.20$, net rate of increase $\text{Lambda} = 1.23$, net reproductive rate $R_0 = 17.8$, mean generation time $T = 14.1$ days, y doubling time $TD = 3.3$ days. Eggs were the preferred stage of prey for *T. tenuiscutus* larvae, and larvae were preferred by the other developmental stages. The average consumption of prey during development from larva to adult was 6.3 eggs, 16.2 larvae, 4.2 nymphs, and 1.7 adults of *M. caribbeanae*.

Data were collected from Mesa-Bellotti mass-rearing cultures of *T. tenuiscutus* to "validate" the model. Predictions of the model deviated from the observed experimental results (Fig.

4.1.14). Various logical adjustments to the model were explored to see what changes would make the best fit to the experimental results. Increasing the level of immature phytoseiid mortality, as a function of adult phytoseiid population permitted a very close fit to the results. This suggests that cannibalism is an important component that was not included in the original model. Based on this discovery we are initiating experiments to measure the rate of cannibalism, which should eventually help us to increase the efficiency of mass-rearing.

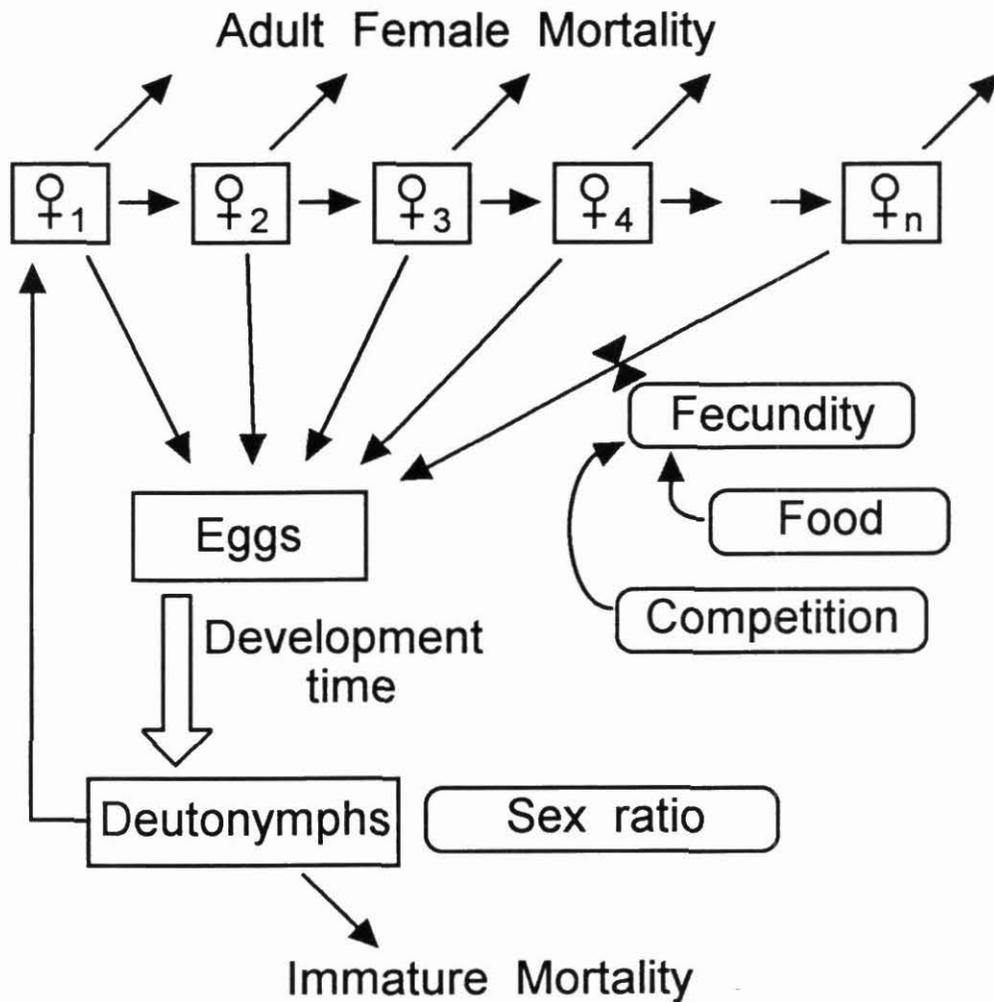


Figure 4.1.13. Diagram of stage-structured computer simulation model to help optimize mass-rearing of phytoseiid mites.

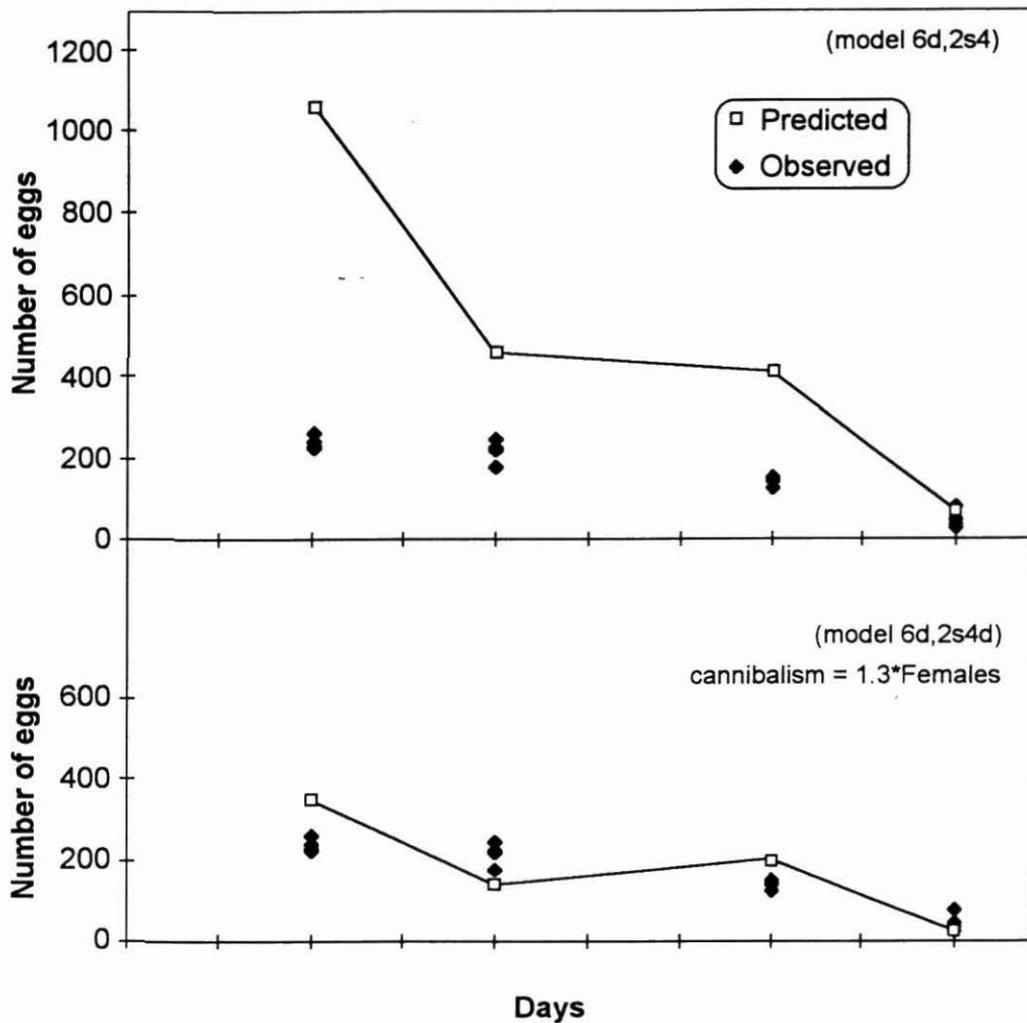


Figure 4.1.14. Comparison of predicted and observed fecundity of the phytoseiid *Typhlodromalus tenuiscutus* in "Mesa-Bellotti" mass-rearing chambers showing that adding a parameter for cannibalism of eggs by females greatly improves the computer simulation model.

Phytoseiid taxonomy

Taxonomic key to common species

A taxonomic key to 30 phytoseiid species commonly found on cassava in northern South America was developed in collaboration with Gilberto de Moraes (CNPMA/EMBRAPA, São Paulo, Brazil) (Table 4.1.8). Phytoseiids collected in the field during explorations and experiments were identified and curated. The taxonomic collection is being reviewed to verify old identifications and correct the computer database.

Table 4.1.8. List of phytoseiid species included in a dichotomous taxonomic key.

<i>Amblyseius (Amblyseius) aeralis</i>	<i>Neoseiulus anonymus</i>
<i>Amblyseius (Amblyseius) chiapensis</i>	<i>Neoseiulus californicus</i>
<i>Amblyseius (Amblyseius) herbicolus</i>	<i>Neoseiulus idaeus</i>
<i>Amblyseius (Amblyseius) largoensis</i>	<i>Neoseiulus neotunus</i>
<i>Amblyseius (Iphiseioidis) zuluagai</i>	
<i>Amblyseius (Typhlodromalus) aripo</i>	<i>Phytoseiulus macropilis</i>
<i>Amblyseius (Typhlodromalus) limonicus</i>	
<i>Amblyseius (Typhlodromalus) manihoti</i>	<i>Phytoseius purseglovei</i>
<i>Amblyseius (Typhlodromalus) peregrinus</i>	
<i>Amblyseius (Typhlodromalus) rapax</i>	<i>Proprioseiopsis cannaensis</i>
<i>Amblyseius (Typhlodromalus) tenuiscutus</i>	<i>Proprioseiopsis mexicanus</i>
<i>Amblyseius (Typhlodromips) bellottii</i>	<i>Proprioseiopsis neotropicus</i>
<i>Amblyseius (Typhlodromips) dentilis</i>	
<i>Amblyseius (Typhlodromips) manglae</i>	<i>Typhlodromina tropica</i>
<i>Euseius alatus</i>	<i>Typhlodromus (Anthoseius) transvaalensis</i>
<i>Euseius concordis</i>	
<i>Euseius ho</i>	
<i>Galendromus annectens</i>	
<i>Galendromus helveolus</i>	

Genetic variability of Phytoseiid "species"

Amplified fragment length polymorphism (AFLP) analysis of genetic variation showed that geographic strains of *N. idaeus* were very homogeneous whereas those of *T. manihoti* differed greatly. These results are similar to the genetic variability shown in isozyme analysis of esterases. This suggests that *T. manihoti* is either a very plastic species or may represent a group of cryptic species. Further studies on genetic crosses between strains of the species are being conducted.

Cassava CD-ROM

We scanned and formatted 11 major documents (2,800 pages) including recent CIAT Cassava Program annual reports, CIAT Working Documents, scientific meeting proceedings and Cassava Newsletters to be included in a CD-ROM being published by IITA and the University of Florida, Gainesville (Table 4.1.9).

Table 4.1.9. List of documents scanned and formatted in Wordperfect for conversion to hypertext to include in a CD-ROM being developed in collaboration with IITA and the University of Florida.

Casava Bulletin (issues 1990-1995) - 80 p.

Yuca Buletin (issues 1990-1995) - 80 p.

CIAT Cassava Program Annual Reports:

1987-1989, Working Document No. 91. - 621p.

1987-1991, Working Document No. 116. - 473p.

1992, Working Document No. 142. - 292p.

1993, Working Document No. 146. - 324p.

CIAT Working Documents:

BRAUN, A.R. (ed.). 1993. Bases fundamentales para investigacion sobre los acaros plagas y sus enemigos naturales en el Ecuador. CIAT Documento de Trabajo No. 126. 172 p.

HENRY, G. (ed.). 1993. Trends in CIAT Commodities 1993. CIAT, Cali, Colombia. Working Document No. 128. July 1993. 221p.

HENRY, G.; IZQUIERDO, D. A.; GOTCRET, M. V. 1994. Proyecto integrado de yuca en la Costa Atlantica de Colombia. CIAT Documento de Trabajo No. 139. 79p.

HERNANDEZ R., L.A.; ASHBY, J.; HERSHEY, C.; IGLESIAS, C.A. 1993. Evaluación de nuevas variedades de yuca con la participación de agricultores. Documento de Trabajo No. 130. 1993. 105p.

IGLESIAS, C.A. (ed. tec.) 1994. Memorias de la Tercera Reunion Panamericana de Fitomejoradores de Yuca. Interfase entre los programas de mejoramiento, los campos de los agricultores y los mercados de la yuca en latinoamerica. CIAT Documento de Trabajo No. 138. 279p.

IGLESIAS, C.A.; FUKUDA W.G. (eds.) 1992. Memorias de la Segunda Reunion Panamericana de Fitomejoradores de Yuca. CIAT Documento de Trabajo No. 112. 184p.

ROCA, W.M.; THRO, A.M. (eds.) 1993. Proceedings of the first international scientific meeting of the Cassava Biotechnology Network, Cartagena, Colombia, 25-28 August 1992. Working Document No. 123. 496p.

4.2 Entomology, by Anthony C. Bellotti

4.2.1 Mealybug

The cassava mealybug *Phenacoccus herreni* causes severe crop losses, especially in Northeast Brazil where in certain areas farmers have abandoned cassava production due to mealybug attacks. Three parasitoids from Colombia and Venezuela have been introduced and released into the Brazilian States of Bahia, Pernambuco and Ceará. All three parasitoids, *Aenasius vexans*, *Apoanagyrus diversicornis* and *Acerophagus coccois* have become established, and two of the species *A. coccois* and *A. diversicornis* are dispersing rapidly. Mealybug control has been so

effective due to these parasitoids that cassava production has returned to some of the areas where it had been abandoned.

In the Llanos of Colombia and Venezuela, cassava is attacked by *P. herreni*, causing crop losses. Studies to evaluate the effectiveness of biological control were conducted in Villavicencio in collaboration with CORPOICA/La-Libertad. This research formed part of a MS thesis requirement for a Colombian student from Georg-August University, Gottingen, Germany.

Field experiments were initiated by artificially infesting cassava plants with *P. herreni* to insure adequate mealybug populations. Three parasitoid species, *A. vexans*, *A. diversicornis* and *A. coccois* were subsequently released in 3 cassava fields; one parasitoid species per field. In a fourth field all 3 species were released. In control plots, cassava growing tips were protected from parasitoids with a gauze sleeve cages. The percentage of parasitism was obtained by dissecting 1,887 2nd, 3rd and 4th instar mealybugs.

The study resulted in parasitism rate of 15.5% and a level of encapsulation of 37.6%. In this experiment low mealybug densities produced higher percentages of encapsulation, while a higher mealybug population caused lower percentages. The parasitoid *A. vexans* had the highest percentage of parasitism. In addition, parasitism rates were highest in the field where all three species were released.

Chemical Mediated Searching Behavior of Parasitoids.

Plants when attacked by insects often release volatiles that can attract natural enemies. Previous results from this on-going research have indicated that two parasitoid species *A. vexans* and *A. diversicornis* respond significantly to infested plants. *A. coccois* was not attracted more to infested plants than to uninfested ones, though it was attracted by cassava plants in general. More recent research using Y-tube olfactometer experiments have tried to determine the cause of the insect attraction: "synomones" released by infested cassava plants or odors emitted by the mealybug and its by-products. The following choices were offered to *A. vexans* and *A. diversicornis*: infested vs. washed infested leaves (Cl), Blank vs. Cl, and healthy vs. Cl. To obtain clean leaves (Cl), mealybugs and exuviae were removed from two infested cassava leaves and leaves were thoroughly washed with wet cotton to remove honey dew and fungi. For the Mb

treatment, all products originating from the presence of mealybugs were removed from two infested cassava leaves. Mealybugs and exuviae were deposited on wet cotton, and honeydew and fungi were collected.

Initial results indicate that both parasitoid species are attracted to the infested leaves (CI) over the mealybug (Mb). A significantly higher proportion of female parasitoids of both species chose the infested but cleaned leaves over the blank (no leaf) or the mealybug and its exudates. There was no significant differences in the CI vs. healthy leaves or the CI vs. infested leaves, indicating that plant volatiles from mealybug-damaged leaves attract the parasitoids.

Field experiments, to determine if a bean intercrop will have an influence on the capacity of two parasitoids *A. vexans* and *A. diversicornis*, to parasitize *P. herreni* on cassava were initiated during 1996. Three treatments were tested, plots with only cassava plants, plots with mixed cassava and beans, and plots with only bean plants. Eight potted cassava plants infested with second and third instar mealybugs, were placed in each plot and 20 parasitoid females of one species were released. After five days, plants were brought into the greenhouse and parasitism was determined.

Initial results show that the parasitism rate of *A. vexans* is higher than *A. diversicornis*, but the difference may not be significant. In addition *A. vexans* appears to have a better searching capacity in pure cassava stands, while *A. diversicornis* appears to prefer the bean crop, but more data is needed to verify these results.

4.2.2 Whiteflies

Aleurotrachelus socialis, *Bemisia tuberculata* and *Trialeurodes variabilis* are the most common species of whiteflies attacking cassava in Colombia. Previous surveys show that *A. socialis* is the predominant species, and can cause severe cassava yield reductions when high populations occur. For the past two years, considerable survey work has been undertaken to more accurately define the pest distribution and its associated natural enemy complex.

Results from these Colombian surveys (the 3 zones extensively surveyed: the Andean zone, the Llanos, and the North Coast (Caribbean) are described in the 1995 Annual Report) show that *A. socialis* and *B. tuberculata* have the widest distribution and are found in all areas surveyed. They are usually found in lower altitudes, sharing the same climatic conditions and altitudes most favorable for the cassava crop. *T. variabilis* was not observed in the Llanos (eastern savanna). High altitudes above 1,000 meters such as the Andean region favored this species. Neither temperature nor relative humidity were factors related to populations of the three species. *A. socialis* was found mostly in those sites with temperatures above 35°C.

Numerous microhymenopteran parasitoid natural enemies found associated with each species of whitefly were collected and identified. Several were new species recorded from Colombia, and others were unrecorded species. The highest level of parasitism was found on *A. socialis* in the Llanos at 15.3% of the pupae parasitized. The highest levels of parasitism for *T. variabilis* and *B. tuberculata* were in the Andean Region with 12.1% and 13.9%, respectively.

Ten species of parasitoids were recovered from the three whitefly species (Table 4.2.1). Seven of these were recorded for the first time in Colombia, and four of these are new, undescribed species, recorded for the first time to science. *Encarsia hispida* (Aphelinidae) was the predominant parasite with 64.2% of the total parasites collected. Three species of the genus *Eretmocerus* are new undescribed species (referred to as *Eretmocerus* sp. "a", "b" and "c"). *Eretmocerus* sp. "b" was the second most numerous parasitoid collected, comprising 16.9% of all the individuals collected. The two species *E. hispida* and *Eretmocerus* sp. "b" were the most widely distributed throughout the three regions surveyed. They were the most abundant species in the Llanos with 79.3% and 69.1 %, respectively. In the Andean zone they represented 20% and 24%, respectively.

Some whitefly host specificity for the different species of parasitoid was also noted. *A. spiniferus* was only obtained from *A. socialis*; *E. hispida* and *Eretmocerus* sp. "b" were only collected from *A. socialis* in the North Coast and the Llanos, and it highly preferred *A. socialis* in the Andean zone. Parasitoid species obtained from *B. tuberculata* were rare in all three regions. *Eretmocerus* sp. "c" was the most frequent species found on *B. tuberculata* in the Andean Zone.

The other new species *E. bellottii* was found parasitizing *A. socialis* and *T. variabilis* in the Andean region.

These results further indicate the great potential for arthropod biodiversity that exists in agroecosystems, that is yet undiscovered and need to be further explored. Research on the behavior of these parasitoids has been initiated.

Table 4.2.1 Natural enemies associated with whiteflies on cassava from surveys of three ecological zones (Andean, Llanos and North Coast) of Colombia.

Species	N	Proportion (%)
Parasitoids:		
Aphelinidae (Family)		
<i>Encarsia hispida</i>	1845	64.2
<i>Encarsia pergandiella</i>	26	0.9
<i>Encarsia bellottii</i>	13	0.5
<i>Encarsia sp.</i>	48	1.7
<i>Eretmocerus sp. "a"</i>	30	1.0
<i>Eretmocerus sp. "b"</i>	485	16.9
<i>Eretmocerus sp. "c"</i>	12	0.4
<i>Eretmocerus sp.</i>	178	6.2
Platygasteridae (Family)		
<i>Amitus spiniferus</i>	159	5.5
<i>Amitus sp.</i>	79	2.7
Total	2875	100.0
Predators:		
Coccinellidae (Family)		
<i>Delphastus sp.</i>	143	

4.3 Plant pathology, by Elizabeth Alvarez and John B. Loke

In collaboration with PROFISMA, CNPMF/EMBRAPA, EBDA, CORPOICA, Brazilian and Colombian Universities, a work plan was designed to investigate cassava root rots and leaf blight. The objective is to develop simple integrated strategies to control soilborne pathogens, embedded

in a farming and agro-ecological management approach. In 1996 the main CIAT activities were isolation of root and stem rot pathogens and development of isolation and inoculation methods and molecular technologies. Biological control experiments were conducted to select strains of *Trichoderma* spp. and *Pseudomonas* spp. antagonistic to fungal pathogens.

4.3.1 Disease survey and isolation of the pathogens

The major limiting factor of cassava root and blight research is understanding of the pathogens that cause rot diseases. Therefore the following methodology was developed to isolate *Phytophthora* spp.

In 1996, 34 farms of the Colombian departments Valle, Quindio, Cauca and Bolivar were visited. The survey revealed that root and stem rots caused by *Phytophthora* spp. are a major problem. In these regions root rots have been known to cause losses in excess of 80% of potential production. At 26% of the farms visited, cassava was badly affected by root and stem rots. For the first time the impact of root rots on cassava yield was analyzed (Fig. 1.1.). Despite full chemical treatment of stem cuttings (planting material), cultivation on ridges and the relative long dry periods and crop rotation at CIAT, cassava is severely affected by root rots. Average root yield reduction by root rots was calculated to be 7.1 T/ha per growing cycle for the period 1981-1995 by analysis of 60 field experiments.

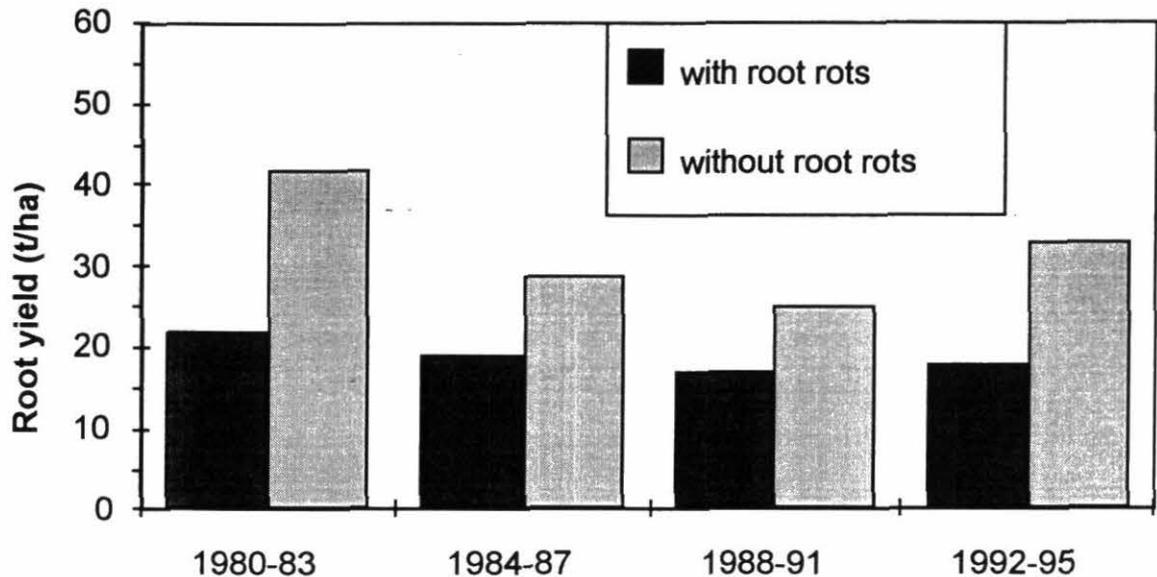


Figure 4.3.1. Root rot development of cassava cultivar MBRA-12 at CIAT.

Random samples were taken from affected plants at the farms visited during the survey of 4 departments in Colombia. *Phytophthora* spp. was isolated by direct plating of diseased plant tissue on selective media and by baiting with a selective host. For direct plating small fragments were placed on V8 juice agar (V8A) and Potato-Dextrose Agar (PDA) amended with thiabendazole (0.5 $\mu\text{l/ml}$), benomyl (20 $\mu\text{g/ml}$), ampicillin (250 $\mu\text{g/ml}$), penicillin (400 $\mu\text{g/ml}$) and rifampicin (10 $\mu\text{g/ml}$). The baiting technique was as follows: soil or macerated plant tissue was suspended in sterile deionized water, and cassava sprout fragments of approximately 2 cm were placed in the suspension. About 1 wk after incubation, fragments of the sprouts were placed on the described selective media. Table 4.3.1 presents a summary of the successful isolations.

Table 4.3.1. Phytophthora root and stem rot diseases observed at 4 Colombian departments and their isolation.

Collection Site	Number of Farms Visited	Positive Recovery ¹			Number of Isolates
		Root	Sprout	Rhizosphere	
Cauca	10	11	1	4	7
Quindio	15	3	2	12	41
Valle	10	3	0	14	25
Bolivar	1	2	0	0	2

¹ number of farms from which *Phytophthora* spp. were isolated.

Isolation of *Phytophthora* spp. was effective using the described techniques, and 75 isolates were obtained. Preliminary inoculations of some of the isolates confirmed their pathogenicity to cassava. A rapid and accurate method to detect these pathogens in plant tissue, soil and water samples is required. Research is ongoing to develop a polymerase chain reaction (PCR) method.

Culture of cassava *Phytophthora* species

It is necessary to have information about the laboratory culturing of *Phytophthora* species to determine the identity of species, to study their responses to ecological factors, and to develop inoculum for use in pathogenicity tests. *Phytophthora* spp. isolated from cassava grow and sporulate best on V8A compared with PDA, at 27°C, with or without light.

Phytophthora cultures established from the field are always contaminated with bacteria. Therefore the following techniques were tested to obtain bacteria-free cultures: 1) placing a small piece of fungal growth on the inside of a glass Van Wiegham cell that has been placed on a selective medium; 2) inverting the entire agar culture in a petri plate; 3) selective weak medium (medium diluted with water). The most efficient method is transferring fungal discs to water agar that contains appropriate antibiotics.

To check if a *Phytophthora* isolate is free from bacterial contamination a fungal plug was transferred to a test tube with liquid medium (g/l: peptone, 5; casamino acids, 0.5; malt extract, 0.5; pH 7.2). If the culture is contaminated with bacteria, the liquid becomes cloudy within 48 h

of incubation at 27°C, but if bacteria-free, the liquid remains clear. *Phytophthora* spp. were successfully transferred from medium to V8A.

Single-zoosporic isolation was readily accomplished by isolation of a single germinated cyst or non-cysted zoospore after a highly concentrated fungal suspension had been spread thinly on the surface of 3% water agar on a microscope slide. After 12-24 h, germinated spores were detected on the surface of the agar by use of a microscope with a magnification of 400x. With a bent Pasteur pipet, a germinated spore was marked, and by use of a needle a plug was placed on V8A. Activities to identify and to conserve strains have been started.

Development of bioassays to study pathogen variability and to test cassava germplasm for resistance

The results of known bioassay methods are not well correlated to field resistance of cultivars. Therefore 5 new *in vitro* and greenhouse methods of inoculation, described in Table 4.3.2, were designed to inoculate cassava plants with pathogen strains. To compare the efficiency of the methods, 22 cultivars were inoculated with 4 *Phytophthora* spp. isolates. Linear extent of root rot and stem necrosis caused by *Phytophthora* spp. was recorded.

Table 4.3.2. Fungal bioassays tested for root and sprout inoculations.

Parameter	Root	Root & Sprout		Sprout	
Plant material	slice of swollen root ^a	cylinder of swollen root ^b	potted plant of 3 wk ^c	potted plant of 4 wk ^c	excised shoot of 22 wk ^c
Technique	disc in center of each slice	cylinder placed vertical in suspension ^d	application of suspension ^d to soil; flooding of soil and removing foliage	fragment on 5 mm wound of stem	cut sprout in suspension ^d
Incubation conditions	cellophane in incubator	plastic box in lab.	screenhouse	greenhouse	glass tubes in lab.
Evaluation period ^e	14	14	14	28	28
Efficiency ^f	24	24	15	15	60

^a 1.5 cm x approximately 5 cm.

^b 1.5 cm x 3 cm.

^c propagated by stem cutting.

^d 1.0×10^5 spores/ml.

^e d after inoculation.

^f average number of inoculations per field plant.

All tested *Phytophthora* strains infected the roots and stems severely in the proven bioassay systems. Results are specified in Table 4.3.3. Inoculated roots were characterized by a yellow discolorization, tissue decomposition, soft texture and pungent smell. Between cultivars there were only small differences in amount of rot obtained.

On young cassava plants, infection by *Phytophthora* spp. produces necrotic lesions on the stems, and the infected stem appears shrunken. Inoculated plants presented light brown or almost black discoloration of the stems. The first observable symptoms are brown patches on the petioles, and the older leaves turn yellow, then there is a loss of turgidity and a general dieback of the whole plant occurs. Leaves eventually turn brown and do not fall off. Significant differences in lesion expression caused by *Phytophthora* spp. was detected by inoculating cassava stems. Tolerant cultivars exhibited shorter or less intense lesions than the susceptible cultivars. The plants of some cultivars were killed, while others recovered by new sprouts.

Table 4.3.3. Results of laboratory and greenhouse bioassays of *P. drechsleri* and *P. nicotianae* (*P. parasitica*) inoculated on cassava cultivars.

Pathogen & cassava cultivar	Root ¹			Sprout ¹				Field Reaction ²
	Whole	Slice	Cylinder	Soil	Soil	Wound	Excised Sprout	
<i>P. drechsleri</i>								
CG 1355-2	R	S	S	S	R	R	R/S	
MCOL 2280	R	S	S	R	R	R/S	R	
MCOL 1505	S	S	— ³	R	S	S	R	
MBRA 1045		S	S	R	R	R		R
MBRA 532 ⁴	S		S			R/S	R/S	R
<i>P. nicotianae</i>								
MCR 54	S	S	R	S	R/S	R/S	R/S	
MCOL 2280	R/S	R	S	S	R/S	S	S	
CG 1355-2		S	S	S	R/S	R/S	S	
MCOL 1505		S	S	S	S	S	S	
MBRA 532 ⁴		S	R			S	R/S	R

¹ S, susceptible (high pathogenicity); S/R, intermediate pathogenicity; R, resistant (low pathogenicity).

² According to farmers.

³ Not sufficient plant material available.

⁴ Variety Ossoduro.

Resistance to root rot not necessarily implies resistance to leaf blight and vice versa. To be sure of an adequate root inoculation test other field resistant and susceptible cultivars have to be identified and inoculated.

Cassava infected by *Phytophthora* spp. at CIAT

Phytophthora root rot and leaf blight were not previously considered to be important limiting factors for cassava stake production at CIAT. By use of advanced isolation methods it was discovered that both diseases are present at CIAT, and severely reduce stake quality. In 1996, the following observations were made about stem cutting and seedling infections by *Phytophthora* spp. Greenhouse plants propagated from a CIAT cassava field showed disease expression,

varying from stem necrosis to death. Out of 200 cultivars, all in the core germplasm collection, 61 expressed *Phytophthora* leaf blight disease. In plants a general blight (yellow leaves and/or necrotic lesions on sprouts) was observed 3-5 wk after planting stem cuttings. Cassava plantlets in the greenhouse can be predisposed to the severe expression of stem rot if the soil in the pots is allowed to dry to near the wilting point before excessive watering.

About 3 ha of commercial cassava production was severely affected by root and stem rots, the field was infested with *Phytophthora* spp. which was isolated from irrigation water, soil and cassava sprout tissue. In plants a general dieback was observed 3 months after planting stem cuttings, 7 months later the swollen roots were rotten. Under less favorable conditions for the fungus, the plants recuperated. Most plants at all stages of growth from germination to maturity showed symptoms. It is concluded that under severe conditions or severe infection plants may be killed, but in plants that survive the stakes are infected.

The disease was observed in seedlings propagated by botanical seeds in seedling containers. It affected seedlings severely, and 90 % of the plants died. The infected stem shrivels, causing wilt and death of the seedling within 10 d.

Phytophthora spp. can be considered to be the main factor causing cassava root rot in Colombia. From the above it can be concluded that systemic transmission of these pathogens is caused by stake planting material. To obtain high yields of cassava under marginal conditions, it is important to start with good sanitary quality of propagation material. Severe outbreaks can be expected in the near future because cassava production continues to become more intensive, at CIAT as well as outside.

In course of 1996, various control methods, e.g. thermotherapy of stem cuttings and chemical control, have been evaluated to reduce infection at CIAT. From 3 heat treatments (dry heat, vapor and hot water bath), immersion of stakes in water of 50°C for 20 min was the most efficient. Stake germination and plant vigor following this treatment were similar to control plants. Development of rapid diagnosis, like PCR, of the diseases has started. A more detailed study to know if *Phytophthora* diseases are transmitted by cassava stem cuttings has to be initiated.

Molecular variation in strains of *Phytophthora* species affecting cassava stake quality and root yield

Phytophthora strains were obtained from 34 cassava farms of the Colombian departments Valle, Quindio, Cauca and Bolivar. Ten strains were compared in virulence tests. Virulence was determined by inoculating cassava plants by stem wounding. The effectiveness of this procedure in distinguishing the severity of disease reactions produced by different strains was evaluated. Considerable variation in lesion size was observed. The symptoms caused by individual strains of *Phytophthora* species were very similar, although symptom severity differed among strains and species. The virulence testing of 76 strains is ongoing.

Amplification of the ITS (Internal Transcribed Spacer) region of rDNA was obtained with template DNA from *P. drechsleri*, *P. nicotianae* and *P. erythroseptica* using extracted DNA or by scraping mycelium. The amplified product for the ITS region of all species was the same. Restriction Digest with *AluI* of the product amplified for the ITS region showed 3 different restriction patterns, which corresponded to the 3 species tested. DNA from 76 *Phytophthora* strains will be digested with *CfoI*, *MspI*, *MseI* restriction enzymes to differentiate the species. In addition, RAPD analysis will be conducted to determine genetic variation among strains.

4.3.2 Biocontrol of Root and Stem Diseases

Antagonism of *Trichoderma* spp. and *Pseudomonas* spp., potential biocontrol agents, to *Phytophthora* spp., *Diplodia manihotis* and *Fusarium oxysporum*, the causal agents of cassava root and stem rot diseases in America and Africa, was investigated.

Extracellular production of antifungal compounds by *Trichoderma* spp.

Trichoderma spp., isolated from the rhizosphere of cassava plants were screened *in vitro* for antifungal activity against a range of fungal pathogens: 2 isolates of *P. drechsleri* and *P. nicotianae*, *D. manihotis* and 1 of *F. oxysporum*. A control treatment with *Thielaviopsis* spp., a causal agent of bud rot of oilpalm, was included.

The activity of 14 one-week-old cultures differed among the pathogens. Filtrates from liquid cultures of *Trichoderma* isolate 11TSM-4 caused marked reduction in growth of the pathogen *D. manihotis* after 24 h of incubation. Filtrates of 14PDA-4 were most effective in reducing the growth of *F. oxysporum*. The culture filtrates of this *Trichoderma* spp. isolate caused 40% growth inhibition of the pathogen. They were toxic in each replication until the end of the experimentation period of 1 week. Experiments are ongoing to study the effect of isolates on pathogen spore germination and germ tube elongation, and the effect of application of pure and mixed extracts of these isolates on infected cassava plants.

Evaluation of *Pseudomonas* spp. by excised shoot bioassay

Biocontrol strains from the genera *Pseudomonas* were tested for antifungal activity in bioassays *in vitro*. This study is undertaken to select strains to control *Phytophthora* spp. Shoots of cassava cultivar CG 1-37 in an excised shoot bioassay were bacterized, and 3 d later inoculated by a *Phytophthora* spp. suspension. Preliminary observations indicate that from 9 isolates, 1 antagonist inhibited lesion development of the cassava sprouts.

Production *Trichoderma* spp. in liquid medium

Production of *Trichoderma* spp. using a liquid culture system on media based on molasses, V8 juice and mineral solutions was studied. These media were incubated at 30°C for 1 wk under darkness of shake culture. Of the media evaluated, a mineral salt medium supported maximum conidia production for 27 *Trichoderma* spp. strains. High antifungal activity was observed by culture filtrates prepared with the medium. Field plots and cassava stem cuttings, infested by *Phytophthora* spp., were inoculated by strains grown in the identified medium.

4.3.3 Study on Stem Rot Disease Caused by *Fusarium* Species

Fusarium solani and *F. oxysporum* strains were obtained from 86 cassava samples including stem cuttings, swollen and secondary roots, and soil obtained from different zones in Colombia

(Maria La Baja, Carimagua, Media Luna, Pivijay, Calarcá, Palmira, Villavicencio, Alcalá, Pescador, Granada and Quilichao).

In addition, 4 methods of classification were tested for differentiation of the strains, including morphology, growth, pathogenicity and site directed (rDNA) PCR. These species were compared in pathogenicity tests, in which the symptoms caused by individual strains were very similar, although symptom severity differed. Infected seedlings exhibited necrotic stem lesions compared with healthy controls. The majority of strains were not clearly differentiated on PDA by colony type, growth or pigment production.

Amplification of the ITS region of the rDNA was obtained with template DNA from 10 strains using extracted DNA. Restriction digest with *AluI*, of the product amplified for the ITS region showed 2 different restriction patterns, which corresponded to the 2 species tested. The PCR analysis was more reliable in differentiating the strain than the other methods. In addition rDNA analysis offered a distinct advantage by requiring less time for distinguishing among the species. The simplicity of the PCR analysis presented in the study, and the accuracy of their predictions make them acceptable for identification of *Fusarium* species on cassava. The most pathogenic of the *F. solani* and *F. oxysporum* strains will be distinguished from the other *Fusarium* strains by RAPD markers.

Furthermore, a screening of 51 cassava cultivars has been conducted by inoculating 28 *Fusarium* strains to evaluate resistance.

4.3.4 Study on Stem Rot Caused by *Diplodia manihotis*

A survey in cassava growing areas in Colombia was conducted in 1995-1996, and samples including cassava stem cuttings and roots were collected. 55 strains of *D. manihotis* were isolated and monosporic cultures were obtained.

Three inoculation methods were used for pathogenicity studies, they included stem injection, stem wounding and root cylinder inoculation. Clear symptom development was observed by stem wounding. From a total of 55 strains, 8 were pathogenic on cassava plants. A pathogenic specialization study has been initiated. At the moment, 20 cassava cultivars have been inoculated with 40 strains selected from the *D. manihotis* collection.

4.4 Virology, by Lee A. Calvert

When the project first began there only one major virus disease was known in the northeast of Brazil. Cassava vein mosaic virus was widespread but its importance as a pathogen had not been documented. The characterization of the virus has progressed, and it is clear that this pathogen is causing major losses throughout the semiarid zone. Farmers still do not recognize the seriousness of this disease, and more work is needed to make the cassava producers aware of the methods and reason for controlling this disease.

During the course of this project, the disease caused by cassava frogskin virus has been expanding its range. It is now endemic in parts of the state of Bahia. This probably occurred because of the major drought in 1992, that caused to farmers to import cassava stem cuttings from neighboring states that border the Amazon region which have wetter climates. Cassava frogskin disease can cause the complete failure of a crop and is an important new pathogen that needs to be controlled.

In Uganda, African mosaic disease has reduced cassava production by 50%. In this project, we collaborated with other institutions in England and Uganda and determined that the epidemic is being driven by a novel geminivirus. The Ugandan African mosaic disease is caused by a virus that is a hybrid of African cassava mosaic virus and East African mosaic virus. This discovery is leading to the development of germplasm solutions that will mitigate the losses of this devastating epidemic.

4.4.1 Cassava vein mosaic virus (CVMV)

Characterization of CVMV.

The molecular characterization of the genome of one isolate of CVMV was completed, and an article was published during 1995. It was determined that CVMV is a member of the family of plant pararetroviruses but was not as closely related to the cauliflower mosaic virus group as previously thought. Because this virus is so unique, a study was made to determine the diversity of isolates from the different states in the northeast of Brazil, to confirm that all isolates could be

identified using the diagnostic methods developed during the molecular characterization and to confirm the genomic structure.

Development of rapid detection methods for CVMV.

Samples were taken from plants collected from several states in Brazil that showed typical CVMV symptoms. Nine isolates of CVMV were amplified using PCR. The isolates were from Fortaleza, Crato and Tiangua in Ceara, Araripina and Petrolina in Pernambuco, Bento Fernandes in Rio Grande do Norte, Piritiba and Itaberaba in Bahia, and Seripe in Alagoas. The diagnostic test used specific PCR primers for two regions of the genome. All of the samples were positive for CVMV using both sets of primers. The virus complex at this level appears to be conserved, and the PCR detection can be considered a reliable detection method over the range of CVMV. Fresh material is more reliable for detection, but if the samples are not for immediate analysis, it is important to desiccate the leaves rapidly. This method of detection is currently being used in Brazil and at the seed health laboratory of CIAT.

Survey of virus isolate diversity.

The analysis of CVMV isolate diversity was done by molecular analysis of some isolates used for the confirmation of the PCR detection method. The CVMV PCR products were cloned into bacterial plasmids, sequenced, and analyzed for homology among the CVMV isolates. For the majority of the isolates, two distinct regions were analyzed. These were the part of the genome that encodes the replicase gene and the region containing the putative coat protein and movement protein. A comparison of the region that encodes the replicase gene reveals that the percentage of homology was always more than 95%. This area appears to be highly conserved with little variation between the isolates.

The region encoding the coat protein and movement proteins has a higher level of variation. While the homology levels were generally more than 90% over the entire region, the junction between the two proteins showed a high degree of diversity. One of the key differences is that the coat protein does not read through to the movement protein as a polyprotein. Because these changes are important in understanding the pathogen diversity and the genomic structure,

additional isolates are being characterized. It can be concluded that, over the range of CVMV, there is a significant degree of molecular diversity and that care should be taken not to spread the isolates via transportation of infected germplasm.

CVMV yield loss field trials.

One of the important questions regarding Cassava Vein Mosaic Virus (CVMV) is the effect on root yield. Since the absence of symptoms is not a reliable indicator that the cassava clone is free of CVMV, *in vitro* plants were used for this yield trial experiment. The plants were grown in the EMBRAPA station near Petrolina Pernambuco. A randomized block design was used. The yield was determined, and the roots were analyzed for starch content. The yields of the infected plants averaged 10% less than the healthy controls. The starch content of the roots from infected plants was 20% less than that of the healthy controls. By combining the decrease in yield and starch content, the losses attributed to CVMV are more than 25%. This disease is endemic over most of the northeast of Brazil and the incidence in the field is often very high, therefore it appears that the economic importance is much higher than previous estimates.

The challenge is to educate the farmers in the region of the losses that they are incurring from this invisible pathogen. The introduction of health planting material is one method that the farmer can rapidly adopt to mitigate losses. This activity needs to be combined with rapid multiplication technologies that deliver improved varieties that can be tested for suitability using farmer participatory methods.

4.4.2 Cassava frogskin disease (CFSD)

Extent of CFSD in the northeast of Brazil.

During the course of this project, cassava producers started reporting a new disease of their plants that was very severe. The roots did not expand and lip-shaped fissures appeared on the cortex of the roots. This disease has been confirmed to be cassava frogskin disease. This disease is known to be widespread throughout the Amazonian region of Brazil, and this is the first report of the disease in the northeast of Brazil.

Analysis of the signs of CFSD.

Cytological studies of the roots of normal and CFSD-affected roots were made using both light and electron microscopes. There were dramatic differences in the root periderm and cortex in the accumulation of starch. In CFSD-affected roots, the starch granules were either absent or apparently empty. The cell layers were much more densely packed in the CFSD-affected roots, and this is due to the absence of starch granules and the hyperplasia.

The other characteristic of CFSD is the enlarged lip-shaped fissures on the surface of the roots. These studies suggest that these fissures are caused by a hyperplasia of the cells in the root cortex. The root cortex is much thicker than normal, yet there are no starch granules in these cells. The hyperplasia is not equal all along the cambium meristem, thus causing the lip-shaped fissures.

These studies were made to identify characteristics that can be used in screening for resistance to cassava frogskin disease (CFSD). The problem with using just root weight is that there is a high degree of variability within the cassava core germplasm collection because some of the materials are better adapted than others to the growing conditions in Cauca, Colombia. It is important to be able to distinguish between poor root growth due to poor adaptation to the growing conditions as compared to the affects caused by CFSD. This assay using the light microscope is relatively rapid and inexpensive and can determine if the starch accumulation and cytology of the cells is abnormal. This assay will be used in the screening of the core collection for tolerance to CFSD.

Screening the core collection for resistance or tolerance to cassava frogskin virus.

During 1995, a field experiment was begun to analyze the 630 accessions of the cassava core germplasm collection for tolerance or resistance to cassava frogskin virus (CFSV). The methodology is to inoculate plants of the core collection using a single source of CFSV. Characteristics including yield and starch accumulation will be analyzed and compared with CFSD-free accessions. Over 500 of these accessions are currently in the field in either the first or second cycle of evaluation. Most of the remaining accessions have been inoculated and will be planted during this season. The number of infected plants per accession will be increased and

screened over several years. The most promising clones will be tested in multilocational trials. This is a continuing activity that will take at least three to five years to get reliable results. Any resistant or tolerant accessions will be used as parents in the development of resistant germplasm gene pools.

Control Strategies.

Until resistant germplasm is available, the use of health planting material is the best method to mitigate losses due to CFSD. This can be combined with the control strategy for CVMV (cassava mosaic virus) and is also useful in controlling root rots. The next phase of the project needs to develop a strategy of getting the farmers to produce health cassava stem cuttings to replant their fields and to trade with the adjacent communities.

4.4.3 African cassava mosaic disease epidemic in Uganda.

African cassava mosaic virus (ACMV) serotype "A" is endemic throughout Uganda and causes substantial, albeit manageable, yield losses. Since 1990, there has been a severe epidemic of African cassava mosaic disease (ACMD) that has moved from the northern regions of Uganda towards the south at the rate of 10-20 km per year. The symptoms in the plants after the epidemic arrives at a site are very severe, with the leaves becoming extreme small and deformed. When infected stem cuttings are used in the following planting cycle, the plants produce roots of no value, causing yield losses of nearly 100%. This epidemic has caused nearly a 50% decrease in total production throughout Uganda and total loss of production in many regions.

In collaboration with IITA, SCRI (Scottish Crop and research Institute, Dundee) and NARO (Uganda NAR), the molecular analysis of the "A" component of the severe Ugandan isolate was made. This represents 50% of the genome of the geminivirus, and the result is that the Uganda isolate appears to be a hybrid between ACMV (serotype A) and East Africa cassava mosaic virus (EACMV, serotype B). The coat protein of this isolate is almost identical to ACMV and cannot be distinguished from ACMV using the currently available monoclonal antibodies specific to serotype A. The common region and the replicase gene are more similar to EACMV.

Since there is such severe deformity of the leaves, the whiteflies move from severely infected plants to cassava that is not yet affected by the severe strain of ACMD in an epidemic front. When this front moved through the NARO experiment station near Kampala, some NARO/IITA cassava varieties showed a fair degree of resistance or tolerance to the Ugandan geminivirus. Many of these varieties are high in cyanide, and, while they are good for making gari, they are not suitable for the Ugandan market where fresh cassava is eaten. Breeding efforts by IITA and NARO are addressing this problem to produce resistant varieties that are low in cyanide. The fact that the epidemic is caused by a distinct geminivirus, and not by simply higher levels of whiteflies, confirms the policy of NARO to insist that only varieties resistant to the virus are replanted in the areas behind the epidemic front.

4.5 Crop management, by Mabrouk El-Sharkawy, Luis Fernando Cadavid, Sara Mejía de Tafur, and Manuel Gerardo Cayón

Long-term response of cassava to NPK levels in acid infertile soils at Santander de Quilichao

Table 4.5.1 contains data on the long-term response (13th year) to nitrogen-phosphorus-potassium (NPK) fertilizer in infertile acid soils at Santander de Quilichao, Cauca, Colombia. In absence of NPK fertilizer application, dry root yield and top (aerial plant parts) biomass were significantly lower in both cassava clones (M Col 1684 and CM 91-3). After 13 consecutive cropping cycles in this soil, dry root yield was about only 4 t/ha. With application of 100 kg/ha each of NPK, root yields were dramatically increased (about 10 t dry root/ha). The largest observed response was for K as indicated by much lower yields in absence of K application and with 100 kg/ha of N and P. Lesser yield responses were observed for N and P. These findings again demonstrate that K is the most limiting nutrient in these soils, which have high organic matter content (ca. 7%). To sustain reasonable productivity, K fertilizer must be applied annually since most K uptake (> 60%) is removed in the harvested roots.

Table 4.5.1 Effect of NPK rates on dry root yield and dry plant top (t/ha) of two cassava varieties during the 1995-1996 season (13th cycle) at Santander de Quilichao, Cauca, Colombia¹.

Fertilizer treatment (kg/ha)			M Col 1684		CM 91-3	
N	P	K	Dry root	Dry top	Dry root	Dry top
0	0	0	4.4	2.0	3.9	2.3
50	50	50	9.1	3.5	7.8	3.9
0	100	100	10.4	3.3	5.2	2.9
50	100	100	10.1	4.4	7.8	3.8
100	100	100	10.5	4.3	9.2	3.9
100	0	100	9.8	4.0	8.9	4.4
100	50	100	13.0	4.2	8.4	3.8
100	100	0	7.5	3.0	5.2	3.1
100	100	50	10.3	4.0	7.6	4.1

¹ LSD 5% (Tukey) for comparison between NPK treatments for dry root 3.75, for dry top 1.57.

Long-term effects of surface mulch, fertilizer and tillage on cassava productivity in poor sandy soil in northern Colombia

Table 4.5.2 presents data on root yield and plant top biomass as affected by plant surface mulch (dry grass), NPK fertilizer and type of tillage on sandy soils at Media Luna, northern Colombia. Both dry root yield and top biomass were significantly increased by the application of either mulch or NPK fertilizer at a moderate level. On the other hand, conventional tillage increased yield only with NPK application in absence of mulch. The combination of surface mulch and zero tillage gave the highest root yield with and without fertilizer. These findings indicate that consecutive cultivation of cassava in these poor soils would lead to very low yields unless appropriate measures are followed to maintain soil fertility. The choice between application of NPK fertilizer or surface plant mulch would depend on various factors related to the socio-economic conditions of the local farmers. However, the potential economic return due to fertilization is high in this case. Moreover, the application of fertilizer would ensure enough quantity of planting material with high quality.

Table 4.5.2 Dry root yield and dry plant top (t/ha) of cassava (cv. M Col 1505) at Media Luna, Magdalena, Colombia during the season 1995-1996 (8th cropping cycle).

Treatment	Fertilized ¹		Unfertilized	
	Root	Top	Root	Top
Conventional tillage	4.3bc ³	2.4b	1.2b	1.4b
Conventional tillage + mulch ²	5.9ab	3.6ba	4.6a	2.7a
Zero tillage	3.3c	2.3b	1.4b	1.2b
Zero tillage + mulch	6.9a	4.2a	4.9a	2.8a

¹ Fertilized with 330 kg/ha 15-15-15 NPK (50 N; 21.6 P; 41.7 K kg/ha).

² Mulch, 12 t/ha dry grasses applied annually.

³ Values in columns followed by the same letters are not significantly different at 5% by Duncan's test.

Table 4.5.3 contains data on the response of cassava to both plant mulch and NPK fertilizer in the soils of Santander de Quilichao. Although these soils contain greater organic matter than the sandy soils of northern Colombia, mulch application increased root yield even in the first year of the trial. Again, these findings indicate the beneficial effect of plant residues as mulch. Fertilizer application also increased root yield in these soils which have been under cassava cultivation for several years. Both plant top biomass and root dry matter content were also enhanced by applications of surface mulch and NPK fertilizer. When native weeds were repeatedly cut, without uprooting, and used for mulch, yields were depressed in absence of NPK fertilizer. This finding suggests that weeds significantly compete for nutrients in this case. Previous research (Cassava Program Annual Reports, 1987-1992) indicated that using post-emergence herbicides at an advanced stage of weed growth enhanced cassava productivity as compared with no weed mulch.

Table 4.5.3 Dry root yield and dry top (t/ha) and root dry matter (%) (cv. CM 523-7 and CM 507-37) at Santander de Quilichao, Cauca, Colombia. First season (1995-1996).

Treatment	CM 523-7						CM 507-37					
	Unfertilized			Fertilized ¹			Unfertilized			Fertilized		
	Root	Top	Root dry matter (%)	Root	Top	Root dry matter (%)	Root	Top	Root dry matter (%)	Root	Top	Root dry matter (%)
Without mulch	9.7	3.9	40.9	12.2	5.3	40.8	10.3	3.9	34.8	13.9	5.7	36.9
Brachiaria mulch ²	11.0	5.7	41.3	13.8	5.5	41.1	12.6	4.4	36.8	16.3	7.5	38.2
Weed mulch ³	7.1	2.8	40.7	13.4	5.1	41.3	8.3	2.8	33.4	13.3	5.1	36.3
Mean	9.3	4.1	41.0	13.1	5.3	41.1	10.4	3.7	35.0	14.5	6.1	37.1
LSD 5% (Tukey)	2.65	1.06	2.91									

¹ Fertilized with 500 kg/ha 10-20-20 NPK (50 N; 44 P; 83 K kg/ha).

² Mulch of *Brachiaria decumbens*, 12 t/ha dry applied annually.

³ Mulch of dry weed, 4.44 t/ha; top biomass of *in situ* weeds were used through repeated cuttings.

Effects of quality of planting material and water stress on cassava productivity

Figures 4.5.1, 4.5.2 and 4.5.3, present data on leaf photosynthesis, stomatal conductance and midday leaf water potential as affected by stake quality and water stress. In this trial, planting material of M Col 1684 were obtained from the long-term NPK trial at Quilichao on the 12th cropping cycle. Mother plants with the following NPK levels were used: 0, 0, 0 (T1); 50, 50, 50 (T2); 100, 100, 100 (T3); and 100, 100, 0 (T4) kg/ha N, P, K. The trial was conducted in the field lysimeter at Quilichao, where half of the experimental area was deprived of water at 65 days of planting by covering the ground with plastic sheets for 3 months. Leaf net photosynthetic rate (Fig. 4.5.1) was similar in both the control and the water-stressed crops, irrespective of the origin of the planting material. Apparently, the stake quality did not affect leaf photosynthesis. Stomatal conductance was significantly reduced by water stress in all stake origins. However, this effect was observed only at 60 days after stress was initiated (Fig. 4.5.2). The same trend was observed in midday leaf water potential (Fig. 4.5.3). These data indicate that cassava tolerates prolonged water stress, and its leaves maintain a high level of photosynthesis during the water stress period. Moreover, cassava conserves water by partially closing its stomata during prolonged water stress.

Evaluation of crop/soil management practices in farmers fields in northern Colombia

In collaboration with CORPOICA, several field trials were established in April-May 1996 in various sites to evaluate the effects of the application of mulch and fertilizer on productivity of the cassava/maize intercropping system in farmers' fields. These trials are located in Los Palmitos, Dept. of Sucre; in Plato, Dept. of Magdalena; Pueblo Nueva, Dept. of Córdoba; Pivijay, Dept. of Magdalena; Chiriguana, Department of César; and in El Carmen de Bolivar, Dept. of Bolivar. These trials will be harvested in Feb./March 1977.

In addition to the research mentioned above, the Associate, Luis F. Cadavid, had participated in two training workshops for specialists of CORPOICA in regions 2 and 3. He prepared and dictated two courses on integrated crop/soil management. The first course was in Barranquilla, Atlantico in May 1996, and the second was in Turipana, Montería, Córdoba in August 1996.

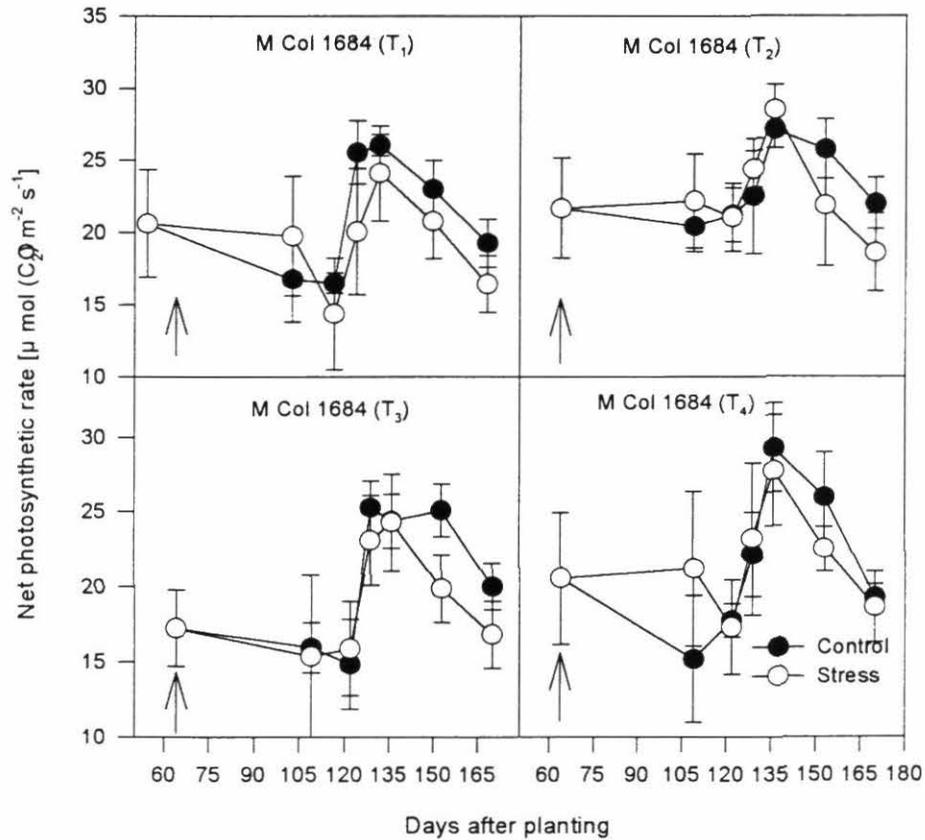


Figure 4.5.1 Effects of the quality of planting material and water stress on leaf photosynthesis. Arrow, Initiation of stress. Stakes were obtained from mother plants on the 13th cycle of a long-term NPK trial that received annually the following treatments: T₁=0,0,0 NPK; T₂=50,50,50 kg/ha NPK; T₃=100,100,100, kg/ha NPK; T₄=100,100,0 kg/ha NPK. (bars, $\pm\text{sd}$).

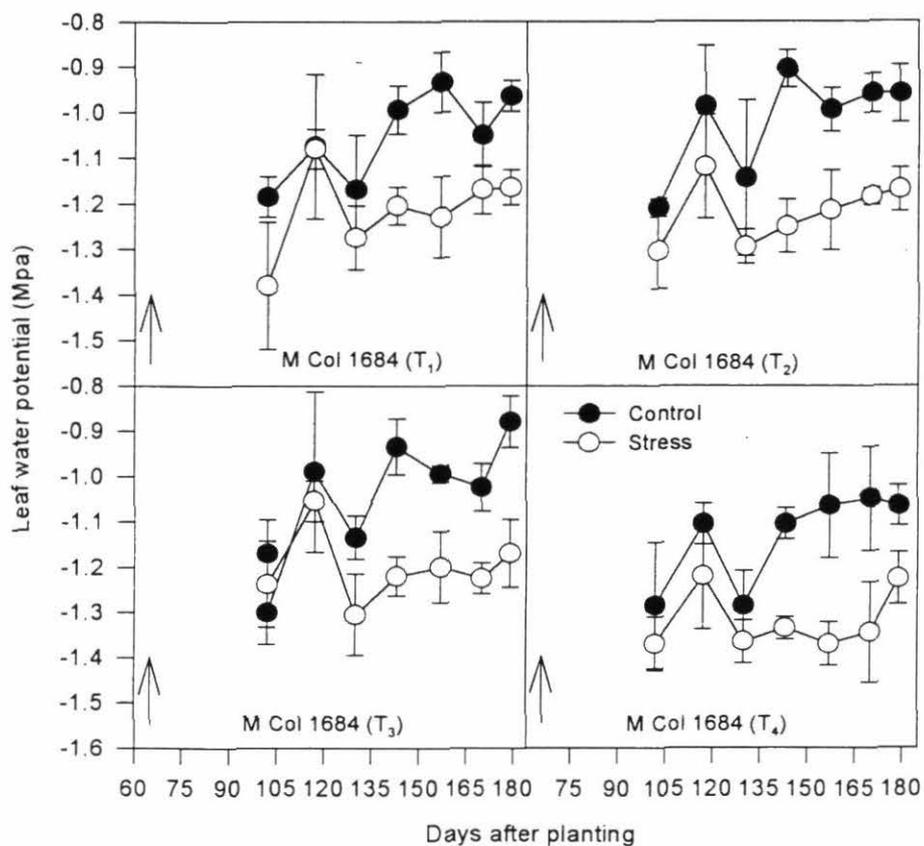


Figure 4.5.2 Effects of the quality of planting material and water stress on leaf conductance. Arrow, initiation of stress. Stakes were obtained from mother plants on the 13th cycle of a long-term NPK trial that received annually the following treatments: T₁=0,0,0 NPK; T₂=50,50,50 kg/ha NPK; T₃=100,100,100, kg/ha NPK; T₄=100,100,0 kg/ha NPK. (bars, \pm sd).

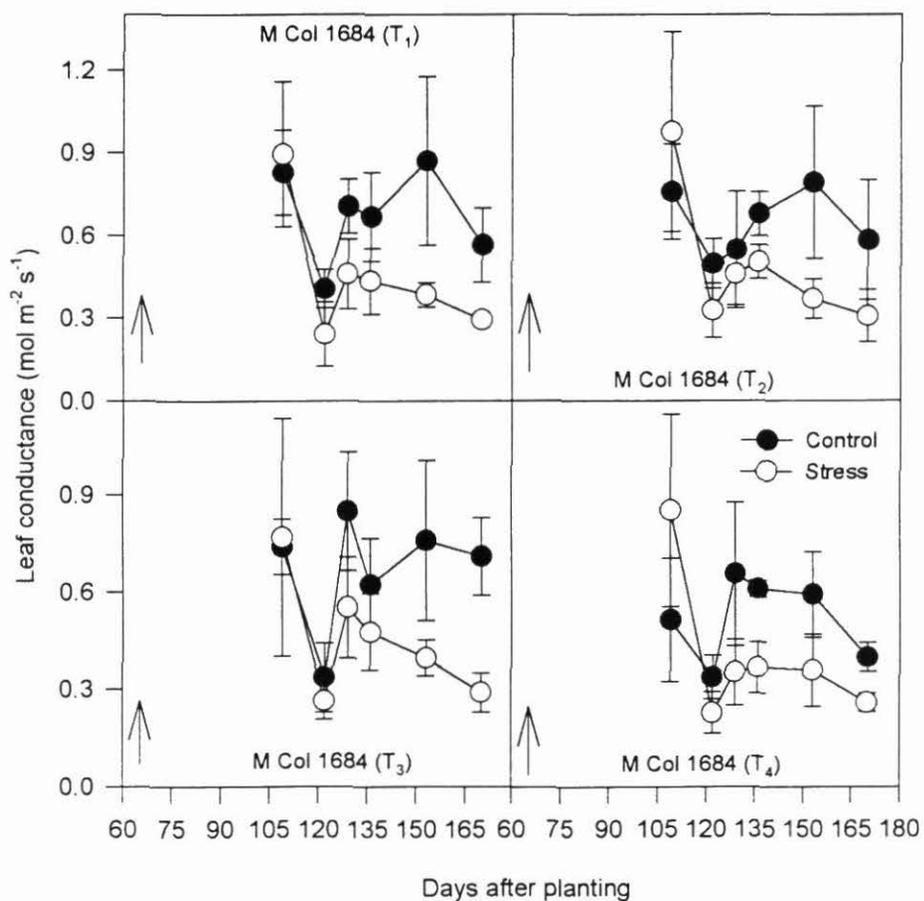


Figure 4.5.3 Effects of the quality of planting material and water stress on midday leaf water potential. Arrow, initiation of stress. Stakes were obtained from mother plants on the 13th cycle of a long-term NPK trial that received annually the following treatments: $T_1=0,0,0$ NPK; $T_2=50,50,50$ kg/ha NPK; $T_3=100,100,100$, kg/ha NPK; $T_4=100,100,0$ kg/ha NPK. (bars, $\pm \text{sd}$).

5 STRATEGIC RESEARCH AT CNPMF

5.1 Biological Control of the Cassava Green Mite, by Aloyseia Cristina S. Noronha

Introduction of phytoseiid mites

Individuals of *Typhlodromalus tenuiscutus* McMurtry & Moraes were collected in Puerto Cayo, Manabi Ecuador and sent from CIAT to the “Costa Lima” Quarantine Laboratory, EMBRAPA/CNPMA, municipality of Jaguariúna, State of São Paulo. During 1996, five shipments of *T. tenuiscutus* were made from EMBRAPA/CNPMA to EMBRAPA/CNPMF and a total of 2,150 individuals were sent, 75.48% of them arrived alive. As stated in PROFISMA’s Annual Report 1995, *T. tenuiscutus* shipments from CIAT during 1996 were with recently established colonies.

Maintenance rearing

Using an adaptation of McMurtry & Scriven’s phytoseiid rearing method, it was possible to obtain a monthly average production of about 2,500 individuals of *N. californicus* per tray on jack bean (*Canavalia ensiformis*) detached leaves, under laboratory conditions, at EMBRAPA/CNPMF (Table 5.1.1).

Mass rearing

T. tenuiscutus rearing was performed under laboratory conditions on *M. tanajoa* on detached cassava leaves. Mass rearing of *N. californicus* was carried out in a screenhouse. Individuals were fed on *Tetranychus urticae* on jack bean plants. Such a methodology enabled the production of 33,500 individuals of *N. californicus* starting from a single colony, in a one month period.

Release and monitoring of phytoseiid mites

Phytoseiid mites were released in cassava fields located in the States of Bahia and Pernambuco, as shown in Table 5.1.2. Releases were performed late afternoon, by attaching phytoseiid infested cassava leaves to cassava branches. A total of 10,813 individuals of *T. tenuiscutus* and 34,020 individuals of *N. californicus* were released during 1996.

Table 5.1.1. Maintenance rearing of *Neoseiulus californicus*, on detached leaves of *Canavalia ensiformis*, under laboratory conditions.

Colonies	Female Initial N°	Evaluation Date	Number			
			Larvae	Nymph+male	Female	Total
01	100	16/Sept/96	215	973	970	2,518
		16/Oct/96	83	490	1,006	1,579
		15/Nov/96	340	1,205	2,021	3,566
02	100	18/Sept/96	178	1,064	882	2,124
		16/Oct/96	151	1,103	1,536	2,790
		15/Nov/96	111	960	1,819	2,890
03	100	25/Oct/96	12	1,203	1,960	3,175
		27/Nov/96	168	885	1,399	2,452

Table 5.1.2. Field releases of phytoseiid mites.

Species	State	Municipality	Field	Individuals	Releases
<i>T. tenuiscutus</i>	Bahia	Juacema	Juacema	600	1
		Jaguari	Jaguari	10,213	50
Total				10,813	
<i>N. californicus</i>	Bahia	C. Almas	EAUFBA	3,000	1
		Ichu	Ichu	8,070	2
		Jaguari	Jaguari	19,950	1
	Pernambuco	Petrolina	Ad	3,000	4
Total				34,020	

Monthly monitoring recovered 1,085 mites from all cassava fields where releases were performed. One hundred and forty-two slides were examined under microscope enabling preliminary identification of 28 phytoseiid mites, resembling *N. californicus*; slides were sent to Dr. G. de Moraes to confirm the identifications.

Effect of neem, *Azadirachta indica* A. Juss, on mites

This study has been carried out aimed at developing an alternative method for the cassava green mite control. Preliminary results showed a decrease on survival of *T. urticae* fed on

jack bean leaf discs previously immersed in neem extract (Table 5.1.3). On the other hand, *N. californicus* survival ability was not seriously affected by neem extract (Table 5.1.4). Although preliminary, these results are very promising. Studies on the effect of neem *M. tanajoa* are under way at the Entomology Laboratory, EMBRAPA/CNPMPF.

Table 5.1.3. Survival ability of *Tetranychus urticae* on jack bean leaf discs immersed in neem aqueous extract.

Treatment	Survival ability (%)	
	24 h	48 h
Neem treated leaf disc/immediately after immersion	55	18
Neem treated leaf disc/1 h after immersion	28	24
Distilled water treated leaf disc/immediately	90	86
Distilled water treated leaf disc/1 h after immersion	90	86

Table 5.1.4. Survival ability of *Neoseuilus californicus* on eggs of *Tetranychus urticae*/ jack bean leaf discs immersed in neem aqueous extract.

Treatment	Survival ability (%)	
	24 h	48 h
Neem treated leaf disc + <i>T. urticae</i> eggs	100	70
Neem treated leaf discs + <i>T. urticae</i> eggs (1 h later)	80	60
Distilled water treated leaf disc + <i>T. urticae</i> eggs	100	80
Distilled water treated leaf disc + <i>T. urticae</i> eggs (1 h later)	100	90
Neem treated leaf discs	70	80
Neem treated leaf discs (1 h later)	80	80
Distilled water treated leaf disc	100	100
Distilled water treated leaf disc (1 h later)	100	100

***In vitro* selection and production of *Neozygites* sp. by Italo Dellalibera Jr.**

Studies were carried out aiming at selecting the most efficient isolates of *Neozygites* sp. for controlling CGM in semiarid zones of Northeast Brazil, as well as for sending to Africa. The bank of *Neozygites* sp. isolates contains currently 21 isolates from several regions; however nine of them lost their viability, i.e. they do not sporulate any more. Since the *in vitro* maintenance of this pathogen is not yet possible and there is very little information on

its storage, all the isolates have been multiplied *in vivo* (Table 5.1.5). Some isolates have been stored for up to 20 months and remain still viable (sporulating). The isolates from the municipalities of Ipirá, Cruz das Almas, and São Miguel das Matas, State of Bahia, stored for 31, 28 and 28 months, respectively, are still viable, while those from Piritiba and Senhor do Bonfim, State of Bahia, and Caruaru and Exu, State of Pernambuco, lost their viability after storing for 29, 20, 6, and 20 months, respectively. Although the cause of losing the viability of some isolates is unknown, it seems that the quality of the collected mummy, and the period of time that the infected mite remains in the field before being collected may constitute determinant factors on the storage period.

Table 5.1.5. *Neozygites* sp. isolates held at CNPMF.

Number	Isolate	Place of collection	Collection date	Last multiplication
1	BIN01	Piritiba, Bahia	May/11/1993	Mar/30/1996
2	BIN04	Ipirá, Bahia	Aug/27/1993	Mar/30/1996
3	BIN08	Cruz das Almas, Bahia	Nov/05/1993	Mar/31/1996
4	BIN09	São Miguel das Matas, Bahia	Nov/22/1993	Apr/03/1996
5	BIN10	Tianguá, Ceará	Nov/11/1994	Apr/14/1995
6	BIN11	Rio Real, Bahia	Mar/14/1996	Apr/01/1996
7	BIN12	Cristinápolis, Sergipe	Jun/14/1994	Feb/15/1996
8	BIN13	Porto Real do Colégio, Alagoas	Jun/14/1994	Apr/01/1996
9	BIN14	Teotônio Vilela, Alagoas	Jun/14/1994	Apr/01/1996
10	BIN15	Vitória de Santo Antão, Pernambuco	Jun/15/1994	Apr/01/1996
11	BIN16	Gravatá, Pernambuco	Jun/15/1994	Feb/26/1996
12	BIN18	Caruaru, Pernambuco	Jun/15/1994	Apr/16/1995
13	BIN19	Croatá, Ceará	Jun/18/1994	Feb/15/1996
14	BIN20	Itapagé, Ceará	Jun/18/1994	Mar/25/1996
15	BIN21	Floriano, Piauí	Jun/22/1994	Mar/26/1996
16	BIN22	Araripina, Ceará	Jun/23/1994	Mar/23/1996
17	BIN25	Itaberaba, Bahia	Jun/29/1994	Apr/02/1996
18	BIN26	Lambari, Minas Gerais		Apr/03/1996
19	BIN28	Mandiocaba, Paraná	Oct/23/1995	
20	BIN29	Jaguariúna, São Paulo*	Mar/11/1996	
21	BIN30	Cali, Colombia*		

*Isolate from *T. urticae*.

The conidial production of six isolates multiplied *in vivo*, under laboratory conditions, in May, 1995 and in March, 1996 was evaluated in November, 1996. The results showed that the isolates 20, 22 and 23 produced a higher number of conidia when stored up to 8 months than when the storage period was 18 months. On the other hand, no effect of storage period was observed in isolates 4, 15, and 21 (Table 5.1.6).

Table 5.1.6. Conidial production of six *Neozygites* sp. isolates, stored for eight and eighteen months at 4°C/5% RH (total number of conidia produced per single mummified *M. tanajoa*, after 24 hours under dark conditions and 100% RH).

Isolate	Total conidia produced per CGM mummy ¹	
	8 month storage period	18 month storage period
4	569±95 a	417±210 a
15	356±70.9 a	430±261 a
20	1284±231 a	132±229 b
21	393±195 a	197±319 a
22	748±133 a	0±0 b
25	963±191 a	261±312 b

¹ Values followed the same letter, in the same column, do not differ significantly (P=0.05).

Due to indications of storage effect on *Neozygites* sp. isolates, a study was carried out in order to evaluate several storage conditions on the viability of two isolates of the pathogen, one from Piritiba and the other from Cruz das Almas. Treatments were as follows: -10°C, 4°C/5% relative humidity (RH), 4°C/21% RH, 4°C/37% RH, 25°C/50% RH. Relative humidity levels of 5%, 21% and 50%, were obtained by using glycerol solutions, while 37% correspond to the RH inside a refrigerator. As shown in Figure 5.1.1, both isolates lost their viability after one month of storage period at 25°C/50% RH. The isolate from Piritiba showed no significant difference related to sporulation after eight month storage period except for 4°C/21% RH storage conditions. On the other hand, the isolate from Cruz das Almas showed no effect of storage period on sporulation when stored under -10°C. It is interesting to call the attention for the fact that there was no decrease in sporulation of both isolates, stored under -10°C, after the eight-month storage period, thus indicating that

freezing mummyfied mites seems to be a very good storage condition, while under 25°C/50% RH the pathogen viability decreased rapidly. This is a very interesting observation since 25°C and 50% relative humidity are not uncommon environmental conditions in cassava growing areas of Northeast Brazil.

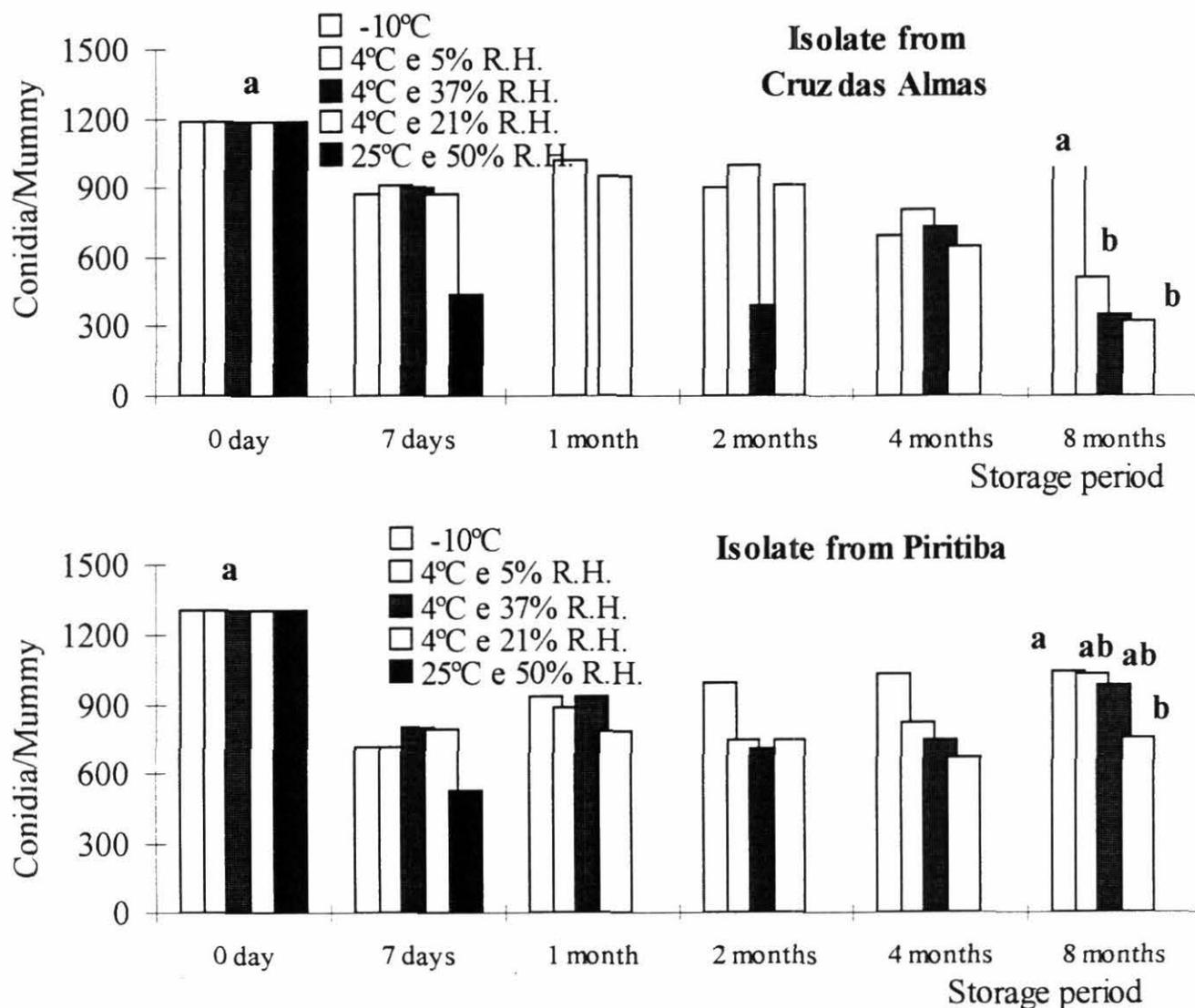


Figure 5.1.1. Viability of two *Necyphites* sp. isolates stored under several conditions.

Phenotypic characterization of *Neozygites* sp. isolates

Since no information is available in the literature regarding to phenotypic characterization of *Neozygites* sp., basic research was conducted aiming at developing the methodology necessary to carry out that activity. Studies were initially directed to determine the effect of relative humidity on sporulation, and length of time of conidial production of *Neozygites* sp. isolates. It was observed that the isolates from Cruz das Almas and Piritiba do not sporulate relative humidity lower than 95% at 24°C (Figure 5.1.2). Consequently, 95% RH was chosen to be used in further studies of selecting *Neozygites* sp. isolates.

Exploratory experiments, regarding the selection of *Neozygites* sp. isolates, consisted in evaluating conidial production of 12 isolates at 24°C and 100% RH, to be later compared with results obtained at 24°C and 95% or less RH. Although significant differences were detected among isolates, a high variation in conidial production by mummies of the same isolate was also observed, as evidenced by the standard deviation (Table 5.1.7). Such a variation may be due to the fact that the mummified mites had been stored for 18 months before using in the experiment. Isolates stored for shorter period of time will be used in further studies of selecting *Neozygites* sp. isolates.

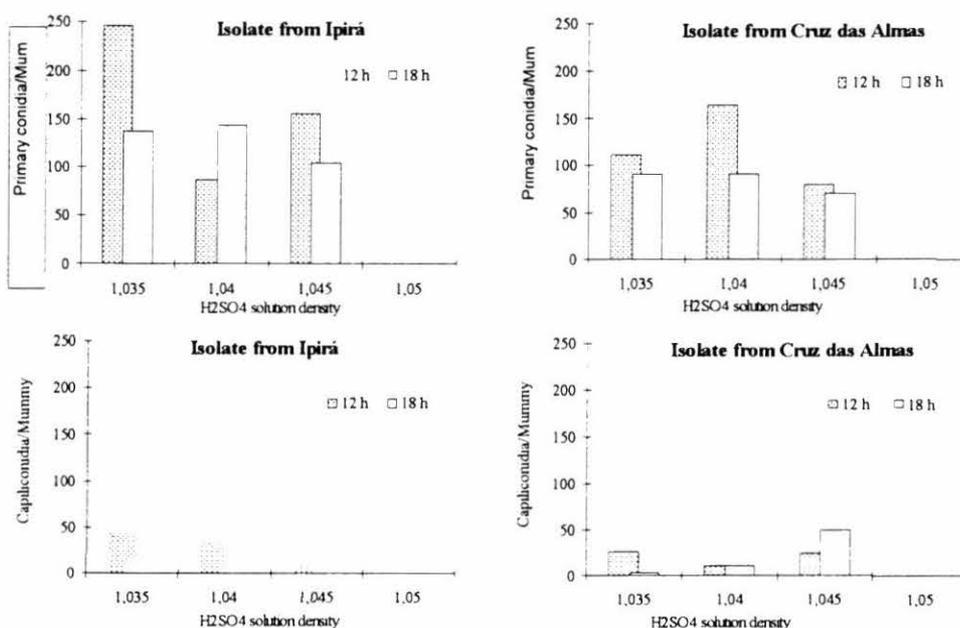


Figure 5.1.2. Effect of humidity levels on conidial production of two *Neozygites* sp. isolates.

Table 5.1.7. Conidial production of 12 *Neozygites* sp. isolates kept under 24°C and 100% RH¹.

Isolate	6 hours		9 hours		12 hours	
	Primary conidia	Capilliconidia	Primary conidia	Capilliconidia	Primary conidia ¹	Capilliconidia
18	0.0±0.0	0.0±0.0	9.4±20.3	0.0±0.0	1.5±4.7 c	0.0±0.0
22	2.5±7.9	0.0±0.0	23.1±57.4	0.0±0.0	1.8±3.9 c	0.0±0.0
04	0.0±0.0	0.0±0.0	49.7±136.2	0.0±0.0	13.6±22.7 bc	0.0±0.0
10	0.6±1.9	0.0±0.0	58.4±121.6	0.0±0.0	14.8±31.3 bc	0.0±0.0
01	47.9±149	0.0±0.0	90.8±129.3	0.0±0.0	168.5±226.8ab	0.0±0.0
08	10.3±31.2	0.0±0.0	57.1±83.5	0.0±0.0	203.6±205.7a	2.4±7.6
11	0.0±0.0	0.0±0.0	3.0±9.5	0.0±0.0	39.6±102.6 bc	0.0±0.0
21	0.0±0.0	0.0±0.0	25.0±55.8	0.0±0.0	5.0±15.8 bc	0.0±0.0
15	0.0±0.0	0.0±0.0	51.3±108.5	0.0±0.0	52.1±113.4abc	0.0±0.0
20	1.0±3.2	0.0±0.0	35.4±57.6	0.0±0.0	64.9±73.8abc	3.8±12.0
25	0.0±0.0	0.0±0.0	65.1±157.1	0.0±0.0	72.6±112.6abc	0.0±0.0
13	0.0±0.0	0.0±0.0	45.6±82.5	0.0±0.0	76.8±94.2abc	0.0±0.0

¹ Values followed by the same letter, in the same column, do not differ significantly (P=0.05).

Another parameter used in studies of phenotypic characterization of *Neozygites* sp. isolates was the pathogen specificity. Nineteen isolates were evaluated for pathogenicity to *M. tanajoa* and *T. urticae*. The results showed that the isolates from *T. urticae*, collected in Cali, Colombia, and in Jaguariuna, State of São Paulo, Brazil, were pathogenic to both *T. urticae* and *M. tanajoa*; on the other hand, the isolates from *M. tanajoa*, collected in Brazil, were pathogenic only to *M. tanajoa*, thus showing a very high specificity (Table 5.1.8).

Table 5.1.8. Specificity of *Neozygites* sp. isolates to *Mononychellus tanajoa* and *Tetranychus urticae*.

Isolate collecting place	Infection	
	<i>M. tanajoa</i>	<i>T. urticae</i>
Cali, Colombia*	+	+
Caruarú, Pernambuco	+	-
Cristinápolis, Sergipe	+	-
Croatá, Ceará	+	-
Cruz das Almas, Bahia	+	-
Gravatá, Pernambuco	+	-
Ipirá, Bahia	+	-
Itaberaba, Bahia	+	-
Itapajé, Ceará	+	-
Jaguariúna, São Paulo*	+	+
Lambari, Minas Gerais	+	-
Mandiocaba, Paraná	+	-
Piritiba, Bahia	+	-
P. Real do Colégio, Alagoas	+	-
Rio Real, Bahia	+	-
São M. das Matas, Bahia	+	-
Teotônio Vilela, Alagoas	+	-
Tianguá, Ceará	+	-
Vit. Sto. Antônio, Pernambuco	+	-

* Isolate from *T. urticae*

***Neozygites* sp. inoculation under field conditions**

An experiment was installed in May, 1996 in Piritiba, Bahia, with the objective of evaluating the efficiency of field inoculation of *Neozygites* sp. to control CGM. Ten inoculations were performed from May 22 to June 24, using 40 mummyfied mites as source of inoculum. Dispersion was evaluated by both visual detection of infected mites and by using spore traps; final evaluation was carried out early August. Very low levels of *Neozygites* sp. infected mites were observed from May to August. The pathogen was found in five evaluations in the inoculated area and only once in the control (Figure 5.1.3). The first infected mites were observed by July 14 in the control field while in the inoculated area the pathogen was detected by July 17, that is 23 days after the last field inoculation (Figure

5.1.3). It is important to call attention to the fact that the CGM population was low, around 25 individuals/leaf, over all the evaluation period. Infected mites were found up to 45 m away from the inoculation site, and by August 2, the pathogen had spread all over the field. These results do not allow ruling out the efficiency of field inoculations with *Neozygites* sp. to control CGM. The number of mummified mites used as source of inoculum seems to be low. Methodological adjustments will be made in order to overcome this problem in further experiments.

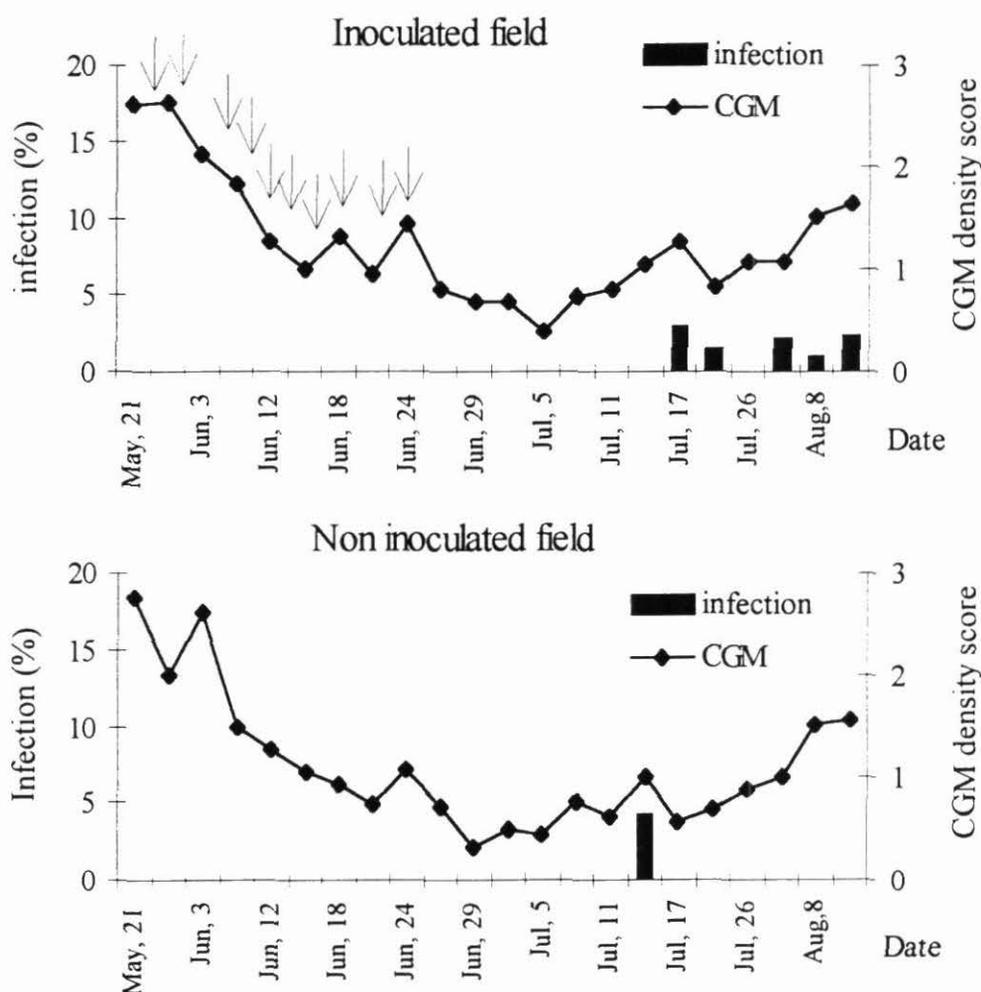


Figure 5.1.3. Population dynamic of *Neozygites* sp. and *Mononychellus tanajoa* in two cassava fields located in Piritiba, Bahia (↓ Indicate inoculation date). CGM density score 1= <25, 2= >25<200 and 3= >200 mites/leaf.

5.2 BIOLOGICAL CONTROL OF THE CASSAVA MEALYBUG (CM), by José Mauricio Simões Bento

Mass rearing and releasing parasitoids of the cassava mealybug

Apoanagyrus (= *Epidiniocarsis*) *diversicornis*, *Acerophagus coccois* and *Aenasius vexans*, all parasitoids of the cassava mealybug, *Phenacoccus herreni*, were introduced from CIAT, through the "Costa Lima" Quarantine Laboratory, EMBRAPA/CNPMA, to EMBRAPA/CNPMF where those parasitoids were mass-reared. A total of 111,610 parasitoids were produced, 35,950 of which were released in three cassava fields in the State of Bahia and three cassava fields in the State of Pernambuco, showing high level of incidence of the cassava mealybug (Table 5.2.1).

Table 5.2.1. Number of cassava mealybug parasitoids produced under laboratory conditions, at EMBRAPA/CNPMF, and released in cassava fields located in the States of Bahia and Pernambuco.

Species	1994			1995			1996		
	Released			Released			Released		
	Produced	BA	PE	Produced	BA	PE	Produced	BA	PE
<i>E. diversicornis</i>	6,190	3,010	310	5,235	3,720	-	5,901	640	-
<i>A. coccois</i>	9,950	1,950	-	11,566	500	750	53,229	11,100	4,050
<i>A. vexans</i>	-	-	-	9,043	2,980	2,000	10,496	3,020	1,900
Total	16,140	4,960	310	25,844	7,200	2,750	69,626	14,760	5,950

Parasitoid dispersion

A. diversicornis showed higher dispersion ability than *A. coccois* and *A. vexans*, reaching 130 km from the releasing site by January 1995, 234 km by September 1995, and 304 km by April/1996. *A. coccois* dispersed slowly, reaching 180 km from the releasing site by September/1995, while *A. vexans* has not dispersed in the State of Bahia as of November 1996 despite the large number of releases already performed. Parasitoid releases of *A. coccois* and *A. vexans* started in the State of Pernambuco in August 1995, and by November 1996 a total of 9,010 individuals had been released. Individuals have already been recaptured 40 km away from the release site.

Biological control of *P. herreni*

Paratoid monitoring has been carried out every two weeks, since the first release back in 1994. Results show a significant decrease on number of CM per plant due to parasitoid action. A positive correlation between number of parasitoids and CM control has also been observed for both *A. diversicornis* and *A. coccois* in the State of Bahia (Figure 5.2.1). Field observations have demonstrated that *A. diversicornis* has high dispersal and survival abilities, but it is less aggressive than *A. coccois* which demonstrates a very high ability to control the cassava mealybug, especially when at high populations. Such a situation has been observed in Itaberaba, State of Bahia, where *A. coccois*, probably because of its ability to attack early stages (instars) of the pest, has shown a better biocontrol effect than *A. diversicornis*.

Biological and economical effects of exotic parasitoids

Studies on biological and economical effects of introduced parasitoids for the cassava mealybug control started by October, 1996. Data have been collected every two weeks and results will be presented in the 1997 Annual Report.

Hyperparasitism

After releasing the exotic parasitoids, *A. diversicornis*, *A. coccois*, and *A. vexans*, a significant increase of hyperparasitoid populations, mainly *Pachyneuron* sp. and *Prochiloneurus* sp, was observed. These hyperparasitoids have been able to reduce the parasitoid populations when they reach high levels under field conditions.

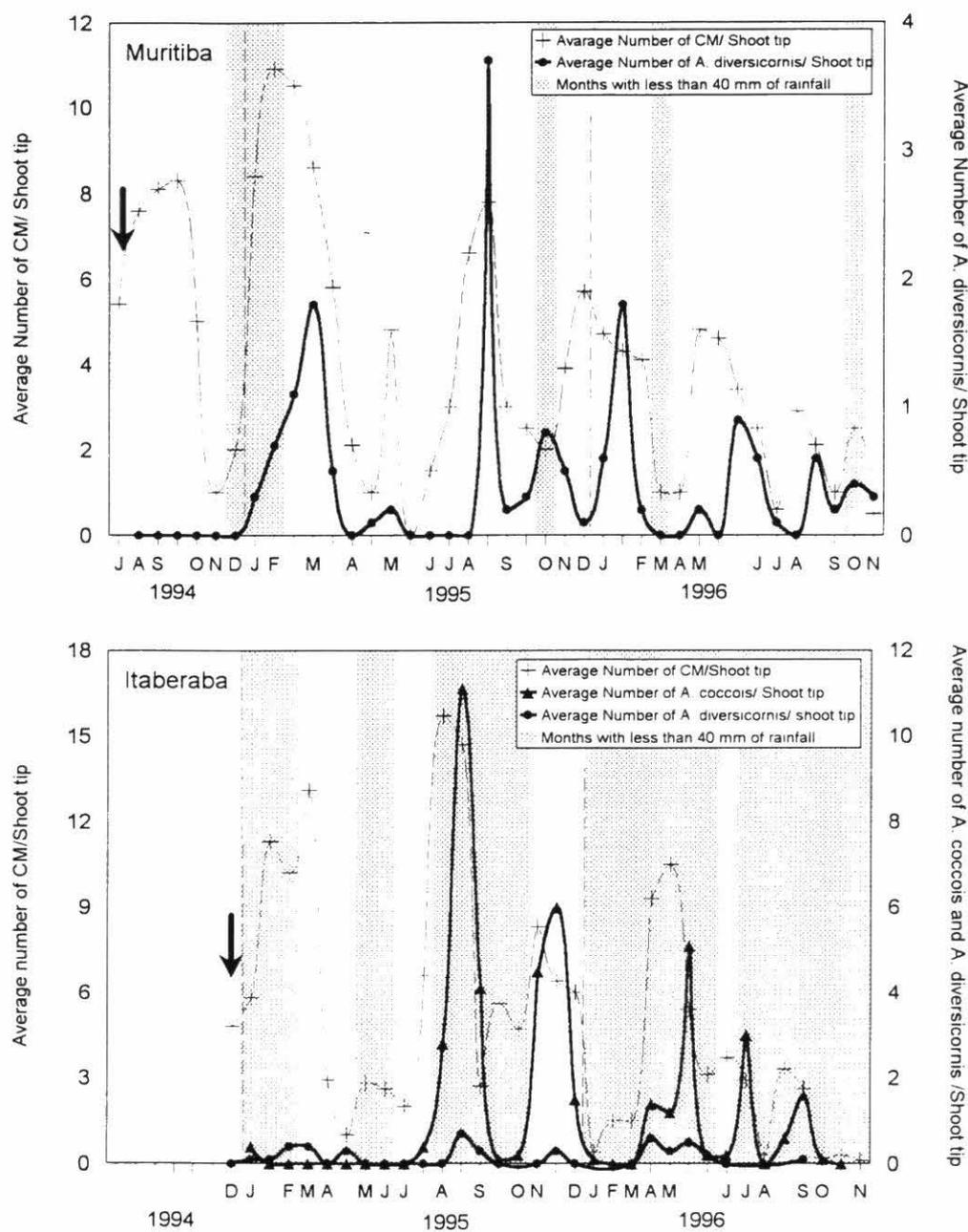


Figure 5.2.1. Population dynamics of cassava mealybug (CM) *Phenacoccus herreni* (second to fourth instars) in cassava fields where *Apoanagyrus* (= *Epidinocarsis*) *diversicornis* and *Acerophagus coccois* were released from 1994 to 1996 (Arrow indicates first release).

5.3 CASSAVA CROP MANAGEMENT, by José Eduardo Borges de Carvalho

Effect of weeds on cassava yield

Weed competition in the cassava crop may cause yield reduction as high as 40%, for both roots and aerial plant parts, if weeding is not performed at the right time during the crop cycle, mainly in ecosystems where the water availability is deficient in the soil. Weeds also act as source of food and shelter for arthropods, either pest or beneficial organisms, as shown in Table 5.3.1 and in previous PROFISMA's Annual Reports.

Table 5.3.1. Relative distribution of various classes of arthropods on weeds commonly found in cassava fields in the municipalities of Itaberaba and Piritiba, State of Bahia.

Weed species	Cassava pests (%)	Other crops pests (%)	Natural Enemies (%)	Other Insects (%)
<i>Blainvillea rhomboidea</i>	-	55.5	33.3	11.2
<i>Acanthospermum hispidum</i>	14.3	57.1	28.6	-
<i>Diodia teres</i>	22.2	55.5	22.3	-
<i>Croton lobatus</i>	25.0	25.0	25.0	25.0
<i>Croton gladius</i>	16.7	33.3	16.7	33.3
<i>Passiflora cincinnata</i>	16.7	33.3	33.3	16.7
<i>Mitracarpus hirtus</i>	16.7	50.0	33.3	-
<i>Polygata violacea</i>	20.0	60.0	20.0	-
<i>Borreira sp.</i>	-	57.1	20.6	14.3
<i>Eupatorium ballataefolium</i>	-	40.0	-	60.0
<i>Wissandula subpeltata</i>	-	57.1	14.3	28.6
<i>Cassia rotundifolia</i>	16.7	33.3	33.3	16.7
<i>Setaria vulpiseta</i>	14.3	57.1	28.6	-
<i>Acanthospermum australe</i>	25.0	50.0	25.0	-
<i>Sida cordifolia</i>	33.3	33.3	-	33.4
<i>Eupatorium laevigatum</i>	42.8	28.6	-	28.6
<i>Solanum erianthum</i>	62.5	12.5	-	25.0
Average	21.5	33.0	22.6	22.9

Studies on weed effects on cassava yield, carried out in past years in Itaberaba and Piritiba municipalities, State of Bahia, showed that in Itaberaba the cassava root yield was very low, and consequently no significant difference for treatment was observed. In the

experiment installed in Piritiba, the control treatment (without weed control) produced only 3.35 metric tons/ha, a root production significantly lower than the other treatments evaluated in that region (Table 5.3.2). Although still low, root yield in Piritiba was higher than in Itaberaba. Such a low root yield, and no significance for treatment in the experiment installed in Itaberaba, seems to be due to a very drastic decrease in rainfall, starting from September, similar to previous observations (PROFISMA's Annual Report 1995). Besides the low rainfall, an unpredicted hornworm attack may have also affected the cassava root yield in the experiment installed in Itaberaba. The results from the effect of weeds on cassava yield give a strong indication that weeding can start 30 days after cassava germination and continue up to five months.

Table 5.3.2. Joint analyses of cassava root yield as affected by weed competition (1995/1996 growing seasons).

Treatments	Yield (t/ha) ¹	
	Itaberaba	Piritiba
Weed control from germination		
For 90 days	2.09 a	8.42 a
For 120 days	2.82 a	8.68 a
For 150 days	3.41 a	9.16 a
For 180 days	3.02 a	8.39 a
For 210 days	2.96 a	7.60 a
Weed control beginning 30 days after germination		
For 60 days	2.54 a	7.91 a
For 90 days	2.72 a	9.53 a
For 120 days	2.43 a	8.33 a
For 150 days	3.06 a	8.67 a
For 180 days	2.36 a	8.44 a
Weed control throughout crop cycle	2.71 a	8.22 a
Without weed control	0.70 a	3.35 b

¹ Values followed by the same letter in the same column, do not differ significantly (P=0.05), according to Tukey's test.

Effect of cover crops on cassava yield, arthropod population and erosion control

Studies have been carried out aiming at evaluating the effect of cover crops on soil fertility/yield, erosion and arthropod populations in the ecosystems of Cruz das Almas,

Itaberaba and Piritiba. It is well known that, due to its initial slow growth, a large portion of a cassava field is exposed to the impact of rain which causes soil loss by surface run-off. Regarding erosion control, a very low soil loss resulted from planting either jack bean or cowpea between cassava double rows (PROFISMA's Annual Report 1995). Another trial was planted in the same area during the 1996 growing season in order to continue this type of evaluation. Due to a very drastic drought in Piritiba, no soil loss by surface run-off was recorded. Raining resumed by late December 1996, and results will be presented in the 1997 Annual Report.

Table 5.3.3 shows data related to the effect of cover crop on arthropod populations in cassava field. Results show that cover crops, either legume or weeds, act as source of food and shelter for both natural enemies and pests. The high amount of natural enemies found on weeds (22.6%) suggests the importance of weed management on setting up biocontrol strategies for cassava pests.

Table 5.3.3. Frequency of arthropods on cover crops in cassava fields in the municipalities of Cruz das Almas, Itaberaba, and Piritiba, State of Bahia.

Arthropods	Frequency (%)	
	Legume	Weeds
Cassava pests	16.0	21.5
Other crop pests	75.0	33.0
Natural enemies	9.0	22.6
Others	0.0	22.9
Total	100.0	100.0

Regarding to root yield as affected by cover crops, the joint analyses of the 1995-1996 growing season presented in Table 5.3.4 show that weed control over entire crop enabled the highest root yield in Cruz das Almas, and in Piritiba. On the other hand, weed control within double rows/weed between double rows, resulted in the lowest yields in both ecosystems, probably due to competition for water during period of low water availability in the soil. No significant decrease on cassava root yield by neither pigeon pea (*Cajanus cajan*), jack bean nor cowpea was observed in Piritiba. In Cruz das Almas, jack bean depressed cassava root

yield, but this behavior seems to be due to other problems that caused yield reduction in replicate two rather than competition by the cover crop. Although showing the highest root yield, weed control over entire crop raised production costs over 30% as compared with treatment four (weed control within double rows/jack bean between double rows).

Table 5.3.4. Joint analysis of cassava root yield for the 1995/1996 growing seasons, in Piritiba and Cruz das Almas.

Treatments	Root yield (ton/ha) ¹	
	Cruz das Almas	Piritiba
Weed control over entire crop	9.93 a	8.10 a
Weed control within double rows; weeds between double rows	2.22 c	3.54 b
Weed control within double rows; pigeon pea between double rows	7.27 ab	6.04 ab
Weed control within double rows; jack bean between double rows	3.97 bc	6.24 ab
Weed control within double rows; cowpea between double rows	6.10 abc	7.51 a
Standard error (%)	47.44	29.61

¹ Values followed by the same letter, in the same column, do not differ significantly (P=0.05), according to Tukey's test.

During the 1995/1996 growing season a very drastic drought occurred in Itaberaba and, due to that, the cover crop experiment installed in that ecosystem was completely lost.

Effect of bagana mulching on cassava yield and weed control.

By Genário Marcolino de Queiroz

This study aims at determining the lowest amount of bagana/ha able to increase significantly the cassava root yield, to maintain soil moisture, and to reduce weeding frequency over the cycle of the cassava crop. In order to reach these objectives, experiments have been carried out in the cassava growing areas of Acaraú and Russas, State of Ceará. On farm trials installed in Acaraú showed that adding bagana at 180 m³/ha, or more, increased significantly the cassava root yield (25.05 ton/ha) in comparison with the

treatments with bagana at 45 m³/ha (10.49 ton/ha) and without bagana (8.11) (Table 5.3.5). Regarding weed incidence in the cassava field with bagana treatment, a predominance of weeds was able to sprout out of the roots, such as “Catanduba” (around 90%), “Cipauba”, “Catingueira” and “Cipó-pau” (about 10%).

Table 5.3.5. Effect of the bagana mulching on the cassava root yield, aerial part and starch content at Acaraú, State of Ceará.

Bagana levels (m ³ /ha)	Yield (ton/ha) ¹		Starch content (%)
	Root	Aerial part	
315	28.98 a	37.10 a	24.76 ab
270	23.52 a	30.46 ab	25.34 a
225	22.12 ab	26.82 abc	24.99 ab
180	25.05 a	32.73 ab	25.05 ab
135	18.64 abc	21.18 bcd	25.34 a
90	17.14 abc	14.70 cd	22.82 b
45	10.49 bc	10.89 d	23.99 ab
0	8.11 c	8.41 d	24.89 ab

¹ Values followed by the same letter, in the same column, do not differ significantly (P=0.05), according to Tukey's test.

The results obtained from the on farm trial installed in Russas, in February 1995, showed no effect of bagana mulching on the cassava root yield (Table 5.3.6). This might be due to the rainfall that occurred from January to July 1995, corresponding to the period of initial growth of the crop (Table 5.3.7), or to soil characteristics as well. It is interesting to mention that previous experiments installed in Russas, using bagana as mulching for cassava fields, were lost due to drought and/or high soil temperatures (previous PROFISMA Annual Reports).

Table 5.3.6. Effect of bagana mulching on the cassava root yield, aerial part and starch content at Russas, State of Ceará.

Bagana levels m ³ /ha	Cassava yield (ton/ha)		Starch content (%)
	Root	Aerial part	
315	11.46	23.47	24.34
270	10.93	24.40	25.46
225	9.98	21.20	25.05
180	13.45	27.62	24.92
135	13.85	27.69	26.37
90	12.16	23.97	28.40
45	7.71	23.43	26.43
0	15.53	24.76	24.36

The experiment installed in Acaraú, in February 1996, shows normal development; on the other hand the experiment in Russas presented problems related to germination probably due to high soil temperature in the bagana covered plots. Both trials will be harvested by July August, 1997.

Table 5.3.7. Rainfall (mm) at Russas, State of Ceará.

Month	Year			
	1993	1994	1995	1996
January	45.2	75.9	45.6	74.2
February	0.0	79.4	104.7	147.4
March	76.0	160.1	129.0	250.0
April	34.6	129.6	329.2	190.3
May	4.4	127.4	213.4	48.6
June	0.0	115.1	45.6	0.0
July	13.2	24.0	38.6	25.6
August	0.0	0.0	0.0	2.0
September	0.0	0.0	0.0	
October	0.0	0.0	0.0	
November	0.0	0.0	0.0	
December	0.0	41.0	0.0	
Total	173.4	752.5	906.1	738.1

5.4 PLANT PATHOLOGY, by Chigeru Fukuda

5.4.1 Cassava Witches' Broom Disease

Cultural Control of Cassava Witches' Broom Disease

The cassava witches' broom disease (CWBD), caused by a phytoplasma, is a very serious cassava problem in the "Serra da Ibiapaba", State of Ceará, growing area. An important achievement on the research of CWBD was the observation that "cleaning-up" (elimination of infection) infected planting material of susceptible varieties enabled high root yield. Despite the effectiveness of the cleaning-up technique, it is only a matter of time for the disease-free susceptible planting material to become re-infected by the CWBD pathogen under field conditions, thus requiring continuous cleaning-up procedures.

Genetic control of CWBD

The study on genetic control of CWBD started in 1986, as part of the EMBRAPA/CNPMPF Cassava Breeding Program, aiming at identifying cassava genotypes resistant/tolerant to that disease. By 1993, this study began to be an EMBRAPA/CNPMPF Cassava Breeding Program/PROFISMA joint activity. From 1986 to 1993 eight hundred and seventy cassava clones from the EMBRAPA/CNPMPF Cassava Breeding Program were tested, through farmer participatory methods, in the Serra da Ibiapaba growing area, where there are about 5,000 ha planted with cassava, most of them planted with the cassava variety Cruvela, highly susceptible to the CWBD pathogen. After several years of evaluation, seven promising clones were selected. In 1994 these seven clones were then evaluated in comparison with the local variety Cruvela, susceptible to the pathogen, and the CWBD resistant variety Bujá, was recommended by the State of Ceará Agricultural Research Institution (EPACE). On-farm trials were installed in 60 rural communities, located in seven municipalities in the Serra de Ibiapaba cassava growing region. Evaluation at harvest time, carried out by cassava growers in 53 rural communities enabled the selection of the clones 8709-2 and 8911-16 for harvesting at both 12 and 18 months after planting, clone 8952-06 only for harvesting at 18 months and clone 8740-10 for harvesting at 12 months after planting.

Results of a simplified phytosanitary diagnostic survey, carried out during 1994 in the Serra da Ibiapaba cassava growing region, showed that one of the most important factors involved in the CWBD spread throughout the region is the sharing of planting material, a very common practice among cassava growers. Such a behavior gives an indication that the selected CWBD resistant clones would be dispersed within rural communities by sharing planting material. A very strong support to this assumption is the observation that, the stem of several selected clones had already been removed by cassava growers prior to harvest and, according to them, used for either replacing their own field or donating to other cassava growers.

It is interesting to mention that growers' preferences for the evaluated cassava clones/varieties were $\geq 60\%$ for the clones 8709-2, 8952-6 and 8911-16 as compared with 15% to 35% for the local variety Cruvela. Increase in adoption level of the selected clones is expected through action of the EMATERCE by diffusion methodologies. Considering 60% adoption of the CWBD resistant clones, this would imply in a replacement of about 3,000 ha of the susceptible variety Cruvela. Considering the root yield potential of the selected clones, around 20 metric ton/ha, such replacement would represent a 60,000 metric ton yield. Considering the average price of USD \$41.80/cassava root metric ton, this would give an estimated USD \$2,508,000.00 production value increase.

5.4.2 Root Rots, by Chigeru Fukuda and Aristoteles Pires de Matos

Cassava root rot disease survey in the State of Bahia

A survey for the cassava root rot incidence was conducted in three cassava growing regions of the State of Bahia. Evaluations, performed in nine rural communities of five municipalities, showed that the disease incidence varies from region to region. The highest levels of root rot incidence, 50%, were observed in the rural community Saco, municipality of Cachoeira, and in a cassava field located in the municipality of Irará. In the rural community Chapada, municipality of Aporá, the disease incidence was 29%. Very low root rot incidence, 1%, was observed in the rural community Buri, municipality of Crisópolis. Despite growers information on the occurrence of the cassava root rot disease in the rural community Umbuzeiro, only one infected plant was found. This is probably due to the

prevalent dry season during the survey. Isolations carried out under laboratory conditions showed that the pathogens involved in the cassava root rot development in the evaluated rural communities in the State of Bahia were *Fusarium* sp., *Phytophthora* sp., or *Scytalidium lignicola*.

Genetic control of the cassava root rot

Field evaluation of 30 cassava hybrids, from the EMBRAPA/CNPMF Cassava Breeding Program, in the municipality of Umbauba, State of Sergipe, enabled the selection of five of them that were evaluated again in an on-farm trial in a randomized block design, with three replicates. Data in Table 5.4.2.1 show that hybrids 90.175-7 and 90.117-5 expressed 11.54% of root rot infection, 90.185-1 showed 14.29% of infection while the hybrids 90.113-2 and 90.115-5 showed over 30% of root rot infection. Besides showing field tolerance to root rot disease, the hybrid 90.175-7 showed root weight of 1.12 kg/plant, a little lower than the susceptible hybrid 90.113-2, which yielded 1.24 kg of root/plant. All the other hybrids produced root weights varying from 0.58 to 0.94 kg/plant.

Table 5.4.2.1. Reaction of cassava hybrids to natural infection of root rot pathogens, 12 months after planting, in Umbauba, State of Sergipe.

Hybrid	Infected plant (%)	Total root weight (kg)	Average root weight (kg/plant)
90.113-2	31.03	26	1.24
90.115-5	39.29	17	0.94
90.117-5	11.54	14	0.58
90.175-7	11.54	28	1.12
90.185-1	14.29	20	0.80

Eighteen cassava varieties/hybrids were evaluated for root rot incidence in a on farm trial carried out at the COPAL Quitéria, Municipality of Alagoa Grande, State of Paraíba, in an area naturally infested by root rot. As shown in Table 5.4.2.2 the hybrid 148-2 expressed field tolerant reaction to root rot pathogen with 9.5% of infected plants and only 2% of infected roots. Moderate infection was observed in the hybrids 46-03 and 128-8, respectively 23.8% and 38.1%, while all the other evaluated genotypes showed high root rot infection,

including the local varieties. The hybrid 148-2, besides expressing tolerant reaction to the cassava root rot, also showed a good root yield, 2.6 kg/plant, higher than all the other evaluated varieties/hybrids.

Table 5.4.2.2. On-farm evaluation of 18 cassava genotypes for root rot incidence, in the rural community “Engenho Mares”, municipality of Alagoa Grande, State of Paraíba.

Genotypes	Infected root (%)	Infected plant (%)	Total root weight (kg)	Average root weight (kg/plant)
179-4	51.0	100.0	12.6	0.60
32-06	89.0	100.0	1.2	0.06
A. Bravo Branco	67.0	100.0	11.7	0.56
Bujá	100.0	100.0	0.0	0.00
M. México.59	100.0	100.0	0.0	0.00
Maria Pau	62.0	100.0	6.3	0.30
Monge*	100.0	100.0	0.0	0.00
Passarinha	100.0	100.0	0.0	0.00
Osso Duro*	83.3	90.5	4.4	0.21
Isabel de Souza	56.0	80.9	21.0	1.00
Olho Verde	29.2	80.9	10.0	0.48
Fio de Ouro	11.0	61.9	26.0	1.24
Paraibana*	50.0	61.9	9.6	0.46
128-8	22.0	38.1	29.8	1.42
100-08	55.0	23.8	17.6	0.84
46/03	20.0	23.8	21.8	1.04
Cedinha*	48.0	23.8	21.0	1.00
148-2	2.0	9.5	54.0	2.60

* Local varieties

One hundred and seventy-nine hybrids, from the EMBRAPA/CNPMF Cassava Breeding Program, were evaluated in on-farm trials in the municipality of Viçosa, State of Alagoas, where root rot incidence was relatively high. Thirty hybrids (16.76%) showed no root rot infection and were selected for further evaluations under field conditions while the others expressed several degrees of root rot incidence. High susceptibility to root rot pathogens was expressed by seventy hybrids that showed over 50% of infected roots (Table 5.4.2.3).

The trial installed in the COPAL Chapada was aimed at evaluating the effect of planting systems on the behavior of two cassava varieties regarding root rot incidence. It was observed that disease incidence is lower when cassava stakes are planted in ridges in comparison with planting in holes. The results also show that planting the susceptible local variety *Cemitério* in ridges, despite having both plant and roots infected by root rot pathogens, yielded the highest root weight, 1.122 kg/plant. Such a good behavior of *Cemitério* explains the cassava growers preference for this variety.

Table 5.4.2.3. Field evaluation of cassava hybrids, 1992 family, for resistance to root rot pathogens, in the municipality of Viçosa, State of Alagoas.

Class of root rot incidence	Plants/class	
	Number	Percentage
No infected roots	30	16.76
0.1 to 10%	27	15.08
10.1 to 25%	20	11.17
25.1 to 50%	32	17.88
Over 50%	70	39.11
Total	179	100.00

In 1995 a high incidence of cassava root rot was observed in the rural community Chapada do Aporá, municipality of Aporá, State of Bahia. Two on-farm trials were installed in that rural community aiming at evaluating the behavior of cassava varieties and promising hybrids regarding root rot incidence. As shown in Table 5.4.2.4, despite showing 45% infected plants and 15.2% infected roots, the local variety *Cemitério* had a very good performance regarding to root yield, 1.44 kg/plant, just behind *Paraibana*, that yielded 1.52 kg/plant, while *Olho Verde* showed the lowest root weight, only 0.285 kg/plant. As can be concluded from data in Table 5.4.4, root rot incidence was very low throughout the experiment, except for the local variety *Cemitério*. It is possible that the root rot incidence in *Cemitério* may be due to the use of infected planting material rather than infection from contaminated soil.

Table 5.4.2.4. Reaction of cassava genotypes to root rot pathogens under field conditions in rural community of Chapada do Aporá, municipality of Aporá, State of Bahia.

Genotypes	Infected root (%)	Infected plant (%)	Total root weight (kg)	Average RW (kg/plant)	Starch content (%)
Rosinha	0.0	0.0	22.20	1.11	28.98
128/8	0.0	0.0	14.40	0.78	28.75
Cedinha	0.0	0.0	25.00	1.25	28.41
Paraibana	0.0	0.0	30.40	1.52	26.61
Milagrosa	0.0	0.0	22.70	1.14	30.28
Olho Verde	0.0	0.0	5.70	0.29	26.84
Olho de Porco	0.0	0.0	24.60	1.37	23.68
Cemitério	15.2	45.0	28.80	1.44	

A very low root rot incidence was observed in the trial where a total of 25 cassava hybrids from the EMBRAPA/CNPMP cassava breeding program were under field evaluation (Table 5.4.2.5), despite the high incidence of the disease in the previous growing season. Such an erratic cassava root rot incidence in the same area, from season to season, has been a problem for evaluating/selecting cassava germplasm for root rot resistance under field conditions. Due to that it was decided, parallel with field evaluations, to develop evaluation techniques, under controlled conditions, aiming at identifying cassava root rot resistant/tolerant genotypes.

Table 5.4.2.5. Reaction of the local cassava variety and hybrids, from the EMBRAPA/CNPMF Cassava Breeding Program, to root rot pathogens under field conditions, rural community of Chapada do Aporá, municipality of Aporá, State of Bahia.

Variety/hybrid	Infected root (%)	Infected plant (%)	Average root weight (kg/plant)
076	2.6	5.0	1.060
056	0.0	0.0	0.471
576	0.0	0.0	1.820
537	26.7	31.58	0.505
478	0.0	0.0	1.158
260	5.0	5.26	0.568
896	0.0	0.0	0.978
554	0.0	0.0	1.856
967	0.0	0.0	0.481
9660.0	0.0	0.0	0.775
858	0.0	0.0	0.374
859	0.0	0.0	1.594
1183	0.0	0.0	0.835
1189	0.0	0.0	0.826
538	0.0	0.0	1.100
142	9.5	16.67	1.578
116	0.0	0.0	1.325
812	0.0	0.0	1.620
053	0.0	0.0	0.500
645	0.0	0.0	2.142
613	8.3	6.25	0.694
649	0.0	0.0	0.800
604	0.0	0.0	0.442
387	0.0	0.0	0.450
598	0.0	0.0	0.920
Cemitério	0.0	0.0	2.071

Studies on cassava root rot under controlled conditions

In order to establish a feasible method for evaluating cassava germplasm for resistance to root rot disease caused by *Fusarium* sp. several inoculation techniques were tested under

laboratory and greenhouse conditions. No disease development was observed when healthy planting material was planted in artificially contaminated soil prior to planting. Although showing disease development, the pathogen inoculation in branches proved to be time-consuming since it takes about 60 days from planting to recording data. Stake inoculation, under laboratory conditions, either by adding a conidial suspension to a wound or by transferring a disk of colony to wounded tissue, gave the best response since evaluation may be performed one to two weeks after inoculation.

Several isolates of *Fusarium* sp. were obtained from infected cassava tissue collected in cassava growing areas in Northeast Brazil. Pathogenicity tests carried out with these isolates inoculated in stakes of susceptible varieties showed that, although pathogenic, the virulence of the *Fusarium* sp. isolates was not high. On the other hand, isolates of *Scytalidium lignicola* expressed high virulence, inciting higher lesion development than *Fusarium* sp. (Figure 5.4.2.1). *S. lignicola* was also able to incite disease development even when inoculated at low inoculum concentrations, such as 10^3 conidia/mL (Figure 5.4.2.2).

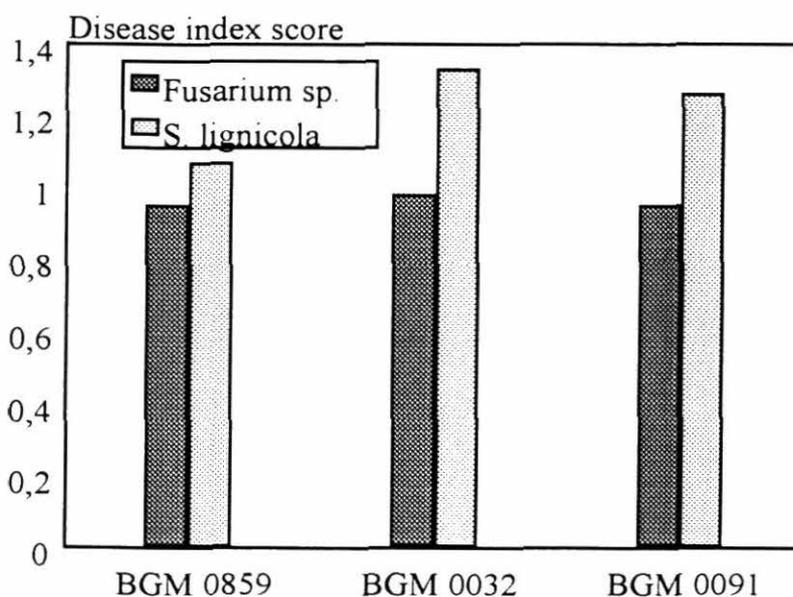


Figure 5.4.2.1. Reaction of three cassava genotypes to stake inoculation with *Fusarium* sp. and *Scytalidium lignicola*.

Preliminary results regarding reaction of 30 cassava genotypes to inoculation with *Fusarium* sp. showed that the average area of lesion, one week after inoculation, was very small, varying from 0.29 cm² up to 1.57cm², while the control variety Correnteza, that shows high susceptibility to root rot pathogens under field conditions, showed only 0.45 cm² average area of lesion, indicating again the low virulence of *Fusarium* sp. to the cassava crop.

Although preliminary, the pathogenicity tests performed up to now seem to indicate that, in Northeast Brazil, *S. lignicola* might be a more important pathogen to the cassava crop than *Fusarium* sp., and *P. drechsleri* seems to be even more virulent than *S. lignicola*. Studying the effect of inoculum concentration on disease development incited by *S. lignicola* in stakes of three cassava genotypes showed that the pathogen was able to cause some infection even at very low concentrations such as 10³ conidia/mL. Increasing inoculum concentration increased lesion development up to 10⁵ conidia/mL. Inoculum concentration of 10⁶ conidia/mL did not significantly increase lesion development (Figure 5.4.2.2).

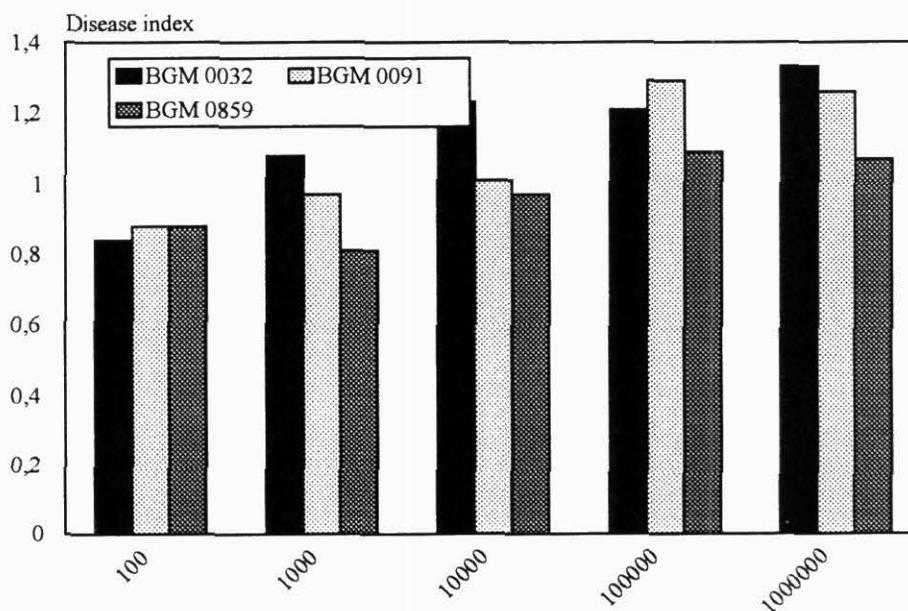


Figure 5.4.2.2. Effect of inoculum concentration of *Scytalidium lignicola*, on disease development of three cassava genotypes.

Biological control of cassava root rot disease

The effect of the antagonistic fungi *Trichoderma* spp. was evaluated on the biocontrol of the cassava root rot caused by either *P. drechsleri* or *S. lignicola*. As shown in Figure 5.4.2.3 the isolate T 11, *Trichoderma polysporum*, when simultaneously inoculated with *P. drechsleri*, was able to antagonize the pathogen, as expressed by the significantly smaller size of the lesion in root of both varieties Maria Pau (BCG 118) and Manteiga (BGM 801). The isolates T 25, *T. harzianum*, and T 26, *T. pseudokoningii*, did not show antagonistic effect to *P. drechsleri* in inoculated cassava roots. Data in Figure 5.4.2.4 show that the isolates T 11, *T. polysporum*, T 25, *T. harzianum*, and T 26, *T. pseudokoningii*, significantly decreased the size of lesion, when simultaneously inoculated with *S. lignicola* in cassava stakes, variety Manteiga, thus indicating antagonistic effect to the pathogen. On the other hand, no antagonistic effect was observed when inoculations were performed in stakes of the cassava variety Maria Pau.

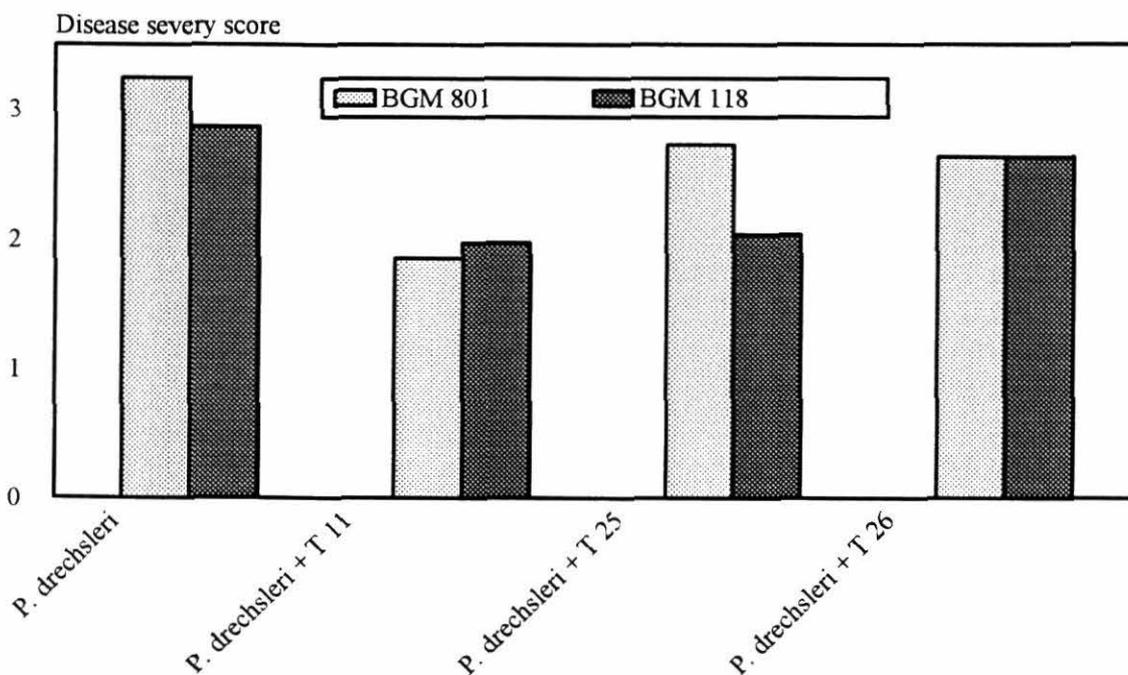


Figure 5.4.2.3. Effect of three *Trichoderma* spp. isolates, simultaneously inoculated with *Phytophthora drechsleri*, in cassava roots, under laboratory conditions.

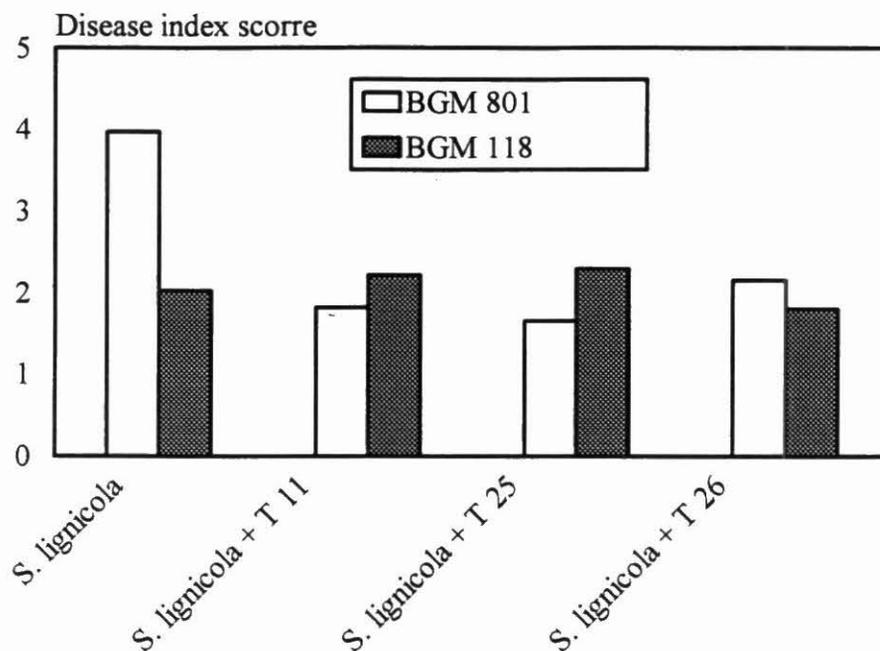


Figure 5.4.2.4. Effect of *Trichoderma* spp. isolates, simultaneously inoculated with *Scytalidium lignicola* in cassava stakes, under laboratory conditions.

5.4.3 Virology, by Celia Camara do Vale, Jose Albercio Lima, Chigeru Fukuda

The main objective of the virology activity during 1996 was to determine root yield losses due to the cassava vein mosaic virus (CVMV). An experiment to study such a kind of problem requires a large amount of virus-free planting material, but the PCR technique did not perform well as a diagnosis method for large numbers of plants. Due to that, the production of CVMV-specific antiserum became the main objective since this technique is reliable and more easy to perform.

Several modifications on the CVMV purification technique, such as viral separation through polyacrylamide gel electrophoresis, were made aiming at overcoming the presence of host protein in the purified virus. Regardless of all modifications, host plant protein was still present in the “purified” virus preparation. It was then decided to study the CVMV host range in order to identify a host from which the virus could be successfully purified. The following plants were evaluated: *Amaranthus caudatus*, *Capsicum annun*, *Cassia*

occidentalis, *Canavalia ensiformis*, *Chenopodium amaranticolor*, *C. murale*, *C. quinoa*, *Citrullus vulgaris*, *Cucumis anguria*, *C. melo*, *Gossypium hirsutum*, *Heliantus annun*, *Lactuca sativa*, *Lycopersicon esculentum*, *Nicotiana benthaminana*, *N. rustica*, *N. glutinosa*, *N. tabacum*, *Sesamum orientale* and *Manihot esculenta*. Several inoculation techniques were tested, including injection of purified virus into plant tissue, and grafting. Symptom development was observed only in cassava plants inoculated by grafting. This study will continue by evaluating other potential host plants.

5.5 CASSAVA INTENSIVE DIAGNOSTIC SURVEY IN NORTHEAST BRAZIL,

by Jose Humberto Almeida de Cerqueira and Carlos Estevão Leite Cardoso

Land property and usage

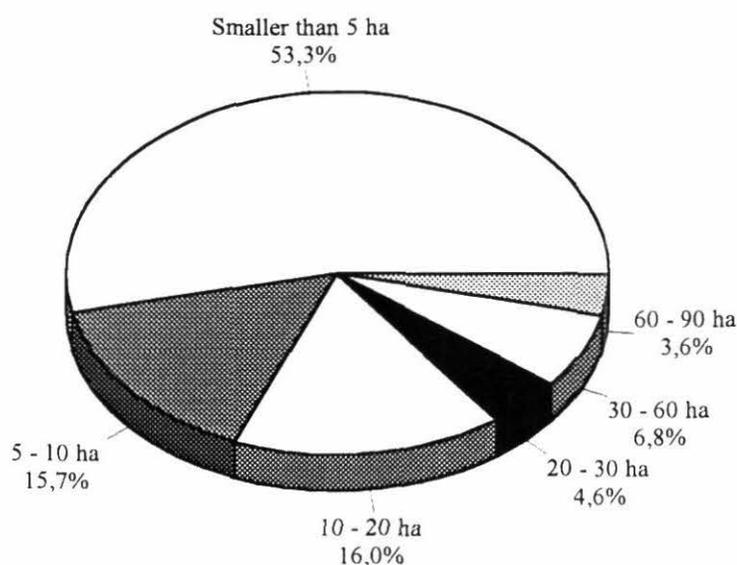
About 59% of the surveyed cassava growers in Northeast Brazil have land property title, and the average area for them is 12.9 her; 16.7% of the cassava growers, average land area around 14 her, have land property document other than title; other 18.9% own their land, 5,7 her average area, but without property title; 17.1% of the surveyed growers are sharecroppers with an average land area of 1.5 her; and 3.2% are renters with 1.4 her average land area. The large amount of cassava growers without land property title, over 40%, constitutes a big problem regarding to agricultural credit.

The average land area of the cassava growers surveyed in Northeast Brazil, varies from 1.9 to 72 her (Table 5.5.1). Figure 5.5.1 shows land area distribution within the surveyed population. It was found that over 53% of the growers have land area smaller than 5 her and for almost 70% the land area is smaller than 10 her, and only 3.6% have land area larger than 60 her.

Table 5.5.1. Land area of the surveyed cassava growers in Northeast Brazil.

Land extracts	Average		Frequency	
	land area (her)	Absolut (number)	Relative (%)	Accumulated (%)
Smaller than 5 her	1.9	150.0	53.4	53.4
Larger than 5 her, smaller than 10 her	6.8	44.0	15.7	69.0
Larger than 10 her, smaller than 20 her	12.6	45.0	16.0	85.1
Larger than 20 her, smaller than 30 her	24.4	13.0	4.6	89.7
Larger than 30 her, smaller than 60 her	48.6	19.0	6.8	96.4
Larger than 60 her, smaller than 90 her	72.0	10.0	3.6	100.0

¹ Total is higher than 100% because some growers have areas in more than one land usage.

**Figure 5.5.1. Distribution of average land area of cassava growers in Northeast Brazil.**

Data in Table 5.5.2 show that all surveyed cassava growers use part of their land, 3.9 her average area, to plant either annual or perennial crops. It is interesting to call the attention for the land used as pasture, 29.9%, left under fallow, 29.9%, and with natural vegetation, 13.2%, all of them generally used for grazing. This suggest a high importance of grazing for the surveyed cassava growers.

Table 5.5.2. Land usage and average land area in each kind of use by cassava growers in Northeast Brazil.

Land usage	Relative participation¹ (%)	Average land area (her)
Either annual or perennial crops	100.0	3.9
Pasture	29.9	5.0
Fallow	29.9	7.4
Native vegetation	13.2	10.2
Forest	11.4	16.4
Non productive area	9.6	0.7
Lowland area	7.8	7.0

¹ Total of relative participation is higher than 100 because all the cassava growers surveyed have their land occupied with several uses.

Rural family

Table 5.5.3 shows family composition in rural communities in Northeast Brazil where cassava is one of the main crops. Over 96% of the families are composed by parents son, and daughters, while 3.1% of them include also other relatives.

The educational level of family members in the surveyed rural communities is very low. The majority of the youth does/did not attend school, that results in 89.02% of the daughters and 80.64% of the sons without any school education. It was found that 57.2% of the parents and 76.8% of the family members including parents did not attend school, while 23.2% are either elementary or high school educated.

The head of the family is predominantly a man older than 18 years; 52.5% of them are 50 year old or less, and 70.7% are around 35 year old. Most of the cassava growers, 86.4%, live in the rural community while the remaining 13.6% live in other places.

Approximately one third of the family members, that is 35.8%, does not work in the rural community: they are either school age children or young people whose activities are other than rural labor. A little high percentage, 36.5%, work only in their land, and 28.7%, besides working in their land, also sell their labor force in order to make extra money and increase the family income.

Table 5.5.3. Family composition and educational level of the cassava growers in Northeast Brazil.

Members	Family composition		Educational level			
	Frequency	(%)	School educated		Not school educated	
			Number	(%)	Number	(%)
Head	318	15.6	182	57.23	136	42.77
Wife	301	14.7	196	62.12	105	34.88
Daughter	674	33.0	600	89.02	74	10.98
Son	687	33.6	554	80.64	133	19.36
Son in law	14	0.7	9	64.29	5	35.71
Sister	6	0.3	4	66.67	2	33.33
Brother	4	0.2	4	100.0	-	-
Mother	12	0.6	5	41.67	7	58.33
Granddaughter	6	0.3	3	50.00	3	50.00
Grandson	16	0.6	8	66.67	4	33.33
Daughter in law	5	0.2	2	40.00	3	60.00
Father	1	0.0	-	-	-	-
Cousin	1	0.0	1	100.00	-	-
Niece	2	0.1	1	50.00	1	50.00
Nephew	1	0.0	1	100.00	-	-

Cassava production technology

According to the recorded data, 18.9% of the cassava growers in Northeast Brazil use to hoe the area as part of the soil preparation activities; for most of the growers, 63.7%, soil preparation is performed only with hoe. Seventy three percent of the cassava growers in Northeast Brazil use stakes of their own to start a new planting, and 42% of them said they carry out any kind of stake selection prior to harvesting. The apical and basal portions of the stem are discarded and only the medium portion is used as planting material as stated by 82.9% of the Northeastern cassava growers; the stake must be 10 to 20 cm long and show a diameter higher than 2 cm according to 63.8%. Most of the growers (82.6%) transport the cassava stem as a tied up bundle, 61.2% plant the stake in a horizontal position, and 63.2% of them plant in a shallow hole. If necessary, the cassava planting material may be stored for up to 60 days according to 50.5% the cassava growers, and 38.8% of them said that they store the planting material under a tree shade. Fertilizer and correction of soil pH by liming is not a usual practice among cassava growers in Northeast Brazil, since 65.5% of them said they do

not carry out these practices. For 63.3% of the growers cassava planting is performed by the beginning of the raining season. Regarding to the topography of the area to be planted, 70% said they use flat areas, 27.5% of them plant in the slope area, and only 2.5% install their cassava fields in lowland areas.

Pest and disease incidences

As shown in Figure 5.5.2, leaf-cutting ants (29.4%) is the most important cassava pest in Northeast Brazil, followed by root rot disease (20.7%), hornworm (16.5%), cassava green mite (13.0%), whiteflies (8.2%), witches' broom disease (CWBD; 3.4%), stake weevil (3.1%), and other pests and diseases (5.8%). Except for the cassava root rot disease that causes root infection, all the other cassava pests and diseases damage to the leaves and branches as shown in Table 5.5.4. Leaf cutter ants have been considered a very important cassava pest in the last years, followed by CGM, hornworm, whiteflies, and root rot. In general, cassava pests are responsible for around 22% decrease of root yield, except for witches' broom and root rot diseases that cause, respectively, 71.9% and 51.8% root yield losses. The stage of plant development at the time of pest/disease detection varies from 1-3 month after planting for ants, and up to 12 months for root rot.

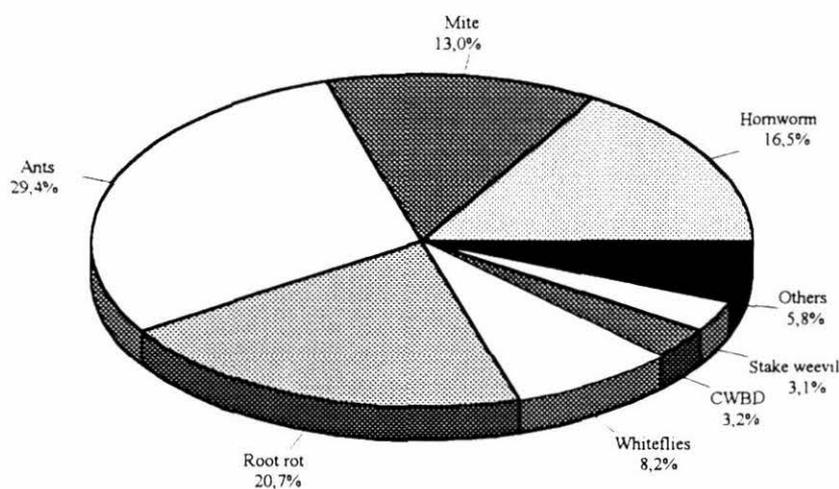


Figure 5.5.2. Main pests and diseases of the cassava crop in Northeast Brazil, as identified by cassava growers.

Table 5.5.4 Cassava pests and diseases in Northeast Brazil and their importance to the crop, according to growers.

Pest/disease	Infection (%)	Infected organ (%)		Period of observation (year)	Yield loss (%)
		Root	Aerial part		
Hornworm	16.5	-	98.3	15	22.7
Mite	13.0	-	81.5	16	21.6
Leaf-cutting ants	29.4	-	99.0	19	22.3
Root rot	20.7	99.3	-	11	51.8
Whiteflies	8.2	-	96.6	13	22.9
Witches' broom	3.4	-	100.0	5	71.9
Stake weevil	3.1	-	95.5	7	33.5
Others	5.8	-	-	-	-

The cassava growers' action to solve these problems are presented in Table 5.5.5. The data show that cassava growers in Northeast Brazil do not apply any control measure for CGM, termites, whiteflies, stake weevil nor witches' broom disease. On the other hand, attempts to control cassava pests by growers has been based on insecticide application. For cassava root rot disease, growers have been using lime and ash as soil amendment aiming at controlling the disease.

Commercialization

Looking at the source of information from where the surveyed growers know about prices of cassava products, it was found that, for cassava flour, starch and fresh root, the information comes from the local market, while for cassava chips the information comes from the extension personnel (Table 5.5.6). The majority of the cassava production in the Northeast, 76.8%, is sold as cassava flour, 19.1% is sold as fresh root, 1.6% is used as animal feeding, and only 0.7% is transformed to chips (Table 5.5.7). Regarding the manner of commercialization, 21.17% of the cassava flour, 0.05% of "tapioca", and 1.03% of the starch products are sold to wholesalers; 0.37% of the "beiju" production is sold directly to the consumer; 0.37% of the chips is commercialized through growers associations; and 5.85% of the root production is sold to truck drivers (Table 5.5.8).

Table 5.5.5. Control measures for pests and diseases used by cassava growers in Northeast Brazil.

Pest Disease	Growers (%)	Kind of Control	Month when control measures are applied											
			1	2	3	4	5	6	7	8	9	10	11	12
Mites	100	None	-	-	-	-	-	-	-	-	-	-	-	-
Hornworm	50	None	-	-	-	-	-	-	-	-	-	-	-	-
	50	Chemical	+	+	++	++	++	++	++	+	+	+	+	+
Ants	1.5	none	-	-	-	-	-	-	-	-	-	-	-	-
	98.5	chemical	+	++	++	++	++	++	++	+	+	+	+	+
Termites	100	none	-	-	-	-	-	-	-	-	-	-	-	-
Whiteflies	100	none	-	-	-	-	-	-	-	-	-	-	-	-
Stake weevil	100	none	-	-	-	-	-	-	-	-	-	-	-	-
Anthracnose	50	none	-	-	-	-	-	-	-	-	-	-	-	-
	50	chemical	+	+	++	++	++	++	++	+	+	+	+	+
Root rot	50	none	-	-	-	-	-	-	-	-	-	-	-	-
	33.3	lime	+	+	++	++	++	++	++	+	+	+	+	+
	16.7	Ash	+	+	++	++	++	++	++	+	+	+	+	+
CWBD	100	none	-	-	-	-	-	-	-	-	-	-	-	-

Table 5.5.6. Source of information regarding price of cassava products.

Source of information	Processed product			
	Chips (%)	Flour (%)	Starch (%)	Fresh root (%)
Growers association	2.5	1.4	1.1	2.5
Extension worker	6.8	8.2	4.3	7.8
Other grower	3.6	31.3	9.6	17.4
Wholesaler	1.1	31.3	10.7	24.9
Radio	1.4	6.0	1.1	1.8
Newspaper/TV	1.1	1.4	0.0	0.7
Local market	2.5	64.8	12.1	17.4
None	0.4	1.4	1.1	0.4

Table 5.5.7. Destination of the cassava production in Northeast Brazil.

Destination	Percentage
Sell as fresh root	19.1
Cassava flour	76.8
Chips	0.7
Animal feeding	1.6
Others	1.8

Table 5.5.8. Destination of cassava roots and cassava products in Northeast Brazil.

Destination	Flour (%)	“Beiju” (%)	“Tapioca” (%)	Starch (%)	Chips (%)	Roots (%)
Consumer	5.64	0.37	0.02	0.05	0.00	1.87
Truck drivers	6.28	0.24	0.00	0.02	0.09	5.85
Wholesalers	21.17	0.03	0.05	1.03	0.00	5.50
Sellers	15.90	0.36	0.00	0.76	0.00	1.01
Associations	0.00	0.00	0.00	0.00	0.37	0.78
For the family	26.46	1.73	0.96	2.42	0.04	0.98
Total	75.45	2.73	1.03	4.28	0.50	15.99

Regarding to “casa de farinha”, the place where cassava roots are processed to flour and other cassava products, such as “tapioca” and “beiju”, the results of the survey showed that 39.5% of the cassava growers use a communal “casa de farinha” to process their harvest; 33.1% of them rent the “casa de farinha” from somebody else; 14.9% have their own “casa de farinha”; 6.0% does not pay for using somebody else’s “casa de farinha”; and 6.4% did not give a specific answer.

Results related to utilization of the aerial part of the cassava plant indicate that, although 38.3% of the aerial part is used for animal feeding, about the same amount, 37.5%, is not utilized by the growers, and only 23.3% is used as planting material. Selling the aerial part of the cassava plant is not a common practice among cassava growers in Northeast Brazil, since only 1.0% of them does it.

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7 ACRONYMS

CGM	Cassava green mite
CIAT	Centro Intenacional de Agricultura Tropical
CM	Cassava mealybug
CNPMA	Centro Nacional de Pesquisa de Monitoramento e Avaliação de Impacto Ambiental
CNPMPF	Centro Nacional de Pesquisa de Mandioca e Fruticultura Tropical
CNPq	Conselho Nacional de Desenvolvimento Científico e Tecnológico
COPAL	Comitê de Pesquisa Agrícola Local
CPATC	Centro de Pesquisa Agropecuária dos Tabuleiros Costeiros
CPATSA	Centro de Pesquisa Agropecuária do Trópico Semi-Árido
CVMV	Cassava Vien Mosaic Virus
CWBD	Cassava Witches' Broom Disease
EBDA	Empresa Baiana de Desenvolvimento Agrícola
EMATERCE	Empresa de Assistência Técnica e Extensão Rural do Ceará
EMATERPB	Empresa de Assistência Técnica e Extensão Rural da Paraíba
EMATERPE	Empresa de Assistência Técnica e Extensão Rural de Pernambuco
EMBRAPA	Empresa Brasileira de Pesquisa Agropecuária
EMDAGRO	Empresa de Desenvolvimento Agropecuário de Sergipe
EMEPA	Empresa Estadual de Pesquisa Agropecuária da Paraíba S/A
EPACE	Empresa de Pesquisa Agropecuária do Ceará
ESCaPP	Ecologically Sustainable Cassava Plant Protection
FPR	Farmer Participatory Research
IFAD	International Fund for Agricultural Development

IICA	Instituto Interamericano de Cooperación para la Agricultura
IITA	International Institute of Tropical Agriculture
IPA	Empresa Pernambucana de Pesquisa Agropecuária
PROFISMA	Proteção Fitossanitária Sustentável da Mandioca na América Latina e África
PROSERTÃO	Projeto de Apoio às Famílias de Baixa Renda da Região Semi-Árida de Sergipe
UFAL	Universidade Federal de Alagoas
UFC	Universidade Federal do Ceará
UNICAMP	Universidade de Campinas

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