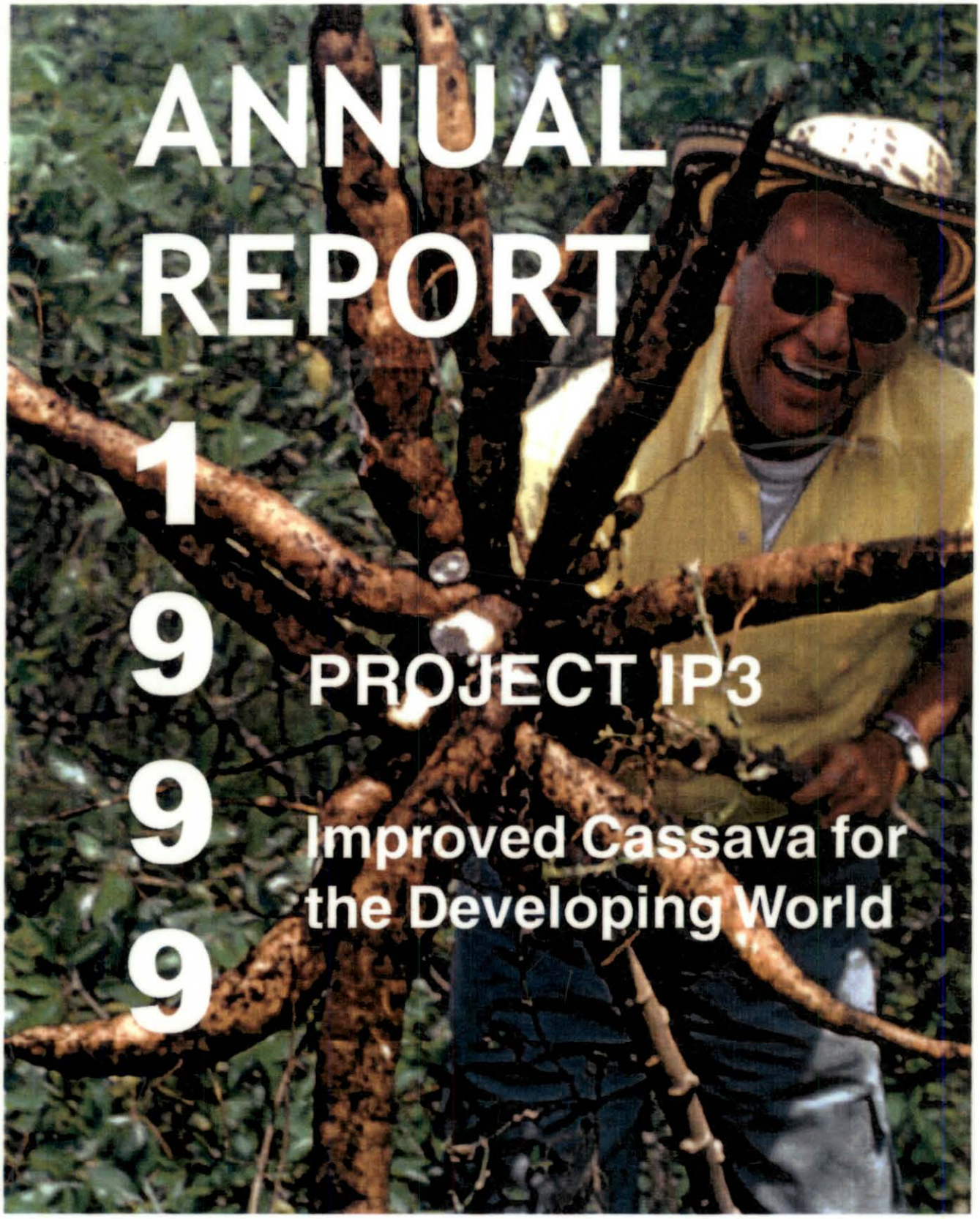


ANNUAL REPORT

1999

PROJECT IP3

Improved Cassava for the Developing World

CIAT

Centro Internacional de Agricultura Tropical
International Center for Tropical Agriculture

PROJECT IP3

Improved Cassava for the Developing World

Annual Report, 1999

INTRODUCTION

The major goal of out project is to contribute in increasing and stabilizing cassava production in diverse environments and for different markets, by developing improved gene pools in cooperation with national programs. The purpose of our project is to generate basic understanding, tools and improved cassava germplasm for sustainable enhancement of cassava production and the diversification of end-uses in relevant ecosystems. The most important ecosystems are: the semi-arid (below 800 mm/year, unimodal rains); the sub-humid (800-1500 mm/year, bimodal rainfall distribution); and the acid soil savannas (1500-3000 mm/year, short dry period and soils with low pH and high Al concentrations). The humid tropical lowlands: the mid-altitude tropics; the high-altitude tropics and the subtropical environments represent ecosystems of secondary importance in terms of area and total production.

As the breeding stages progress, we give emphasis to traits of lower heritability, because more planting material for each genotype is available, and the evaluation can be conducted in bigger plots with replications and at more than one location. Certain selection criteria are of general importance across ecosystem (i.e. yield potential, dry matter content in the roots, etc.), while others are specific for each ecosystem (i.e. pest and diseases). Besides the selection of superior genotypes a major research priority is the development an d use of research tools that will shorten the breeding cycle and increase its efficiency, such as molecular marker assisted selection, and farmer participatory evaluation at early stages of the breeding cycle. New sources of resistance to major biotic and abiotic constraints, and favorable alleles for root quality traits are constantly being incorporated through recombination and selection.

CIAT has been actively involved with CORPOICA and CNPMF/EMBRAPA in the development and implementation of methodologies for the evaluation and selection of cassava germplasm with the participation of end-users. Also with CNPMF/EMBRAPA, a project has been implemented developing germplasm adapted to semi-arid conditions. In Thailand, CIAT works with FCRI, resulted in the diffusion of improved cultivars that have diversified the genetic base for cassava in the region, which has had tremendous impact on cassava production in Asia. CIAT and IITA have actively collaborated in the development and introduction of germplasm into Africa, combining elite Latin American genotypes with sources of resistance to African Cassava Mosaic Virus. This interaction is very important for helping African farmers, through the valuable in situ work by IITA, to increase cassava yield potential in Africa. It also helps Latin America by introducing sources of resistance to ACMV, a disease which is not present in the continent, but whose spread is now feasible upon the finding of the insect vector in the region.

In Latin America, there has been an increased involvement of the private sector in cassava research, not only for the traditional starch (native and fermented) and feed production, but also processed cassava for human consumption (pre-cooked frozen croquettes, fried cassava chips for the snacks market, etc.). As a result of the globalization of the economies, there is an increased interest in tropical regions in the exploitation of local resources, including cassava. There has been an interesting shift in Latin America and Africa (as seen by IITA scientist) to move cassava from the low-input, low value product towards a higher-inputs, higher-values, cash crop. This new scenario originates in the private sector (mainly processor or end-users), who has recognized the advantages cassava offers, but who also demands for special traits in the crop. This trend had consolidated in Asia few years ago, particularly Thailand, but now also in China and Indonesia.

HIGHLIGHTS

- Creation of **CLAYUCA** (Latin American Consortium to support research on cassava involving Bolivia, Colombia, Cuba, Ecuador and Venezuela).
- Creation of the first ***"Ingenio Yuquero"*** as an ingenious alternative to solve constraints in cassava utilization: root bulkiness and marketing.
- Promotion of the idea that dried cassava can **compete** with maize for animal feed in tropical countries. Concept supported by the private sector of different countries: poultry and swine growers associations, and animal feed industry.
- To support the concept of cassava as a cash-crop, for different value-added processes, several genotypes, with high drymatter **yield potential**, have been identified for each ecosystem.
- Cassava competitiveness also depends on **reducing production costs**: **a)** mechanization of planting and harvesting; **b)** genetic transformation (i.e. for herbicide resistance); **c)** organic fertilization with poultry and swine manure.
- **Molecular markers** for resistance to ACMV and early bulkiness have been identified. 186 SSR markers developed for a better saturation of the molecular genetic map of cassava.
- Promising varieties, with complementary **disease resistance** genes, identified for the acid soil savannas. Genotypes, resistant to root rots, selected by indigenous women farmers.
- Good sources of **resistance** to root rots were found in field and greenhouse screenings in Brazil. Two highly resistant cultivars were found at CIAT.
- IITA and CIAT have agreed to develop a **joint research agenda** for areas of common interest: heterosis among African and Latin American germplasm; resistance to ACMV; interaction with private sector; processing technology; molecular markers and biotechnology.
- Good progress made in the areas of genetic resistance and biological control of **whiteflies**. Crosses made and evaluations underway, to determine the inheritance of resistance.
- Two different antixenosis mechanisms identified to explain resistance to **green mites** in cassava.
- Evaluation of germplasm bank collection for **nutritive and starch quality** properties of roots and/or leaves.
- Basic research for understanding the **biochemical basis** of post-harvest physiological deterioration.

PERSPECTIVES

- Emphasize breeding and research for the development of high dry matter yielding genotypes for industrial uses of cassava, while maintaining the activities for more traditional types of cassava production.
- Take advantage of the information available and incorporate molecular markers assisted selection into the project.
- Carry out a massive research effort (using diallel crosses analysis) for three different ecosystems to produce information on the genetics of several traits of agronomic or economic relevance whose inheritance has not been studied yet.
- Evaluate and contribute to the mechanization of cassava production, which results in reduced costs of production and increases yields.
- To strengthen the interaction with IITA by developing new joint research projects and presenting a common stand in the Global Cassava Strategy Forum at Rome, early in 2000.
- Apply the protocols already developed for genetic transformation of cassava for the introduction of herbicide resistance and the Bt-gene.
- Carry out environmental studies to evaluate the advantages and risks that the deployment of genetically modified herbicide resistant cassava may represent: reduction of soil erosion (and weeding costs), and presence of herbicide residues in watersheds and/or cassava roots.
- Rapid introduction of resistance to ACMV into Latin American elite germplasm using sources provided by IITA and in collaboration with this Institute.
- Continue with the revitalization of the physiology laboratory by searching of funds for research projects in cassava and also interacting with other institutions that could make use of the services provided by the laboratory.
- Continue searching for the natural occurrence of apomixis in the cassava germplasm bank and in collaboration with IITA
- Continue searching for novel starch types in the cassava germplasm bank and in wild *Manihot* relatives.

Contents:

cont. Output 5 Page

IMPROVED CASSAVA FOR THE DEVELOPING WORLD

OUTPUT 1: Genetic base of cassava and *Manihot* species evaluated and available for cassava improvement.

The overall objective of this project is to improve the nutritional status of people living in marginal environments of the tropics, by selecting and promoting cassava genotypes with high and good bio-availability of micronutrients and vitamins.

Activity 1.1. ***Evaluation of genetic diversity and heritability for vitamins and mineral content in cassava leaves and roots.***

Specific Objectives:

a) to screen 600 cassava landraces from CIAT's germplasm collection for B-carotene and ascorbic acid contents in roots and leaves;
b) to correlate B-carotene and ascorbic acid contents in both tissues;
c) to correlate vitamin contents with physiological post-harvest deterioration.

Rationale: Most of the emphasis in relation to cassava breeding has been centered on increasing root production and concentration of starch. Since cassava is a staple in regions where there are severe deficiencies of micro-nutrients; the crop can be used as a vehicle to deliver vitamins and minerals in higher concentrations. Improving the efficiency with which cassava acquires micro-nutrients and accumulates them in the roots and leaves can have an enormous potential not only in terms of human nutrition; but also in terms of crop production.

The short post-harvest storage life of cassava is a characteristic that limits the marketability of the root and necessitates either consumption or processing shortly after harvesting. Post-harvest physiological deterioration (PPD) of cassava roots begins within 24 hours of harvest, and results in crop and product quality losses, high marketing margins and risks, and restricted management flexibility for farmers, traders and processors. The reduction of PPD has been identified as a priority target for strategic research. In many respects, PPD resembles wound responses found in other better studied plant systems but cassava appears to lack the wound healing capacity which is normally associated with the inhibition of wounding responses. An important component of these wound responses are the oxidative processes. Ascorbic acid and carotene are known to have antioxidant properties. Therefore, PPD was measured in a sample of genotypes to evaluate the potential correlation between these two vitamins and PPD.

Materials and Methods: Root and leaves samples of plants from each of the 601 accessions of the core collection from the cassava germplasm bank, were taken and evaluated for carotene and ascorbic acid contents.

Carotene concentration measurements: the extraction procedure outlined by Safo-Katanga et al. (1984) was adjusted by extracting root parenchyma with petroleum ether. The extraction protocol for leaves had to be modified due to the presence of tannins and chlorophyll. The adjusted protocol included several washing steps with methanol in order to minimize the interference from the other pigments that were present in the leaves. A sample of 5 g was taken out of the root or leaves, taken at random 10 to 11 months after planting. The quantification was done by ultraviolet spectro-photometry using a Simadzu UV-VIS 160A recording spectrophotometer. UV detection was done at λ = 455nm for root extracts and at λ = 490nm for leaves extracts.

Ascorbic acid concentration measurements: the protocol for the determination of ascorbic acid by Fung and Luk (1985) was adjusted for cassava leaf and roots taking as base the procedure outlined and involved the following steps:

a) Homogenization of 1 g of fresh leaves or 6 g of fresh roots in a turrax with 20 ml of extraction buffer (3% phosphoric acid and 8% glacial acetic acid).
b) Centrifugation for 5 minutes at 10 °C and 3000 rpm.
c) Separation of supernadant and vortex of 1 ml of the extracts with 2 ml of 10% hydrochloric acid. Reading was taken inmediately with an UV-VIS spectrophotometer. UV detection was done at λ = 245nm. Quantification was done using previously decomposed extract with 1M sodium hydroxide solution as blank. During the whole process, samples were protected from air to avoid oxidation.

PPD measurement: post-harvest physiological deterioration was measured (six days after harvest) on 30 genotypes whose ascorbic acid and carotene root concentrations were known.

Results: a total of 601 accessions, of core collection of the cassava world germplasm bank collection held at CIAT, were evaluated for the traits described. In a few cases, because plants did not grow adequately or did not develop roots, measurements could not be taken. Those missing data points will be completed during the current year. The origin of the accessions evaluated is described in Table 1.1.

Ascorbic acid concentration in leaf tissue ranged from 1.68 to 419.25 mg/100g FW (Table 1.2). Data was obtained from a total of 568 samples, with a mean concentration of 120.16 mg/100g FW and a standard deviation of 84.14 mg/100 g FW. Concentration of ascorbic acid in the roots ranged from less than 1.00 to 39.52 mg/100 g FW of fresh roots (Table 1.2). Data was obtained from samples of 530 accessions, showing a mean of 9.52 mg/100 g FW, and a standard deviation of 6.48 mg/100 g FW. Both distributions showed a strong skewness, with values concentrating on the left, and long right tails. In this type of distribution the median is a better measurement of central tendency than the arithmetic mean.

It is obvious that ascorbic acid concentrates on the leaves, rather than in the roots. Its mean concentration on leaves was more than 12 times larger than the mean concentration on the roots. There was no correlation between the ascorbic acid concentration on leaves and roots ($\rho = 0.045$, based on 514 data points, Table 1.4).

Table 1.1. Origin and number of accessions from CIAT's Core Collection evaluated for Vitamin C and Carotene in roots and leaves[¶].

Origin	Ascorbic acid		Carotene	
	Leaves	Roots	Leaves	Roots
CIAT Elite Clones	33	32	32	33
Argentina	7	7	5	7
Bolivia	2	2	1	2
Brazil	98	91	94	94
China	2	2	2	2
Colombia	124	106	116	95
Costa Rica	19	19	18	11
Cuba	18	17	17	16
Dominican Republic	5	5	5	5
Ecuador	26	24	26	23
Fiji	2	2	2	2
Guatemala	12	12	14	12
Indonesia	7	7	7	7
Malaysia	14	15	14	13
Mexico	17	17	15	15
Nigeria	3	3	3	3
Panama	9	6	9	7
Paraguay	33	36	32	34
Peru	68	69	68	67
Philippines	2	2	2	2
Puerto Rico	7	5	7	4
Thailand	4	4	4	4
U.S.A.	2	4	4	4
Venezuela	52	44	50	42
TOTAL	**566**	**529**	**547**	**504**

[¶] Samples of some accessions could not be obtained or measured because missing plants, small roots and/or damaged roots.

Table 1.2. Ascorbic acid concentration in leaves and roots of 551 and 530 cassava accessions, respectively [¶].

Data from leaves		Data from roots	
Range (mg / 100 g FW)	Frequency	Range (mg / 100 g FW)	Frequency
0.0 - 14.9	47	0.00 - 1.49	26
15.0 - 29.9	43	1.50 - 2.99	49
30.0 - 44.9	40	3.00 - 4.49	38
45.0 - 59.9	30	4.50 - 5.99	56
60.0 - 74.9	37	6.00 - 7.49	71
75.0 - 89.9	41	7.50 - 8.99	54
90.0 - 104.9	34	9.00 - 10.49	49
105.0 - 119.9	38	10.50 - 11.99	43
120.0 - 134.9	32	12.00 - 13.49	30
135.0 - 149.9	27	13.50 - 14.99	25
150.0 - 164.9	35	15.00 - 16.49	22
165.0 - 179.9	32	16.50 - 17.99	16
180.0 - 194.9	18	18.00 - 19.49	11
195.0 - 209.9	26	19.50 - 20.99	6
210.0 - 224.9	23	21.00 - 22.49	12
225.0 - 239.9	16	22.50 - 23.99	3
240.0 - 254.9	16	24.00 - 24.49	2
255.0 - 269.9	8	25.50 - 26.99	5
270.0 - 284.9	5	27.00 - 28.49	2
285.0 - 299.9	4	28.50 - 29.99	3
300.0 - 314.9	3	30.00 - 31.49	2
315.0 - 329.9	5	31.50 - 32.99	0
330.0 - 344.9	1	33.00 - 34.49	3
345.0 - 359.9	0	34.50 - 35.99	0
360.0 - 374.9	1	36.00 - 37.49	1
375.0 - 389.9	1	37.50 - 38.99	1
390.0 - 404.9	4	39.00 - 40.49	0
405.0 - 419.9	1	40.50 - 41.99	0
Minimum [§]	0	Minimum [§]	0
Maximum	419.25	Maximum	37.52
Total	568	Total	530
Median	109.30	Median	8.09
Skewness[†]	0.69	Skewness[†]	1.28
Mean	120.16	Mean	9.48
St.Dev.	84.14	St.Dev.	6.50

[¶] Samples of some accessions could not be obtained or measured because missing plants, small roots and/or damaged roots.

[§] Measurement below detection threshold of equipment.

[†] Skewness test ranges from negative values (left tales); to 0.0 (perfect symmetry); to positive values (right tales). Larger magnitudes imply larger asymmetry.

Carotene concentration on leaves ranged from 23.28 to 86.22, with a mean of 48.26 mg / 100 g FW (Table 1.3). As in the case of vitamin C, carotene distribution was also skewed to the left, but to a lesser degree. Three samples (≥ 72.76 in Table 1.3) were different from the general population, showing 75.09 (MECU 104), 80.00 (MECU135),

and 86.22 (MCOL 1522), mg / 100 g FW. Carotene concentration on roots showed a strongly skewed distribution to the, ranging from 0.102 to 10.40, and a mean of 0.232 mg / 100 g FW. Following the same trend observed with ascorbic acid, carotene concentrated 100 times more on leaves than in roots, illustrating once again the excellent nutritive value of cassava leaves. There was no correlation between carotene concentration on leaves and roots (Table 1.4).

Table 1.3. Carotene concentration in leaves and roots of 551 and 530 cassava accessions, respectively [¶].

Data from leaves		Data from roots	
Range (mg / 100 g FW)	Frequency	Range (mg / 100 g FW)	Frequency
≤ 27.25	2	0.100-0.135	14
27.26-29.00	1	0.136-0.170	106
29.01-30.75	2	0.171-0.205	249
30.76-32.50	6	0.206-0.240	44
32.51-34.25	9	0.241-0.275	5
34.26-36.00	17	0.276-0.310	15
36.01-37.75	15	0.311-0.345	9
37.76-39.50	26	0.346-0.380	10
39.51-41.25	33	0.381-0.415	10
41.26-43.00	40	0.416-0.450	9
43.01-44.75	41	0.451-0.485	3
44.76-46.50	49	0.486-0.520	5
46.51-48.25	45	0.521-0.555	0
48.26-50.00	43	0.556-0.590	7
50.01-51.75	41	0.591-0.625	3
51.76-53.50	37	0.626-0.660	2
53.51-55.25	31	0.661-0.695	2
55.26-57.00	33	0.596-0.730	3
57.01-58.75	15	0.731-0.765	1
58.76-60.50	13	0.766-0.800	1
60.51-62.25	14	0.801-0.835	0
62.26-64.00	8	0.836-0.870	0
64.01-65.75	7	0.871-0.905	1
65.76-67.50	2	0.906-0.940	0
67.51-69.25	3	0.941-0.975	1
69.26-71.00	4	0.976-1.010	1
71.01-72.75	4	1.011-1.045	2
≥ 72.76	3	1.044-1.080	1
Minimum [$]	23.28	Minimum[$]	0.102
Maximum	86.22	Maximum	1.040
Total	544	Total	504
Median	47.72	Median	0.185
Skewness[†]	0.48	Skewness[†]	3.26
Mean	48.26	Mean	0.232
St.Dev.	8.61	St.Dev.	0.137

[¶] Samples of some accessions could not be obtained or measured because missing plants, small roots and/or damaged roots.

[†] Skewness test ranges from negative values (left tales); to 0.0 (perfect symmetry); to positive values (right tales). Larger magnitudes imply larger asymmetry

Table 1.4. Correlations between ascorbic acid and carotene content (mg / 100 g FW) in leaves and root tissues of several genotypes[¶] from CIAT's cassava germplasm collection.

	Ascorbic acid/Leaves	Carotene/Roots	Carotene/Leaves
Ascorbic acid /Roots	0.045 (514)	-0.108 (475)	-0.015 (497)
Ascorbic acid/Leaves	-.-	-0.087 (499)	-0.060 (439)
Carotene/Roots	-.-	-.-	-0.008 (486)

[¶] Number of observations used to estimate correlations is shown between parenthesis.

Table 1.5. Relationship between ascorbic acid content on roots and leaves, and physiological deterioration (PPD), in a sample of 32 cassava genotypes.

GENOTYPE	Ascorbic acid content (mg /100 g FW)		PPD[¶]
	Roots	Leaves	(%)
MBRA 73	26.65	65.36	14.33
MBRA 110	26.78	175.80	66.90
MBRA 191	36.92	41.18	69.14
MBRA 217	20.00	104.86	71.80
MBRA 328	20.00	8.13	56.38
MBRA 416	30.94	8.13	54.45
MBRA 507	21.01	37.15	24.99
MBRA 542	22.96	118.57	16.43
MBRA 702	30.94	201.60	60.17
MBRA 915	37.52	208.85	18.89
MCHN 1	28.26	141.94	6.43
MCOL 40	21.41	233.04	64.57
MCOL 451	33.16	96.80	68.57
MCOL 474	23.02	245.13	79.10
MCOL 490	25.24	204.02	89.05
MCOL 511	33.83	81.49	15.00
MCOL 1185	20.00	216.11	55.71
MCOL 1795	22.29	143.56	57.50
MCOL 1853	21.75	212.08	60.95
MCOL 1968	21.21	239.48	95.35
MECU 47	21.75	35.54	83.21
MECU 82	21.61	75.84	92.85
MECU 135	26.31	42.79	70.57
MMAL 38	21.08	66.17	56.46
MPER 488	23.56	187.09	42.14
MPER 484	22.02	45.21	27.86
CG 5-79	27.32	105.67	80.8
CM 507-37	28.59	17.00	45.71
CM 523-7	22.35	209.66	95.35
CM 922-2	27.39	69.39	48.22
CM 3306-9	29.06	37.95	70.86
CM 2772-3	34.16	83.10	35.14
MEAN	**25.22**	**129.16**	**56.09**

[¶] Measured six days after harvest.

In general there were poor, irrelevant correlations between post-harvest physiological deterioration and vitamin contents on roots (Tables 1.5 and 1.6). Correlation between PPD and vitamin C and carotene contents on roots were, respectively, –0.169 and –0.30. Therefore, the hypothesis that vitamin content can help to reduce PPD (through their antioxidant capacity), seems to be supported by these results.

Table 1.6. Relationship between carotene content on roots, and post-harvest physiological deterioration (PPD), in a sample of 30 cassava genotypes.

GENOTYPE	Carotene content (mg /100 g FW)	PPD (%)[¶]
MARG 6	0.67	55.81
MBRA 85	0.57	17.14
MBRA 191	0.47	58.62
MBRA 311	0.62	3.81
MBRA 337	0.58	0.00
MBRA 467	0.97	5.71
MBRA 475	0.58	2.86
MBRA 507	0.88	13.30
MBRA 512	0.75	53.93
MBRA 509	1.04	6.78
MBRA 522	1.04	17.14
MCOL 72	0.43	43.22
MCOL 638	0.43	3.80
MCOL 764	0.46	53.71
MCOL 2144	0.47	20.95
MCOL 2306	0.58	43.71
MCOL 2353	0.42	11.79
MECU 31	0.52	51.43
MECU 33	0.58	31.42
MMAL 29	0.60	25.99
MMAL 63	0.45	71.43
MPER 297	0.98	5.14
MPER 458	0.44	35.36
MPER 589	0.70	50.47
MPER 593	0.51	5.71
MPHI 3	0.48	72.14
MUSA 4	0.57	67.62
CM 1999-5	0.43	12.14
CM 2772-3	0.66	26.46
CM 5286-3	0.42	31.53
MEAN	**0.63**	**29.97**

[¶] Measured six days after harvest.

The samples evaluated do not represent adequately the core collection. Both samples (for ascorbic acid and carotene) showed considerably higher mean contents (Tables 1.5 and 1.6) than the population (Tables 1.2 and 1.3). Therefore, if the range of variation in vitamin content is widened, it is expected that the correlations with PPD will also increase. There was a positive association between degree of root deterioration and ascorbic acid on leaves.

Activity 1.2. Evaluation of the stability of vitamins and mineral content after different processing procedures or growth in different environmental conditions.

Specific Objectives:

a) *To measure vitamins content after three different processing procedures (boiling, sun drying and oven drying).*
b) *To measure variability for vitamin C and carotene content measurement using different sources of variation.*

Rationale: cassava is processed before consumption using heat treatments that can eventually affect the ascorbic acid and carotene contents. Solar or oven drying of cassava flour, and cooking fresh roots for several minutes are common processing procedures used by diverse cultures and for different end uses of the product. It is, therefore, important to know about the stability of vitamin content after those cooking procedures. Genotype by interaction is particularly important in cassava. Vitamin contents in cassava tissues are dependent on the environmental conditions where the crop is grown. Macro-environmental effects (i.e. between locations) have been measured in the past. However, little is known on the effect of the micro-environmental variation (i.e. within locations) on vitamin contents in cassava

Materials and Methods: The stability of vitamin content after i) solar drying of cassava flour; ii) oven drying of cassava flour; and iii) cooking fresh roots for 30 minutes, was measured on 26 and 33 genotypes for carotene and ascorbic acid, respectively. Another study was carried out to determine inter and intra-plant sampling variation for carotene and vitamin C, as well as for the measurement procedures. Ten plants (from two replications with five plants each) of two different genotypes were used for this experiment. A sample of five roots from each plant was taken and vitamin concentrations measured on each root individually. Also, five measurements from the same root sample were taken in three plants of the two genotypes.

Results: Carotene content was considerably more stable than ascorbic acid after the different processing procedures evaluated. On average 53.2% of the original carotene content remained after boiling or drying the roots, whereas only 14.4% of the ascorbic acid was recovered after the same treatments (Table 1.7).

Carotene contents were similar after boiling or drying with an oven (≈ 61% of the original), but were considerably lower after solar drying (≈ 40%). This would suggest that light had a detrimental effect on the endurance of carotene on roots. Ascorbic acid, on the other hand, showed a strong dependency to the different processing procedures. Boiling allowed maintaining the highest level of vitamin C (36.6 % of the original), whereas oven drying and solar drying reduced its content to 6.2 and 0.005 % of the original levels, respectively.

Table 1.7. Carotene and ascorbic acid contents (mg / 100 g FW) in roots and after different processing procedures.

Carotene					Ascorbic Acid				
Genotype	Roots (control)	Boiling	Oven drying	Solar drying	*Genotype*	Roots (control)	Boiling	Oven drying	Solar drying
MARG 6	0.67	0.40	0.46	0.32	MBRA 73	26.65	9.60	0.00	0.00
MBRA 85	0.57	0.30	0.33	0.22	MBRA 110	26.78	15.51	0.00	0.00
MBRA 191	0.47	0.28	0.28	0.16	MBRA 172	21.21	11.01	0.00	0.00
MBRA 335	0.58	0.47	0.50	0.23	MBRA 191	36.92	3.97	0.00	0.00
MBRA 337	0.58	0.56	0.51	0.41	MBRA 217	20.00	6.65	6.78	0.00
MBRA 400	0.45	0.32	0.39	0.18	MBRA 507	21.01	12.82	18.19	2.76
MCOL 72	0.43	0.24	0.21	0.17	MBRA 542	22.96	17.08	0.00	0.00
MCOL 638	0.43	0.25	0.27	0.15	MBRA 702	30.94	8.06	0.00	0.00
MCOL 764	0.46	0.25	0.25	0.16	MBRA 915	37.52	4.70	0.00	0.00
MCOL 2144	0.47	0.26	0.41	0.19	MCOL 40	21.41	3.93	0.00	0.00
MCOL 2353	0.42	0.37	0.24	0.15	MCOL 474	23.02	12.09	1.28	0.00
MECU 33	0.58	0.35	0.39	0.28	MCOL 511	33.83	19.27	0.00	0.00
MMAL 29	0.60	0.39	0.46	0.26	MCOL 1185	20.00	11.48	0.00	0.00
MMAL 63	0.45	0.23	0.18	0.16	MCOL 1240	20.41	15.71	1.01	0.00
MMEX 71	0.65	0.31	0.35	0.25	MCOL 1795	22.29	2.96	2.09	0.00
MPER 368	0.67	0.23	0.32	0.21	MCOL 1853	21.75	1.85	8.13	0.00
MPER 390	0.42	0.25	0.26	0.19	MCOL 1968	21.21	1.75	1.08	0.00
MPER 458	0.44	0.25	0.25	0.16	MCHN 1	28.26	1.35	2.42	0.00
MPER 556	0.49	0.25	0.24	0.16	MECU 47	21.75	1.45	3.36	1.08
MPER 589	0.70	0.43	0.65	0.36	MECU 82	21.61	12.39	0.00	0.00
MPER 593	0.51	0.19	0.16	0.14	MECU 135	26.31	11.62	0.00	0.00
MPHI 3	0.48	0.27	0.23	0.19	MGUA 7	24.77	11.21	1.41	0.00
MUSA 4	0.57	0.27	0.21	0.16	MMAL 38	21.08	8.63	0.00	0.00
CM 5286-3	0.42	0.32	0.24	0.16	MMAL 63	28.06	8.93	0.00	0.00
CM 2772-3	0.66	0.32	0.45	0.28	MMEX 54	19.87	14.30	0.00	0.00
CM 1999-5	0.43	0.25	0.27	0.19	MPER 484	22.02	12.02	5.91	0.00
-.-	-.-	-.-	-.-	-.-	MPER 488	23.56	14.77	0.00	0.00
-.-	-.-	-.-	-.-	-.-	CM 507-37	28.59	9.27	0.00	0.00
-.-	-.-	-.-	-.-	-.-	CM 523-7	22.35	5.41	0.00	0.00
-.-	-.-	-.-	-.-	-.-	CM 922-2	27.39	3.03	0.00	0.00
-.-	-.-	-.-	-.-	-.-	CM 2772-3	34.16	10.81	0.00	0.00
-.-	-.-	-.-	-.-	-.-	CM 3306-9	29.06	11.08	0.00	0.00
-.-	-.-	-.-	-.-	-.-	CG 5-79	27.32	10.54	0.00	0.00
MEAN	**0.52**	**0.31**	**0.33**	**0.21**	**MEAN**	**25.27**	**9.25**	**1.57**	**0.12**

Stability of vitamin C and carotene was not highly dependent on sampling. Measurements of plant to plant variation, and of root to root variation within the same plant (Table 1.8) were not significantly larger than those derived from the experimental procedure (variation of measurements for the same root sample).

Table 1.8. Standard deviations from several sources of sampling for ascorbic acid and carotene concentrations in roots from two contrasting cassava genotypes.

	Vitamin C		Carotene	
	CM 2772-3	MCOL 1505	CM 2772-3	MCOL 1505
Between roots/within plant [§]	4.43	6.66	0.07	0.02
Between plants[¶]	3.88	2.47	0.06	0.02
Measurement error [‡]	4.33	5.37	0.02	0.01
Mean concentration	29.12	31.92	0.74	0.34

[§] Mean standard deviation of measurements from five roots (average of ten plants per genotype).

[¶] Standard deviation of the mean concentration (from five roots) of 10 plants per genotype (two replications with five plants each).

[‡] Mean standard deviation of five measurements from the same root (sample of three plants per genotype).

Therefore, there is no need to measure vitamin contents from several roots obtained from several plants in order to guarantee the proper evaluation of each genotype. It was decided that a homogenized sample of 2-3 roots from just one plant would be adequate enough to provide accurate measurements of the genetic characteristics of the respective genotype. However, whenever possible (most of the cases), a sample of 2-3 roots from 2-3 plants, should be harvested.

Activity 1.3. Evaluation of starch content and traits among elite germplasm and/or core collection entries.

Specific Objectives:

a) To measure other root quality traits in addition to vitamin contents.
b) To increase the interaction with the private sector to explore new avenues for cassava starch and other cassava products utilization.

Rationale: CIAT is currently evaluating germplasm bank and elite cassava clones for vitamins content. Obtaining the samples constitutes a major component for the costs of this particular project. Also the cryopreservation research currently conducted at the Center may allow us in the near future to stop growing the large germplasm bank in the field season after season. It was, therefore, decided to take advantage that tissue samples were being obtained to extract more information from those samples. The kind of data that is or will be taken includes: **1)** dry matter content; **2)** total carbohydrates content; **3)** amylose and amilopectin contents; **4)** starch content; **5)** sugar content; **6)** starch paste clarity; **7)** pasting properties; **8)** acidity resistance; **9)** freezing resistance; **10)** syneresis; **11)** welling power and solubility of granular starches; **12)** starch bound phosphates; and **13)** quantitative measurement of cyanide content.

Materials and Methods: this research was started recently and only partial results will be presented. Most of the traits will be measured in CIAT's Laboratory at AGROEMPRESAS. However, for at least one variable (starch bound phosphates), further training and equipment will be required for the proper evaluation. The same germplasm evaluated for vitamins and PPD will also be evaluated for these traits. The methodology to be used for each trait is the standard published in the literature and will not be described herein.

Results: a proposal to National Starch & Chemical Company (U.S.A.) for exploring the genetic variability of cassava starch and roots properties has been positively accepted. It is likely that this private company will support the evaluation of the entire germplasm collection (≈ 6000 accessions). Additionally a hundred elite genotypes have been evaluated for starch traits as described in Table1.9. There are indeed good evidences that ample genetic variability exist for cassava root traits as to justify further evaluations of the germplasm collection.

Table 1.9. Results (using descriptive statistical parameters) from the evaluation of one hundred elite genotypes for roots and leaves quality traits.

	Dry Matter Leaves (%)	HCN in leaves (ppm)	Dry Matter Roots (%)	HCN in roots (ppm)	PPD (%)	Total Sugars (%)	Reduc. Sugars (%)	Starch (%)	Sugar / Starch	Amilose (%)
Min	22.1	216.0	30.6	31.6	4.5	1.0	0.03	78.0	1.25	10.1
Max	38.0	,712.0	54.3	922.9	83.1	7.2	1.58	89.0	8.83	17.8
Mean	33.0	834.9	42.2	210.0	28.7	2.6	0.44	82.9	3.13	14.4
Median	33.3	770.9	42.4	162.5	26.1	2.5	0.35	82.5	2.98	14.5
St.Dev.	2.1	389.5	3.5	158.8	16.1	1.0	0.32	2.5	1.21	1.6

Activity 1.4. Other ongoing activities related to root quality research.

Specific Objectives:

a) *To evaluate segregating progenies for carotene content, yellow colored roots and post-harvest physiological deterioration.*
b) *Select a set of materials for further genetic, physiological and/or molecular studies related to carotene content, color of the root and PPD.*
c) *Select a set of materials for evaluating their performance and potential for the fried cassava chips (snack market).*

Materials and Methods: seed from several crosses between cassava genotypes with yellow and/or white roots were planted early in 1999. The surviving plants will soon be harvested and evaluated for color, carotene content, and PPD. Table 1.10 lists the materials to be harvested in October. Some crosses had a few plants and therefore, will be of little use. However, several segregating progenies are made up of an adequate number of plants for drawing valid genetic conclusions. They are also more likely to show genotypes where the linkage between color of root parenchyma and carotene content is broken.

Similarly, there are few crosses involving RDOM 5 (highly resistant to post-harvest physiological deterioration) with other contrasting genotypes. Upon the harvest of the segregating progenies (Table 1.11) they will be analyzed to confirm the resistance in RDOM 5 and, hopefully, elucidate the inheritance of this important trait.

Table 1.10. Crosses between cassava genotypes with yellow roots (high carotene) and / or white roots (low carotene) to study the inheritance of root color and carotene content.

Cross ID	Female parent	Root color	Male parent	Root color	Number of plants
CM 7062	CM 2772-3	Y	HMC-1	W	7
CM 9149	CM 8371-7	Y	CM 2772-3	Y	2
CM 9150	CM 8371-23	Y	CM 8371-7	Y	3
CM 9153	CM 8371-23	Y	CM 2772-3	Y	13
CM 9248	SM 980-4	W	CM 2772-3	Y	14
Cm 9249	SM 1551-18	W	CM 2772-3	Y	41
CM 9319	SM 1551-18	W	SM 980-4	W	111
CM 9629	CM 2772-3	Y	MPER 183	W	17
CM 9640	CM 6740-7	W	CM 8371-23	Y	30
CM 9641	CM 6740-7	W	SM 1551-18	W	10
CM9679	SM 1551-18	W	CM 8371-7	Y	48
CM 9680	HMC-1	W	CM 8371-7	Y	53
CM 9681	CM 8371-7	Y	MPER 183	W	30
CM 9683	SM 1551-18	W	CM 8371-23	Y	57
CM 9684	CM 8371-23	Y	MPER 183	W	5
CM 9707	HMC-1	W	SM 980-4	W	148
CM 9708	SM 980-4	W	MPER 183	W	28
CM 9712	HMC-1	W	SM 1551-18	W	125
CM 9713	SM 1551-18	W	MNGA 1	W	57
CM 9714	MPER 183	W	SM 1551-18	W	129
CM 9722	MCOL 2298	Y	CM 8371-23	Y	9
CM 9730	HMC-1	W	CM 2177-2	Y	42
CM 9731	HMC-1	W	CM 8371-23	Y	20
CM 9733	HMC-1	W	MPER 183	W	17

Table 1.11. Progenies segregating for resistance to post-harvest physiological deterioration from crosses of RDOM 5 (resistant) with three contrasting cassava genotypes.

Cross ID	Male parent (resistant)	Female parent (susceptible)	Number of surviving plants
CM 9726	RDOM 5	CM 523-7	62
CM 9727	RDOM 5	SM 627-5	65
CM 9728	RDOM 5	SM 985-9	77

Results: To take advantage of this evaluation in the field, a proposal has been submitted, together with the private food processing company CONGELAGRO (Santafé de Bogotá) to evaluate the performance and potential of yellow rooted cassava for the fried cassava chips (snack market). The possibility of introducing the product for the fresh market as a new, highly nutritive product, will also be considered.

OUTPUT 2: Genetic stocks and improved gene pools developed and transferred to national programs.

The overall objective of this activity is to produce genetically improved cassava germplasm, by recombining selected parental genotypes and then evaluating the segregating progenies under adequate environmental conditions. Recombinant seed and/or vegetative propagules from elite clones are then shipped to our collaborators in Africa, Asia and Latin America.

Activity 2.1. ***Selection of parental material based on previous cycle results, and the information obtained from the other outputs (i.e. resistance/tolerance, root quality traits, etc.).***

Specific Objectives:

a) *Based on information based on evaluation trials at several locations, and the new objectives defined for the project, a set of elite clones are identified for recombination, to start a new cycle of selection.*
b) *For this particular cycle, the parental genotypes have been selected and will be crossed so that enough seed is produced to carry out a complete diallel study at each of the three most important eco-zones.*

Rationale: The selection of parents to build populations for future breeding work represents the core of our improvement efforts, since it will determine the genetic progress we will achieve in the future. There are two types of populations developed: open pollinated and control crosses. We usually used open pollination (polycrosses) to develop populations for target ecosystems. We have consistently developed polycrosses for the sub-humid tropics, acid soil savannas, semi-arid tropics, mid-altitude and highland tropics, and sub-tropics. In the case of controlled crosses, we used them to develop progenies for specific traits, special studies or the combination of elite experimental material with local landraces that need to be improved.

There is very little knowledge about the genetics of cassava. Most of the information is related to particular traits, not necessarily of agronomic relevance. Because of the mode of reproduction in cassava, and the very limited work to obtain homozygosity in the crop, it is very difficult to produce segregating materials suitable for relevant genetic studies on agronomically important traits. Diallel studies involve large number of crosses, but allow for the simultaneous analysis of many traits and do not require homozygosity of the parental lines. Therefore, for this particular cycle of selection, controlled crosses will be made for the target ecosystems. Enough seed will be produced among parental lines for a complete diallel analysis. The evaluations will serve to produce valuable data about the genetics on many traits in cassava, but also they will be part of the normal selection process to identify genetically superior germplasm.

Materials and Methods: Only genotypes that have been selected over 2 consecutive years in advanced yield trials are selected to participate as parents for the following generation. Among those genotypes, we select those with outstanding performance for the most important agronomic traits. After the analysis of variance is conducted with data across 2 years, those genotypes exceeding at least one standard deviation from the overall mean are considered as parents for the next generation. Sometimes we also include landraces or already released cultivars that can contribute special features to the progenies generated. Several parental lines have been selected to produce seed for the diallel analysis,-1Xeparately for the northern coast, the acid soil savannas, and the mid-altitude valleys. It is expected that, for some parents, there will be some missing crosses or not enough seed. Therefore, we have selected more parents that will actually make up each diallel study. Only after the crosses are made and the resulting seed harvested, we will know the exact list of parental materials used for each diallel. The three most important ecosystems (sub-humid tropics; acid soil savannas; and mid-altitude tropics) will be considered in this study.

The information provided by Pathologists, Entomologists and Quality Specialists in relation to sources of resistance or special traits is used to select genotypes for control crosses. These control crosses are developed upon specific requests from National Programs that want their main landrace or released variety crossed to genotypes with specific traits; or requests from CIAT's scientists that want to pyramid genes, or develop segregating progenies for gene tagging.

Results: The parents selected for the development of gene pools targeted to specific ecosystems using a diallel scheme is presented in Table 2.1. The agronomic performance of these materials is described further down in this document. Seed will be harvested from July, 1999 through March, 2000. F1 plants will grow until the planting of the trials early in 2001.

Table 2.1. Parental lines to be used in crosses for diallel studies in each of three different ecosystems relevant for cassava production in the world.

#	Sub-humid tropics	Acid-soil savannas	Mid-altitude tropics
1	CM 523-7	CM 4574-7	CM 5655-4
2	CM 6754-8	CM 6740-7	CM 6740-7
3	SM 805-15	CM 7033-3	SM 1219-9
4	SM 1219-9	SM 1219-9	SM 1278-2
5	SM 1411-5	SM 1565-15	SM 1741-1
6	SM 1565-17	SM 2219-11	HMC 1
7	SM 1657-12	*Brasilera* (MCOL2737)	MECU 72
8	SM 1665-2	HMC 1	MPER 183
9	SM 2192-6	MPER 183	-.-
10	MTAI 8	MTAI 8	-.-

The criteria for selecting parental lines reflect the new emphasis given to the project, with an increasing interest on industrial uses of cassava. Therefore, many of these genotypes are characterized by the high dry-matter productivity per hectare (i.e. MTAI 8, SM 1565-15 and SM 1219-9). Other parents have been selected for their excellent characteristics for industrial uses in the food industry (MPER 183, HMC 1 and *Brasilera*), recognized good combining ability (CM523-7, SM 805-15 and SM 1565-17), or special traits such as resistance whiteflies (MECU 72) or resistance to root rots (CM 4574-7).

Many genotypes have been selected for their performance in specific traits and have taken part of our control crosses during the last 3 years. This group represents a genetically broad population with outstanding performance *per se* and/or through their progenies.

Other crosses have been planed for incorporating desirable genes into popular varieties in different countries, diversifying the genetic base for resistance to biotic and abiotic stresses, and in few cases, produce adequate genetic materials to study the inheritance of the trait and for the potential identification of molecular markers. For instance a few new sources of resistance to ACMV have been identified at IITA and are currently being prepared for shipment to CIAT.

Achievements:

- An average of 115% advantage of the best selected parents over check varieties.
- Sources of resistance to pest and diseases as well as specific traits selected for the development of control crosses.
- New criteria have been incorporated to select parental genotypes that reflect the new emphases given to the project.
- The crosses produced will serve the dual purpose of providing valuable genetic information and generating a promising group of segregating progenies.

Activity 2.2. Establishment of crossing blocks and production of recombinant seed from previously established blocks.

Specific Objectives:

a) To produce large number of seed by sexual crosses (either polycrosses or controlled) recombining desirable traits from selected parental materials, and deliver them to NARS in Africa, Asia and Latin America.

Rationale: Populations developed for specific ecosystems represent the basis for our cooperation with National Programs and IITA. The development of genetic stocks is gaining importance through the years. Genetic stocks are produced based on the recombination of a set of genotypes that excel for a particular trait, and we would like to upgrade that trait beyond its natural range of variation (i.e. look for transgressive segregation in broader adaptation). Stocks developed for inheritance studies or to support molecular mapping of specific traits are constructed by the recombination of contrasting genotypes (i.e. resistance to ACMV, African Cassava Mosaic Virus). Often times our aim is to pyramid genes responsible for different sources of resistance (i.e. bacterial blight). As we shift our emphasis from applied breeding to more basic research supporting breeding (i.e. molecular marker assisted selection or MAS) genetic stocks will become even more important.

A key trait where MAS will be applied is for the introgression of resistance to ACMV (partially recessive) into elite Latin American germplasm. The vector of this virus has recently been found in the Americas, which makes the apparition of ACMV now a feasible possibility. If ACMV shows up in this continent it would have devastating consequences, because Latin American cassava germplasm lacks resistance to the virus. Traditional breeding for resistance to ACMV in the Americas has been extremely difficult because the virus is not present here yet, it is an almost recessive resistance, and the transfer of materials between Africa and Latin America is expensive and time consuming.

Parental population development in the future will concentrate more in targeting specific crosses between genotypes selected by NARS and complementary sources of genetic information from our genetic enhancement program or our global germplasm collection.

Materials and Methods: For polycrosses we use the design developed by Wright 1965 for polycrosses in forage species. For this type of design there is a need to have a number of clones equal to a prime number minus one (i.e. 12, 16, 18, etc.). The design allows for each genotype to have the same probability of being surrounded by any other genotype of the selected group. Knowledge on flowering capacity is important in order to select a group of materials with synchronized flowering. When there are considerable differences we have to implement delayed planting and/or pruning of the earliest flowering genotypes. At harvest the seed from different plants of the same genotype are combined together and named as a half-sib family (SM).

For control crosses, we plant 10 to 20 plants depending on the flowering capacity of the genotype in question. Each flower has the potential to produce 3 seeds, but in average we obtain no more than 1 seed per cross. This is due to the sensitivity of the stigma to the manipulation during pollination. Seeds from the same cross are mixed together and name as a full-sib family (CM).

Results: A total of 117442 recombinant seeds were produced during the period 1998-99 (Table 2.2). The number of materials shipped to different regions is presented on Table 2.3. Genetic stocks are being built for traits of high priority in our project. In the

case of Africa and ACMV resistance, we are developing "backcross" populations between F1 crosses (African x Latin American), and a different African parent. The strategy is to have progenies with 75% African background. Stocks for root quality traits represent a large proportion of our efforts. For cyanide, white flies, post harvest deterioration and bacterial blight we have crossed genotypes representing the extremes in performance in order to map and tag the genetic factors responsible for the inheritance of those traits. In the case of dwarf genotypes we are developing a population with combining genetic factors determining short plant type, in order to start breeding for other agronomic traits with that population.

Table 2.2. Recombinant seed produced within the project (January 1998 - September 1999).

Parental population	Controlled crosses	Polycrosses	Total
Broad adaptation	8201	42680	50881
Africa	3977		3977
Asia	1477		1477
Dwarf plant type	437		437
Resistance to:			
Bacterial Blight	1395		1395
Mites	213	8884	9097
White flies	249		249
Post harvest	1125		1125
Root rot	1781	949	2730
African Mosaic Virus	454		454
Cooking quality	64		64
High carotene content	4611		4611
Adaptation to drought	812		812
Tetraploid cassava	25		25
Crosses to wild species	493		493
Sub-humid tropics		17485	17485
Acid soil savannas		3999	3999
Mid-altitude tropics		11922	11922
Highland tropics	616	5593	6209
Total	25930	91512	117442

Achievements:

- Considerable amount of recombinant seeds produced.
- Large proportion of our work shifted to specific traits, through the development of genetic stocks, and pre-breeding populations.
- More targeted crosses with landraces build during this period, upon request from National Programs.

Activity 2.3. Generation and distribution of advanced breeding materials for Asian National Programs.

Rationale: Breeding for Asia has mainly centered on the issue of increased productivity of dry matter per hectare. Yield and root dry matter concentration have been the primary traits for selection, with almost none emphasis given to pests and diseases, or cooking quality. The work developed in Asia for 15 years, has revealed the possibility to select for broader adaptation of genotypes. We have the case of Rayong 60 and Kasetsart 50 with good performance in a range of Asian countries. The production of germplasm for Asian has been moved from Thailand to Colombia due to budget constraints. However, because of the attained by several NARS in Asia, the provision of recombinant material from Colombia can satisfy their needs. A CIAT soil scientist based in Thailand still coordinates the cassava network for Asia.

Materials and Methods: The same approaches as the ones implemented for other regions of the word (polycrosses and controlled crosses) have been implemented, but a greater proportion of segregating progenies from controlled crosses is usually produced. Elite germplasm identified from the evaluations across the Asian region is periodically sent back to Colombia, to be used as a parental material in new cycles of selection.

Results: Close to 100,000 were produced during the last 3 years of activities combining the activities in Thailand and Colombia. About thirty percent of that seed was transferred to 4 National Programs in the region and to CIAT-HQ (Table 2.4). The retirement of our cassava breeder stationed in Thailand, implies that starting in 1998 an increasing proportion of recombinant seed originated in CIAT-HQ. However, it will take longer for Asian National Programs to receive materials from CIAT. In the future, we foresee that the flux of improved germplasm between CIAT-HQ, and the Thai breeding program will continue, and it will be through us that other National Programs will receive progenies involving the latest selections in Thailand.

Table 2.3. Shipments of recombinant seed produced within the project (Jan. 1998 – Jul. 1999).

Continents	Genotypes in-vitro	Crosses (families)	Plants (in-vitro)	Seeds in shipment
Latin America				
In-vitro	52		135	
Hybrid seed		125		13793
Asia				
In-vitro	23		112	
Hybrid seed		245		28011
Africa				
In-vitro				
Hybrid seed		77		37000
Europe + NA				
In-vitro	1		5	
Stakes	9[¶]		168[¶]	-.-
Total				
In-vitro	74		252	
Stakes	9[¶]		168[¶]	
Hybrid seed		447		62876

[¶] Stakes sent to England for quarantine evaluation and then shipment to IITA in Africa or for studies of post-harvest physiological deterioration at the University of Bath (England).

Table 2.4. Cassava F1 hybrid seeds from CIAT (Thailand and/or Colombia), distributed to Asian programs (1996-98).

Country	1996	1997	1998	1999	Total
Indonesia	2740	1250	4621	5893	8611
China	2,292	1,500	4955	2667	8747
Vietnam	3,309	2,940	3176	3176	9425
Thailand			4955	3523	
Total	**9691**	**6790**	**12752**	**15259**	**29233**

Achievements:
- Unrestricted support and collaboration from the Thai breeding program.
- Use of the most elite genotypes in crosses.
- Distribution of segregating progenies of high value for Asian National Programs.

Activity 2.4. Shipment of the best performing varieties in Asia to Colombia, to continue the breeding program from Asia, at CIAT-Palmira. Visits to the region.

Specific Objective:

a) to assume the breeding activities for the Asian Region in CIAT-HQ, and partially replacing the activities carried out by the breeder previously based in Thailand.

Rationale: Asia has benefited considerably from CIAT's breeding activities. The work had the support of a scientist based in Thailand. Starting in 1998, it was decided to concentrate the breeding activities at HQ. Both financial limitations and the growth of Asian NARs supported this decission. A CIAT soil scientist, however, is still based in Thailand, coordinating a cassava research network and serving as a liason between Asian and CIAT-HQ. Asia remains an important cassava growing area with valuable and dinamic experience in uses of this crop for industrial processes.

Materials and Methods: During this year we continued the shifting process started in 1998. Arrangements have been made for introducing back the best performing clones in Asia to Colombia, so they can be used as parental materials for new crosses. Several outstanding genotypes are already in Colombia, including MTAI-8, which is the best performing genotype in the Northern Coast of Colombia (see results of Activity 2.5) and has been included as a parent, not only for germplasm targeted for Asia, but also for Latin America. MTAI-8 is an excellent example of the advantages and feasability of doing breeding work in one region which is then transferred to other regions. The same concept is true for the exchange of germplasm between Latin America and Africa. In every case, however, particular local biotic stresses (ACMV, Bacterial Blight, Thrips, etc.) may limit in some cases the direct usefulness of an introduced clone in a given region.

Results: At this point we already have most of the elige Asian germplasm at HQ. The very newest clones to be brought back will be identified and arrangements made during the nexs Regional Cassava Meeting that will take place in Viet Nam this February. This event will also serve as an opportunity for meeting personally the most influential asian scientist working with cassava. It will also be a perfect event for implementing an efficient strategy for delivery and distribution of new germplasm, developed at CIAT-HQ in Asia.

Achievements:

- We are now in the situation of producing new recombinant seed specially targeted for the Asian needs, using elite germplasm for that region.
- A recognition to CIAT contributions to cassava production in Asia was given through two special awards to our scientists in the region (K. Kawano). The awards were given by the Thai (K. Kawano) and Chinese Governments (R. Howeler), respectively.

Activity 2.5. Selection of recombinant progenies for broad and specific adaptation within major agro-ecosystems (sub-humid; semi-arid; highland and acid soil savanna).

Specific Objectives:

a) *Modify the evaluation procedure to make it more efficient and to adapt it to the new breeding objectives.*
b) *Evaluate superior germplasm adapted to particular ecosystems.*

Rationale: Our strategy for cassava germplasm development is centered on the development of improved gene pools for specific edapho-climatic zones with importance for cassava production, as defined in Table 2.5. The most relevant ecosystems are the semi-arid and sub-humid tropics, for which we devote the majority of our efforts. The main selection activity is conducted in sites selected to represent the conditions of the target ecosystem. For every genotype that was tested in those sites a copy was maintained at CIAT-HQ. This location is considered to be free of bacterial blight and some important viruses, and to maintain that condition, the introduction of vegetative material from other areas is restricted. In case vegetative material has to be brought to HQ, then it has to pass through quarantine, which usually takes more than a year.

Materials and Methods: For each of the zones we conduct a recurrent selection program, with a progressive set of stages as described in **Figure 2.1.** As the stages progress, we give more emphasis to traits of lower heritability, because we have more planting material for each genotype, and the evaluation can be conducted in bigger plots with replications. Certain selection criteria are of general importance across ecosystem (i.e. yield potential, dry matter content), while others are specific for each ecosystem (i.e. pest and diseases).

Traditionally, the progenies generated from the crossing blocks (***F1***) were planted in screen houses and transplanted to the field after 2 months at CIAT. At 6 months after planting, 2 stakes were harvested from each plant and given a consecutive number according to the plant. One of the stakes was planted at CIAT, the other one was planted at the main selection site (***F1C1***). Selection was conducted at harvest on individual plants at the main selection site. Planting material taken from the selected genotypes, at CIAT, was used to establish a non-replicated, 6-plant plot, both at CIAT

and at the main selection site (***clonal evaluation*** stage). Evaluation was done using the central 3 plants. Selections were transferred to the following stage (***preliminary yield trial)*** and planted in non-replicated, 20-plant plots. Evaluation was done in the central 6 plants, and selections were then passed to the advanced yield trials at 1 or 2 sites, with 3 replications of 25-plant plots. Genotypes selected over 2 consecutive years at the advanced yield trial level were consider as **"*elite genotypes*"** and incorporated in the germplasm collection and the crossing blocks. Since each year a new breeding cycle was initiated, all the stages were simultaneously being conducted in each site.

Some modifications will be implemented for the F1 plants already in the field. A major constraint of the traditional evaluation methodology was that the first two stages of selection (F1C1 and the clonal evaluation) were based on non-replicated plots. In addition large amount of material was maintained at HQ just to have duplicates of the very few materials that each cycle would reach the status of *"elite genotype"*. Therefore some changes are currently being implemented that will speed up the selection process and allow for the evaluation of larger number of progenies. The main changes are as follows:

1) The F1 plants will be grown for 10 months rather than 6. At that age they can produce up to 8-10 stakes. The stakes will be sent to the proper evaluation site for the *clonal evaluation*. This implies that the F1C1 stage is eliminated and that no duplicate of each genotype is maintained at CIAT-HQ.

2) The *clonal evaluation* will be based on eight plants, rather that six as before. An important modification is that this *clonal evaluation* will be carried out in two stages: at the normal harvest time only two plants will be harvested to measure % of dry matter. This trait varies considerably with the time of harvest or age of the plant. Therefore to estimate it correctly, the plants have to be harvested at the proper time. The remaining six plants of each plot will be harvested just prior to normal planting time (one week before). Yield potential will be estimated visually (as had been done traditionally at the F1C1 and clonal evaluations stages), based on the volume of roots produced by the six plants. Few other traits will also be taken using visual scores: plant architecture, foliar health (for insects and diseases separately), above ground biomass (for a rough estimate of harvest index), and root aspect. A selection index software developed at CIMMYT (G. Edmeades and J.Crossa, personal communication) will be used to make an efficient and fast selection of the approximately 2500 genotypes evaluated at this stage for each ecosystem.

3) The changes described above allow taking stakes from six plants, rather than three, as in the past. These six plants will produce more that 30 cuttings, which will be used for the first replicated trial based on three replications and two row plots with ten plants per plot. It is recognized that this evaluation will result in some competition effect among neighboring plots. However the number of replications will neutralize most of these effects. Also, row spacing between plots can be increased and the plant to plant distance within the plot reduced. This will maintain the density unchanged, while favoring competition among plants from the same genotype.

4) A final important modification to the evaluation process is that data will be taken and analyzed for **all** the progenies evaluated. In the past, data was taken only for those families that went beyond the *clonal evaluation* stage. Therefore it was impossible to estimate combining ability of parental materials, because most of the crosses did not produce data (they had been discarded in the field before any data was taken). The changes introduced will allow us to base the selection of the parental materials on its breeding value (combining ability) rather that its performance *per se*, or empirical appreciation of their potential as progenitor.

The main advantages of the new evaluation scheme can be summarized as follows:

- The duplication of materials maintained at CIAT-HQ is avoided until they reach status of "elite genotype".
- The selection of large number of segregating progenies, at the F1C1 stage, which is based on single plant observations, is avoided.
- The time required to reach the stage of replicated trials is minimized.
- The total length of each cycle of selection reduced by almost a year.
- Data records will allow for selecting parental material based on combining ability.
- The total cost for each cycle of selection should be reduced.
- Selection will be less subjective by using appropriate software (specifically developed for that purpose).

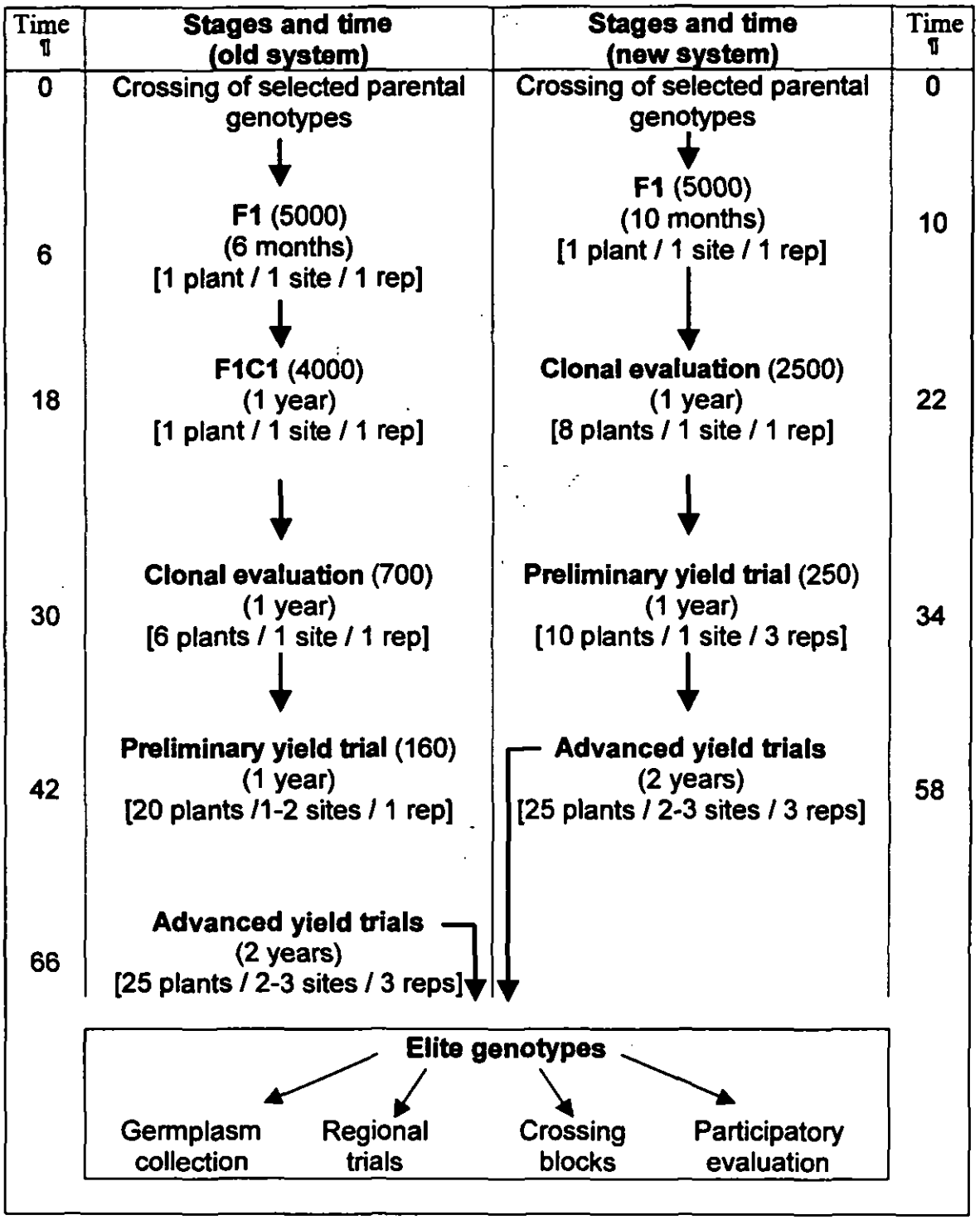
Results: In order to summarize results from the last year of work, we present a description of the main activities at the four most important ecosystems: semi-humid tropics (Tables 2.6 to 2.15); acid-soil savannas (Tables 2.16 to 2.21); mid-altitude tropics (Tables 2.22 to 2.27); and highland tropics (Tables 2.28 to 2.30).

An increasing emphasis has been given to dry matter yield (t/ha) in the last few selection cycles. On the other hand, less weight has been given to HCN. These changes are a response to the demand from the industry. Dry matter yield is the resultant from a relatively low heritability trait (root yield) and an intermediate to high heritability trait (root dry matter %).

Table 2.5. Main ecosystems for cassava production, representative production regions, and main breeding sites.

Description	Representative Countries/Regions	Breeding Sites
Sub-humid tropics (800- 1500 mm /year, bimodal rainfall distribution)	Colombia (Atlantic Coast & Santanderes); NE Brazil; NE Thailand; Domin. Rep.; N. Venezuela; Mexico (Yucatan Peninsula); subhumid belt of Africa;	Media Luna Sto Tomás Betulia
Acid soil savannas (1500 – 3000 mm/year, short dry period, low pH)	Plains of Colombia & Venezuela; Brazil (Cerrado); Mexico (Tabasco); Cuba; W Africa savannas; Philippines; Panama (Ocu)	La Libertad Matazul Sder de Quilichao
Humid tropical lowlands (above 3000 mm/year, no clear dry period)	Amazon basin (Brazil, Colombia, Peru); West Java & Sumatra; Malaysia; S. Vietnam; Equatorial West Africa	La Libertad
Mid-altitude tropics (800-1400 masl)	Andean zone; central Brazilian highlands; mid-altitude areas of Nigeria, Cameroon, East Africa	Palmira Sder de Quilichao
High-altitude tropics (1400-2000 masl)	Andean zone; Rwanda; Burundi	Popayán Mondomo
Subtropics (latitudes higher than the tropics)	S Brazil; Argentina; China; N Vietnam; Cuba; Paraguay; S Africa	Sta Cat. (Brazil)
Semiarid (below 800 mm/year, unimodal)	NE Brazil; NE Colombia; (Guajira) semiarid belt of West Africa; Tanzania; Mozambique; Ecuador (Coast)	Guajira Sto Tomas NE Brazil

Figure 2.1. Basic cassava breeding scheme applied for each of the priority ecosystems.



¶ Time, in months, after the harvest of recombinant seed.

Table 2.6. Trials conducted in the sub-humid ecosystem during the 1998-1999 season.

Trial	Location	N° of Genotypes	N° of Reps	C.V. (%) (for yield)
F1C1	Caracolí	491 (1)	1	-.-
Clonal evaluation	Caracolí	251 (6)	1	-.-
Preliminary yield trial	Media Luna	87 (20)	1	-.-
Advanced yield trials	Caracolí	100 (25)	2	13.97
	Caracolí	35(25)	2	12.23
	Media Luna	80 (25)	2	22.97
	Chinú	80 (25)	2	25.98
Regional trials	Caracolí	30 (25)	3	13.28
	Media Luna	30 (25)	3	31.69
	Sahagún	27 (25)	2	41.01
	Chinú	30 (25)	3	20.30
	Cereté	27 (25)	2	40.27
Seed increase	Various	45	10 ha	-.-

Table 2.7. Best performing genotypes from the **regional trial** conducted at **Caracolí** (sub-humid tropics) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
TAI-8	0.75	38.0	35.7	**13.5**	8.0
SM 1411-5	0.61	35.1	35.0	**12.3**	8.0
CM4919-1	0.79	36.7	33.1	**12.1**	8.0
CM6754-8	0.77	33.5	35.0	**11.7**	7.0
Trial Mean	0.64	35.8	28.0	**10.1**	6.8
Checks	0.62	35.9	21.4	**7.6**	7.0

Table 2.8. Best performing genotypes from the first of two **advanced yield trials** conducted at **Caracolí** (sub-humid tropics) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
TAI-8	0.79	38.3	38.8	**14.8**	8.5
SM 1411-5	0.64	35.6	35.8	**12.8**	8.0
SM 1438-2	0.64	38.9	32.8	**12.8**	6.5
CM 6754-8	0.81	33.6	36.7	**12.3**	7.0
Trial Mean	0.64	36.0	26.7	**9.6**	6.6
Checks	0.61	34.1	23.8	**8.1**	6.6

Table 2.9. Best performing genotypes from the second of two **advanced yield trials** conducted at **Caracolí** (sub-humid tropics) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
SM 2192-6	0.71	37.8	34.7	**13.1**	9.0
TAI-8	0.80	36.5	34.1	**12.4**	9.0
SM 1778-50	0.67	38.3	32.2	**12.3**	8.0
SM 1521-10	0.69	33.9	36.0	**12.2**	4.5
Trial Mean	0.62	34.7	25.2	**8.7**	7.3
Checks	0.61	32.3	22.4	**7.2**	7.7

Table 2.10. Best performing genotypes from the **regional trial** conducted at **Media Luna** (sub-humid tropics) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
TAI-8	0.69	33.6	20.4	**6.9**	9.0
SM 1411-5	0.63	36.6	17.4	**6.3**	8.0
CM4919-1	0.76	34.9	19.4	**6.8**	7.5
CM6754-8	0.71	30.3	19.7	**6.0**	8.0
Trial Mean	0.65	34.0	14.7	**5.0**	6.5
Checks	0.64	34.0	12.7	**4.3**	5.5

Table 2.11. Best performing genotypes from the **advanced yield trial** conducted at **Media Luna** (sub-humid tropics) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
SM 1565-17	0.72	30.8	31.0	**9.6**	6.0
SM 2081-34	0.70	31.6	29.8	**9.6**	8.5
TAI-8	0.71	32.1	26.2	**8.5**	9.0
SM 1656-7	0.65	35.7	23.3	**8.4**	6.5
Trial Mean	0.66	33.1	17.5	**5.8**	6.9
Checks	0.66	30.8	14.6	**4.5**	6.9

Table 2.12. Best performing genotypes from the **regional trial** conducted at **Chinú** (sub-humid tropics) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
TAI-8	0.61	36.0	26.7	**9.6**	9.0
CM 3306-19	0.65	34.8	24.9	**8.7**	5.5
CM 6754-8	0.72	33.6	24.3	**8.2**	7.0
CM 8027-3	0.60	34.1	23.6	**8.2**	8.5
Trial Mean	0.52	35.3	16.7	**5.9**	6.6
Checks	0.52	35.3	13.6	**4.8**	5.4

Table 2.13. Best performing genotypes from the **advanced yield trial** conducted at **Chinú** (sub-humid tropics) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
SM 1652-21	0.62	33.9	30.9	**10.5**	9.0
SM 1521-10	0.63	34.0	29.2	**10.0**	7.0
SM 1657-12	0.57	35.8	23.7	**8.4**	8.5
SM 1511-14	0.50	35.5	22.9	**8.1**	9.0
Trial Mean	0.49	34.6	14.9	**5.2**	7.3
Checks	0.51	32.7	11.5	**3.8**	6.8

Table 2.14. Best performing genotypes from the **regional trial** conducted at **Cereté** (sub-humid tropics) during the 1998-99 season.

Genotype[¶]	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
MBRA 384	0.57	34.7	41.6	**14.5**	5.0
CM 3555-6	0.42	36.8	38.3	**14.1**	6.0
CM 6754-8	0.43	36.0	37.6	**13.6**	7.5
CM 6182-8	0.51	37.8	36.8	**13.9**	4.0
Trial Mean	0.47	36.0	25.9	**9.5**	6.2
Checks	0.42	36.7	24.3	**9.3**	5.0

[¶] MTAI-8 was not included in this trial due to lack of seed.

Table 2.15. Best performing genotypes from the **regional trial** conducted at **Sahagun** (sub-humid tropics) during the 1998-99 season.

Genotype[¶]	Harvest Index (%)	Dry matter (%)	Fresh. Roots (t/ha)	**Dry matter (t/ha)**	HCN: (1=low 9=high)
SM 1438-2	0.58	38.1	14.0	**5.3**	9.0
CM 3555-6	0.62	37.7	14.5	**5.0**	5.5
CM 6754-8	0.76	34.8	13.3	**4.6**	9.0
SM 805-15	0.63	37.1	12.4	**4.6**	8.5
Trial Mean	0.56	36.2	8.0	**4.6**	7.8
Checks	0.50	36.4	5.3	**1.9**	6.9

[¶] MTAI-8 was not included in this trial due to lack of seed.

Sub-humid tropics: A group of selected genotypes (Tai-8, SM 1411-5, CM6754-8 and CM4919-1) consistently showed an outstanding performance in the different environments where the trials were carried out. The consistency of their superiority reinforces the conclusion that these clones are, indeed genetically superior, and therefore they have been included as parents in the diallel study (Table 2.1). In two cases, however, (CM4919-1 and CM 8027-3) promising genotypes were not included because they fail to flower under normal conditions at Palmira. For the advanced yield trials and the regional trials the average superiority of the best four clones over the checks was 95.1 and 100.6%, respectively. Likewise, the mean superiority of all the genotypes evaluated in those trials over the respective checks was 26.1 and 29.1%. This suggest that good progress will be attained as a result of these selections.

Acid-soil savannas: As in the case of sub-humid environments the superiority of some genotypes was also very consistent and clear for this ecosystem (CM 6740-7 and CM 4574-7). Those clones have been included as parental material for the diallel study targeting the acid-soil savanna environment. Mean superiority of the best four genotypes and of all the genotypes included in the regional trials over the checks were, 129.2 and 22.5 %, respectively. It was found that CM 4574-7 posses good resistance to root rots, based on the regional trial carried out at Granada. It should be pointed out that the checks used for these trials (Brasilera and Catumare are, respectively a CIAT introduction and a CIAT variety released for the region).

Table 2.16. Trials conducted in the acid-soils ecosystem during the 1998-1999 season.

Trial	Location	N° of Genotypes	N° of Reps	C.V. (%) (for yield)
F1C1	La Libertad	309 (1)	1	-.-
Clonal evaluation	La Libertad	458 (6)	1	-.-
Preliminary yield trials	La Libertad Matazul	161 (20)	1	-.-
Advanced yield trials	La Libertad Matazul	74 (25)	3	28.4
Regional trials	San Martín	20 (25)	3	21.7
	Matazul	20 (25)	3	16.7
	Granada	22 (25)	3	43.4¶
	La Libertad	20 (25)	3	21.0
	La Libertad	25 (25)	3	§
Seed increase	La Libertad	35	2.5 ha	-.-

¶ High incidence of root rot. Allowed for the identification of a genotype with excellent level of resistance.

§ Trial not analyzed yet.

Table 2.17. Best performing genotypes from the first of two **regional trials** conducted at **La Libertad** (acid-soil savannas) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
CM 6740-7	0.61	35.0	31.4	**11.0**	5.7
CM 6921-3	0.62	36.6	29.2	**10.7**	7.3
SM 1821-7	0.71	34.4	30.7	**10.6**	6.7
CM 4574-7	0.63	37.0	27.8	**10.3**	8.0
Trial Mean	0.57	34.8	17.6	**8.5**	6.6
Checks	0.71	35.7	27.0	**9.7**	6.7

Table 2.18. Best performing genotypes from the second of two **regional trials** conducted at **La Libertad** during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
SM 2219-11	0.64	35.3	32.2	**11.4**	6.5
SM 1152-13	0.59	37.5	29.2	**11.0**	7.0
SM 1363-11	0.57	38.1	28.1	**10.7**	9.0
CM 6740-7	0.59	37.0	28.1	**10.4**	4.5
Trial Mean	0.56	34.5	19.0	**6.6**	7.0
Checks[¶]	0.55	31.9	13.4	**4.4**	6.5

[¶] Best checks: Brasilera (7.3 t/ha dry matter) and Catumare (8.2 t/ha dry matter).

Table 2.19. Best performing genotypes from the **regional trial** conducted at **Granada** (acid-soil savannas) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
CM 4574-7[¶]	0.41	31.0	15.4	**4.8**	8.0
CM 6740-7	0.38	30.5	9.4	**2.9**	6.5
SM 1697-1	0.33	29.2	7.1	**2.1**	7.5
BRA 97	0.32	28.8	7.2	**2.1**	6.5
Trial Mean	0.21	28.3	4.1	**1.2**	7.2
Checks [§]	0.16	27.4	3.0	**0.8**	6.4

[¶] High incidence of root rot, allowed for the finding that CM 4574-7 posses high level of resistance to this problem.

[§] Best checks: Brasilera (0.5 t/ha dry matter) and Catumare (1.3 t/ha dry matter).

Table 2.20. Best performing genotypes from the **regional trial** conducted at **San Martín** (acid-soil savannas) during the 1998-99 season [¶].

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
CM 4574-7	0.60	34.6	29.3	**10.1**	8.0
CM 6438-14	0.58	35.9	28.0	**10.1**	7.0
SM 627-5	0.64	32.9	29.8	**9.8**	6.3
CM 6975-14	0.59	33.7	26.4	**8.9**	6.0
Trial Mean	0.58	33.7	22.2	**7.6**	6.2
Checks [§]	0.57	33.4	22.6	**7.6**	6.4

[¶] CM6740-7 was not planted at this location due to lack of seed.

[§] Best checks: Brasilera (9.3 t/ha dry matter) and Catumare (8.9 t/ha dry matter).

Table 2.21. Best performing genotypes from the **regional trial** conducted at **Matazul** (acid-soil savannas) during the 1998-99 season.

Genotype [¶]	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
SM 1794-18	0.63	38.2	27.3	**10.4**	8.0
SM 1821-7	0.69	34.4	24.1	**8.3**	8.3
CM 6438-14	0.59	35.6	20.5	**7.3**	8.0
SM 1697-1	0.64	32.8	21.0	**7.0**	6.0
Trial Mean	0.57	33.7	15.2	**5.2**	7.3
Checks [§]	0.54	32.7	12.2	**4.1**	6.7

[§] Best checks: Brasilera (3.6 t/ha dry matter) and Catumare (6.8 t/ha dry matter).

[¶] Genotypes CM6740-7 (4.0 t/ha) and CM4574 (5.5) were 14th and 11th, respectively.

Table 2.22. Trials conducted in the mid-altitude ecosystem during the 1998-1999 season.

Trial	**Location**	**N° of Genotypes**	**N° of Reps**	**C.V. (%) (for yield)**
F1 Seed planted	Palmira	15011 (1)	1	-.-
F1C1	Palmira	800 (1)	1	-.-
Clonal evaluation	Palmira	645 (6)	1	-.-
Preliminary yield trial	Palmira Quilichao	212 (20) 32 (20)	1 1	-.- -.-
Advanced yield trials	Palmira (A) [§] Palmira (B) [§] Quilichao	128 (25) 90(25) 48 (25)	3 3 3	18.3 [¶] 40.2
Regional trials	Palmira	26 (25)	3	16.9
Seed increase	Palmira Quilichao	100 5	12 ha 3.5 ha	-.- -.-

[§] (A) and (B) refer to plantings of the first and second semester, respectively.

[¶] Trial not harvested yet.

Mid-altitude tropical region: this ecosystem shares many characteristics with the acid-soil savannas. Acid soils are frequently found (i.e. Quilichao), and the two most prevalent diseases (Bacterial Blight and Super Elongation) are common to both

environments. It is not surprising, therefore, that CM 6740-7 is also an outstanding in this region. For this ecosystem we are selecting for high dry yield matter (for which SM 1219-9 has shown an outstanding performance) but also for good cooking quality (for which MPER 183 has a very promising future). Tables 2.26 and 2.27 present the yield potential obtained at semi-commercial/commercial plots from private sector collaborators. Those results are mainly for industrial cassava clones. The genotype SM 1219-9 produced around 90 t/ha of fresh roots in a seed increase plot at Almidones Nacionales (not shown in Table 2.26). It has shown excellent performance in several trials (see Table 2.23), therefore, it was decided to use it as a parental genotype for the next cycle of selection, not only for the mid-altitude, but also for the acid-soil savannas and sub-humid tropics (Table 2.1).

Table 2.23. Best performing genotypes from the **regional trial** conducted at **Palmira** (mid-altitude tropics) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
SM 1219-9	0.61	39.3	46.0	**18.1.**	8.3
SM 1602-13	0.56	37.8	42.3	**16.0**	5.3
MPER 183	0.59	34.6	42.9	**14.9**	4.3
SM 1565-17	0.55	35.6	41.8	**14.9**	4.3
Trial Mean	0.48	38.4	31.3	**12.0**	5.6
Checks	0.46	36.7	26.2	**9.3**	5.3

Table 2.24. Best performing genotypes from the **advanced yield trial** conducted at **Quilichao** (mid-altitude tropics) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
SM 1562-11	0.61	40.5	19.7	**7.9**	4.0
SM 1557-26	0.52	41.8	18.1	**7.6**	5.0
CM 6438-14	0.60	40.1	18.7	**7.5**	3.5
SM 1871-26	0.68	41.1	18.3	**7.5**	7.0
Trial Mean	0.59	35.9	11.9	**4.3**	6.6
Checks	0.57	33.4	7.8	**2.6**	6.9

Table 2.25. Best performing genotypes from the **advanced yield trial** conducted at **Palmira** (mid-altitude tropics) during the 1998-99 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
SM 2141-1	0.55	40.3	48.7	**19.6**	8.6
SM 1557-26	0.62	39.0	42.8	**16.7**	6.5
CM 6438-14	0.57	41.2	39.1	**16.2**	8.0
SM 1871-26	0.55	37.9	41.6	**1.57**	5.0
Trial Mean	0.48	37.8	29.0	**11.0**	6.3
Checks	0.41	33.1	19.5	**6.6**	6.5

Table 2.26. Yield performance of promising industrial cassava clones in commercial *production* at Almidones Nacionales farm in Quilichao.

Genotype	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
CM 6740-7¶	37.0	51,70	**19.13**	5.1
CM 5655-4	42.0	42.71	**17.94**	4.7
SM 653-14	40.6	43.50	**17.66**	4.6
MCOL 1505	38.6	44.13	**17.03**	5.0
SM 909-25	36.0	47.20	**16.99**	4.0
"Parrita"	37.0	44.64	**16.52**	4.0
SM 653-14	35.6	43.00	**15.31**	3.0
SM 1557-17	32.0	42.50	**13.60**	6.0
Check	38.0	27.0	**10.26**	6.0

¶ Clone CM 6740-7 has an outstanding performance in the *acid-soil savannas*.

Table 2.27 Yield performance of promising industrial cassava clones in seed multiplication plots in two locations in the North of Valle del Cauca.

Genotype	La Unión		Roldanillo		Average	
	Fresh Roots (t/ha)	**Dry matter (t/ha)**	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)	Culinary quality
SM1741-1	58.4	**23.5**	55.8	**21.3**	5.0	Intermediate
SM1278-8	68.8	**27.0**	36.2	**13.4**	2.5	Poor
SM 636-8	56.8	**25.0**	36.6	**14.7**	6.5	Very poor
HMC-1	55.8	**21.8**	45.8	**17.7**	2.0	Good
Checks	**49.2**	**18.3**	**40.9**	**14.0**	**4.5**	**Good**

Table 2.28. Trials conducted in the highland tropics during 1998-99.

Trial	Location	N° of Genotypes	N° of Reps	C.V. (%) (for yield)
Clonal evaluation	Cajibío-98	189 (6)	1	-.-
	Cajibío-99	159 (6)	1	-.-
Preliminary yield trial	Cajibío-98	75 (20)	1	-.-
Advanced yield trial	Cajibío-98	35 (25)	3	21.2
	Cajibío-99	46 (25)	3	33.1
Seed increase	Cajibío-98	13	0.5 ha	-.-
	Cajibío-99	12	0.5 ha	-.-

Table 2.29. Best performing genotypes from the **advanced yield trial** conducted at **Cajibío** (highland tropics), during the 1998 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
SM 1498-4	0.51	36.5	39.5	**14.4**	4.0
SM 1835-15	0.61	35.5	38.5	**13.7**	4.0
SM 1834-20	0.55	34.2	39.2	**13.4**	7.5
SM 616-22	0.45	31.8	39.8	**12.7**	7.0
Trial Mean	0.45	34.2	25.3	**8.7**	4.4
Checks	0.47	32.7	20.7	**6.7**	4.4

Table 2.30. Best performing genotypes from the **advanced yield trial** conducted at **Cajibío** (highland tropics), during the 1999 season.

Genotype	Harvest Index (%)	Dry matter (%)	Fresh Roots (t/ha)	**Dry matter (t/ha)**	HCN (1=low, 9=high)
SM 1058-13	0.56	38.5	26.2	**10.1**	3.5
SM 1834-40	0.57	35.1	26.5	**9.3**	2.5
SM 998-3	0.58	34.1	25.7	**9.0**	2.0
CM 7438-14	0.58	33.5	25.9	**8.6**	2.5
Trial Mean	0.53	34.7	16.5	**5.8**	3.2
Checks	0.55	32.1	12.3	**3.9**	3.5

Highland tropics: because there is relatively less cassava growing in highland environments, the breeding activities for this kind of ecosystem is smaller than for the other three environments. Cassava is generally harvested at 18 months of age. Because of low radiation and lower temperatures, yields tend to be lower than for lower altitude environments. The best four genotypes yielded an average of 120% more than the checks, whereas the mean performance of all the genotypes evaluated yielded 39.3 % more than the checks. These results demonstrate that there are excellent possibilities for improving the genetic component of cassava production in the highlands. It is also important to emphasize that these cassava growing areas are generally characterized by poverty and by few alternatives for the farmers, especially in Colombia.

Finally, Table 2.31 present a list of the genotypes that recently have been postulated to become elite germplasm (1999), separately for each adaptation zone. Most of these materials originate in crosses made around the beginning of the decade. It is important to emphasize that crosses of parental lines specifically oriented for the industry have not reached the regional trial evaluation stage. It is expected, therefore, that in 2 or 3 years a clear increase in dry matter yield will characterize the newly emerging elite clones.

Achievements:

- Several outstanding, genetically superior varieties have been identified for each ecosystem.
- Selected genotypes show excellent stability, by performing above the population mean in most (or all) the trials where they were evaluated.
- The new emphasis for clones specially designed for industrial uses, has been implemented, and the first *industrial genotypes* identified.

Activity 2.6. Distribution of improved germplasm to partner is Latin America and to IITA.

Specific Objective:

a) to deliver genetically improved germplasm (seed or in vitro) to partners in Latin America and Africa (mainly through IITA)

Summary: CIAT has a worldwide mandate for cassava, and has developed a strong capability for developing and delivering technology to partners around the world. An important component of this collaboration is the provision of vegetative and seed materials. Table 2.3 already described the amount, type and destination of different deliveries made since January, 1998. IITA has been a major recipient of these materials which is proving to be extremely useful when introgressed into African germplasm.

Table 2.31. List of genotypes recently nominated as "elite germplasm" from CIAT evaluation trials in different ecosystems.

Genotype	Progenitors		Harvest index	Root yield (t/ha)	% Dry matter	Dry matter yield t/ha	HCN (1= Low; 9= high)
	Female	Male					
Z01 (Sub-humid tropics)							
SM 1657- 12	CM 4209-3		0.71	26.3	34.6	9.1	8.0
SM 1657- 14	CM 4209-3		0.65	24.7	34.5	8.5	8.0
SM 1665- 2	M COL 1505		0.72	22.5	32.1	7.2	8.0
SM 1669- 7	SG 731-4		0.66	19.4	36.2	7.0	7.0
CM 3306- 4			0.50	13.9	35.8	5.0	6.0
CG 1141- 1			0.51	15.3	34.5	5.3	7.0
Z02 (Acid soil savannas)							
CM 6975- 14	CM 2772-3	CM 523-7	0.54	23.9	35.4	8.5	7.0
SM 1779- 8	CG 1355-2		0.51	22.5	33.7	7.6	6.0
SM 1920- 1	CM 4402-4		0.50	19.5	35.1	6.8	7.0
SM 1820- 8	M VEN 77		0.49	18.3	33.8	6.2	8.0
SM 1822- 12	CG 1450-4		0.55	17.3	37.3	6.5	7.0
BRASILERA			0.55	15.6	33.7	5.3	6.0
CM 523- 7			0.51	15.3	35.5	5.4	7.0
Z04 (Mid-altitude tropics)							
SM 1602- 13	CM 489-1		0.60	36.5	37.3	13.6	5.0
CM 8370- 11	CG 1372-6	CM 523-7	0.65	36.2	38.0	13.8	7.0
PER 183			0.59	35.9	33.0	11.9	5.0
SM 1660- 4	M BRA 12		0.59	35.1	37.1	13.0	6.0
SM 1543- 16	CG 1450-4		0.61	30.9	36.3	11.2	9.0
SM 1559- 28	CM 278-3		0.51	30.5	38.2	11.7	7.0
SM 1636- 24	M MEX 1		0.57	30.0	39.4	11.8	6.0
SM 1673- 10	CG 1450-4		0.63	28.7	40.5	11.6	8.0
BRA 12			0.48	23.2	33.6	7.8	7.0
CM 523- 7			0.53	25.4	37.4	9.5	6.0
Z05 (Highland tropics)							
SM 1053- 23	M COL 1522		0.58	25.9	32.2	8.3	5.0
CM 7595- 1	M COL 2261		0.59	24.1	34.5	8.3	4.0
SM 850- 1	M COL 2257		0.47	22.9	34.2	7.8	3.0
SM 998- 3	CG 481-3		0.58	22.2	34.4	7.7	4.0
CM 7138- 7			0.52	20.2	35.5	7.2	3.0
COL 1522			0.39	17.2	31.4	5.4	5.0
CG 402- 11			0.58	23.2	31.3	7.3	6.0
Quilichao							
SM 1557- 26	CM 4729-4		0.54	21.8	38.1	8.3	7.0
CM 523- 7			0.60	24.5	36.5	8.9	7.0

There has been a self imposed limitation on the distribution of vegetative material to collaborators around the world. We had discovered some contamination of materials coming from Brazil with Vein Mosaic Virus. During the first semester of the year CIAT developed an indexing protocol to make sure that vitro-plants are not carrying the virus. Upon developing the protocol, we started delivering vegetative materials by August, 1999.

Activity 2.7. Initiation of physiological studies in collaboration with the **Universidad Nacional de Colombia.**

Specific Objectives:

a) *To develop an agreement of collaboration with* the Universidad Nacional de Colombia, *so we can develop join projects for the benefit of both institutions.*
b) *To reopen the Cassava Physiology Laboratory, which is very well equipped but has not been used after the retirement of CIAT's cassava physiologist.*

Summary: There has always been a very productive association between CIAT and the *Universidad Nacional de Colombia*. However, there is still some room for increased collaboration between the two institutions. We developed and agreement that allows a Professor from the University to invest part of her working hours at CIAT. The agreement allows her to use the physiology laboratory and to develop special projects to take advantage of this well equipped facility. An evaluation of light interception, as a measure of canopy development, has already been completed. In addition, two students have been selected to carry out physiological studies. The areas of research can be summarized as follows: **1)** Flowering induction of recalcitrant genotypes through hormonal treatments; **2)** Monitoring root growth using a capacitance meter (van Beem et al. 1998. Agron. J. 90:566-570); and **3)** Simulation of erosion reduction expected through the use of herbicide resistant cassava varieties.

Achievements:
- An agreement between CIAT and the Universidad Nacional de Colombia has been signed.
- A person had been jointly hired by the two institutions for cleaning and calibrating the different equipment at the lab.
- Two students have been selected to start doing physiological studies in cassava, using the lab and under the supervision of the professor from the University.

Activity 2.8. Evaluation of M. esculenta and related species from the germplasm collection for useful traits, particularly for the natural occurrence of apomixis.

Specific Objectives:

a) To start searching for the natural occurrence of apomixis in the germplasm bank collection.
b) To evaluate the germplasm collection for useful genetic variability.

Summary: CIAT's cassava germplasm bank constitutes a rich source of genetic variability. However, it has not been properly exploited. One of the reasons is that we know little about the properties of the over 6000 accessions making up the bank. A proposal to a private company from the starch sector to finance the evaluation of starch properties (as well as other agronomic traits) in the germplasm collection is well advanced and likely to be accepted. In that case several root traits will be evaluated for the first time.

Apomixis has always been an interesting process to cassava breeders. There has been some reports of apomixis occurrence in the *Manihot* genus (Nassar et al., 1998. Genetics and Molecular Biology 21:527-530). This reproductive abnormality is likely to occur in cassava germplasm introgressed with wild species. The germplasm bank posses accessions collected in areas where the likelihood of natural crossing between *M. esculenta* and other *Manihot* species is high (especially from the Amazon basin). Therefore, we have already started to bag clusters of female flowers, searching for a genotype that will produce seed without pollination. Having apomictic cassava would greatly simplify maintaining genetic stocks unchanged, and also facilitate the exchange of germplasm almost without the risks of introducing diseases.

Achievements:

- We have started to look into the germplasm collection for the natural occurrence of apomixis.
- It is likely that a proposal will be accepted for evaluating (during three years) the germplasm collection for root starch properties, as well as for other agronomic traits.

OUTPUT 3: Work with NARs in Latin America and Asia for the selection, multiplication and dissemination of elite cassava germplasm and diversification of uses.

The overall objective of this activity is to maintain and improve our relationship with NARs. In the last few years there has been an increased interaction with the private sector, this trend is to be strengthened in the future.

Activity 3.1. ***Support planning and execution of germplasm development in collaboration with other research institutions.***

Specific Objectives:

a) *To contribute to the development of cassava research from different NARs by interacting with official institutions..*
b) *To carry out, jointly with NARs, research projects in differently cassava growing regions of the world.*
c) *To establish a regional evaluation trial around Montenegro (Quindío)*

Summary: A major role and strategy for CIAT's success is to interact with different NARs for jointly delivering technology to cassava growers. Without the strong support of NARs, CIAT cannot possibly satisfy many of its goals. It is therefore very important to maintain and, whenever possible, to strengthen the relationship with collaborators around the world. The description below jointly summarizes several of the activities that have been described separately in the project work plan.

During the current year several visits were made to different countries in the region (Brazil, Cuba, Ecuador, Nicaragua, Panamá, Paraguay and Venezuela), as well as in Africa (Nigeria and Malawi). During these visits CIAT's scientist interacted with local researchers inquiring about needs, new developments, results of evaluation trials involving CIAT's germplasm, etc. Some of the trips also served to promote the concept of CLAYUCA (Latin American Consortium for Cassava Research and Development). One trip to the North East of Brazil was to participate for a week in the harvest of different trials of cassava adapted to semi-arid conditions. This is an important activity because this germplasm has been extensively shipped to IITA in Nigeria, with excellent results. The same strategy is followed by the scientist based in Thailand and in charge of interacting with Asian countries.

An important interaction takes place with Colombian institutions. Several activities are carried out jointly with CORPOICA (Corporación Colombiana de Investigación Agropecuaria) mainly at La Libertad (Acid-soil savannas), Montería and Cármen de Bolívar (sub-humid tropics). As a result of this close interaction a joint project on Participatory Plant Breeding is currently carried out by the two institutions (see below

Output 4). Also, four new varieties, originated at CIAT, are being released by CORPOICA this year: CM3555-6; CM3306-19; SGB765-2 and SGB 765-4. The last two varieties were developed using the concept of participatory plant breeding.

Other frequent interaction with local government institutions in Colombia is for the establishment of evaluation trials of elite germplasm in many different sites, throughout Colombia. This modality is very effective for distributing and multiplying elite germplasm from CIAT, disseminating crop cultivation and post-harvest technologies. A list of representative agreements already signed is presented below:

a. Cooperativa Agroindustrial del Tolima (COOPALTOL): October 1998-October 1999.
b. Departamento del Casanare – FENAVI: November 1998 – November 1999.
c. Departamento del Tolima – FENAVI: December 1998 –December 1999.
d. UMATA del Municipio de Montenegro: September 1999 – September 2000.
e. Departamento del Putumayo: October 1999 – October 2000.
f. Fed. Nac. de Cafeteros de Colombia, Antioquia: September 1999, September 2000

CIAT's contribution to cassava production in Africa has been traditionally in collaboration and through IITA (International Institute of Tropical Agriculture) in Nigeria. Three CIAT scientist visited IITA this year and developed several strategies for increasing the collaboration between the two centers. This was particularly important because of the now feasible risk of ACMV (African Cassava Mosaic Virus) appearing and spreading through the Americas. One major goal is to speed up the exchange of cassava germplasm between Africa and Latin America.

A fundamental activity in the interaction between CIAT and NARs, has been the training of scientists in different areas. During the current year, for instance, people from Colombia, China, Nigeria and Venezuela have been trained in different areas of expertise at CIAT. Two Ph.D. students from Africa are doing their research thesis at CIAT, and two other students are starting their work on cassava physiology.

After the serious earthquake that hit the coffee growing area between the cities of Armenia and Manizales, early this year, many initiatives were developed to help this severely affected region. The cassava project was asked to establish a regional evaluation trial, and visit the region to host field days around this trial. CIAT also has collaborated with an important company of cassava producers and processors (Arango Cano y Co., La Tebaida, Quindío). The trial was established in August, 1999.

Achievements:
- CIAT continued the productive collaboration with NARs institutions, not only from the government, but also with NGOs and the private sector.
- There has been a strengthening of CIAT collaboration with IITA, which will allow us to increase (through IITA) our contribution of cassava production in Africa.
- Four new varieties will be released in Colombia.

Activity 3.2. ***Search for financial support for integrated projects in different countries of the region through the establishment of the CLAYUCA consortium.***

Specific Objectives:

a) To contribute to the creation of CLAYUCA.

b) To carry out, jointly with CLAYUCA, the activities to be shared between the two projects.

Summary: By the end of 1998, CIAT decided to invest some resources to encourage the creation of the Latin American Consortium for Cassava Research and Development (CLAYUCA). Members of the consortium contribute with an annual fee (proportional to the importance of the crop in the country). The budget thus obtained is used to satisfy the Consortium own research agenda. It was envisioned that this kind of arrangement would be very effective to complement the activities that the project IP3 cannot carry out because of insufficient funding.

Several trips (within and outside Colombia) were made to promote the idea of CLAYUCA. By April, 1999 the consortium was officially created with the participation of the following countries: Bolivia, Colombia, Cuba, Ecuador and Venezuela; and institutions: CIAT and CIRAD (Center for International Cooperation in Agricultural Research and Development). An interesting characteristic of CLAYUCA is that most of its members are end-users of cassava.

CLAYUCA and CIAT's IP3 project, work tightly united. The main research topics set up by the consortium were: **1)** Mechanization of planting and harvest; **2)** Drying of cassava chips; **3)** Cassava nutrition and fertilization; **4)** Integrated pest and diseases management; **5)** Genetic transformation of cassava to incorporate resistance to herbicide(s) and the Bt gene; and **6)** Production and uses of cassava foliage in animal feed rations.

Achievements:
- CLAYUCA was created and is already operative.
- CLAYUCA has a realistic and ambitious research agenda that, in the end, will benefit cassava growers in many regions of the world.

Activity 3.3. ***Integration of private sector in cassava germplasm development project, particularly in relation with the "Ingenios Yuqueros" (cassava mills) concept.***

Specific Objectives:
a) To contribute to the creation of the first Ingenio Yuquero.
b) To assist with technology and germplasm the operations of the Ingenio Yuquero.

Summary: Commercialization of cassava roots is limited to short distances because of its bulkiness (≈ 65 % water) and its short shelf life (at best one week). Therefore a system was developed that minimized the transport of fresh cassava roots. The main idea of the *Ingenio Yuquero* is the concentration of about 6000 ha of cassava around a drying mill, whose function is to collect the roots, chop them and dry the chips. An important addition to the concept is the association of poultry or swine industries to the *Ingenio*. They would consume the dried cassava chips (replacing up to 50% of the maize currently used for the rations) and return the manure to the system.

By July 1999, the first *Ingenio Yuquero* was created. CIAT had a mayor role contributing to the consolidation of the idea. Many meetings took place in Cali and Bogotá, promoting the idea. Workshops were held at CIAT's facilities, as well. The current share holders of the *Ingenio* are: **Pronaca- Senaca** (Poultry industry, Ecuador); **Monomeros Colombo-Venezolanos** (bi-nacional fertilizer company) ; **COLANTA** Ltda (dairy industry); **GRAVETAL** S. A. (feed industry); **FENAVI** (Poultry Growers Association); **and *Fundación para el Desarrollo Integral del Valle del Cauca*** (representing several farmers).

Currently the Ingenio is financing the development, in collaboration with CIAT, of a pilot artificial drying plant that will serve as a model for the final Ingenio Yuquero. The main characteristics of this drying plant should be: **1)** efficiency in the drying process (cost wise); **2)** adequate release of the cyanogenic glucosides (potentially high in industrial cassava varieties); and **3)** able to dry adequate amount of cassava roots year round. The dried cassava should come to a price equivalent or lower than 70% of the price of maize. To achieve this figure, cassava should produce at least 20 t/ha of fresh roots. This figure is a reasonable one based on results from multiplication plots in different regions of Colombia (Tables 3.5 and 3.6).

The establishment of this drying cassava facility is very important because it will make use of the most advanced technology currently available for cassava. It will be a model for the developing concept of cassava as a cash crop. It will also serve as a model to be used in other areas of Colombia. Currently up to 10 cassava mills have been proposed. Finally, in the case of Colombia, it will allow to replace a substantial fraction of the maize currently imported. For CIAT, the establishment of the *Ingenio* implies the creation of a potentially important source of financial support for our research. For each hectare of cassava related to the *Ingenio*, there will be a fixed contribution of money for CIAT's research and technology development.

Achievements:

- The first "*Ingenio Yuquero*" was created, to a large extent thanks to the commitment and efforts from CIAT.
- Adequate funds have been made available for the development of innovative cassava drying technology.

Activity 3.4. Participation in the development of cassava multiplication scheme in different regions of Colombia.

Summary: The demand for elite germplasm is always high because it represents the basis for research in other disciplines, on-farm testing and regional trials in Colombia. Aside from maintaining all elite genotypes in the germplasm collection, a group of high priority genotypes is kept in plots of at least 50 plants. The ones with the highest demand are multiplied more extensively, reaching plots of up to 1 ha. Our project has been able to supply cuttings to other scientists at CIAT, and to institutions throughout Colombia.

Multiplication plots are established as blocks of 50 or more plants. They are given the best possible management in terms of crop, pest and disease management. The objective is to obtain planting material of prime quality. There are 2 ways to deliver the planting material; one is as 1 m pieces, which gives more security in case there is a dry period ahead and the material needs to be stored; and 20 cm pieces, ready to plant. We recover the production and shipment cost through a basic price.

This multiplication activity is fundamental for the development of the cassava industry. One major drawback of cassava as a crop is its low multiplication rate. Whereas a seed of maize will produce 500 progeny seeds in four months, a cassava plant, on the average, will produce only 8 progeny cuttings in a period of about 10 months. Therefore, the multiplication of elite germplasm has to be planned and financed to accelerate the introgression of new improved germplasm. The list of materials multiplied reflects the strong interest in Colombia, to favor the development of a cassava based industry, both for feed and processed human food.

Tables 3.1 trough 3.4 describe the elite germplasm multiplied in different regions of Colombia, as well as the particular uses given to those genotypes. In Tables 3.5 through 3.10 yield obtained in some of those multiplication plots (up to 11.0 ha) was measured. Results from the Northern Coast of Colombia (Tables 3.5-3.8) demonstrate that the minimum yield of 20 t/ha required by the *Ingenio Yuquero* to produce dried cassava at or lower than 70% of the price of maize is feasible. At Santo Tomás and Caracolí (Tables 3.7 and 3.8, respectively), two samples were taken to estimate the performance of each genotype. Samples were randomly taken and consisted in six neighboring plants. At Chinú the best varieties of the several multiplied were CM 4843-1 and CM6754-8 (See Output 2), which, on average, yielded 28.6 and 27.5 t/ha,

respectively (Table 3.5). CM 4843-1 was the highest yielding material at Santo Tomás (Table 3.7) with 40.3 t/ha of fresh roots. At Media Luna (Table 3.6), one of the best material in the new generation of industrial clones was M TAI 8, with an average yield of 49.5 t/ha. This outstanding clone also performed well at Caracolí (Table 3.8) with a fresh roots yield of about 39 t/ha. CM4919-1 and CG 1141-1 yielded 42.6 and 45.4 t/ha, respectively, at Media Luna (Table 3.6).

Table 3.1. *Seed* multiplication of elite germplasm adapted to the sub-humid tropics in the north coast of Colombia.

Genotype	Sources of seed for elite germplasm			
	MADR	Concent. del Norte	CIAT	Totals
SM 1127-8			0.1	0.1
SM 1433-4	97.5		0.1	97.6
CM 6119-5	9.0		0.1	9.1
CM 3306-19	35.0		0.1	35.1
CM 4919-1		9.0	0.1	9.1
CM 4843-1	6.5	16.5	0.1	23.1
M TAI 8	2.0	34.5	2.3	38.8
M VEN 25		6.0	0.5	6.5
CM 4365-3	5.0		0.1	5.1
CG 1141-1	115.0	28.5	0.1	143.4
CM 3306-4	50.0		0.1	50.1
M COL1505	117.0		0.1	117.1
Brasilera (Betulia)	27.5			27.5
Regional Trials			4.0	4.0
SUBTOTAL	464.5	94.5	7.7	
TOTAL Ha				**566.7**

Table 3.2. *Seed* multiplication of elite germplasm adapted to the acid-soil savannas in the eastern *llanos* of Colombia.

Genotype	Sources of seed for elite germplasm						
	Agriculture Office of the Department		Corpoica	CIAT	Juan Vergara	Marcos Calderin	Totals
	Casanare	Meta					
CM 6740-7	30.0	2.0		3.0			35.0
CM 523-7	20.0	2.0	1.0	2.0		5.0	30.0
HMC-1	150.0						150.0
Brasilera (Granada)	50.0		3.0		50.0		103.0
Regional clones	50.0						50.0
Regional Trials		1.0		5.0	0.5		6.5
SUBTOTAL	300.0		4.0	10.0	50.5	5.0	
TOTAL Ha							**374.5**

Table 3.3. *'Seed'* multiplication of elite germplasm adapted to the mid-altitude tropics in the north coast of Colombia.

Genotype	Sources of seed for elite germplasm							
	Ciat Palmira *Quilichao*	Agrovelez *Jamundí*	Ochoa Hnos. *Buga*	Coopaltol *Tolima*	Cultivares S.A. *La Tebaida*	Almidon. Nacional. *Quilichao*	Other	Totals
CM 849-1		0.4						0.40
CM 5655-4		0.7				10.0		10.70
CM 6740-7	2.0	0.7				20		22.70
CM 7514-7		0.4						0.40
CM 7951-5		0.1						0.10
SM 653-14		0.8				10.8		10.80
SM 909-25		0.4				10		10.40
SM 1210-4		0.7				10		10.70
SM 1219-9		0.7						0.70
SM 1460-1		0.4				10		10.40
SM 1543-16		0.4						0.40
SM 1557-17		0.2				10		10.20
SM 1741-1		0.4						0.40
M BRA 383	1.2	0.8						2.00
M PER 183	1.2	0.2				10		11.40
CM 523-7	1.2	0.2		50		100	1.0	152.40
M BRA 12	1.2	0.2		15		40	5	61.40
CM 3306-4	1.2	0.2		10			10	21.40
MCOL 1505	1			50		18	9	78.00
HMC 1	1		70	40	100	6	54	271.00
Brasilera [1]				35				35.00
Parrita						40		40.00
Bitter						30		30.00
Casa Verde						10		10.00
Chiroza					50			50.00
SUBTOTAL	10	7.9	70	200	150	334	79	
TOTAL Ha								**850.90**

[1] Planted in San Luis (Tolima).

Table 3.4. Description of elite germplasm multiplied in different regions of Colombia.

Genotype	Ecosystem	Root Yield	Dry matter	D. M. yield	HCN	USE
	¶	(t/ha)	%	(t/ha)		
CM 523-7	2 – 3 - 5	20-25 **	38.0	7.6-9.5	-	DP
CM 3306- 4	1 - 3 - 4	20-22 **	36.5	7.3-8.0	-	DP
CM 3306- 9	1 - 3 - 4	20-22 **	36.0	7.2-7.9	+	IND
CM 6740- 7	2 - 4	20-25 **	35.5	7.1-7.8	+	IND
CM 5655- 4	3 - 4	37.1*	37.8	14.6	+	IND
CM 6370- 2	3 - 4	41.3*	35.4	15.1	+	IND
CM 7514- 7	3 - 4	33.6*	41.5	14.2	-	IND
SM 643-17	3 - 4 - 5	31.2*	40.0	12.8	-	IND
SM 653-14	3 - 4 - 5	34.9*	40.5	15.4	-	IND
SM 719- 6	3 - 4	33.0*	38.2	12.7	-	IND
SM 909- 25	3 - 4	39.2*	37.5	14.8	-	IND
SM 1210- 4	3 - 4 - 5	34.2*	40.5	13.5	-	IND
SM 1406- 1	3 - 4	36.9*	37.9	14.4	-	IND
SM1557- 17	2 - 3 - 4	28.7*	36.0	10.2	-	IND
SM 1741- 1	2 - 3 - 4	33.0*	37.9	13.6	+	IND
M BRA 12	3 - 4	20-26 **	35.0	10.0	+	IND
M COL 1505	1 - 3 - 4	20-25 **	37.0	10.2	-	DP
M PER 183	3 - 5	32.9*	32.0	10.5	-	DP
M TAI 8	1	34.2*	33.1	11.3	+	IND
SM 1433-4	1	23.1*	35.1	7.9	+	IND
CM 7514-8	1	21.6*	35.9	8.3	+	IND
SM 1411-5	1	22.8*	34.9	7.9	+	IND
SM 1438-2	1	20.5*	36.1	7.4	+	IND
M BRA 383	4	36.7*	38.1	13.9	-	IND
CM 849-1	3 - 4	39.8*	35.0	13.8	+	IND
SM 1127-8	1	19 – 22 **	33.0	6.3	-	IND
CM 6119-5	1	19.5 – 21.0*	36.1	7.0	+	DP
CM 3306-19	1	21.4 – 24.8*	33.0	7.0	+	DP
CM 4919-1	1	21.3 – 27.0*	34.0	7.2	+	IND
CM 4843-1	1	20.4 – 23.0*	34.0	6.9	+	IND
M VEN 25	1 - 5	20 – 23 **	33.5	9.8	+	IND
CM 4365-3	1	20.6 – 24.0*	34.0	6.5	+	IND
CG 1141-1	1 - 5	20 – 23 **	33.6	6.0	+	DP
CM 7951-5	4	35.0 – 44.0*	34.1	11.9	+	IND
CM 1219-9	4	35.0 – 39.0*	32.3	11.3	-	IND
CM1543-16	4	29.0 – 32.8*	31.7	9.1	+	IND
BRASILERA	2 - 5	22 – 25 **	35.0	7.7	+	DP
PARRITA	4	35 – 40 **	37.0	12.9	-	DP
REG. AMARGA	4	22 – 27 **	38.0	8.3	+	IND
CHIROZA ARM.	6	25 – 30 **	34.0	8.5	-	TBL
HMC1-ICA-P13	3 – 5 - 6	20 – 25 **	33.0	6.6	-	DP

¶ 1 = Northern Coast (Sub-humid tropics); 2 = Eastern savannas (Acid-soil savannas); 3 = Departamento Valle del Cauca (Mid-altitude tropics, non-acid soils); 4 = North of Departamento del Cauca (Mid-altitude tropics, acid soils); 5 = Tolima (Sub-humid, mid-altitude tropics); 6 = Coffee growing region (Intermediate between mid-altitude and highland tropics).

* Root yield based on experimental plots ** Root yield based on pre-commercial or commercial plots.

Table 3.5. Yield of various sources of cassava multiplied in William Guzmán property in the northern coast of Colombia, at Chinú.

Genotype	Area (ha)	Production (t)	Yield (t/ha)
CM 4843-1	0.50	14.341	28.6
CM 6119-5	0.75	9.398	12.5
CM 6754-8	0.25	6.874	27.5
CM 4365-3	0.50	6.977	13.9
Negrita	0.50	11.303	27.6
SM 433-4	0.25	4.928	19.7
Various	0.50	10.688	21.4
Mean/Total	**3.25**	**64.509**	**19.85**

Table 3.6 Yield of various sources of cassava multiplied in Octavio Ropaín (R.I.P.) in the northern coast of Colombia, at Media Luna.

Genotype	Area (ha)	Production (# of sacs)	Yield (t/ha)
M Col 1505	4.00	2040 (55 kg/sac)	28.0
CM 3306-4	5.00	2360 (60 kg/sac)	27.6
CG 1141-1	11.00	7140 (70 kg/sac)	45.4
CM 4919-1	6.00	3600 (71 kg/sac)	42.6
C Col 2215	2.50	123 (71 kg/sac)	3.5
CM 4843-1	3.00	1350 (75 kg/sac)	33.8
M Tai 8	3.25	1760 (65 kg/sac)	49.5
Mean/Total	**34.75**	**-.-**	**32.9**

Table 3.7. Yield of various sources of cassava multiplied at Santo Tomás, in Tomás J. Fontalvo farm. Results are the average of two random samples of six neighboring plants.

Genotype	Dry matter (%)	Fresh root yield (t/ha)	Dry matter yield (t/ha)
CM 4843-1	34.1	40.3	13.8
CG 1141-1	37.5	36.3	13.6
CM 6119-5	37.7	36.1	13.6
CM4365-3	36.9	36.5	13.5
SM 1422-4	36.4	31.1	11.3
MBRA 384	34.3	31.5	10.8
CM 7514-8	31.9	33.0	10.5
MIND 39	34.6	30.3	10.5
SM 805-15	35.5	29.3	10.4
SM 1411-5	36.1	28.6	10.3
SM 1438-2	37.2	27.3	10.2
CM 7514-8	37.3	27.2	10.1
MCOL 1505	36.1	27.9	10.1
CM 8027-3	34.2	29.0	9.9
CM 6758-1	34.5	28.0	9.7
SM 1431-2	36.0	24.4	8.8
CM 6754-8	30.7	28.2	8.6
CM6182-8	37.0	23.2	8.6
SM 1201-5	36.1	23.2	8.4
CM 3306-4	37.6	22.1	8.3
Mean/Total	**35.6**	**29.7**	**10.5**

Table 3.8. Yield of various sources of cassava multiplied in CIAT-managed nursery at Caracolí. Results are the average of two random samples of six neighboring plants.

Genotype	Dry matter (%)	Fresh root yield (t/ha)	Dry matter yield (t/ha)
M TAI 8	38.2	40.4	15.4
M TAI 8	35.0	37.6	13.2
M VEN 25	34.3	36.2	12.4
CM 4919-1	36.5	33.1	12.1
CM 4843-1	33.0	28.3	9.4
CM 4365-3	37.1	25.0	9.3
CM 3306-4	36.9	23.9	8.8
CG 1141-1	35.3	20.2	7.1
M COL 1505	35.0	20.0	7.0
Mean/Total	**35.7**	**29.4**	**10.5**

Information from multiplication plots in the eastern acid-soil savannas was also gathered. According to the data provided in Table 3.2, three and two hectares of CM 6740-7 and CM523-7 were planted and maintained under CIAT's management. In each multiplication plot six random samples of nine plants each were taken and measurements made to estimate fresh root yield, dry matter content and dry matter yield. The information thus obtained is presented in Table 3.9. It is interesting to note the consistency of data from different samples of the same clone. It is also worth mentioning the superiority of the new clone (CM 6740-7), over a widely adapted material (CM 523-7), yielding about 28.5 % more dry matter. As in other regions of Colombia, data from semi-commercial multiplication plots suggest that fresh root yield potentials were above 20 t/ha. This is the threshold minimum for cassava to be able to compete with maize as source of energy in animal feeds.

Table 3.9. Yield of various two elite varieties multiplied at Corpoica La Libertad, in Villavicencio (eastern acid soil savannas). From large multiplication plots six random samples of nine plants were taken and production measured.

Sample	CM 523-7			CM 6740-7		
	Fresh roots yield (t/ha)	Dry matter content (%)	Dry matter yield (t/ha)	Fresh roots yield (t/ha)	Dry matter content (%)	Dry matter yield (t/ha)
1	16.4	39.7	6.53	16.4	38.9	6.40
2	17.6	39.5	6.93	25.6	38.8	9.91
3	20.9	40.5	8.45	22.8	38.0	8.66
4	16.7	40.9	6.82	23.3	37.3	8.71
5	18.3	41.0	7.52	27.3	38.8	10.59
6	16.1	40.8	6.57	27.7	39.0	10.80
Mean	**17.7**	**40.3**	**7.14**	**23.9**	**38.5**	**9.18**

Achievements:

- Almost 1800 ha of elite industrial varieties of cassava have been planted as seed multiplication plots.
- New varieties yielding more than 20 t/ha of fresh roots, for each ecosystem have been identified and are currently being multiplied. Several materials produce in excess of 10 t/ha of dry matter.
- The Colombian government and the private sector recognize the strategic importance of this activity and have decided to finance it.

Activity 3.5. ***Collaboration with different NARs in Asia, particularly Thailand, Vietnam, Indonesia and China.***

Summary: After the retirement of the cassava breeder posted in Thailand, there is a need to start producing germplasm and collaborating with Asian NARs from Colombia, but with the help of the CIAT scientist, based in Thailand, and coordinating the Asian Cassava Network. It has not been possible to visit the region during the current year. Part of the reason is the Asian Cassava Workshop planned for early next year. It was suggested that this would be the ideal conditions to start planning and developing the strategies to satisfy Asian germplasm needs from Colombia. Other issues related to collaboration will be discussed during that meeting.

In the meantime, a scientist from CATAS (Chinese Academy of Tropical Agricultural Sciences (Hainan, China), visited CIAT to receive training in rapid multiplication techniques, tissue culture, seed health and quarantine, and breeding.

OUTPUT 4: Contribute with efforts for evaluation and development of participatory plant breeding in the Northern Coast of Colombia, jointly with CORPOICA.

The overall objective of this activity is to contribute to the amelioration of farmers' living conditions through the increase and improvement of genetic variability in production systems involving cassava in different cropping arrangements.

Activity 4.1: Evaluation and participatory selection of cassava germplasm in five farmers fields and at an experimental station in the Northern Coast of Colombia.

Specific Objectives:

a) *Compare the efficacy of the selections made by farmers and breeders both in farmers' fields and at the experimental station.*
b) *Reduce the time between crossing of selected parental material and the adoption of new cassava varieties.*

Rationale: The past experience in developing a methodology for participatory development of cassava varieties between 1986 and 1991 allowed for the identification of several mechanisms for back-feeding the breeding programs and improving the research. For instance, gaps in the methodology were identified because farmers' participation was not always real and effective. Also, the period of time between the crossing of parental genotypes and the eventual adoption of new varieties by the farmers was found to be too long. This period of time can be reduced if farmers' evaluations were also performed early in the selection process. Based on these observations early evaluation of segregating progenies were performed The method for variety selection used has been developed jointly by ICA, CORPOICA and CIAT.

In this report results from the evaluations on one cycle of selection are presented as the initial stages of the project "Participatory plant breeding with women and small farmers in Africa and Latin America" , financed by DFID.

Materials and Methods: Current participatory breeding research benefits from previous experience in the same region. Five locations were selected based on the criteria established by the methodology. At each of these locations, CADETs (Agriculture Commitee for Technological Development) have been operational for few years, within CORPOICA's new model for participatory research. A description of those sites is summarized in Table 4.1.

Table 4.1. Description of the five locations used for the joint evaluation with farmers of cassava clones adapted to the Caribe Region of Colombia in 1999.

CADET's location	Geographic Location (State)	Fisiography and soils.	Farm size	Land ownership	Main use for cassava
La Colorada	Pivijay (Magdale-na)	Aluvial plains. Moderately to deep soils.	Small farms, up to 5 ha.	Owner.	Fresh market and dried chips
Pasaco-rriendo	Plato (Magdale-na)	Aluvial plains or rolling hills with 7-50 % slope. Moderately deep soils.	Small farms, up to 5 ha.	Owner and rented.	Fresh market.
Bajo del Arroyo.	El Carmen (Bolívar)	Aluvial plains or rolling hills with up to 75 % slope. Very shallow to moderately deep soils.	Small farms, up to 5 ha.	Owner.	Fresh market.
San Jaime	Los Palmitos (Sucre)	Rolling hills with 3-25 % slope. Shallow soils.	Small farms, up to 5 ha.	Owner.	Fresh market.
El Salado	Ciénaga de Oro (Córdoba)	Aluvial plains. Shallow to moderately deep soils.	Small to medium sized farms, up to 10 ha.	Rented.	Dried chips. Starch.
C. I. El Carmen	EL Carmen (Bolívar)	Aluvial plains. Moderately deep soils.	Experimental station	Govmnt property.	Research

Thirty genotypes are evaluated simultaneously by farmers and other user groups, as well as by the breeders. Evaluations are being carried out in the five locations described in Table 4.1. At each site there will be a local check chosen by the farmer, which may vary from site to site and a common regional check (ICA-Costeña). The remaining 28 clones are promising materials developed by CORPOICA or CIAT (Table 4.2). Among these new materials, some are mainly for the fresh market and other for dried cassava chips and/or starch production. At each farmer's trial the normal cultural practices are followed (Table 4.3). All the trials were planted during June, 1999, with relatively good germination (Table 4.4), except for the trial at C. I Carmen de Bolivar, were the stakes from CORPOICA had poor germination percentages. Germination data from the Pivijay area (La Colorada) are not available due to late social order problems that made the area unsafe to visit.

Table 4.2. Clones evaluated in six different locations to compare conventional and participatory breeding in the Caribe Region of Colombia. CORPOICA - CIAT 1999.

Genotype	Origin : year (Institution)
SM 2277-4	Yield Trial: 1998 (CIAT)
CM 8787-5	Yield Trial: 1998 (CIAT)
CM 8777-3	Yield Trial: 1998 (CIAT)
SM 2276-3	Yield Trial: 1998 (CIAT)
CM 7518-1	Yield Trial: 1998 (CIAT)
SM 2275-10	Yield Trial: 1998 (CIAT)
CM 8288-46	Yield Trial: 1998 (CIAT)
SM 2278-2	Yield Trial: 1998 (CIAT)
SM 8472-5	Yield Trial: 1998 (CIAT)
CM 8628-2	Yield Trial: 1998 (CIAT)
CM 8796-1	Yield Trial: 1998 (CIAT)
SM 2777-11	Yield Trial: 1998 (CIAT)
SM 2275-3	Yield Trial: 1998 (CIAT)
SM 2277-14	Yield Trial: 1998 (CIAT)
SMB 2445-9	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
SMB 2277-3	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
SMB 2448-3	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
SMB 2451-5	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
SMB 2447-2	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
SMB 2445-3	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
SMB 2446-8	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
CMB 8472-1	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
CMB 8472-3	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
CMB 8791-11	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
CMB 9024-1	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
CMB 9021-2	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
SMB 2447-8	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
SMB 2278-9	Preliminary Yield Trial: 1998 (CORPOICA, El Carmen)
ICA COSTEÑA	Common check from CORPOICA
Local check	Farmer

Results: The importance of a uniform germination and the problems associated with management of stakes can be clearly seen in data from Table 4.4. Across all five locations, the stakes from CORPOICA (SMB or CMB clones in Table 4.4) had a mean lower germination level than those from CIAT (64.42 vs. 82.8 %). Similarly, across genotypes, the mean germination percentage from farmers' field was considerably higher than at C.I. Carmen de Bolivar (82.4 vs. 38.6 %). The two problems are probably related. Plant development at Carmen de Bolivar may result in stakes produced at the station having lower germination percentage either due to undesirable physiological conditions and/or disease contamination. That there may be some adverse growing condition at the Experimental Station is also reflected by the fact that the lowest germination percentage (9.4) was precisely for materials developed and planted at the station. However, there is another important reason to explain the low germination percentages at Carmen de Bolivar: harvesting was done at the time farmers usually harvest their plots. Unfortunately, planting usually has to wait some time, until the beginning of the rains. During this period of time, the stakes gradually dehydrate and loose germinability. CIAT's stakes had a better germination because a second shipment of stakes was possible.

Table 4.3. Planting systems used in the comparison between conventional and participatory breeding in the Caribe Region of Colombia. CORPOICA - CIAT 1999

Location	Planting systems				Planting date
	Associations	Ridge	Soil preparation	Distances (m)	
La Colorada	Monoculture	No	Disc Harrowing	1.2 x 1.0	June 26
Pasacorriendo	Monoculture	No	Manual. Localized	1.2. x 1.2	June 22
Bajo del arroyo	Cassava + maize	No	Disc plowing and harrowing.	1.2 x 1.0	June 10
San Jaime	Monoculture	No	Disc plowing	1.2 x 1.0	June 3
El Salado	Monoculture	No	Heavy disc Harrowing	1.2 x 1.0	June 8
CI El carmen	Monoculture	No	Disc plowing and harrowing.	1.0 x 1.0	June 20

Table 4.4. Germination percentage of 28 experimental clones and two checks at five locations in the Caribe Region of Colombia. Plantings from June, 1999.

Clone	Pasa-corriendo	Bajo del Arroyo	San Jaime	El Salado	C.I. El Carmen
SM 2277-4	77	90	97	53	96.6
CM 8787-5	100	80	97	90	80
CM 8777-3	68	83	47	80	80
SM 2276-3	100	97	90	90	93.3
CM 7518-1	70	83	90	83	40
SM 2275-10	83	100	90	97	93.3
CM 8288-46	42	80	100	60	50
SM 2278-2	83	100	97	73	100
CMB 8472-5	100	90	93	80	0
CM 8628-2	68	97	90	87	89.9
CM 8796-1	85	90	97	67	70
SM 2277-11	80	93	93	83	100
SM 2275-3	83	97	90	87	93.3
SM 2277-14	70	83	100	97	96.6
Mean	**84.9**	**90.2**	**90.8**	**80.5**	**67.8**
SMB 2446-9	80	80	97	97	0
SMB 2277-3	97	97	100	100	0
SMB 2448-3	90	83	100	97	0
SMB 2451-5	100	97	93	90	70
SMB 2447-2	37	20	97	80	0
SMB 2445-3	100	97	100	90	0
SMB 2446-8	57	80	90	97	0
CMB 8472-1	90	50	33	97	0
CMB 8472-3	n.a.	90	100	70	16
CMB 8791-11	40	77	100	80	0
CMB 9024-1	53	83	87	63	0
CMB 9021-2	53	60	77	60	45
SMB 2447-8	47	43	87	60	10
SMB 2278-9	67	33	97	57	0
Mean	**70.1**	**70.7**	**89.9**	**82**	**9.4**
ICA-Costeña	93	97	100	70	75
Local Check	100	93	100	80	65

A sociologist is carrying out a users group study to define different types, which will then be used through the selection process. The definition of the different types of cassava users will be complete by the end of the year.

Output 5: Disease resistance in cassava.

The overall objective of this output is develop cassava germplasm resistant to different diseases by evaluation and selection of segregating progenies; screening for resistance in farmers fields; developing screening and diagnostic methodologies; studying genetic variability of the pathogens; and interacting with farmers through participatory research.

Activity 5.1. ***Evaluate fifty cassava genotypes and characterize their reaction to twelve CBB pathotypes*** **(Xanthomonas axonopodis** ***pv.*** **manihotis)** ***under greenhouse conditions.***

Specific Objectives:

a) to evaluate the response to CBB of 50 different cassava genotypes at three different locations in the acid soil savannas environments.
b) to evaluate the same response under greenhouse conditions against a set of 12 X. axonopodis isolates.
c) to measure the virulence of the 12 pathotypes.
d) to identify potential sources of resistance to CBB.

Rationale: cassava bacterial blight (CBB) is one of its most important and damaging diseases. CBB is induced by *Xanthomonas axonopodis* pv. *manihotis*, and it is found in many countries in Latin America and Africa. The disease is not prevalent in Asia. CBB is disseminated by infected stakes and favored by humid environments. The occurrence of frequent rains facilitates the spread of the disease from plant to plant. The bacterium shows large genetic variability for virulence, making it difficult to develop clones that are uniformly resistant to all pathotypes.

Materials and Methods: fifty genotypes were characterized for their reaction to cassava bacterial blight (CBB) under greenhouse conditions. Each genotype was inoculated with 12 *Xanthomonas axonopodis* pv. *manihotis* pathotypes from different edaphoclimatic zones in Colombia, Brazil, and Venezuela. In the greenhouse, 30-day-old cassava plants were inoculated by stem injection with a bacterial suspension of 1×10^5 cfu/mL. Disease severity was recorded 10, 24, and 38 days after inoculation.

The same cassava genotypes were also evaluated for their field reaction to CBB, during two different growing cycles (1997 and 1999) at three locations in the acid soil savannas of Colombia (Matazul, Carimagua and Villavicencio), all in the Meta Department. No artificial inoculation was used for these evaluations. Evaluations were based on 5-plant plots and three replications per site. Every five rows, susceptible genotypes were planted to stimulate disease development and spread.

Table 5.1. Disease reaction[a] of cassava genotypes to 12 isolates of *Xanthomonas axonopodis* pv. *manihotis*, causal agent of cassava bacterial blight.

Genotype	Field reaction[a]			Isolates[c]												Total[d]			R + I (%)[e]
	Ca	Ma	Vi	V2	V132	V372	ST1A	ST1B	C10	C261	C277	C285	C421	C616	C763	R	I	S	
CM 3311-4	*R*	-	*R*	I	I	I	I	I	S	I	I	I	I	I	I	0	11	1	91.7
CM 7772-13	*R*	-	*R*	R	R	I	R	I	S	I	R	I	I	I	I	4	7	1	91.7
M Col 2465	-	-		I	R	I	I	R	S	I	R	I	I	I	R	4	7	1	91.7
CM 7670-2	*R*	-	*R*	S	I	I	R	I	S	I	I	I	I	I	I	1	9	2	83.3
M Col 2409	-	-	*I*	I	S	I	R	I	I	R	I	S	I	I	I	2	8	2	83.3
M Ecu 82	*I*	*I*	-	I	I	I	R	I	S	I	I	S	I	I	I	1	9	2	83.3
SG 104-74	*I*	*I*	-	I	I	I	I	S	S	I	I	I	I	I	R	1	9	2	83.3
CM 7661-12	*R*	-	*R*	S	I	I	I	S	I	I	S	I	I	I	I	0	9	3	75.0
M Bra 512	*I*	*I*	*S*	I	S	I	I	I	S	I	I	I	R	S	R	2	7	3	75.0
M Col 2424	-	-	-	I	I	I	I	I	I	S	S	S	I	I	I	0	9	3	75.0
CM 7052-2	*I*	-	*R*	I	S	I	I	S	S	S	I	I	R	I	I	1	7	4	66.7
CM 7666-31	*R*	-	*R*	I	I	S	R	S	S	R	I	I	I	S	R	3	5	4	66.7
M Bra 703	*I*	*I*	-	S	S	I	I	I	I	S	I	I	S	S	I	0	7	5	58.3
M Col 707	*S*	-	*I*	I	I	I	I	S	S	S	I	I	S	I	S	0	7	5	58.3
CM 2177-2	-	*I*	*I*	S	I	S	I	S	S	S	I	I	S	I	I	0	6	6	50.0
CM 7661-15	*R*	-	*R*	R	R	S	I	S	S	S	S	I	I	I	S	2	4	6	50.0
CM 7772-11	*I*	-	*R*	I	S	S	I	S	S	I	S	S	I	I	I	0	6	6	50.0
CM 7772-2	*I*	-	*R*	I	I	I	I	S	S	S	S	I	S	S	I	0	6	6	50.0
CM 7803-1	*R*	-	*R*	I	I	I	I	I	S	S	S	S	S	I	S	0	6	6	50.0
CM 7666-25	*I*	-	*R*	I	S	I	I	S	S	I	S	R	S	S	S	1	4	7	41.7
M Col 2538	-	-	*I*	I	I	I	R	I	S	S	S	S	S	S	S	1	4	7	41.7
CG 1367-1	*I*	-	-	I	I	S	I	S	S	S	I	S	S	S	S	0	4	8	33.3
CM 6975-8	*I*	-	*R*	S	I	S	S	S	I	S	S	I	S	I	S	0	4	8	33.3
CM 7670-4	*R*	-	*R*	I	I	S	R	S	S	S	I	S	S	S	S	1	3	8	33.3
M Col 1030	*S*	-	-	I	S	S	I	S	S	S	S	I	S	I	S	0	4	8	33.3
M Col 2435	-	-	-	S	S	I	I	S	S	S	I	S	I	S	S	0	4	8	33.3
M Col 2441	-	-	-	S	I	S	R	S	S	S	I	R	S	S	S	2	2	8	33.3
SM 1479-8	*I*	*I*	*S*	S	I	S	R	R	S	I	S	S	S	S	S	2	2	8	33.3
SM 909-25	-	-	-	I	-	R	I	-	S	S	S	S	S	S	S	1	2	7	30.0
M Col 2428	-	-	-	I	S	S	I	S	S	S	S	I	S	S	S	0	3	9	25.0
M Cub 74	*S*	*S*	*S*	S	S	I	I	S	S	S	S	I	S	S	S	0	3	9	25.0

Table 5.1 (cont.)

Genotype	Field reaction [a]			Isolates [c]												Total [d]			R + I
	Ca	Ma	Vi	V2	V132	V372	ST1A	ST1B	C10	C261	C277	C285	C421	C616	C763	R	I	S	(%)[e]
CM 7666-10	*R*	-	*R*	S	S	S	S	S	S	S	S	S	I	S	R	1	1	10	16.7
CM 7811-15	*R*	-	*R*	I	S	S	I	S	S	S	S	S	S	S	S	0	2	10	16.7
M Col 2066	-	-	*S*	S	I	S	I	S	S	S	S	S	S	S	S	0	2	10	16.7
M Col 2446	-	-	-	S	I	S	S	S	S	S	S	S	I	S	S	0	2	10	16.7
SM 643-17	-	-	-	S	-	R	S	-	S	S	S	S	S	S	S	1	0	9	10.0
SM 653-14	-	-	-	S	-	R	S	-	S	S	S	S	S	S	S	1	0	9	10.0
SM 1479-8	-	-	-	S	-	R	S	-	S	S	S	S	S	S	S	1	0	9	10.0
Brasileira	*S*	*S*	*I*	S	S	S	I	S	S	S	S	S	S	S	S	0	1	11	8.3
CM 6306-11	*R*	-	*R*	I	S	S	S	S	S	S	S	S	S	S	S	0	1	11	8.3
M Bra 917	*I*	*I*	-	S	S	S	S	S	S	S	I	S	S	S	S	0	1	11	8.3
M Col 2436	-	-	-	S	S	S	I	S	S	S	S	S	S	S	S	0	1	11	8.3
M Col 2490	-	-	-	S	S	S	I	S	S	S	S	S	S	S	S	0	1	11	8.3
M Col 1505	*S*	*S*	*S*	S	S	S	S	S	S	S	S	S	S	S	S	0	0	12	0.0
M Col 2438	-	-	-	S	S	S	S	S	S	S	S	S	S	S	S	0	0	12	0.0
CM 5655-4	-	-	-	S	-	S	S	-	S	S	S	S	S	S	S	0	0	10	0.0
SM 1210-4	-	-	-	S	-	S	S	-	S	S	S	S	S	S	S	0	0	10	0.0
SM 1406-1	-	-	-	S	-	S	S	-	S	S	S	S	S	S	S	0	0	10	0.0
SM 1557-17	-	-	-	S	-	S	S	-	S	S	S	S	S	S	S	0	0	10	0.0
CM 2967-8	-	-	-	S	-	S	S	-	S	S	S	S	S	S	S	0	0	10	0.0
Totals:																			
Resistant				2	3	4	9	2	0	2	2	2	2	0	5				
Intermediate				22	19	19	26	10	5	11	16	19	14	17	12				
Susceptible				26	19	27	15	29	45	37	32	29	34	33	33				
Virulence [f]				52.0	46.3	54.0	30.0	70.7	90.0	74.0	64.0	58.0	68.0	66.0	66.0				

a. Disease reaction: R = resistant; I = intermediate; S = susceptible; - = not determined.

b. Disease reaction during different crop cycles between 1997 and 1999 under field conditions at sites in Carimagua (Ca), Matazul (Ma), and Villavicencio (Vi), respectively (*under field evaluation*)

c. Isolates, respectively, are: V2, V132, V372 = Villavicencio 2, Villavicencio 132, Villavicencio 372 of Meta, Colombia; ST1A, ST1B = Santo Tomás 1A, Santo Tomás 1B of Atlántico, Colombia; CIO 10 of Brasília, Brazil; CIO 261, CIO 277, CIO 285 of Monagas, Venezuela; CIO 421 of Cauca, Colombia; CIO 616 of Meta, Colombia; CIO 763 of Vaupés, Colombia.

d. Total of isolates to which each genotype shows either a resistant (R), intermediate (I), or susceptible (S) reaction.

e. Percentage of isolates to which each genotype shows either a resistant (R) and intermediate (I) reaction.

f. Percentage of genotypes susceptible to each isolate.

Results: the percentage of virulence of 10 pathotypes toward cassava genotypes was more than 50%. The three most aggressive isolates were CIO 10, CIO 261, and Santo Tomás 1B. Twelve varieties were intermediate or resistant to 66.7–91.7 % of the pathotypes used (Table 5.1). No cassava genotype showed resistance to all and every pathotype. Clones CM 3311-4; CM7772-13; and Mcol 2465 showed good levels of resistance (91.7%) with intermediate or resistant reactions to all pathotypes, except for their susceptible reaction to C10. This isolate originates from Brasilia (Brazil). It is unlikely that the segregating materials in the selection process carried out in the acid soil savannas of Colombia have ever been exposed to this isolate. This may explain the generalized susceptibility (only five of the fifty clones showed an intermediate reaction to the isolate). Little is known about the resistance in cassava to *X. axonopodis*. No single major gene has been identified, to support the gene for gene hypothesis. However, some specificity, as revealed by data in Table 5.1 is apparent. The kind of results obtained suggest that horizontal (additive, polygenic) resistance is prevalent, but this horizontal resistance contrary to what occurs in other plant/pathogen associations, shows some degree of specificity. Molecular studies to characterize both *X. axonopodis* virulence and cassava resistance, are currently carried out in the SB-2 project.

Activity 5.2. ***Evaluate sixty elite cassava cultivars for resistance to different Phytophthora species under greenhouse conditions.***

Specific Objectives:

a) *to evaluate the response to six different* Phytophthora *species of 60 elite cassava genotypes under greenhouse conditions.*
b) *to identify potential sources of resistance to* Phytophthora *root rots.*

Rationale: root rots caused by different *Phytophthora* species can be devastating in cassava fields, causing plant death and/or the rotting of roots. CIAT continuously carries out screenings for resistance to this disease. Root rots are favored by soils lacking adequate drainage, with frequent wet soils and cooler temperature. Overly matured cassava varieties are more susceptible to this type of rotting. Typically, many of the smaller feeder and the storage roots are dead and necrotic. As the roots start to decay, the entire plant becomes wilted, defoliates and dies. Infected roots emit pungent foul odors.

Materials and methods: 60 genotypes were inoculated with six isolates from different *Phytophthora* species collected from important cassava-growing regions around Colombia: **P4** is a *P. arecae* isolate, and **P12** is considered to be *P. vignae*. Isolates 27 and 44 were from Barcelona (Quindío Department); 66 from Palmira and 69 from Buenaventura (both from Valle del Cauca Department). Isolate 44 has been identified as belonging to *P. citricola*, based on ITS sequence analysis. Evaluations were carried out under greenhouse conditions, following the methodology developed at CIAT. A randomized complete block design with four repetitions per isolate was used (one plant per repetition).

Table 5.2. Cassava elite genotypes evaluated for tolerance of different *Phytophthora* species under greenhouse conditions [a].

Genotype	Isolate [b]						Resistance (%)
	27	44	66	69	P4	P12[c]	
CM 2772-3	S	S	MR	I	S	R	17
M Col 1522	I	S	I	R	MR	MR	17
SM 1697-1	R	MR	MR	MR	I	MR	17
CG 1141-1	R	MR	R	MR	MR	MR	33
CG 402-11	I	S	R	MR	HS	R	33
CM 489-1	R	MR	R	I	HS	S	33
CM 2967-8	R	MR	MR	R	MR	MR	33
CM 6740-7	MR	MR	MR	R	MR	R	33
CM 7951-5	R	MR	R	MR	MR	MR	33
CM 8024-2	R	MR	MR	R	MR	MR	33
SM 643-17	R	I	R	MR	MR	MR	33
SM 653-14	R	MR	R	MR	MR	S	33
SM 805-15	R	MR	R	MR	MR	MR	33
SM 1406-1	R	MR	I	I	R	MR	33
SM 1407-3	MR	S	R	R	I	I	33
SM 1627-16	R	MR	R	MR	MR	MR	33
CG 1-37	R	MR	R	R	MR	MR	50
CG 165-7	R	R	MR	R	MR	I	50
CM 2766-5	R	S	R	MR	MR	R	50
CM 3306-4	R	MR	MR	R	MR	R	50
CM 7514-8	R	MR	R	MR	R	MR	50
M Bra 489	R	MR	R	R	MR	I	50
M Col 1468	R	MR	MR	R	MR	R	50
M Col 1684	R	I	MR	R	MR	R	50
M Col 2066	R	I	MR	R	R	MR	50
M Col 2215	R	I	R	R	MR	MR	50
M Mal 2	R	R	R	MR	MR	MR	50
M Tai 1	R	I	R	MR	R	MR	50
SM 1483-1	R	I	R	MR	I	R	50

Table 5.2 (cont.)

Genotype	Isolate [b]						Resistance (%)
	27	44	66	69	P4	P12[c]	
SM 909-25	R	HS	R	R	I	I	50
SM 1201-5	R	S	R	I	MR	R	50
SM 1433-4	R	I	R	R	MR	S	50
SM 1438-2	R	MR	R	R	MR	MR	50
SM 1479-8	R	MR	R	MR	R	MR	50
SM 1511-6	R	MR	R	MR	MR	R	50
SM 1562-11	R	I	R	R	MR	MR	50
CG 915-1	R	MR	R	R	I	R	67
CM 4365-3	R	MR	R	R	MR	R	67
CM 6182-8	R	MR	R	R	MR	R	67
CM 7395-5	R	MR	R	R	MR	R	67
CM 7685-5	R	MR	R	R	R	I	67
M Col 2329	R	MR	R	MR	R	R	67
SM 1210-4	R	MR	R	R	MR	R	67
SM 1411-5	R	I	R	R	R	MR	67
SM 1619-3	R	MR	R	R	MR	R	67
SM 1624-2	R	MR	R	R	MR	R	67
CM 5655-4	R	MR	R	R	R	R	83
CM 7514-7	R	R	R	MR	R	R	83
Resistance (%)	90	6	75	56	21	42	

a. Reaction to the disease: R = resistant; MR = intermediately resistant; I = intermediate; S = susceptible; HS = highly susceptible.
b. Origin of isolates: P12 = Brazil, P4 = Colombia; 27 and 44 = Quindío, Colombia; 66 and 69 Valle del Cauca, Colombia.
c. P12 was included as a reference isolate

Results: the most tolerant cassava genotypes were CM 5655-4 and CM 7514-7, both well adapted to the mid-altitude, tropical valleys and coffee-growing regions (Table 5.2). Isolate 44 (*P. meadii*) showed the highest virulence (only 6% of the clones showing resistance to it). On the other hand, isolate 27, also coming from Quindío Department had very little virulence (90 % of the cassava genotypes expressing resistance to the isolate). P4, the *P. arecae* isolate, was the second most virulent (only 21 % of the clones showing resistance to it).

As in the case of CBB, *Phytophthora* root rots show a complex relationship with cassava. Resistance seems to have some degree of specificity against certain isolates of the pathogen. However, no clear gene by gene relationship has been established. Good sources of resistance are apparent from data in Table 5.2, and have been or will be incorporated into the breeding program. CM 5655-4 has been included as one of the parents for the diallel study described in Output 2 (Table 2.1).

Activity 5.3. Field evaluation of twenty-five elite cassava cultivars characterized for resistance to Phytophthora root rot at six locations, using participatory approach.

Specific Objectives:

a) *to evaluate twenty five elite cassava varieties for their field response to Phytophthora root rot, using a participatory research approach.*
b) *to validate different Phytophthora root rot control practices.*

Materials and Methods: a participatory research approach was used to develop effective disease management for the control of cassava root rots in four Colombian Departments. Twelve field experiments were established in the Departments of Cauca, Quindío, Valle del Cauca, and Vaupés, representing the different agroecological zones where root rots are endemic. The experiments were conducted with farmers and extension officers.

Table 5.3 shows treatments being currently evaluated in the different locations. The potential control practices were first evaluated in the greenhouse where results had shown that applying potassium chloride significantly reduced disease severity on young plantlets, even when inoculated with the most aggressive *Phytophthora* isolate. Also, the use of thermotherapy did not affect stake germination. At each location one or several different treatments (and levels of treatment) will be compared with the results from susceptible local varieties without any control treatment.

Results: At Santander de Quilichao, 17 cassava genotypes were harvested and evaluated for resistance to *Phytophthora* root rot. These genotypes had been previously selected in the greenhouse for resistance to several *Phytophthora* isolates. M Col 72 was the only genotype that did not perform well in this evaluation, possibly because it is not adapted to this agroecological zone. M Arg 6 and M Arg 9 were highly susceptible to root rots. These two varieties had also been scored as susceptible in the greenhouse evaluations. M Bra 1045 and M Bra 383 did not show any symptoms of this devastating disease. In the same infected field, stakes of the local variety *'Algodona'* (M Col 1522) were treated with the biocontrol agent *Trichoderma* spp. and did not present any diseased plants (Table 5.3, second experiment at Santander de Quilichao). This experiment has also been harvested. No significant yield differences were observed between the chemical and biological treatments used *on 'Algodona'*. Results from the plantings at locations other than Santander de Quilichao are not yet available because they were recently planted.

Table 5.3. During 1998 and 1999, 12 field experiments were established to evaluate the control of *Phytophthora* root rots in Colombia[¶].

Departament	Municipality	Experiment	Validation of practices to control *Phytophthora* roots											
			A	B	C	D	E	F	G	H	I	J	K	L
Cauca	Santander	1	17	-	+	1	-	-	-	-	-	-	-	-
	Quilichao	2	1	2	+	1	-	+	+	+	+	-	-	-
Cauca	B. Aires	1	2	-	+	1	-	+	+	+	+	+	-	-
Valle	Caicedonia	1/2	4	2	+	1	+	+	-	+	+	+	-	-
Quindío	La Tebaida	1	4	1	+	1	+	+	+	+	+	+	-	-
Quindío	Montenegro	1	3	1	+	1	+	+	+	+	-	-	-	-
Vaupés	Mitú	1	2	-	-	-	-	-	-	-	-	-	+	+
		2	2	-	-	-	-	-	-	-	-	-	+	+
		3	2	-	-	-	-	-	-	-	-	-	+	+
		4	2	-	-	-	-	-	-	-	-	-	+	+

[¶] A = Number of resistant genotypes planted and evaluated in the experiment.
B = Hot-water treatment. (-): treatment not evaluated; (1): standard immersion of cassava stakes at 49°C for 49 min in the lab; and (2): two hot water treatment levels: the standard method and the same temperature treatment goal but in the farmers' field using burning wood.
C = Chemical treatment, immersion of stakes in 3 g/L Ridomil (metalaxyl) for 5 min.
D = Biological treatment, immersion of stakes in 1×10^8 conidia/mL *Trichoderma* spp. (isolate 14PDA-4) for 30 min and application of 50 mL of this suspension per plant to the soil.
E = Biological treatment, immersion of stakes in MICOBIOL HE suspension for 30 min and application of 50 mL of suspension per plant to the soil. MICOBIOL HE is a mixture of micro-organisms (*Bacillus thuringiensis, Trichoderma* spp., *Beauveria bassiana, Metarhizium anisople, Paecilomyces lilacinus, P. fumosoroseus, Nomureae rileyi, Entomophthora muscae, Hirsutella thompsonii*, and *Verticillium lecanii*).
F = Potassium chloride (50 or 100 kg/ha).
G = Potassium sulfate (50 or 100 kg/ha).
H = Agropremix, a chemical mixture containing (per kg): 150 g nitrogen, 100 g phosphate, 120 g zinc, 20 g boron, 7.5 g copper, 30 g sulfate, 1 g molybdenum, and 2 g silicon.
I = Sucromag: wine lees with magnesium.
J = Sucrocal: wine lees with calcium.

Activity 5.4. Cassava populations identified to determine the genetic base of resistance to Phytophthora *root rots.*

Specific Objectives:

a) to elucidate the inheritance of the resistance to Phytophthora *root rots.*

b) to screen and introduce for new sources of resistance to Phytophthora *root rots.*

Rationale: little is known about the genetics of the resistance against *Phytophthora* root rots in cassava. A better understanding of the inheritance of resistance would help speeding up the breeding process, making the selection more efficient and/or facilitate the development of coherent strategy for controlling or reducing the damage caused by this disease.

Materials and Methods: several cassava genotypes were evaluated for their reaction to six highly pathogenic *Phytophthora* root rot species obtained from different cassava-growing regions. The cassava genotypes comprised segregating progenies from two specific crosses of varieties that were either susceptible or resistant to the disease: 39 individuals from CM 9582 (M Cr 81 x M Bra 1045), and 43 individuals from CM 9600 (M Cr 81 × M Cr 54). Disease progress was evaluated 7, 14, 21, and 28 days after inoculation by measuring the affected stem area. Evaluations were made in the greenhouse, using a randomized complete block design with five replications and one plant per plot.

Table 5.4. Percentage of segregating cassava genotypes from two crosses (CM 9582 and CM 9600) with different levels of resistance to six *Phytophthora* isolates.

Progenies	Isolates[¶]						Mean
	27	44	66	69	P4	P12	
CM 9582							
Resistant	64	21	72	22	4	53	39.33
Moderately resistant	9	29	16	22	27	16	19.83
Intermediate	23	17	0	3	23	9	12.50
Susceptible	4	21	12	39	27	16	19.83
Highly susceptible	0	12	0	14	19	6	8.50
CM 9600							
Resistant	53	0	29	48	26	38	32.33
Moderalety resistant	13	25	16	5	26	31	19.33
Intermediate	7	12	13	14	16	6	11.33
Susceptible	13	38	13	14	11	12	16.83
Highly susceptible	14	25	29	19	21	13	20.17

[¶] Isolates: P12=Brazil; P4=Colombia; 27 and 44=Quindío, Colombia; 66 and 69=Valle del Cauca, Colombia.

Results: in general, the progeny from CM 9600 was more susceptible than that of CM 9582 (Table 5.4). Several genotypes from cross CM 9582 showed excellent levels of resistance to either three or four *Phytophthora* isolates (CM 9582-4, CM 9582-5, CM 9582-7, and CM 9582-17). The reaction among the population of F1 individuals varied from highly susceptible to highly resistant. Overall, 39.3% and 32.3% of the individuals from CM 9582 and CM 9600 were highly resistant, respectively. The host-pathogen interaction between progenies and individuals was highly significant, suggesting a strong specificity of the sources of resistance present in the study. In general, based on the results observed, there is a clear genetic variability for the resistance to *Phytophthora*, which seems to be mainly polygenic and additive. However, there were some interesting results that would suggest the eventual occurrence of vertical (monogenic) resistance due to major gene(s). That was the case, for instance, of CM 9582 when challenged with isolate 66.

Table 5.5 shows native cassava varieties collected in indigenous settlements from Mitú (Vaupés) that will be introduced to the germplasm collection held at CIAT. Because of the environmental conditions where these cassava varieties are grown, it is expected that they will show some useful degree of resistance to *Phytophthora* root rots. The evaluation of their reaction to different species and isolates of the pathogen will soon be carried out.

Table 5.5. Native cassava varieties collected in indigenous communities from Mitú (Vaupés), Colombia.

Variety	Native name	Pulp color	HCN content
Yuca de Abeja (of the bee)	Mumiá Ducú	White	High
Yuca de Garza (of the egret)	Yajá Ducú	White	High
Yuca de Piña (of pineapple)	Sané Ducú	White	High
Yuca de Rana (of the frog)	Omá Ducú	White	High

Activity 5.5. Evaluation of two heterologous probes for resistance to* Phytophthora *root rot in 150 segregating cassava genotypes from one population.

Specific Objective:
a) Evaluate heterologous probes from different crops, for resistance to Phytophthora *root rots.*

Rationale: it has been already demonstrated, in several cases, a high genetic resemblance between resistance genes from different crops to similar diseases. In the case of cassava, for instance, seven loci have been found to have high similarity with a

rice gene responsible for resistance to *Xanthomonas* bacterial blight. These regions are, therefore, expected to have some role in the expression of resistance to CBB in cassava. Similarly, it is expected that resistance genes from other crops may have close resemblance, and could be used to identify, cassava genes responsible for the resistance to Phytophthora root rots.

Materials and Methods: heterologous probes from different crops were evaluated for their eventual association with cassava genes for resistance to *Phytophthora* spp. The 144 segregating progenies from the cassava K family (CM 2177-2 × M Nga 2) were used for this study. Family K is currently being used to develop the molecular map of cassava at CIAT's Biotechnology Laboratory. In collaboration with Dr. Jan Leach from the Kansas State University, 10 resistance gene analogs, cloned from maize and rice, were obtained to hybridize with DNA from the cassava K family (Table 5.6).

Cassava DNA of K-Family parentals was extracted by the Gilbeston-Dellaporta protocol. Genomic restriction with the enzymes Hind III, Xba I, Eco RV, and Eco RI was done after gel depurination and denaturation. The digested DNA was transferred overnight to a Hybond N+ membrane, using 10× SSC (NaCl and trisodic citric acid) as transferring solution. The DNA was fixed by baking for 2 h at 80°C. For isolating the PGEM-T vector containing the inserts of interest, minipreps were done in five clones from *Escherichia coli*, and conserved in glycerol at –80°C. The next step will be to amplify the inserts, using adequate primers, label the insert with ^{32}P, hybridize the K family DNA, and then, to compare the disease reaction in inoculated plants under greenhouse conditions.

Table 5.6. Disease resistance clones isolated from maize and rice to be used in the search for heterologous genes in cassava.

Accession	Gene name	Clone name	Plant	Vector	Insert size (bp)	Restriction enzyme to excise	Amplified by
		Pic 11	Corn	PGEM-T	518		Ploop - GLPL
13(5-3 FF)	NBS	Pic 12	Corn	PBS	504	Eco RI/Eco RI	
		Pic 14	Corn	PGEM-T	272		Kinase2 - GLPL
		Pic 15	Corn	PGEM-T	930		Ploop - MHD
		Pic 16	Corn	PGEM-T	293		Kinase2 - GLPL
		Pic 18	Corn	PGEM-T	469		Kinase - CFA
133	RP1	Pic 20	Corn	PBS			T3/T7
14 (7'4 F)	NBS	Pic 21	Corn	PUC 19	600	Eco RI	M13 F/R
58(AFO14467)	Peroxidase	POX22-3	Rice	PBS	1306	Eco RI/Xho I	
131	RP1	1700	Corn	PUC 19	1700	Xba I/Xba I	M13 F/R

Activity 5.6. Characterization of cassava germplasm resistance to cassava bacterial blight (CBB),and superelongation disease (SED), under field conditions.

Specific Objectives:

a) to evaluate segregating progenies for their field reaction to CBB, and SED.

b) to select highly yielding genotypes with resistance to the two most prevalent diseases in the acid soil savannas ecoregion.

Rationale: CBB and SED (*Sphaceloma manihoticola*) are the two most prevalent diseases in the acid soil savanna ecoregion. Therefore, every season CIAT carries out screenings in different locations representative of this ecoregion to select elite germplasm with good yield potential and adequate field resistance to these two diseases.

Materials and Methods: 198 and 246 cassava genotypes were characterized for their reactions to CBB and SED, respectively at Villavicencio and Matazul, under natural disease pressure from several pathotypes of each causal agent. Three replications in a randomized complete block design, with five plants per plot, were used in this evaluation. Scores were taken at 4, 7 and 12 months after planting averaging the five plants of each plot.

Results: in Matazul, two genotypes were intermediately resistant to CBB and resistant to SED (SM 1812-72 and SM 1812-72), whereas 79 varieties were intermediately resistant to both diseases. In Villavicencio, 24 varieties were intermediately resistant to CBB and resistant to SED and 76 varieties were intermediately resistant to both diseases. Table 5.7 shows only those genotypes with fresh root yields above 20 t/ha, and reaction to SED between intermediate and resistant. No CBB-resistant varieties were selected in this evaluation cycle. The clones SM 1821-7, SM 1871-32, and SM 2061-1 were intermediately resistant to CBB and SED in both locations.

Eighteen CBB-resistant cassava genotypes selected at Matazul (Colombia) were sent to Agropecuaria Mandioca (Venezuela), as part of an agreement with CIAT. These genotypes were CM 3311-4, CM 6306-13, CM 6306-11, CM 6975-1, CM 6975-8, CM 7052-2, CM 7661-12, CM 7661-15, CM 7666-21, CM 7666-25, CM 7666-31, CM 7670-2, CM 7670-4, CM 7772-2, CM 7772-11, CM 7772-13, CM 7803-1, and CM 7811-15.

Table 5.7. Genotypes selected for resistance to cassava bacterial blight (CBB) and superelongation disease (SED) at Matazul and Villavicencio, Colombia[¶].

Genotype	CBB	SED	Root yield (t/ha)	Genotype	CBB	SED	Root yield (t/ha)
Matazul				**Vallavicencio (cont.)**			
SM 2219-9	I	I	36.0	SM 1871-29	I	I	28.6
SM 1794-18	I	I	34.7	CM 2772-3	I	R	28.3
SM 1345-10	I	I	34.2	SM 1555-18	I	I	28.3
SM 2069-55	I	I	33.8	SM 1828-11	I	R	27.8
SM 1080-1	I	I	33.2	SM 1779-8	I	I	27.6
SM 1553-23	I	I	32.6	SM 1821-7	I	I	27.6
SM 1459-2	I	I	30.6	SM 1920-1	I	R	27.0
SM 1553-28	I	I	30.0	SM 1861-18	I	R	26.7
SM 2069-57	I	I	29.4	SM 1881-17	I	I	26.7
CM 2177-2	I	I	29.2	M Col 707	I	R	25.6
SM 1871-32	I	I	28.3	SM 1673-11	I	I	25.4
SM 1159-6	I	I	28.1	SM 1152-16	I	R	25.3
SM 1920-1	I	I	27.8	Chirosa Morada	I	I	25.0
SM 1215-1	I	I	27.5	SM 1859-26	I	R	24.3
SM 1812-72	I	R	27.1	SM 1689-18	I	I	24.2
SM 1144-4	I	I	26.7	SM 1794-18	I	R	23.9
SM 2069-1	I	I	26.4	SM 1871-32	I	I	23.8
CM 6975-14	I	I	25.6	M Cr 32	I	R	23.5
SM 1826-10	I	I	24.7	CM 8370-14	I	I	23.5
SM 1225-13	I	I	24.0	SM 1583- 8	I	I	22.7
SM 1859-26	I	I	23.1	SM 1545-25	I	I	22.2
SM 2061-1	I	I	21.9	M Col 2307	I	I	21.5
CM 7086-13	I	R	5.6	CM 6055-3	I	I	21.1
CM 3306-4 (check)	S	S	11.2	SM 1361-8	I	I	21.0
CG 915-1 (check)	S	S	6.6	SM 1539-2	I	I	20.4
M Mex 59 (check)	S	I	3.8	SB 0240-8	I	R	20.3
M Cub 74 (check)	S	I	3.0	M Ven 77	I	I	20.2
CM 3306-9 (check)	S	S	2.0	SM 1545-19	I	I	20.0
				SM 1820- 8	I	I	20.0
Villavicencio				SM 667-1	I	R	19.2
M Col 2387	I	I	43.5	SM 1855-9	I	R	18.8
SM 1555-17	I	I	39.9	SM 1363-3	I	R	18.2
SM 1225-12	I	I	36.9	SM 1225-11	I	R	16.3
SM 1152-19	I	I	36.7	CM 6787-9	I	R	15.1
M Bra 489	I	I	36.5	CM 2177-2	I	R	12.5
SM 1460-1	I	I	35.3	SM 1871-39	I	R	12.5
SM 2069-2	I	R	34.6	CM 7086-17	I	R	11.3
SM 1411-5	I	I	34.5	CM 7086-13	I	R	9.9
M Col 2409	I	R	33.8	CM 3320-23	I	R	8.6

Table 5.7 (cont.)

Genotype	CBB	SED	Root yield (t/ha)	Genotype	CBB	SED	Root yield (t/ha)
Matazul				**Vallavicencio**			
M Col 2329	I	R	33.6	CM 3306-4 (check)	S	I	8.7
Brasileira	I	I	33.2	M Cub 74 (check)	S	I	8.3
M Bra 466	I	I	32.6	CG 915-1 (check)	S	I	5.3
SM 1665-5	I	I	30.3	CM 3306-9 (check)	S	S	2.8
SM 1855-15	I	R	29.3				

[1] R = resistant; I = intermediate resistance.

Activity 5.7. Evaluation of three Phythophthora *isolates from Mitú, Colombia, inoculated on cassava root fragments.*

Summary: cassava root fragments from 16 Colombian genotypes were inoculated with fungal discs of *Phytophthora* isolates (MTRA 4, MTRA 6, and MTRA 7) from Mitú. The cassava genotypes had been selected according to previous greenhouse and field evaluations.

The response from the different cassava genotypes was determined by measuring diameter and depth of damage at 2, 4, and 6 days after inoculation. Ward's cluster analysis at 94.5% of significance (given by SAS), formed five variety groups according to their disease reaction (Figure 5.1):

Cluster 1 = corresponding to resistant varieties (M Bra 97, M Bra 532, and M Bra 1045) that showed least disease development.

Cluster 2 = CM 2772-3, HMC 1, and M Bra 222, with intermediate resistance.

Cluster 3 = CG 165-7, M Cr 81, M Bra 1044, and CM 523-7, also showing intermediate resistance.

Cluster 4 = M Bra 311, M Col 1522, CM 2177-2, and M Bra 12, with a susceptible reaction.

Cluster 5 = M Nga 2 and M Col 2066, was also very susceptible

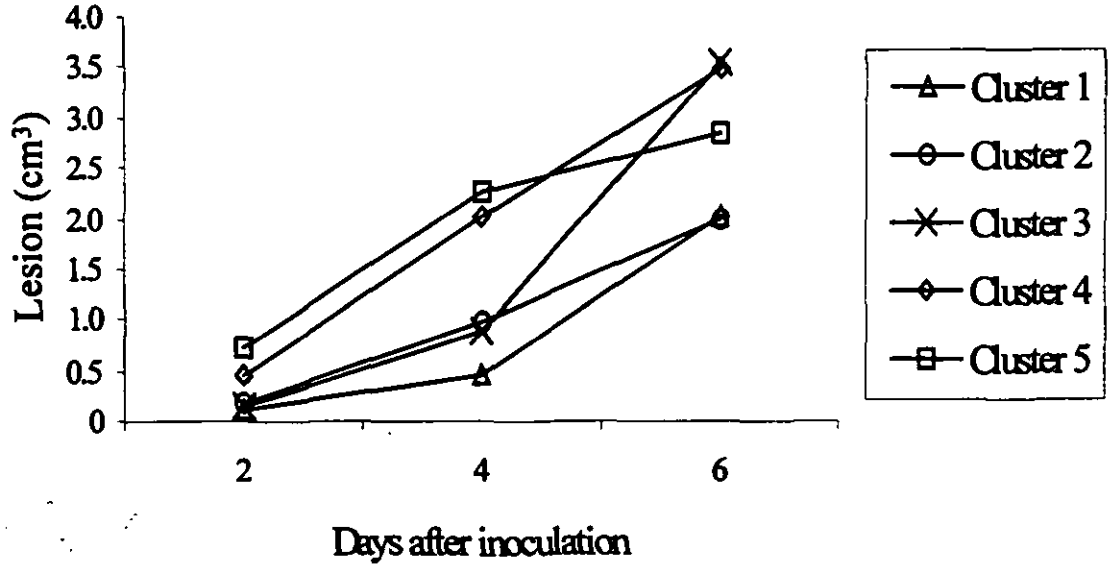


Figure 5.1. Disease curve of five cassava genotypes clusters, after inoculation on root fragments with three *Phytophthora* spp. isolates from Mitú (Vaupés), Colombia.

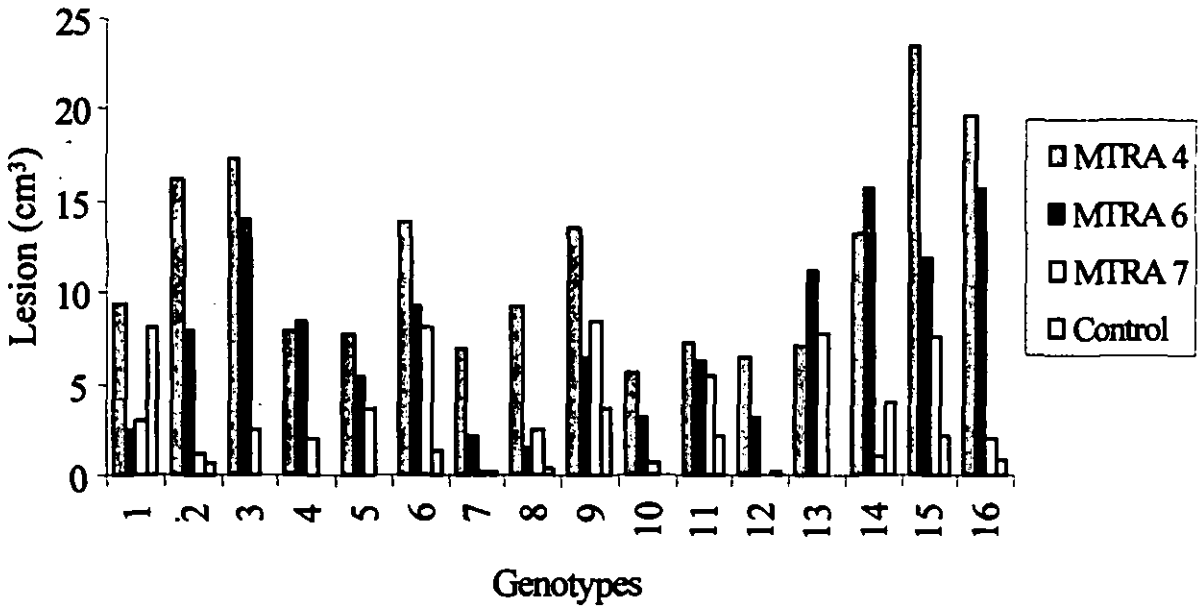


Figure 5.2. Reaction of 16 cassava genotypes to three *Phytophthora* isolates from Mitú, Colombia, 6 days after inoculation on root fragments. Genotypes: 1 = CG 165-7; 2 = CM 523-7; 3 = CM 2177-2; 4 = CM 2772-3; 5 = HMC-1; 6 = M Bra 12; 7 = M Bra 97; 8 = M Bra 222; 9 = M Bra 311; 10 = M Bra 532; 11 = M Bra 1044; 12 = M Bra 1045; 13 = M Cr 81; 14 = M Col 1522; 15 = M Col 2066; 16 = M Nga 2.

Figure 5.2 shows the reaction of the 16 cassava genotypes to three *Phytophthora* isolates from Mitú, Colombia. The most aggressive isolate was MTRA 4, whereas the least aggressive was MTRA 7. The most tolerant genotypes, 6 days after inoculation, were M Bra 222, HMC 1, M Bra 1045, M Bra 532, and M Bra 97.

Activity 5.8. Participatory evaluation of cassava genotypes for adaptation and root rot resistance in Mitú, Colombia.

Specific Objectives:

a) to learn about criteria of selection by native indigenous women farmers both at vegetative stage and at the root processing stages.

b) *to select varieties that, under the Vaupés conditions, show resistance to Phytophthora root rot.*

Rationale: participatory research applied to cassava has been already described in output 4 in breeding activities. It has also been used by pathologists in the Vaupés region (Mitú Department), a part of the Amazon basin.

Materials and Methods: four trials were evaluated and harvested by indigenous women in four *chagras* (small plots of burned rain forest planted with cassava and other crops) in four communities at Mitú (Vaupés), applying participatory research methodologies. The objective was to select varieties that, under conditions of indigenous cultivation, showed resistance to *Phytophthora* spp., adaptation to Amazonian conditions, good yields, and acceptable starch quality.

Women farmers from eight communities close to Mitú were brought together to share their experiences. One woman from each of the four communities, where varieties were evaluated, shared her experiences and trial achievements. Since then, meetings have been organized in different communities, at which women spoke about the trials, and 200 illustrated handbooks on controlling cassava root rots were distributed among the Indians.

Soil chemical, physical, and biological analyses were made on samples taken from different indigenous plots, some with and others without root rots caused by *Phytophthora* spp. Coincidentally, clay layer was found at below 15 cm in plots where root rots was present.

Table 5.8. Criteria used by Indian women farmers for selecting cassava genotypes at the vegetative stage, Mitú, Colombia.

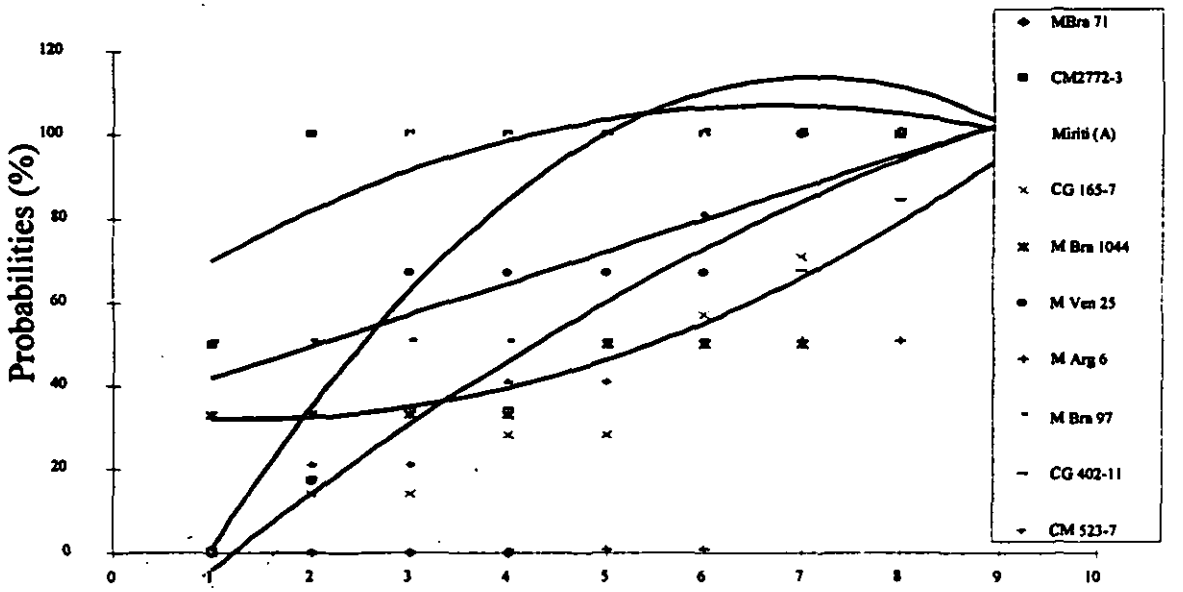
Criteria[a]	Frequency (%)
Height, "fat" stake, nodes, plants per site (cuttings)	27.9
Strong growth (vigor)	23.3
Leaves color: "sad, beautiful" (vigor, health, esthetic)	20.9
Fast yield, short plant with good roots (early maturity)	11.6
Uniform growth (uniformity)	9.3
Dry and rotted leaves, dry branches (health)	6.7
Good root production (yield)	21.4
Leaves' appearance: "sad or beautiful" (vigor, health, etc.)	16.1
Plant height (vigor, yield, cuttings)	15.9
Stem growth (yield, cuttings)	12.5
Hard to scrape by nails (starch content)	10.7
Pests and diseases (health)	5.7
High branches (branching height)	5.0
Pulp color	4.7
Harvest time (earliness)	3.4
Root appearance	2.3
Foliage	1.1
Ease of harvest	1.1

a. Information from four communities (15 Indian women farmers).
b. Information from four communities (18 Indian women farmers).

Table 5.9. Agronomic characteristics of cassava genotypes evaluated in Indian *chagras* in Mitú, using participatory research methodologies.

Genotype	Origin	Pulp color	Food roots (%)	Root rot (%)	Yield (t/ha)	HCN [a]	Plant height (m)	1st branch height (m)	Number of branches	Cuttings per plant	Dry matter (%)	Starch (%)	Preference [b]	
													Harvest	Starch
CM 2772-3	CIAT	Yellow	8.9	0.0	5.4	7	1.5	0.9	2.0	3.9	29.9	28.1	9.5	8.5
Mirití	Mitú	Yellow	14.3	0.0	7.7	8	1.5	0.6	2.3	4.0	31.8	29.7	8.0	8.0
M Bra 97	CIAT	White	9.2	0.0	3.5	6	1.3	0.9	1.3	2.9	33.9	31.8	7.5	7.5
CM 523-7	CIAT	White	22.7	1.5	7.9	7	2.3	1.0	3.6	8.7	35.0	32.8	6.0	8.3
Abeja	Mitú	White	14.0	0.0	3.8	8	2.0	1.0	1.3	6.0	29.9	27.8	7.0	7.0
M Bra 532	CIAT	White	33.0	0.0	10.7	7	1.8	0.9	3.8	7.8	34.5	32.4	7.8	6.0
M Bra 71	CIAT	White	19.7	0.2	7.6	8	2.1	1.1	3.0	5.9	30.7	28.6	6.0	7.7
M Ven 25	CIAT	White	19.8	0.6	6.0	8	1.7	1.1	2.1	5.2	34.5	32.3	6.9	6.8
CG 165-7	CIAT	White	11.9	0.0	4.6	7	1.7	0.9	3.1	3.8	32.3	30.2	5.1	8.0
Blanca	Mitú	White	14.3	0.0	11.1	8	2.1	1.2	2.7	6.7	34.1	32.0	7.0	6.0
CG 402-11	CIAT	White	12.8	4.3	3.9	6	1.7	0.8	2.4	4.7	28.0	26.0	5.3	7.6
Abiyú	Mitú	Yellow	41.0	0.0	8.9	8	1.5	1.1	2.5	3.0	32.1	30.0	7.2	5.0
Lapa blanca	Mitú	White	27.5	3.6	17.3	9	2.8	0.2	0.5	10.0	31.7	29.6	6.4	5.0
M Bra 1044	CIAT	Yellow	15.6	0.0	6.8	7	1.8	1.1	2.8	7.0	36.2	34.1	5.8	4.5
M Arg 6	CIAT	Cream	6.0	0.0	3.0	7	0.9	0.4	1.8	1.3	35.0	32.8	2.5	6.0
Wasaí	Mitú	Yellow	4.0	0.0	3.7	8	1.1	0.5	0.7	3.7	32.1	30.0	5.0	3.0
Brava Blanca	Mitú	White	66.7	0.0	19.9	7	3.5	1.8	3.0	16.7	33.9	31.7	-[c]	4.0
Brava Amarilla	Mitú	Yellow	43.3	0.0	12.2	8	2.9	1.6	4.0	10.0	32.4	30.4	-	2.0
Low cyanide variety	Mitú	White	6.4	1.7	8.1	6	2.2	1.2	3.1	7.8	30.6	28.5	7.0	4.5
White native varieties [d]	Mitú	White	30.6	1.1	8.1	8	2.6	1.1	1.9	9.8	32.4	30.3	6.6	7.0
Yellow native varieties [d]	Mitú	Yellow	25.7	0.0	5.1	8	1.8	0.9	2.4	5.2	32.1	30.0	7.0	4.0

(a) Cyanide content: 1 = low; 9 = very high; **(b)** Preference scale: 1 = low; 10 = high; **(c)** - = not determined; and **(d)** averages of several varieties.



B

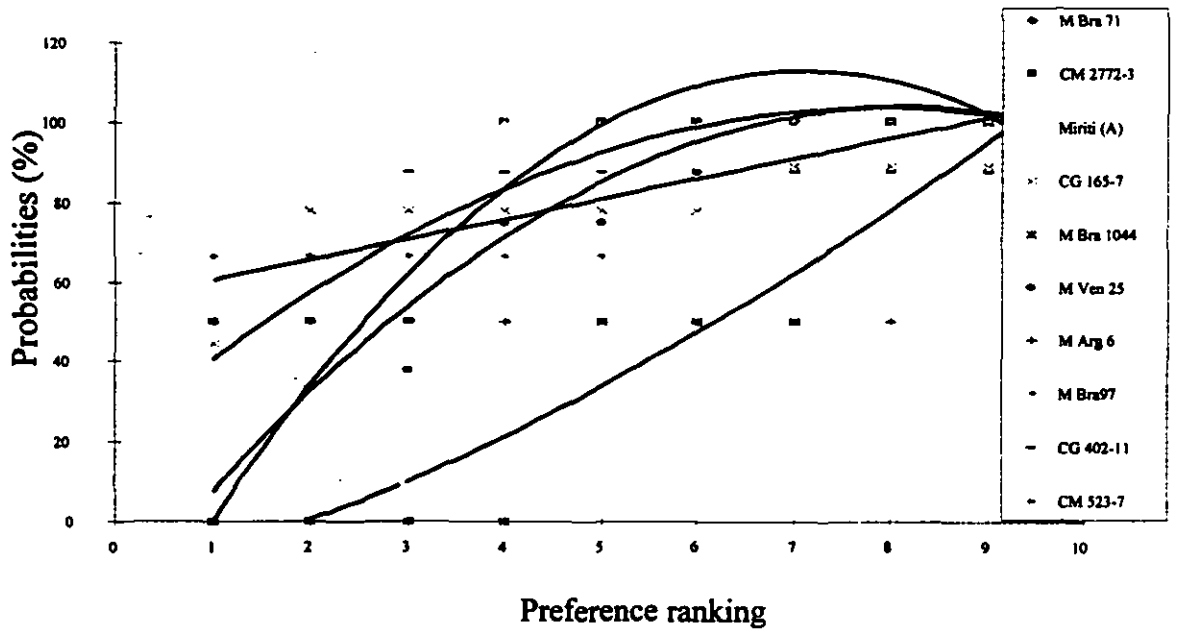


Figure 5.3. Comparison of preference rankings for cassava genotypes evaluated at harvest (A) and by starch quality (B) by Indian women farmers at Mitú (Vaupés), Colombia, applying participatory research methodologies.

Results: based on open evaluations, criteria used by the Indian women in variety selection were identified and a field book was designed for further evaluations to be conducted at Mitú. For cassava in the vegetative stage, vigor, health, plant height, number of stems per plant, and early maturity were the criteria most frequently used. For harvested cassava, participating women looked at yield, starch content and quality, cutting production (i.e., planting material production), root health, and early maturity. Yield tended to be a secondary criterion (Table 5.8).

According to the preference analysis, the women selected the CIAT genotype CM 2772-3 (low cyanide and yellow pulp) at harvest and for starch quality above their native varieties (Table 5.9 and Figure 5.3). Also, a field day was organized in the Cucura Indian settlement, with the participation of 85 Indians.

Soil physical analysis showed compaction, low organic matter, and physical degradation in a soil from Central Seima, which partially explains the low cassava yield observed in this region.

Chemical analyses were made of soil samples taken at the 0-15 and 15-30 cm layers from three indigenous plots. In those plots, a cassava variety adaptation trial was planted and managed under Indian farming practices to select the most adapted varieties. Although soil fertility was low, it was conserved with traditional crop management, which consists of maintaining organic matter on soil and using ash as fertilizer (Figure 5.4).

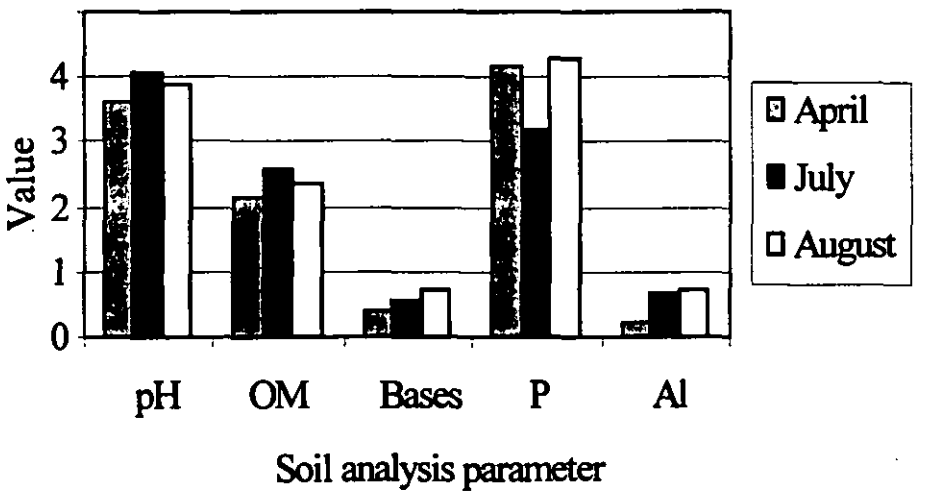


Figure 5.4. Soil fertility dynamics in a cassava crop grown under traditional Indian farming management. pH = units; OM = organic matter (%); Bases = Ca, Mg, and K; Al = meq/100 g; P = phosphorus in ppm.

Activity 5.9. Multiplication of promising cassava varieties to ensure sufficient stake material for experiments in the greenhouse and field at CORPOICA, Palmira.

One hundrend and ninety promising cassava genotypes were multiplied at CORPOICA, Palmira, for experiments to be conducted on resistance to *Phytophthora* spp., *Xanthomonas axonopodis* pv. *manihotis*, and *Sphaceloma manihoticola*, and on disease management.

Output 6: Characterization of cassava germplasm for resistance/tolerance to major pests.

The overall objective of this project is to reduce the damage caused by different arthropod pests that can affect cassava production below economic level by screening and selecting for sources of genetic resistance and/or agents for biological control. It is also an important objective to monitor the evolution of different pest, to anticipate the occurrence of new problems caused or induced by different arthropods.

Activity 6.1. Preliminary studies: Whitefly species diversity on cassava at CIAT, Palmira.

Summary: cassava whitefly populations at CIAT have increased dramatically in recent years, causing crop damage. Concurrently, the incidence of cassava frogskin virus disease, which is whitefly-transmitted has also increased. Traditionally, whitefly infestations at CIAT have consisted of three species; *Aleurotrachellus socialis* has always been the predominant species while *Bemisia tuberculata* and *Trialeurodes variabilis* are found in much lower populations. This follows the pattern observed in most commercial cassava plantations in Colombia (Castillo et al. 1999). Since frogskin disease is transmitted by *B. tuberculata*, and the incidence of the disease has increased in recent years, it was decided to monitor the populations of the whitefly species population on cassava at CIAT.

Leaf samples were taken from cassava plants at 98 randomly selected sites on the CIAT farm. Leaves were removed from the lower portion of the plant, and brought to the laboratory for stereoscope observation and individuals were identified to species.

A total of 13, 194 whitefly individuals were collected from the leaf samples; 12, 987, or 98.5% corresponded to the species *A. socialis*. *B. tuberculata* and *T. variabilis* were represented by 90 (0.68%) and 114 (0.86%) individuals respectively. These results indicate that there has probably been no shift in the relationship of whitefly species populations in recent years. Although populations of *B. tuberculata* are very low, they may be sufficient to cause the increase in the incidence of frogskin disease, especially if there is a high source of disease inoculum. Whitefly populations will continue to be monitored in the future.

Activity 6.2. Development of a colony of Bemisia tabaci *on cassava.*

Summary: *B. tabaci* is the vector of African Cassava Mosaic Disease (ACMD) in Africa. Until recently, the *B. tabaci* biotypes found in the Americas did not feed on cassava. It has been speculated that the absence of ACMD in the Americas may be related to the inability of its vector, *B. tabaci*, to colonize cassava. Since the early 1990's a new

biotype (B) of *B. tabaci*, considered by some to be a separate species (*B. argentifolii*) has been found feeding on cassava in the neotropics. It is considered that ACMD now possess a more serious threat to cassava production, as most traditional varieties in the neotropics are highly susceptible to the disease. In addition the B biotype of *B. tabaci*, as a virus vector, causes heavy crop losses on numerous other crops in the neotropics, and these are often grown in association with cassava, or in the same area. The possibility of virus diseases moving between crops, or the appearance of previously unrecorded viruses has become a potential threat.

For many years we have maintained an active program to identify cassava germplasm resistant to whiteflies and develop resistant hybrids. The species most frequently used in our evaluations has been, and continues to be, *A. socialis*. However, in light of the fact that there is a biotype of *B. tabaci* now capable of feeding on cassava, and its capacity to transmit geminiviruses, research has been initiated to study the potential of *B. tabaci* as a pest or vector on cassava. Therefore, an attempt was initiated to establish a colony of *B. tabaci* biotype found in Colombia, on cassava.

Adults of *B. tabaci* were collected from several hosts, including lettuce, beans, cotton, squash, and cabbage (supplied by the bean entomology project).

Potted, five-week old, cassava (Var. CMC 40) plants were infested by placing 20 to 25 adult whiteflies in small leaf cages, and they were allowed to oviposit 48 and 96 hours (two separate experiments).

Preliminary results indicate that the local *B. tabaci* biotype "B" does not easily survive on cassava. Whiteflies collected from all of the aforementioned hosts were able to oviposit on cassava, and nymphs emerged and initiated feeding on cassava. A total of 898 nymphs were produced in the two experiments; however only 10 adults (1.1%) emerged. In the first experiment, 257 nymphs were produced and resulted in 10 adults. In the second experiment of the 641 nymphs produced, there was 100% mortality during the nymphal stage. Nymphal mortality occurred during both, early or late stages.

Although these results indicate a poor adaptation of *B. tabaci* on cassava, the fact that some oviposition and nymphal development occurred, indicates that populations of *B. tabaci* could move onto cassava. This is already occurring in certain areas (i.e. Brazil), indicating that the local biotype may not yet have had sufficient opportunity to adapt to cassava. We will continue to monitor this situation.

Activity 6.3. Whiteflies: Germplasm evaluations at CIAT, Palmira.

Summary: cassava is often considered more tolerant to pests than most crops because it does not have critical periods that affect the organs on which yield depends on. Nevertheless, research, and field observations, have shown that several pests can

reduce yield significantly when pest populations are high and/or environmental conditions are unfavorable. An estimated 200 species of arthropods feed on cassava in the neotropics and many of these are specific to cassava and have adapted in varying degrees to the array of natural biochemical defenses that include laticifers and HCN content. A successful integrated pest management (IPM) program in cassava will depend on having effective, environmentally sound, low-cost pest management technologies available to cassava farmers in developing countries. Stable host plant resistance (HPR) offers a practical, long term, solution for maintaining reduced pests populations. Sources of resistance have been identified for mites, lacebugs, whiteflies, thrips and burrower bug. The CIAT cassava germplasm bank of approximately 6000 accessions is continually being screened for resistance to arthropod pests. Current emphasis is being given to whiteflies, mites and stem borers.

Whitefly populations at CIAT, especially those of the species *A. socialis*, continued to remain high (see Activity 6.1) during 1999. Whitefly resistance in agricultural crops is rare, however after evaluating nearly 5000 cultivars, several good sources of resistance have been identified and high yielding, whitefly resistant hybrids have been developed. Three of these hybrids are presently being evaluated by CORPOICA and may soon be released to producers.

The clone MEcu 72 has consistently expressed the highest level of resistance. Three additional clones MEcu 64, MPer 335 and MPer 415 have also been selected for high levels of resistance. The progeny from MEcu 72 x Bra 12 cross, CG 489-34, CG 489-4, CG 489-31 an CG 489-23 have consistently displayed moderate levels of whitefly resistance. The families evaluated were as follows:

Hybrid family		Female Parent		Male Parent
CM 8984	=	MCol 1505	x	CG 489-34
CM 8990	=	MCol 2026	x	CG 489-34
CM 8991	=	MCol 2026	x	MBra 12
CM 8995	=	MEcu 72	x	MCol 1468
CM 8996	=	MEcu 72	x	MCol 2246
CM 3317	=	MBra 12	x	MCol 1468
CM 5438	=	MBra 12	x	MCol 1505
CM 7559	=	MNGA 2	x	MBra 12
CM 8891	=	MCol 1468	x	CG 489-4
CM 8884	=	CG 489-4	x	MCol 1468

The clones MCol 1505, MCol 2026, MCol 1468, MCol 2246 and MNGUA are susceptible to whiteflies and MBra 12 is tolerant. These families were planted in fields at CIAT and at CORPOICA, Nataima, Tolima. Plantings were done at both sites in

early November 1998 and whitefly damage on populations was scored during April - May 1999. A 1 to 6 damage scale (1 = no damage, 6 severe damage) is used to measure whitefly damage and a 1 to 6 scale is also used to measure whitefly populations (Table 6.1)

Table 6.1 Population and damage scales for evaluating cassava germplasm for resistance to whiteflies.

Population scale (Nymphs and pupae)
1 = No whitefly stages present
2 = 1 – 200 individuals per cassava leaf
3 = 201 – 500 per leaf
4 = 501 – 2000 per leaf
5 = 2001 – 4000 per leaf
6 = > 4000 per leaf
Damage scale
1 = No leaf damage
2 = Young leaves still green but slightly flaccid
3 = Some twisting of young leaves, slight leaf curling
4 = Apical leaves curled and twisted; yellow-green mottled appearance
5 = Same as 4, but with "sooty mold" and yellowing of leaves
6 = Considerable leaf necrosis and defoliation, sooty mold on mid and lower leaves and young stems.

Results show that whitefly (*A. socialis*) populations at CIAT were extremely high and probably caused excessive selection pressure on the cassava clones. Of the 671 progeny and parents evaluated 637 (or 95%) had damage ratings above 4.1 and 47% between 5.1 and 6.0 (Figure 6.1). Only 7 had ratings below 3.5 and two of these were parents. MEcu 72, our most resistant variety held up fairly well under this heavy selection pressure with a damage rating of 2.0, 3.0 and 3.5 in the three repetitions.

That whitefly populations were extremely high can be seen in Figure 6.2; 658 of the 671 progenies (or 98%) had whitefly population ratings above 4.1, and none had populations below 2.5 on the population scale. If we look at just one family, CM 8990 (MCol 2026 x CG 489-34) 170 of the 179 progenies (95%) had damage ratings above 4.1 and only 1 clone below 3.6 (Figure 6.3). This family is being evaluated in the greenhouse using potted plants and a controlled whitefly population.

Activity 6.4. Germplasm evaluations for whiteflies with CORPOICA, at Nataima, Tolima.

Summary: the cassava cultivars from the crosses between resistant and susceptible cultivars (as described in Activity 6.3) were also planted at CORPOICA, Nataima, Tolima. We have been evaluating germplasm for whitefly resistance at this experiment station for about 15 years. Whitefly populations have traditionally been high, making this an excellent site for HPR evaluations. However in recent years whitefly population pressure has decreased while, as previously noted, populations have increased dramatically at CIAT. The reasons for this phenomenon are not fully understood.

Evaluations at CORPOICA, Nataima, contrast with those taken at CIAT, on the same cultivars, in that damage levels were considerably lower. Of the 554 genotypes tested at Nataima, 290 or 52% had damage ratings of 2.5 or lower (Figure 6.4), while at CIAT, no variety had damage ratings below 2.5. At Nataima, only 53 cultivars (9.6%) had damage ratings above 4.0; at CIAT, 637 genotypes, or 95% had damage ratings above 4.0.

Data recorded on whitefly (*A. socialis)* populations show a higher population level at CIAT, than at Nataima (Figures 6.1 and 6.5).At CIAT, 658 cultivars (or 98%), had whitefly population rating above 4.0 and 173 (25.8%) of these were between 5.5 and 6.0 (Figure 6.1). At Nataima, only 82 genotypes (14.8%) had population rating above 4.0 and none above 5.5 on the 0 to 6 scale (Figure 6.5).

A comparison of the progeny from the family CM 8990 between the two evaluation sites presents similar results (Figures 6.3 and 6.6). At Nataima, the cultivars were well distributed in their rating from 1.0 to 5.5, and no genotype in the 5.5 to 6.0 range (Figure 6.6). Of the 104 cultivars, 49, nearly one half (47%), were rated in the 1.0 to 2.5 range; while at CIAT no variety fell within this range (Figure 6.3). In addition, at CIAT, 74 genotypes (41.3%) received a rating of 5.1 to 5; at Nataima only 13 cultivars (12.5%) received a damage rating above 4.0.

The susceptible check in these trials was CMC-40 (MCol 1468) and at Nataima this cultivar was planted at systematic intervals through the trial area. This facilitates "measuring" the levels, intensity and distribution of whitefly populations and damage levels. CMC-40 received a moderate to high damage (3.0 to 5.0) and population (3.0 to 4.5) throughout the trial. At CIAT, the high CMC-40 damage rating and whitefly population rating was 5.5 and 4.5 respectively; at Nataima it was 5.0 and 4.5 (Table 6.2), slightly lower than at CIAT but also indicating that there were adequate whitefly populations.

Whitefly damage and population ratings were consistently higher at CIAT. Table 6.2 contains a representative sample of the ratings at the two sites for selected cultivars. The varieties MCol 1505, MCol 2026 and CMC-40 are susceptible (data from previous trial) and the three had high damage ratings at both sites; MCol 2246, also susceptible, did not follow this pattern. The clones CG 489-23, CG 489-31, CG 489-34, and CG

489-4 (selected resistant progeny from a MEcu 72 x MBra 12 cross), and the resistant cultivar MEcu 72, had low damage ratings, as expected, at Nataima. These results are consistent with several previous trials done over numerous years at this site.

The results from these two trials, one at CIAT, the second at Nataima probably express the difficulty in obtaining an ideal situation, with the optimal amount of pest selection pressure needed to do this type of genetic study. Whitefly populations and consequently cassava plant damage was so great at CIAT that small or moderate differences could not be detected in the cultivars. Whitefly populations at Nataima appear to be too low, resulting in insufficient damage and too many genotypes with low ratings. Controlling whitefly populations in the field will always be difficult; perhaps the greenhouse studies will provide more optimal selection pressure in order to detect differences in resistance from these crosses.

The high whitefly populations/severe damage syndrome at CIAT during early 1999, is an indication of the potential severity of whiteflies as a cassava pest, and reinforces the need to develop highly resistant cultivars.

Table 6.2. Whitefly (*A. socialis*) populations and damage on selected cassava cultivars at two evaluation sites: CIAT, Palmira, Valle and CORPOICA, Nataima, Tolima.

	CIAT		Nataima	
Cultivars	**Damage / Populations**		**Damage / Populations**	
CM 5438-194	6.0	5.9	1.0	2.75
CM 8991-129	5.5	5.7	1.0	1.5
MEcu	3.0	3.5	1.0	1.1
CG 489-4	4.0	4.6	1.5	2.1
CG 489-23	4.5	4.3	1.5	1.6
CG 489-31	3.0	3.4	2.0	2.0
CG 489-34	5.5	4.6	1.0	1.1
CM 8984-60	4.5	4.5	1.0	1.5
MCol 2246	5.0	4.8	1.5	3.0
CM 8995-4	5.0	4.7	1.5	1.8
CM 8991-37	5.5	4.3	4.0	3.6
CM 5438-74	3.0	4.9	1.0	1.8
MCol 2026	6.0	5.6	5.5	5.2
CMC 40 (MCol 1468)	5.5	4.9	5.0	4.5
MBra 12	5.5	5.5	2.5	3.7
MCol 1505	6.0	5.7	3.5	3.8

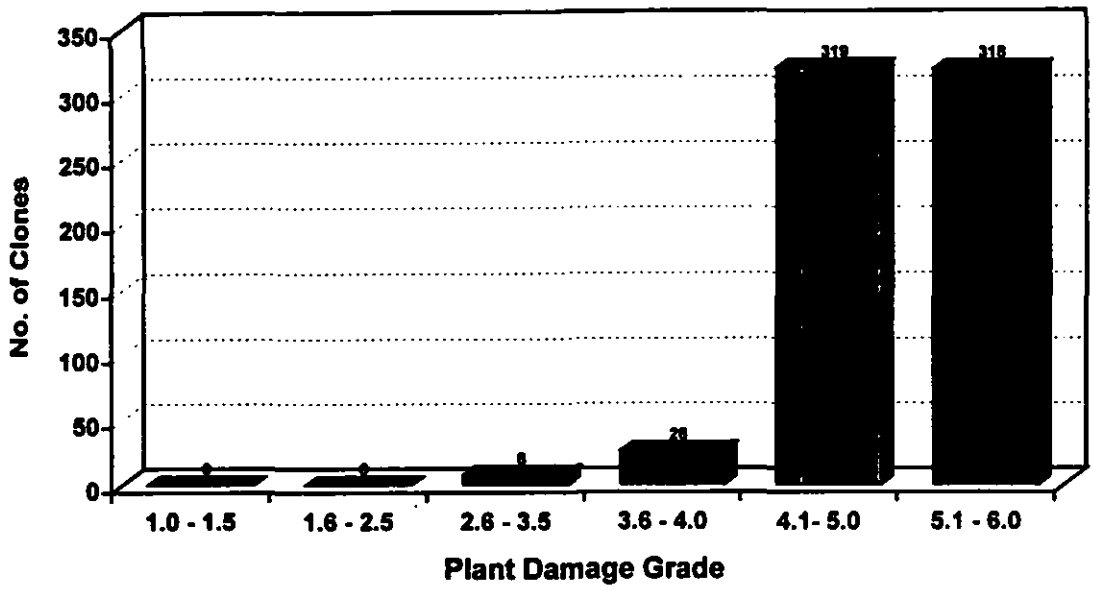


Figure 6.1. Evaluations of cassava clones from crosses for whitefly damage (*A. socialis*) on resistant and susceptible cultivars at CIAT (1998-1999).

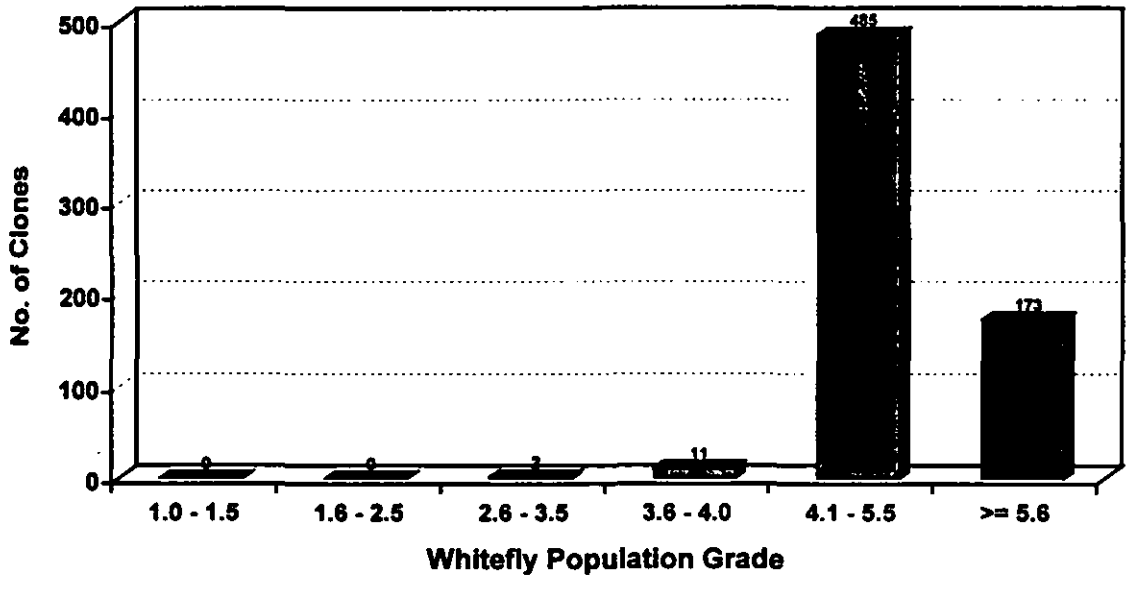


Figure 6.2. Evaluations of whitefly (*A. socialis*) populations on cassava clones derived from crosses between whitefly resistant and susceptible cultivars at CIAT (1998-1999).

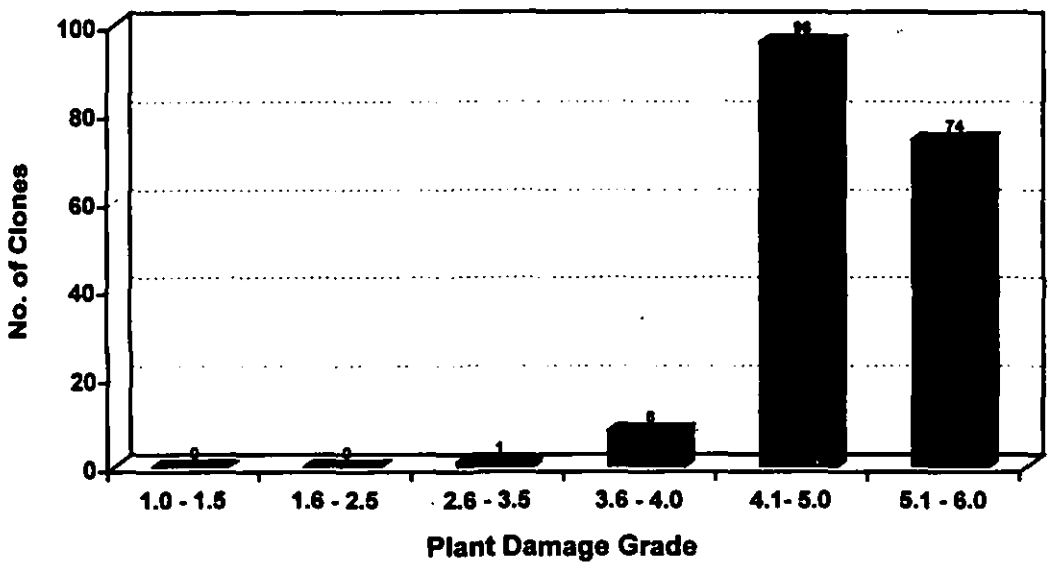


Figure 6.3. Evaluations of whitefly (*A. socialis*) damage on cassava clones resulting from a MCol 2026(S) x CG 489-34(R) cross (Fam. CM 8990) at CIAT, Palmira (1998-1999).

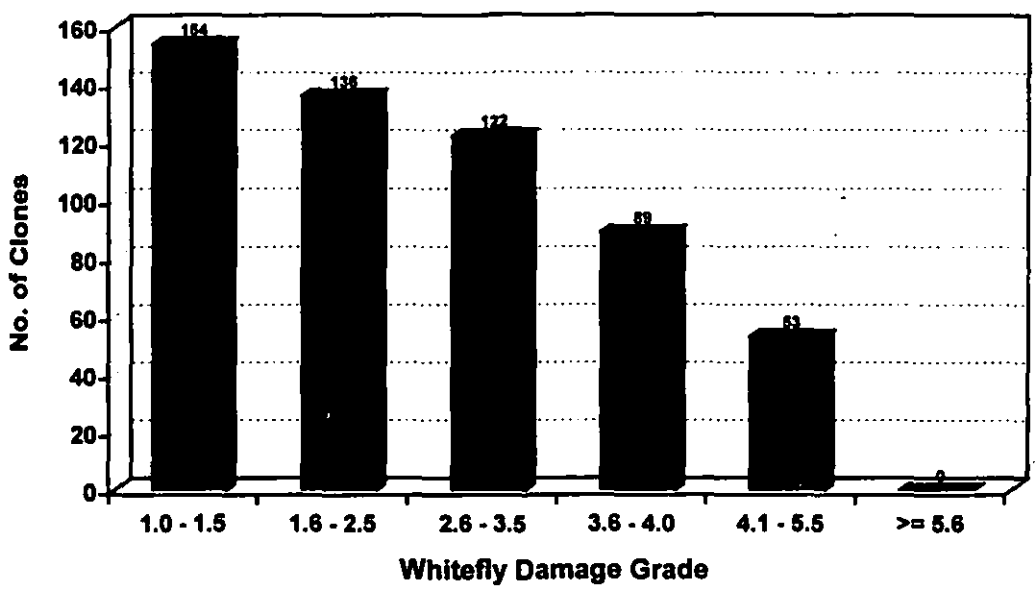


Figure 6.4. Evaluations of whitefly (*A. socialis*) damage on cassava clones derived from crosses of whitefly resistant x susceptible cultivars at CORPOICA, Nataima (1998-1999).

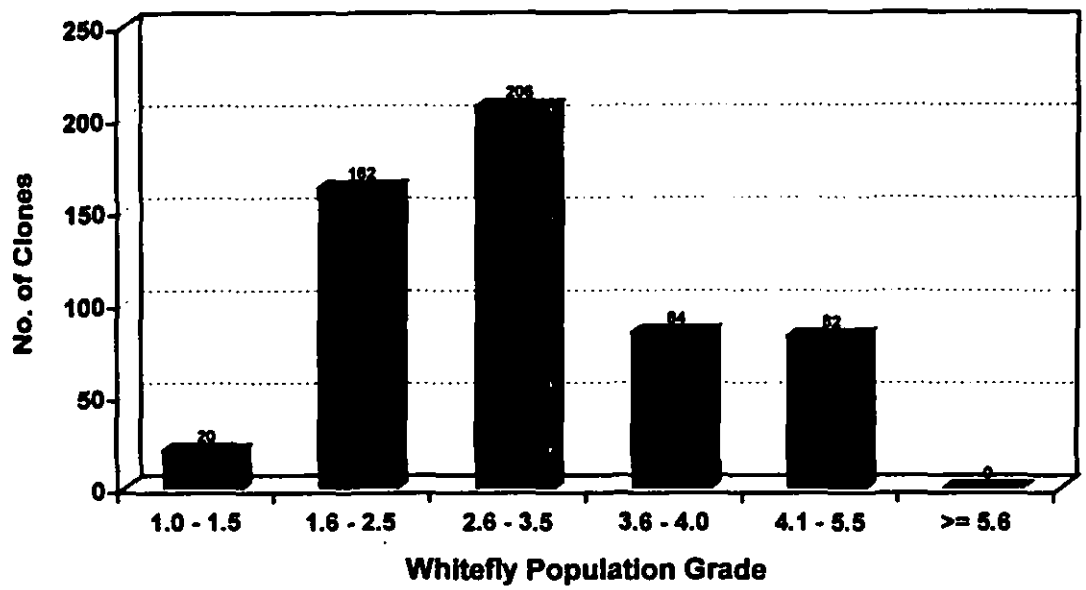


Figure 6.5. Evaluations of whitefly (*A. socialis*) populations on cassava clones derived from crosses of whitefly resistant x susceptible cultivars at CORPOICA, Nataima (1998-1999).

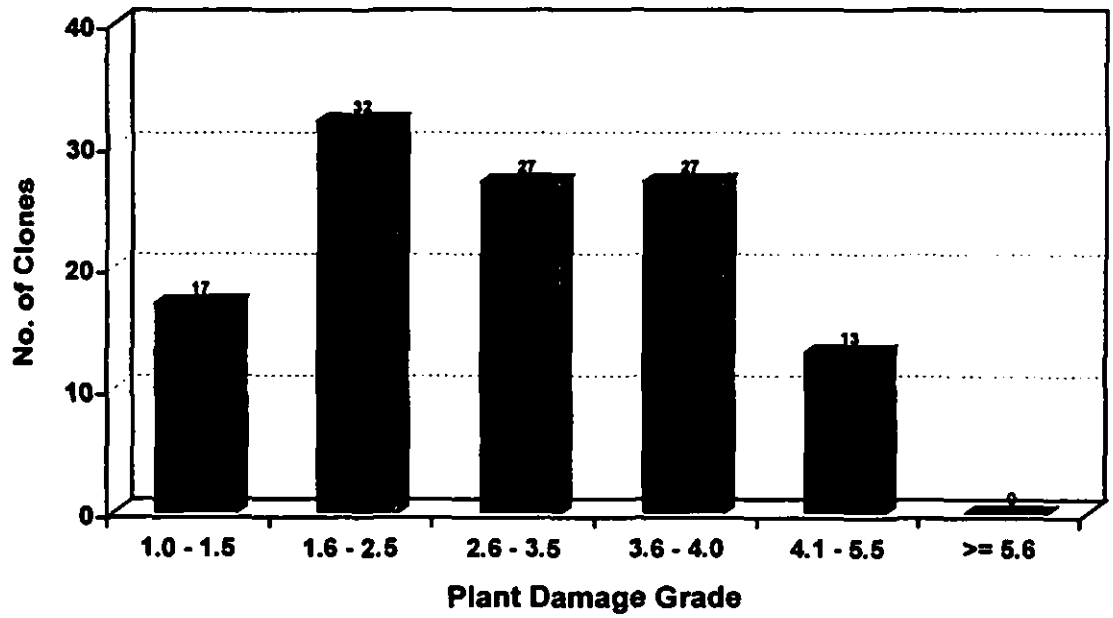


Figure 6.6. Evaluations of whitefly (*A. socialis*) damage on cassava clones resulting from a Mcol 2026(S) x CG 489-34 (R) cross (Fam. CM 8990) at CORPOICA, Nataima (1998-1999).

Activity 6.5. Tritrophic interactions: studies to determine the effect of HPR on whitefly parasitism.

Specific objectives:

a) *to identify, study and evaluate potential sources of biological control of whiteflies that feed on cassava.*
b) *to determine the preferred instar of A. socialis for E. hispida parasitism.*
c) *to determine the effect of four cassava varieties, MEcu 72, CG489-4, MBra-12 and CMC-40, on the emergence and survival of E. hispida parasitizing A. socialis.*

Rationale: Biological control and host plant resistance can offer a low-cost sustainable solution to cassava losses from whitefly damage. Host plant resistance studies at CIAT, especially with the whitefly species *A. socialis* are well advanced and several resistance sources have been identified.

In recent years we have increased our activities in biological control, surveying for natural enemies in several regions of Colombia and Venezuela. Numerous parasitoids have been collected from cassava whiteflies; these will be identified, studied and evaluated. The most frequently observed parasitoid species of *A. socialis* is *Encarsia hispida*. The genus *Encarsia* is recognized as possessing good searching ability, dispersion and adaptation.

Present research is investigating the compatibility between host plant resistance in biological control, the two most important components in an integrated pest management system. Experiments were designed to determine the compatibility of the parasitoid *E. hispida* on the three cassava genotypes.

Materials and Methods: four cassava varieties (MEcu 72, CG489-4, MBra-12 and CMC-40) were selected because of their resistance or susceptibility to *A. socialis*. MEcu 72 has consistently expressed a high level of resistance to *A. socialis;* CMC-40 is a highly susceptible variety (the cassava whitefly colony is maintained on CMC-40); MBra-12 is a tolerant (low levels of resistance) genotype with good agronomic qualities; and CG489-34 is moderately resistant to A. *socialis*. The progeny of a MEcu 72 x MBra 12 cross was also included.

Potted cassava plants of the above mentioned varieties were maintained in the screen house until 4 to 5 weeks of age. They were then transferred to a whitefly infestation chamber in the greenhouse and subjected to *A. socialis* oviposition for approximately 36 hours. Infested varieties were maintained in a growth room for exposure to the parasitoid *E. hispida.*

The *E. hispida* colony was developed by collecting cassava leaves with whitefly parasitized pupae from the field. These leaves were placed in plastic boxes with a paper towel on the bottom and darkened with black cheesecloth. Clear glass gars were

The *E. hispida* colony was developed by collecting cassava leaves with whitefly parasitized pupae from the field. These leaves were placed in plastic boxes with a paper towel on the bottom and darkened with black cheesecloth. Clear glass gars were connected to an opening in the lid of the box, where parasitoids were drawn to the light and collected.

Studies on *A. socialis* instar preference by *E. hispida* were done on the variety CMC-40. Adult whiteflies were placed in small leaf cages on cassava leaves and allowed to oviposit for 8 hours. This was done periodically so that the cassava leaves eventually contained patches of inmatures of instars I, II, III and IV of *A. socialis*. Infested leaves were isolated by placing a nylon mesh "bag" over each leaf. Twenty five (25) females collected from the field were released into each bag.

Studies on the biology of *E. hispida* were done on six-week old plants, and *A. socialis* infestations were done every second day with small leaf cages. When nymphal "patches" reached the third instar, one *E. hispida* parasitoid was introduced to each patch; there were 30 repetitions. The parasitoid was transferred to patches of the same instar three times each week until parasitoid death. The patches, with parasitized nymphs, remained on the plants until parasitoid emergence.

Results: of this experiments show that *E. hispida* prefers to parasitize third and forth instar nymphs (Figure 6.7). There was no significant difference between the two instars. Parasitism of the first instar was negligible, and very low in the second instar. A high number of nymphs had no parasitoid emergence. This phenomena occurred for all instars but was significantly higher in the forth instar (Figure 6.8). These non emerged nymphs were either "non viable nymphs" or, as reported in the literature within the genus *Encarsia*, it is characteristic for the adult parasitoids to feed on its host. This host-feeding characteristic can cause considerable nymphal mortality, especially in the early instars. The fact that the highest number of non-viable nymphs were in the forth instar indicates a possible preference for parasitoid feeding on this instar.

The survival of *E. hispida* does not appear to be adversely affected by any of the four genotypes used in this experiment (Figure 6.9). Female adult longevity was 27, 28, 32 and 35 days respectively on the varieties MEcu 27, CMC-40, MBra-12 and CG 489-34. MEcu 72 and CMC-40, the highly resistant and susceptible varieties respectively gave very similar results related to longevity. Why longevity is about 25% longer on CG 489-34 is not known, unless there are chemical factors in the leaf that are conducive to parasitoid longevity. Leaves of MEcu-72 are highly pubescent whereas those from CMC-40 are non-pubescent; leaves from MBra-12 and CG 489-34 are intermediate. It has been suggested that pubescence might play a role, perhaps a detrimental effect, to parasitoid longevity. These results do not support that hypothesis.

The emergence of *E. hispida* parasitoids from *A. socialis* pupae on the variety CMC-40, indicate that peak oviposition occurs on about the third days after parasitoid emergence, and tapers off rapidly with continued oviposition for about 23 days (Figure

to get results from MBra-12 as plant leaves dried up and dropped during the experiment. These results strongly indicate that whitefly resistant genotypes could have a detrimental effect on biological control agents, especially parasitoids.

It can generally be concluded, from these experiments, that *E. hispida* will parasitize all instars of *A. socialis* but is most successful on 3rd and 4th instars. There was no genotype effect on survival and longevity of *E. hispida* and that leaf tricomes do not alter these factors. However the low parasitoid emergence rate on the resistant cultivars MEcu-72 and CG489-34 indicate a possible negative effect of whitefly varietal resistance on biological control.

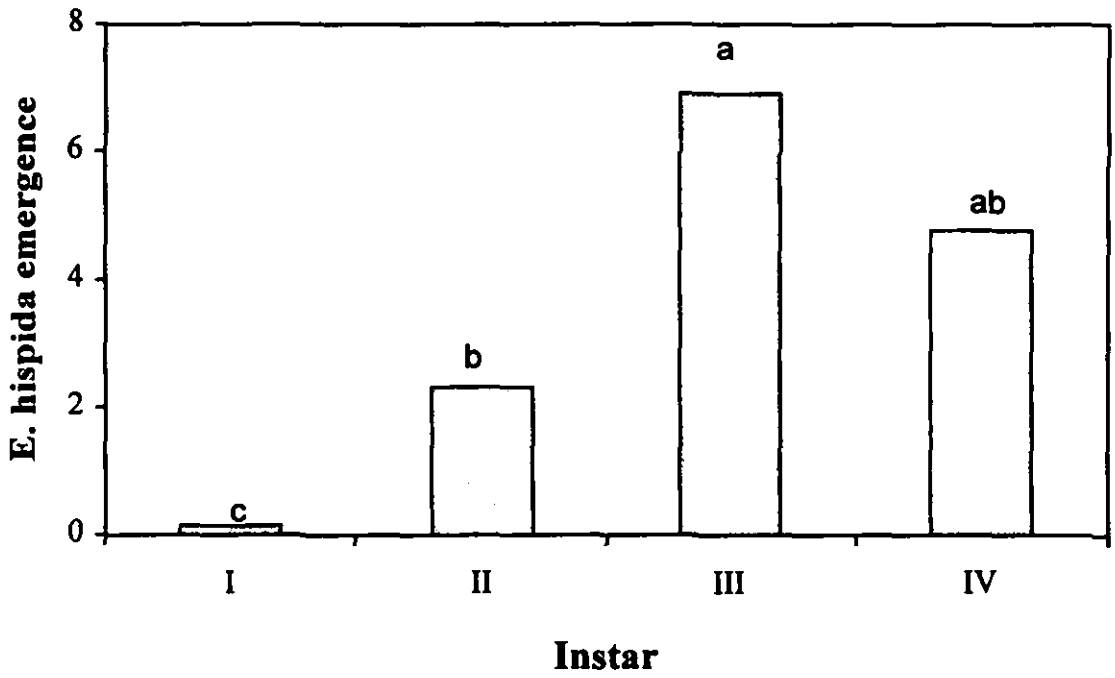


Figure 6.7. Emergence of the parasitoid *Encarsia hispida* from four instars of the cassava whitefly *A. socialis*. Columns with different letters represent statistically different values.

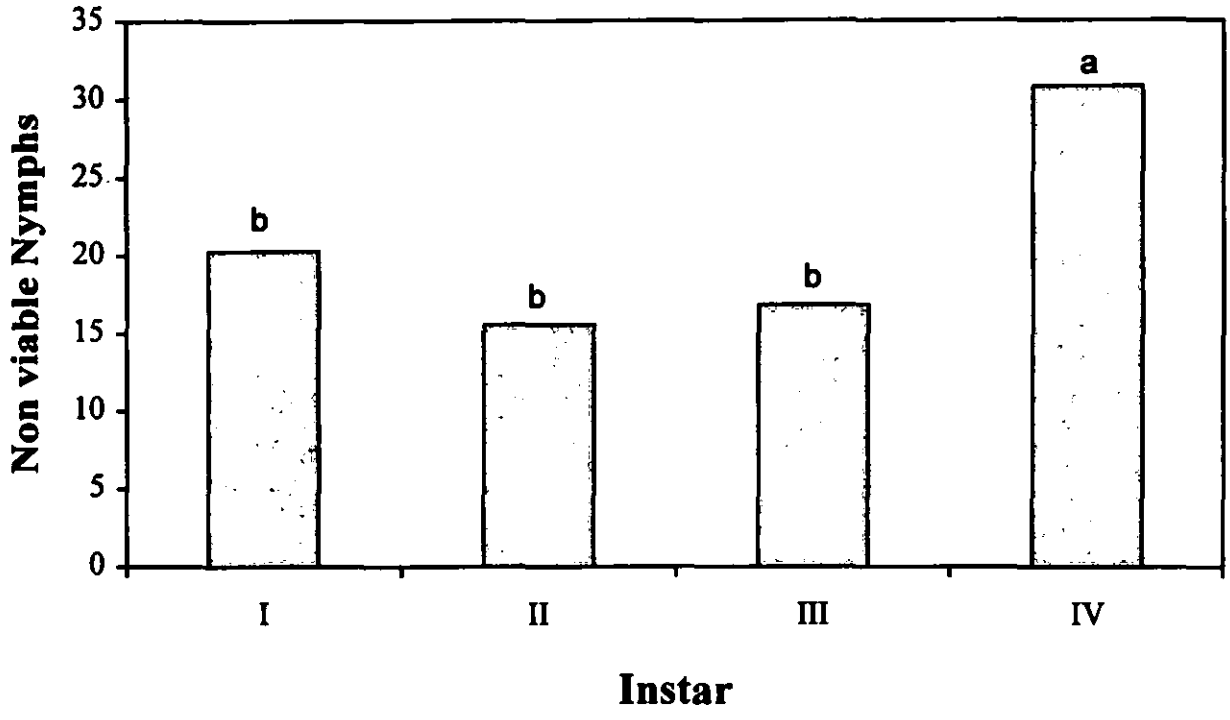


Fig. 6.8. *A. socialis* nymphs that were "non-viable" based on the lack of *E. hispida* emergence. Columns with different letters represent statistically different values.

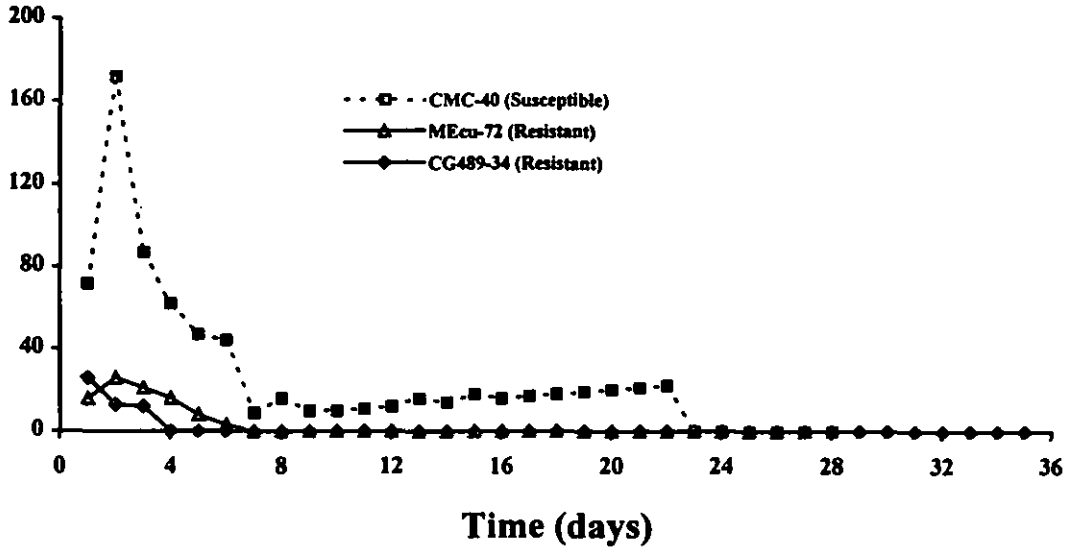


Figure 6.9 The effect of four cassava varieties on the survival of the *parasitoid E. hispida* on the cassava whitefly *A. socialis*.

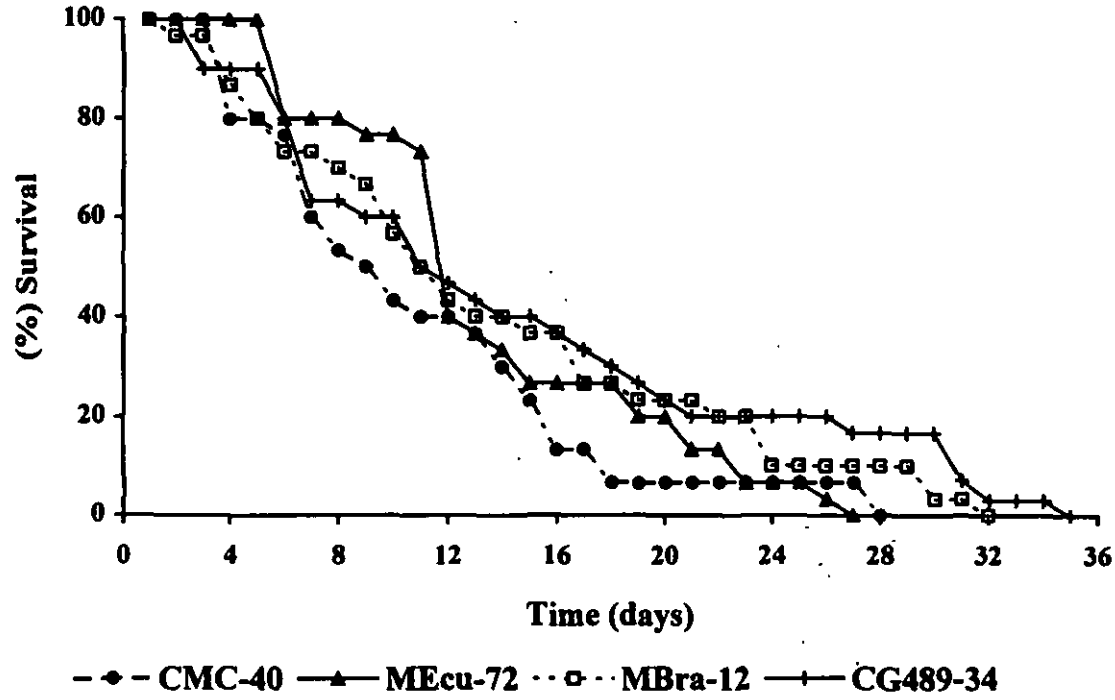


Figure 6.10. The effect of three cassava varieties on the emergence of whitefly parasitoid *E. hispida* from parasitized *A. socialis* pupae.

***Activity 6.6. Screening cassava germplasm for resistance to mites* (Mononychellus tanajoa).**

Specific Objective:

a) To continue the screening and evaluation of cassava germplasm resistant to the green mite.

Rationale: the cassava green mite *M. tanajoa* (syn = *Mononychellus pregresivus*) is native to the neotropics and was first reported in 1938 from NE Brazil. The natives had long known the damage symptoms, which provides the name tanajoa (a sick or diseased plant). It attacks young leaves and meristems of cassava; infested leaves develop yellow spots, lose their normal green color, develop a mottled, bronzed, mosaic like appearance, and become deformed. Under severe attack, shoots lose their green color, turn rough and brown, eventually presenting dieback. Stems and leaves become progressively necrotic from top to bottom; severe damage stunts plant growth and induces branching. It is normally a serious problem only in dry regions.

Yield losses ranging from 20 to 50% have been reported from the Americas. (Bellotti et. al.,1999). The mite first appeared in Africa (Uganda) in 1971; by 1985 it had spread

across most of the African cassava belt, occurring in 27 countries. Today it is one of the principal pests of cassava in Africa, causing estimated root yield losses of 13-80%.

Application of acaricides is not recommended as it is not an economical option for low-income farmers and also because of their adverse effect on natural control agents. Host plant resistance (HPR) and biological control (see AR, Project PE1) offer the most promising approaches to find a practical, long lasting solution. Many years of research at CIAT, IITA, and national research programs (i.e. CNPMF, EMBRAPA, Brazil) has shown that high levels of resistance is probably not available in cassava germplasm. Nevertheless, after substantial effort, cultivars with low to moderate levels of resistance have been developed and are being grown by farmers.

Materials and Methods: approximately 4,560 varieties from the CIAT germplasm bank have now been evaluated for mite resistance; 6.8% (or 310 varieties) of these have been selected as having low to moderate levels of resistance to *M. tanajoa*, after several (2 to 7) evaluation cycles. Low to moderate resistance is indicated by a 0 to 3.5 damage rating on a 0 to 6 evaluation scale.

Mite damage evaluations are traditionally done at two sites, Pivijay, Magdalena, on the Colombian Atlantic Coast and at CIAT, Palmira. Mite populations at Pivijay, because of the prolonged dry season, are usually higher than populations at CIAT where dry seasons tend to be shorter and yearly precipitation is higher. During 1998/99, 398 cassava cultivars were planted at Pivijay; the majority of these had been selected as promising for resistance in previous trials. The susceptible checks planted were MCol 22, MCol 1468 (CMC 40), MCol 2215, MCol 1505, CN 3306-4, and the resistant hybrid CG 1141-1 (ICA-Costeña) was also included.

Results: mite populations during this cycle were low and, in general, plant damage levels were also correspondingly low. Results show that 388 cultivars (85%) had damage ratings of 3.0 or lower (Figure 6.11). However, since most of the varieties evaluated had previously been selected as resistant, these low damage ratings were not unexpected.

Of the 310 varieties that, over numerous years, have been selected as promising for resistance to mites, 72 of these have consistently, over several crop cycles, had damage ratings below 3.0 (Table 6.11). This list consists of varieties originally collected from several neotropical countries, but is dominated by Brazil (14), Colombia (14), Perú (17), Venezuela (7), and Ecuador (6). The list also includes several hybrids. The varieties MEcu 72, MEcu 64 and MPer 415 and the hybrid CG 489-31 have also been selected for resistance to whiteflies.

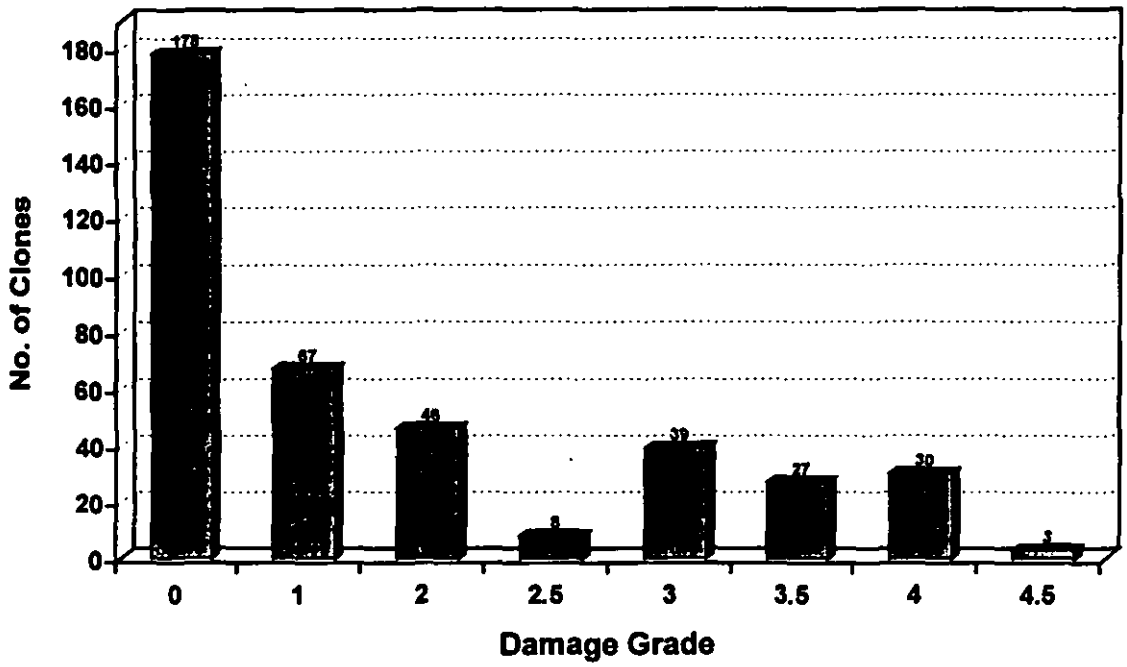


Figure 6.11. Evaluations of 398 selected cassava cultivars for damage caused by the cassava green mite (*Mononychellus tanajoa*) at Pivijay (Magdalena, Colombia) (1998-1999).

***Activity 6.7. Studies on resistance mechanisms for the cassava green mite,* M. tanajoa.**

a) to determine if resistance/tolerance to green mite is based on antixenosis or antibiosis mechanisms.

Rationale: in previous studies, mite resistance in cassava, has usually been expressed as antixenosis (preference vs. non- preference) or antibiosis. A clear understanding of the physiological basis of resistance/tolerance to insects and other arthropods is fundamental for an efficient utilization and exploitation of different mechanisms of control.

Materials and Methods: antixenosis studies for *M. tanajoa* oviposition were carried out on several resistant cultivars. The susceptible check was CMC 40 (MCol 1468). The experimental arena utilized was a petri dish with a layer of moisturized cotton on the bottom. Two leaf discs (3 cm^2), one from CMC 40 and the other from a resistant cultivar were placed, slightly overlapping, on the moist cotton. Five female *M. tanajoa* mites were placed on each disc and allowed to oviposit for 72 hours. There were 30 repetitions of each treatment. In a separate study, *M. tanajoa* mite females were

allowed to oviposit on numerous selected cassava cultivars. This was a "non-choice" study in that females were obligated to oviposit on the cultivar offered and could not migrate to a "preferred" cultivar. The experimental arena was as described above.

Table 6.3. Cassava cultivars with mite (*M. tanajoa*) damage ratings of 3.0 or lower after numerous evaluation cycles in two ecosystems (CIAT, Palmira and Pivijay, Magdalena).

	Damage grade*	
Variety	**Pivijay**	**CIAT**
CG 5-99	2.0 - 3.0 - 3.0	3.0 - 2.5
CG 406-5	1.0 - 2.0 - 2.0	2,5 - 2.0 - 2.5
CG 489-31	2.5 - 1.0 - 1.0 - 2.5 - 0	2.5 - 2.0 - 2.0
CG 489-57	3.0 - 1.0 - 2.0	3.0 - 3.0
CG 502-1	2.0 - 2.5 - 1.0 - 0 - 0	2.0 - 3.0
CG 1141-1	3.0 – 3.0 – 3.0 - 3.0 – 3.0 – 2.0 – 0	3.0 – 2.0 – 1.0
SG 350-23	1.0 - 1.0 - 2.0 - 0	3.0 - 2.0 - 2.0
SG 698-3	3.0 - 3.0 - 1.0	2.5 - 3.0
CM 4574-7	3.0 - 2.5 - 3.0 - 1.0 - 0	3.0 - 3.0 - 3.0
CM 6173-8	3.0 - 3.0	2.0
MBra 64	3.0 - 2.0 - 2.0 - 2.5 - 1.0	2.5 - 2.0 - 2.0
MBra 69	1.0 - 2.0 - 2.0 - 1.0	2.5 - 3.0
MBra 137	2.5 - 2.5 - 1.0 - 0	3.0 - 3.0
MBra 173	3.0 - 2.0 - 1.0 - 2.0 - 2.0 - 0	3.0 - 3.0 - 3.0
MBra 191	3.0 - 3.0 - 3.0 - 1.0 - 2.0 - 0	3.0 - 3.0 - 3.0
MBra 225	2.5 - 3.0	3.0 - 3.0
MBra 235	3.0 - 2.0 - 1.0 - 2.5 - 0	2.5 - 2.0
MBra 245	1.0 - 1.0	2.5 - 3.0
MBra 276	2.5 - 2.0 - 2.0 - 0	3.0
MBra 292	2.5 - 3.0	3.0 - 2.0
MBra 391	2.0 - 3.0 - 2.0 - 0	2.5 - 2.5 - 3.0
MBra 404	1.0 - 2.5 - 2.0 - 0	3.0 - 3.0 - 3.0
MBra 420	1.0 - 3.0 - 0	3.0 - 2.5
MBra 826	3.0 - 3.0	3.0
MCol 282	2.5 - 1.0 - 3.0 - 1.0	3.0 - 3.0
MCol 336	2.5 - 2.0 - 2.0 - 0	3.0 - 2.5 - 3.0
MCol 548 A	3.0 - 2.5 - 2.0 - 3.0	3.0 - 2.5 - 3.0
MCol 576	0 - 0	2.0
MCol 593	1.0 - 3.0 - 0	3.0 - 2.0
MCol 1254	3.0 - 2.0 - 2.0 – 0	3.0 - 2.5 - 3.0
MCol 1336	3.0 - 2.0 - 3.0	3.0 - 2.5 - 3.0
MCol 1373	3.0 - 3.0 - 2.0	3.0 - 2.5 - 2.0
MCol 1432	2.0 - 2.0 – 0	2.0 - 3.0
MCol 1439	2.0 - 2.0 - 1.0 – 0	3.0 - 2.0 - 3.0
MCol 1856	2.0 - 3.0 - 1.0	3.0

	Damage grade*	
Variety	**Pivijay**	**CIAT**
MCol 1926	1.0 - 1.0 – 0	2.0 - 2.5 - 3.0
MCol 2019	3.0 - 2.0 - 3.0 – 1.0 - 0	3.0 - 2.5 - 3.0
MCol 2179	3.0 - 1.0 - 3.0 – 0	3.0 - 3.0 - 2.5
MCR 20	2.0 - 3.0 – 0	3.0 - 3.0
MCR 52	2.0 – 0	2.5 – 3.0 – 3.0
MPer 221	3.0 - 3.0 - 1.0	3.0 - 3.0 - 2.5
MPer 266	2.0 - 3.0 - 2.5 - 1.0	2.5 - 3.0 - 2.5
MPer 320	2.5 - 0	3.0
MPer 324	3.0 - 0	3.0 - 3.0- 2.5
MPer 365	3.0 - 3.0 - 2.0 - 2.0 - 0	3.0 - 3.0
MPer 366	2.0 - 3.0 - 2.0 - 1.0 - 2.0	3.0 - 3.0 - 3.0
MPer 394	2.0 - 2.0 - 2.0 - 0	3.0 - 3.0 - 2.0
MPer 415	3.0 - 2.0 - 1.0 - 0	2.0 - 2.0 - 2.0
MPer 435	3.0 - 2.0 - 3.0 - 0	2.5 - 2.0 - 2.0
MPer 460	1.0 - 0	3.0
MPer 461	2.0 - 1.0 - 0 - 0	2.5 - 2.5
MPer 463	1.0 - 2.5 - 2.0	2.5 - 3.0 - 2.5
MPer 523	1.0 - 1.0 - 3.0	3.0 - 2.5 - 3.0
MPer 562	2.5 - 1.0 - 1.0 - 3.0	3.0 - 2.5 - 3.0
MPer 564	2.0 - 2.0 - 0 - 3.0 - 0	2.5 - 2.5 - 1.0
MPer 608	2.0 - 2.0 - 0	3.0 - 2.0 - 3.0
MPer 611	2.0 - 1.0 - 2.5 - 3.0 - 1.0	2.5 - 2.0
MEcu 48	3.0 - 3.0 - 0	2.5 - 2.5
MEcu 58	2.0 - 1.0 - 1.0	2.5 - 2.0 - 2.5
MEcu 64	2.0 - 2.5 - 1.0 - 3.0 - 1.0	2.5 - 3.0 - 3.0
MEcu 72	3.0 - 3.0 - 3.0 - 2.5 - 2.0 - 0	2.5 - 2.0 - 2.5
MEcu 87	2.0 - 3.0 - 3.0 - 3.0	3.0 - 2.5 - 2.5
MEcu 97A	2.0 - 2.0 - 0	2.5 - 30 - 3.0
MGua 86	3.0 - 1.0 - 1.0 - 0	3.0
MPan 127	3.0 - 1.0 - 3.0	3.0 - 2.5
MVen 54	2.5 - 3.0 - 3.0 - 0	2.5 - 2.5 - 3.0
MVen 68B	2.5 – 3.0 – 3.0 – 3.0	3.0 – 2.5 – 3.0
MVen 146	1.0 - 3.0 - 1.0	3.0 - 3.0
MVen 174	2.5 - 2.5 - 3.0 - 1.0- 2.0 - 0	3.0
MVen 216	2.5 - 3.0 - 0	3.0
MVen 276	2.5 - 1.0 - 2.0 - 1.0 - 3.0	3.0 - 2.0 - 3.0
MVen 291	2.0 - 2.0 - 2.5 - 2.0	3.0 - 2.5 - 3.0
CM 3306-4	3.5 - 4.0 - 4.0 - 4.0 - 4.0	3.5
MBra 12	4.0 - 4.0 - 3.5 - 4.0 - 3.0	2.5 - 3.5
MCol 22	4.5 - 4.0 - 4.5 - 4.5 - 4.5 - 5.0	4.0
MCol 2215	3.5 - 3.5 - 4.0 - 4.0 - 3.0 - 4.0 - 4.0	4.0
MCol 1468 (CMC 40)	4.0 - 4.0 - 3.5 - 4.5	4.5
MCol 1505	3.5 - 3.0 - 2.0 - 4.5 - 4.5 - 4.0	4.5

*Each damage rating represents the maximum rating in each evaluation cycle.

Results: Table 6.3 present the current accumulated results from two sites, of the most promising sources of resistance in cassava to the green mite. The results from Table 6.4 indicate a strong ovipositional preference for *M. tanajoa* to oviposit on susceptible vs. resistant varieties. When CMC 40 was paired with CMC 40, oviposition was nearly equal on the two leaf discs. However, in every trial where CMC 40 was paired with a resistant cultivar, oviposition was considerably higher on CMC 40, the susceptible variety. For example, when paired to MEcu 72, 94.9% of the eggs oviposited were on CMC 40; when paired with MPer 611, 90.9% were oviposited on CMC 40; and when paired with MEcu 64, 88% were oviposited on CMC 40. Mbra 12 is a "tolerant variety" often displaying "field resistance," but laboratory evaluations for antibiosis usually result in little difference when compared to susceptible varieties. These evaluations for ovipositional preference indicate an antixenosis mechanism involved for MBra 12. When paired with the resistant cultivar CG 1141-1, oviposition was similar; however when paired with CMC 40 oviposition was considerably lower. These results may help explain the "field resistance" observed with MBra 12.

Table 6.4. Female mite (*M. tanajoa*) oviposition on resistant cultivars compared to the susceptible cultivar CMC 40, in laboratory studies[1].

Varieties	No. Eggs
CMC 40	731
CMC 40	812
CMC 40	1829
MCol 1505	694
CMC 40	1939
CG 1141-1	605
CG 1141-1	537
MBra 12	695
CMC 40	2460
MEcu 72	133
CMC 40	863
MBra 279	279
CMC 40	976
SM 643-17	513
CMC 40	1551
CM 6173-8	610
CMC 40	1032
MCol 2179	644
CMC 40	1395
MEcu 64	183
CMC 40	1179
MRex 71	436
CMC 40	2247
MPer 611	226
CMC 40	1769
MBra 12	800

[1] 5 female mites/30 repetitions/cultivars/3 days.

Results in this study are varied. CMC 40 the susceptible check had, as expected, a high rate of oviposition, 5.26 eggs per day (Table 6.5). However several other cultivars had similar levels of oviposition (i.e. MMex 71, SM 1181-3, MPer 415, and CG 6119-5) indicating that, in these genotypes, there may not be a strong enough antixenosis factor to actually reduce or prevent oviposition, but strong enough influence a "choice" in oviposition.

Table 6.5 Average daily oviposition of the cassava green mite (*M. tanajoa*) on resistant and susceptible cassava cultivars in laboratory studies[¶].

Cultivar	No. Eggs/day	Maximum damage rating	
		Pivijay	CIAT
CG 502-1	2.48	2.5	3.0
CG 489-4	2.97	3.5	3.5
CG 489-31	3.06	3.0	2.5
CG 489-34	2.87	3.5	3.5
CM 6173-8	4.67	3.0	2.0
SM 643-17	4.33	3.5	4.0
MBra 235	4.60	3.0	2.5
MBra 276	4.20	2.5	3.0
MCol 576	3.60	0	2.0
MCol 1468 (CMC 40)	5.26	4.5	4.5
MEcu 48	3.26	3.0	2.5
MEcu 58	4.25	2.0	2.5
MEcu 64	4.52	3.0	3.0
MEcu 72	2.60	3.0	2.5
MPer 415	5.36	3.0	2.0
MPer 611	4.32	3.0	2.5
MVen 276	4.47	3.0	3.0
CG 6119-5	4.71	3.5	3.0
SM 1181-3	5.62	4.0	3.0
MCol 1351	4.07	4.0	3.0
MMex 71	5.17	3.5	3.0
MVen 116	4.04	2.5	3.5

[¶] 1 female mite/30 repetitions/cultivars/3 days.

However, there is another group of cultivars where the actual numbers of eggs oviposited was reduced considerably (Table 6.5). These include MEcu 72 (2.6 egg/day), CG 502-1 (2.5), CG 489-4 (2.97), CG 489-31 (3.1), CG 489-34 (2.9) and MEcu 48 (3.3). Interestingly CG 489-4, CG 489-31 and CG 489-34 are progeny from a MEcu 72 x MBra 12 cross, and are also resistant to whiteflies (*A. socialis*) where similar experiments have shown an antixenosis mechanism to whitefly oviposition. These

results indicate that there may exist in these cultivars, a strong enough antixenosis mechanism (*substance ?*) as to actually cause a reduction in oviposition.

The results so far obtained suggest that there may be two separate types of antixenosis mechanisms involved. One being a preference for one cultivar over another, the second having an actual deterrent to oviposition. These studies will continue to try to better define this phenomenon.

Activity 6.8. Evaluation of wild Manihot *species for resistance to mites.*

Summary: studies were initiated to evaluate the possibility of obtaining a source of mite resistance from wild *Manihot* species. In these initial studies, *M. tanajoa* oviposition was measured on six *Manihot* species using the methodology described above (Activity 6.7).

The following results were obtained:

Manihot species	Average daily oviposition
M. grahami 3	4.98
M. carthaginensis 15A	4.30
M. chlorosticta 2	4.27
M. carthaginensis 13	3.03
M. Alutacea 10	1.6
M. quinqueloba 4249	2.4

As can be observed, at least two species show very low oviposition, *M. alutacea 10* and, *M. quinqueloba 4249*.

The use of wild Manihot species to obtain resistance to pests, in cases where high level of resistance in *M. esculenta* is not available, is a tempting opportunity. This is particularly true now, that we have the cassava gene map and the distinct possibility of transgenic plants. The future of this area of research will depend, in part, on resources available.

Activity 6.9. Evaluation of cassava germplasm for resistance to the stem borer Chilomima clarkei.

Specific Objective:
a) to identify cassava germplasm with resistance to C. clarkei.

Rationale: a complex of arthropod stem borers feed on, and damage cassava. This complex includes both coleopteran and lepidopteran species. Species of the genus *Lagochirus* (long horned beetles) are worldwide in distribution, but do not appear to cause sufficient damage to result in yield losses. Stem borers are of particular importance in the neotropics where they are reported causing severe damage and yield losses in Colombia and Brazil. Seven species of the genus Coelosternus (Coleoptera: Curculionidae) are reported attacking cassava in the Americas, especially in Brazil where literature reports indicate that they can reduce root production and the quality of planting material. However they are generally reported as causing sporadic or localized damage, not resulting in significant yield losses.

In recent years, especially in the northern part of South America (Colombia and Venezuela), the lepidopteran stem borer, *Chilomima clarkei* (Fam: Pyralidae), is causing considerable crop damage. Recent reports and observations also indicate that the pest is present on cassava in Paraguay and Southern Brazil. Adult females oviposit on cassava stems usually around the bud and upon hatching, first instar larvae feed on the outer bark or stem epidermis. At the 2nd and 3rd instar it bores into the stem where it completes its larval cycle, pupaes and emerges as a winged adult. Numerous borers may occupy or infest the same stem causing extensive tunneling, frequently resulting in stem breakage. Considerable stem breakage (more than 35% of the plants) will result in significant yield loss (45 to 62%). In addition, tunneling damage in the stems may cause rotting and a reduction in the quality of planting material, and the number of cuttings produced.

Once the *C. clarkei* larvae enter into the stem it is very difficult to control and pesticide applications are not effective. The early instar larvae are vulnerable while they are feeding on the outer stem surface and could be controlled in this stage with applications of a biopesticide such as *Bacillus turingensis*. However since there are overlapping generations, several applications would have to be made to achieve effective control and this would probably be too costly for cassava producers.

Adult stem borers are very mobile and this mobility probably accounts for the dissemination of *C. clarkei* into many cassava growing regions. However it has not reached the Cauca Valley and this has limited, to a certain extent, our ability to adequately study this pest. In our research activities we are pursuing two related strategies:

Materials and Methods: *C. clarkei* has now invaded several areas of Colombia including the Atlantic Coast, Tolima, The Llanos Orientales and Caldas. This latter observation indicates that it is getting closer to Valle del Cauca, and probably a matter of time before it reaches it. Highest populations have been observed on the Atlantic Coast, especially in Magdalena where we have been evaluating germplasm for the past several years.

During 1998-1999 approximately 400 cassava clones were evaluated at Pivijay, Magdalena. Evaluations were done by counting the number of stem borer holes and tunnels in cassava stems, and the percentage of broken stems, using the following scale:

Population Grade	# Holes/plant	Damage grade	% Broken stems
0	0		
1	0.1 - 1	0	0
2	1.1 - 2.5	1	1 - 10
3	2.6-5	2	11 - 25
4	5.1 - 10	3	26 - 50
5	> 10	4	51 - 75
		5	76 - 100

Results: of this evaluation indicate that *C. clarkei* populations were moderate to high as 88% of clones had at least one hole/tunnel damage per plant (Figure 6.12). Ninety nine percent of the clones presented *C. clarkei* damage and 1% (or four clones) presented no damage. These four clones were MCol 135, MCol 313, MPer 423 and MVen 320. Forty seven clones (11.8%) had low population/damage levels of less than one hole/larvae per plant; 80% presented high populations and 6.5% had more than 5 holes/larvae per plant.

These results, combined with previous evaluations, are allowing us to build up a small gene pool exhibiting resistance to *C. clarkei*. These selected cultivars will need to be evaluated over several cycles. In the future, however, we will have to identify another evaluation site due to lack of security in the Magdalena region.

Field evaluations of germplasm using natural populations of a highly mobile pest such as *C. clarkei* can often give misleading results. Plants showing no damage may be *"escapes"* (adult females by chance did not oviposit on these clones), rather than be indicative of resistance. The employment of "artificial" infestation from a pest colony, or continued field evaluations in areas of high pest incidence, over a period of several years can offset the effect of chance escapes.

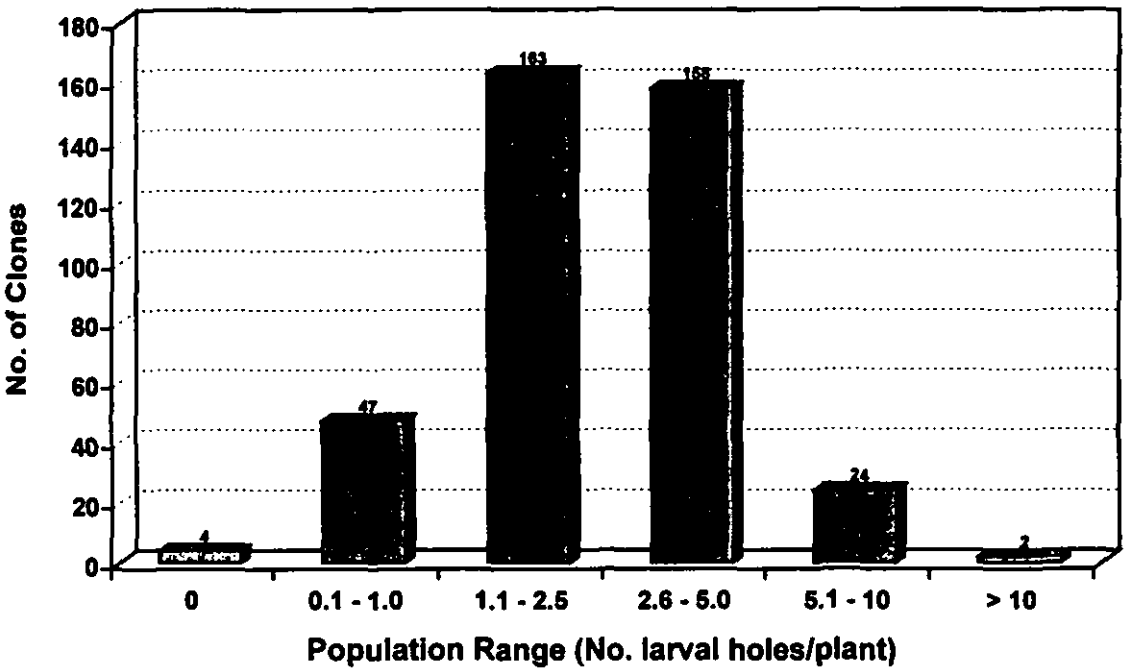


Figure 6.12 Evaluation of cassava clones for damage caused by the stem borer *C. clarkei* at Pivijay, Magdalena, Colombia (1999).

***Activity 6.10. Preparation of artificial diets for laboratory rearing of* C. clarkei.**

Summary: artificial diets for laboratory rearing of insects, such as *C. clarkei*, can often help facilitate research in insect behavior and nutrition, and in studies for host plant resistance and biological control. *C. clarkei* is difficult to rear in adequate numbers in the laboratory when obliged to maintain colonies feeding on cassava cuttings. Due to its long life cycle, manipulation of larvae, transferring them from deteriorating cuttings to fresh cuttings is both time consuming and often causes injury or death of inmatures.

In order to carry out research on host plant resistance (see Activity 6.9) and facilitate bioassays of Bt toxin, studies were initiated to develop an artificial diet for *C. clarkei*. Four different diets were tested: two diets, known to be effective for rearing lepidopterans, were obtained. Diet # 1, used for rearing *Helicoverpa armige* (Noctuidae), has a base of wheat germ, corn flour and corn oil. Diet # 2, effective for rearing *Homoeosoma nebulella* (Pyralidae) differs from the first diet by containing sunflower seed oil instead of corn oil. *C. clarkei* larvae that we received from Pivijay could not feed successfully on either diet.

Diet # 3 and 4 were transformed from Diets # 1 and 2, by substituting "flour" from pulverized cassava stems for corn flour. *C. clarkei* feed on these and can survive for a long period, but eventually died. Diet # 4 which consists of the basic Pyralidae diet plus

cassava stem flour will be further modified and tested until we find a diet that will successfully sustain *C. clarkei* through the adult stage.

To facilitate these studies we have the collaboration of CORPOICA on the Atlantic Coast. They are maintaining a colony of *C. clarkei* on cassava cuttings and are able to send us eggs or early instar larvae to test on the artificial diets. *C. clarkei* reared at CIAT is sacrificed before they reach the adult stage to prevent the introduction of this pest into the region.

OUTPUT 7: Molecular Markers for Gene Mapping and Genetic Diversity Assessment in Cassava

Activity 7.1 Molecular Mapping of Genes Conferring Resistance to the African Cassava Mosaic Disease (CMD) in African Cassava Germplasm.

Specific Objectives:

a) to map genes for resistance to ACMD in cassava.
b) to develop molecular markers for breeding of resistance to ACMD in Latin American and Asian germplasm.

Summary: The Cassava Mosaic Disease (ACMD) is a viral disease of cassava and was first reported in East Africa by Warburg in 1894. It is endemic in all cassava growing regions of Africa and India, with disease epidemics occurring in a cyclic fashion in sub-Saharan Africa. The causal agents of ACMD , the African Cassava Mosaic Virus (ACMV), is a geminivirus , with the white fly, *Bemisia tabaci* as vector. ACMV is not known in the Americas, neither is the white fly biotype vector. However, this biotype has recently invaded the New World. The presence of this pest in the Americas increases the possibility that ACMV or a native geminivirus will cross over to cassava.

A project to develop molecular tags for ACMD resistance for marker-assisted resistance breeding in Latin American germplasm was initiated in collaboration with IITA, with funding from the Rockefeller foundation. A half-sib back-cross population of 276 plants, obtained by crossing five F_1 individuals of the cassava mapping population to TMS 30572 female parent of the mapping population, was established *in vitro* at CIAT as an ACMD mapping population. The population was subcloned, and one copy was shipped to IITA, Ibadan, Nigeria. TMS 30572 represents the major source of deployed ACMD resistance in Africa. The ACMD mapping population was evaluated for field resistance by a randomized complete block design with 3 replicates, 6 genotypes per replicate, in 2 high disease pressure sites at 3 and 6 months after planting.

The half-sib backcross population has been genotyped with 70 RFLP markers from the frame work map of cassava. Currently polymorphic SSR from the 186 Cassava MapPairs SSR markers developed at CIAT, are also being placed on the map to complete the BC_1 frame work map. Parallel to the BC_1 mapping effort, a bulk segregant analysis (BSA) approach was conducted to quickly identify markers linked to CMD resistance. The entire set of 186 SSR markers were screened on bulked DNA from 10 resistant, and 5 susceptible genotypes, from a single family of the half-sib BC_1 population. Ten SSRs were found to be polymorphic in the bulks. The markers were then evaluated in individuals of the bulks, and 1 SSR marker, SSRY40, was polymorphic between the resistant and susceptible genotypes, with one recombinant. Segregation and linkage analysis of SSRY40 in F_1 cassava map progeny placed the marker on linkage group D of the female-derived molecular genetic map of cassava. A considerable portion of linkage group D is thought to be an introgression from *Manihot glaziovii,* as evidenced from a large number of polymorphic markers, significantly

reduced recombination, rare alleles (Fregene et al. 1997). Resistance to CMD in TMS30572 was introgressed from *Manihot glaziovii.*

CMD resistance data of the half-sib back-cross family, at 6 months after planting, was regressed on the marker genotype classes of SSR marker CSY1, and other RFLP markers on Linkage group D. Results reveal that an RFLP marker, GY59, and CSY1 marker flank a region of group D that explains 50% of the phenotypic variance of CMD resistance ($P<0.0001$). These markers, GY59 and CSY1, are separated by a distance of 15cM, and no other markers have been found between them.

During a recent trip to IITA, the CIAT virologist, and cassava geneticist visited the mapping population in the field after the plants were de-topped. The CIAT ACMD mapping population was heavily infested, and a clear-cut range of symptoms was not easily obtained. However an F_1 mapping population, in the same field, developed from crossing a Nigerian land race that represent a new source of ACMD resistance, with a susceptible line showed qualitative expression of resistance, with resistant plants being almost immune, while susceptible plants were heavily infested. Resistant and susceptible plants were counted for the F_1 300 plants, and the ratio was not significantly different from a 1:1 ratio by a Chi square test at a probability level of 0.05. This suggests a single dominant gene heterozygous in the ACMD resistance parent.

Since the source of the ACMD resistance in the CIAT population was found to be inadequate and is known to be recessive, requiring a homozygous state for the gene to be effective, it was decided to expand the gene mapping to the novel sources from IITA. CIAT has invited Dr 'Tunji Akano, IITA post-doc working on the field evaluation of the mapping populations to come over with the particular IITA mapping population, to work on the development of markers for this new source of resistance. The Nigerian land race, source of this novel resistance has been requested from IITA for further use in breeding for resistance to ACMD at CIAT.

Achievements:

- Description of a molecular tag for currently deployed resistance to ACMD.
- Identification of a more durable, and a qualitative novel source of resistance to ACMD.
- Novel source of resistance to ACMD for adapting Latin American cassava gene pools to ACMD.

Activity 7.2 Molecular Mapping of Early bulking, Root Quality Traits and Morphological Characters in Cassava.

Specific Objectives:

a) *to map quantitative trait loci (QTL) controlling Post-Harvest Deterioration, and Starch quantity/quality in cassava.*
b) *to understand the components of Early bulking and map corresponding genes, in cassava.*
c) *to map genes controlling morphological caharcters in cassava.*

Summary: The OTL mapping project of root quality and other agronomic traits at CIAT is a continuation of activities in the construction of a molecular genetic map of cassava and its application to cassava breeding. The objective of this study is to identify markers tightly associated with genes controlling root quality, early bulking, and morphological traits of interest to cassava breeding, as a starting point for marker assisted selection of these traits to increase the efficiency and cost effectiveness of cassava improvement.

The F_1 mapping population comprising 144 genotypes, was established in January 1998 at Palmira and Quilichao in a replicated trial, using a partially balanced triple lattice design with three replicates of 12 blocks each. Plots were made up twenty plants per genotype, per replication. A second year trial of the same population using the same design was planted in January, 1999 at Quilichao and Palmira. Morphological characters (plant height, branching levels height of first branch, leaf shape and length of stem with leaves) were evaluated in October 1998 on 6 non-border plants at 10 MAP. Other traits, including dry matter percent, dry matter yield, starch content, culinary quality, post harvest deterioration were evaluated at 11 MAP using the six central plants of each plot. Post-harvest physiological deterioration was evaluated at 5, 10, and 15 days, scored on randomly selected storage root. Leaf morphology was assessed both as qualitative and quantitative trait. TMS 30572, the female parent of the F_1 progeny has elliptic shaped leaves, while the male parent, CM 2177-2 is linear shaped. Leaf morphology was scored qualitatively based on shape. Quantitative measurement was done using the width/length ratio of the central leaf lobe as trait value.

The early bulking trial to investigate initiation and rate of starch accumulation was set up in December 1998 using 80 genotypes selected from the F_1 population. Based on the result from the first year QTL mapping trial at Palmira in 1998, the best forty genotypes and lowest forty genotypes for dry matter yield at 7 MAP were selected. These genotypes were planted in a randomized complete block design with two replications. Starting from 6 WAP, sequential harvesting was done at three-weekly interval up to 30 WAP. The early bulking trial was evaluated for dry foliage yield, storage dry matter yield, harvest index, storage root diameter, leaf area index, root number, and starch initiation.

Genetic analysis of variance of raw data for all traits in each environment was computed with trait values averaged over replications per genotype, considering all effects as random. Broad sense heritability estimates were calculated from variance components. Correlation coefficients across environments for traits were also calculated. For early bulking studies, correlation and multiple regression of yield with other growth related parameters were also done to identify the most paramount characters influencing early bulking. To detect association between markers and QTLs, adjusted trait means, from the ANOVA were used in a simple linear regression of phenotypic data on marker genotype classes, as independent variable, using the computer package Q gene (Nelson, 1997). A region of the genome was considered to be associated with a QTL for any trait if $P < 0.005$.

Broad sense heritability estimates (H^2) were high for plant height, first branching height, and branching levels, 78, 91, and 68% respectively, and low for length of stem with

attached leaves (48%). Broad sense heritabilty estimates for early bulking, measured as dry matter yield at 7MAP, were 0.6 in Palmira and 0.64 in Quilichao respectively. Three regions of linkage group D were found to bear the most important QTLs for dry matter yield at 7MAP and 11 MAP, dry matter percentage, starch content, branching levels and cooking quality. Together they explain 49 and 37% of dry matter at 7 and 11 MAP respectively in Quilichao. The low level of recombination on linkage group D, relative to the rest of the genome, and the large number of markers on this linkage group suggest it might be an introgressed segment from *M. glaziovii*. A QTL controlling cooking quality was found in the same region as the AGPase small sub-unit gene, suggesting a role for genes involved in starch biosynthesis in cooking quality. Three QTLs linked to RFLP markers GY 120, CDY 131 and GY 138 on linkage groups L, X and U respectively accounted for 8-12% of phenotypic variance of post harvest physiological deterioration. Single point marker analysis revealed a single region of the genome with strong association (P <0.0001) between phenotypic variance for leaf morphology in the F_1 cross and RFLP marker rGY99 on linkage group H of the male-derived linkage map (Figure 1). The association accounted for 79% of the phenotypic variance. Qualitative scoring revealed that linear:elliptic leaf shape segregated in the F_1 population in the ratio of 1:1. Result suggests that a single major gene control leaf shape in cassava, linear shape being dominant to the elliptic shape as in earlier findings (Hershey & Cesar, 1989).

Table 7.1. Markers on the genetic map of cassava showing significant linkage to QTL effects for foliage, harvest index and bulking rate from early bulking studies at Palmira, 1999.

Trait	Marker	Chrom	N	F	RSq	P	AA	SE	Aa	SE
Foliage	rCDY74	NgJ	34	18.43	0.3655	0.0002	197.8	25.26	376.74	32.89
	CDY76	NgJ	40	18.42	0.3265	0.0001	210.2	23.49	373.11	30.30
	CDY131	NgX	36	11.54	0.2534	0.0018	371.69	35.35	224.93	23.90
HI	GY153	NgS	77	18.18	0.1951	0.0001	0.13	0.01	0.20	0.01
	GY34	NgJ	64	11.76	0.1594	0.0011	0.40	0.02	0.50	0.02
	rBEST-2	CmA	41	18.14	0.3175	0.0001	0.35	0.02	0.23	0.02
Bul. rate	GY202	NgL	78	12.19	0.1382	0.0008	23.41	1.66	16.71	1.0
	rP3	NgQ	39	10.95	0.2284	0.0021	24.47	2.04	15.44	1.82

In early bulking studies, for each harvest, correlation coefficient of storage root weight was highly significant (P < 0.002) with root diameter, foliage, dry weight, plant height, harvest index, vigor, starch initiation and root number. Multiple regression of dry root yield with these related traits, showed that, only the regression coefficients for foliage and HI index were significant, indicating these two traits as the most highly associated traits influencing bulking, and are therefore of great primary importance for early bulking improvement in cassava. Single regression analysis identified three markers associated with foliage, rCDY74, CDY76 and CDY 131 on the female derived map explaining 25-

36% of phenotypic variance (Table 7.1). For harvest index, markers GY 153 and GY 34 and rBEST-2 (accounts for 15-31% of phenotypic variance) were found associated with this trait (Table 7.1). Markers GY 34 and CDY 131 were also associated with dry matter yield at 7 MAP in Palmira in 1998. The significant relationship of foliage with yield, underscores the importance of top growth, and thus photosynthetic activity, in assimilate production, while harvest index is a direct measure of assimilate partitioning between the top (source) and the sink strength (bulking). Markers GY 202 and rP3 from the female map were significantly linked to QTLs controlling bulking rate (Table 7.1).

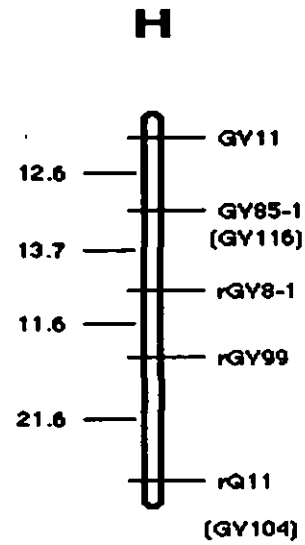


Figure 7.1. Linkage group H showing RFLP marker rGY99 linked to leaf morphology

Achievements:
- QTLs identified for root quality traits in Quilichao.
- Components of early bulking elucidated, and QTLs for these traits identified.
- Position for the gene controlling leaf morphology identified in cassava.

Activity 7.3 Simple Sequence Repeat (SSR) Analysis of Genetic Diversity and their Use in Predicting Heterosis in Cassava.

Specific Objectives:

a) to determine genetic structure of cassava gene pools from Latin America and Africa.

b) to define heterotic pools based on clustreing of gene pools based on SSR markers.

c) to detect linkage equilibrium between useful traits, and SSR markers in farmers' cassava collections selected for different traits and agro-ecologies.

Summary: Crop genetic diversity is the results of thousands of years of natural and farmer selection and it is the basis of crop improvement. The right germplasm in the hand of talented breeders produced spectacular increases in grain yields in Latin America and Asia in the 1950s, 60s, wiping away food deficits and heralding the 'Green Revolution" (Witt 1985). Much before that corn breeders at the turn of the century discovered the phenomenon of hybrid vigor or heterosis and produced unbelievable bumper harvests from the same ancient fields. Discovering useful genetic variation and its exploitation is still the wheel on which agricultural productivity turns.

Basic information on the genetic structure of cassava gene pools is needed before efficient population improvement can be done. Additional information to be gained from a study to characterize genetic diversity of cassava include: an understanding of how total genetic variation is partitioned within the collections, which may provide insight into which accessions should be given priority for use, and conservation given limited resources. Others are linkage disequilibra between markers and agronomic traits, levels of heterozygosity and inbreeding, and how they relate to hybrid vigor, and rate of extinction of rare alleles.

Powerful new marker tools that have recently become available at CIAT, makes the systematic genetic characterization of cassava germplasm, for realizing the crop's full potential, a feasible goal in terms of time and resources. The markers, simple sequence repeat markers are PCR-based, thus easily assayed, highly polymorphic, and lend themselves to automation.

More than 70% of cassava genotypes grown in a major cassava production region of Southern Tanzania, a total of 175 genotypes was sampled as a pilot study . This sampling was done in the Mtwara and Lindi regions of Tanzania in June. The most frequently used parents at CIAT in cassava breeding since the 1970s were included for the molecular analysis. A group of 50 genotypes, representative of the CIAT core collection, studied in an earlier SSR analysis of the core collection.

DNA extraction was by a miniprep modification of the Dellaporta et. al (1983) method (Fregene 1998, unpublished result. Between 0.2 and 0.5g of freeze dried, or oven dried (48°C for 2 days) of plant tissue was used and extraction was in a 1.5 microfuge tube. This method drastically slashes the cost of DNA isolation.

186 SSR markers were recently developed for cassava at CIAT. A survey of the parents of the cassava mapping population reveal that 113 SSR markers were polymorphic in the parents and could be placed on the existing map of cassava. More than 80 SSR markers have been mapped, and genetic mapping of the rest is ongoing. We wish to determine a robust minimum sub-set of SSR markers, with a broad coverage of the cassava genome, for all future genetic diversity study. To do this a total of 82 SSR markers, from all over the genome were chosen for PCR multiplexing. The primers were be assayed on all the approximately 300 genotypes using 12 PCR reactions, each with 3 or 4 flourescently-labelled primer pairs. PCR product was be denatured and electrophoresed on 4% polyacrylamide gels on an automated DNA

sequencer (ABI model 377). Data was analyzed by ABI PRISM Gene Scan analysis software. This aspect was done in collaboration with Professor Steve Kresovich of the Institute for Genome Diversity, Cornell University, with funding from the International Program in the Physical Sciences, Uppsala University, Uppsala, Sweden, and will be concluded in October.

Genetic variation can be described as identities of alleles and their distribution within a defined set of individuals, and can be measured in many ways. The experiment is designed around the following specific null hypothesis:

Ho: Genetically diverse clusters of cassavas exist in African, and Latin American germplasm collections.

Ho: Materials collected in specific regions of Africa are genetically similar to each other and differentiated from others grown in other parts of Africa, and Latin America.

Ho: Cultivars showing strong hybrid vigor are heterozygous at the majority of loci, consisting of approximately 50% heterotic group 1 alleles, and heterotic group 2 alleles.

Ho: Clusters from Africa and Latin America contain approximately equal amounts of genetic variation.

Rejection or acceptance of the null hypothesis will be accomplished through the interpretation of several lines of statistical evidence using various methods to quantify genetic variation.

Sources of error contributing to genetic variation include experimental sampling and genetic drift. Genetic drift will be measured within populations (assuming SSR are selectively neutral), and statistical approaches that take allelic frequency variation due to finite sample sizes into account will be used.

Principal Component Analysis (PCA) (Sneath and Sokal 1973) will be applied (using JMP, SAS Institute 1995) to deduce multivariate relationships amongst genotypes using a variance-covariance matrix of allele frequencies. The stepwise mutation model (SMM) (Valdes, Slatkin, and Freimmer 1993) is preferred for calculating genetic diversity and distance measures using SSR frequency. SMM assumes that an allele mutate in a predictable way and are less biased than other methods. The computer based program MICROSAT (Minch et. al. 1996) allow the assumption of SMM for population genetic analysis of SSR data. The neighbor joining method (Saitou and Nei 1987), available in the software MEGA will be used to draw dendograms of genetic similarity.

The major question to be answered by this research is if SSR markers can be used to identify heterotic groups, and other genetic parameters required for population improvement. SSR markers will be used to search for highly genetically differentiated, clusters, which may have either been isolated reproductively for a long time, or arose from different domestication events. Groupings based on molecular markers will be

tested by diallel crosses and an evaluation for hybrid performance of the progeny to confirm the heterotic pools. These are important issues in a crop such as cassava in which little is known about the genetic basis of successful combining ability. Selecting genetically superior material based on phenotype alone maybe unwise and unnecessarily limiting. It has been shown that alleles from a phenotypically inferior appearing wild relative could be recovered during crosses to enhance agronomic traits in tomato.

Two to four genotypes, representative of the groupings obtained by SSR markers will be chosen to produce diallel crosses. The diallels will be made such that at least half or more of the parent are adapted to one of the five ecological zones in which CIAT conducts its breeding activities. The crosses, and their parents, will be established at CIAT head quarters, Palmira, Colombia (Palmira – highland tropics). Woody cuttings will be obtained from the progenies at 8 months for a replicated trial, on family basis, with the parents, in the appropriate ecological zone. The following traits will be measured on all plants at 10 months after planting (MAP)

a) Dry matter yield (root yield)
b) Dry foilage yield (upper plant canopy)
c) Harvest index: ratio of dry matter yield to total biomass (dry matter + dry foliage yield)
d) Dry matter percentage
e) Amylose/Amylopectin ratio
f) Post-harvest deterioration.

Hybrid performance will be estimated from the performance of the families, based on the above criteria, compared to the performance of the better parent.

Achievements:
- Collection of cassava land races in Tanzania, and isolation of DNA from them, and 100 CIAT cassava genotypes.
- Successful analysis of 300 land races, and improved varieties of cassava with 82 SSR markers in multiplexed.

Activity 7.4 Development of Simple Sequence Repeat (SSR) Markers for the Saturation of the Molecular Genetic Map of Cassava

Specific Objectives:
a) to develop a few hundred SSR markers for the cassava map.
b) to transfer SSR marker technology, as applied to cassava improvement, and genetic diversity evaluation, to collaborators.

Summary: In an attempt to make marker technology widely available in cassava, an effort was embarked upon to place on the cassava map simple sequence repeat (SSR) markers, markers that are PCR-based and highly polymorphic, and best meet the criteria required for marker technology transfer to developing country research systems. SSR markers are abundant and interspersed in eukaryotic genomes

Two enrichment experiments, "Enrichment A" and "Enrichment B", differing essentially in the oligonucleotides used for enrichment and the cloning vectors, were conducted with two cassava elite clones. Total genomic DNA used in "Enrichment A" was from TMS 30572, an improved cassava variety developed at the International Institute of Tropical Agriculture, Ibadan, Nigeria. Enriched libraries were constructed for di-, tri-, and tetra nucleotide repeats. The DNA for "Enrichment B" was from CMC 40, a cassava accession from CIAT's core collection originally collected from Brazil, and enriched libraries were constructed for only di-nucleotide repeats, $(GA)_{15}$,/$(CA)_{15}$.

Two microlitres from the all enriched libraries were transformed into E. *coli*, DH10 (GIBCO BRL Inc), cells by electroporation, according to the Manufacturer's protocol. Electroporated cells were plated out on 100µg/ml ampicillin LB-agar plates and incubated overnight at 37°C. Approximately 6,000 clones from each of the di-, tri-, and tetra- enriched libraries of "Enrichment A" were picked and spotted onto one single 48cm X48 cm high density filters using the QBOT robot (Genetix PLC, UK) of the Clemson University Genome Institute (CUGI). Two thousand, and three hundred clones were handpicked from the $(GA)_{15}$/ $(CA)_{15}$ enriched library of "Enrichment B" and organized manually onto 12 18cm X10 cm filters.

The filters, from both enrichments were screened with the appropriate di, tri, or tetra nucleotide, and end labeled with $\alpha[^{32}P]$dATP. Hybridizations were at 65°C or 45°C for 14- 16 hr. Post-hybridization washes were 6XSSC at 65°C or 45°C for 5 min, twice. Autoradiography was from 2 – 24 hr. Plasmids miniprep of overnight 2ml LB + 100ug/ml ampicillin culture of positive clones was carried out using the QIAGEN (Gmbh) plasmid miniprep kit or the Promega (Inc.) Wizard prep kit. A PCR pre-screen, to determine the position of the SSR repeat in the clone sequence, was carried out using M13 reverse or universal primer, and the corresponding primer made up of the sequence, or complementary, of the appropriate oligo and an anchor. Forward and reverse strands of all positive clones were sequenced using the M13 universal and reverse primers (New England Biolabs, USA and Microsynth, Switzerland) on an automated sequencer (Perkin Elmer/ Applied Biosystems models ABI 373 and 377).

Vector and adaptor sequences were cleaned out of the raw DNA sequence using the GCG (University of Wisconsin) or the Sequencer 3.0 (Gene Codes Corp) software. The software packages were also used align the forward and reverse strands. Duplicate sequences were identified using Local BLAST from http://www.ncbi.nlm.nih.gov. Primers were designed for all unique SSR containing sequences with at least 10 repeats for di-nucleotide repeats, and >6 for tri- and tetra- nucleotides. Primer design was with "Primer3" picking software found at http://waldo.wi.mit.edu/cgi-bin/primer/primer3 (Whitehead Institute for Biomedical Research). Oligonucleotide

primers were synthesized by Research Genetics, USA and designated "Cassava MapPairs".

The female parent of the F_1 cassava mapping population, TMS 30572, one of the accessions employed in the "Enrichment A, and the male parent, CM2177-2, an improved clone from Colombia, were evaluated with all the 172 SSR markers identified, using non radioactive PCR amplifications, and silver stained (Promega Inc.) 6% polyacrylamide sequencing gels. PCR reactions were 50 µl volume containing 50-100ng of genomic DNA, 0.2 µM of each forward and reverse primers, 10mM Tris-HCL (pH 7.2), 50mM KCL, 1.5 or 1mM $MgCl_2$, 200mM od each dNTP, and about 1U of Taq DNA polymerase. Temperature cycling profile was: an initial denaturation step for 5min at 94°C, followed by 30 cycles of denaturation at 94°C for 1min, annealing at 55°C or 45°C for 2 min and primer extension at 72°C for 2 min. A final extension cycle of 5 min at 72°C was added. Between 2 and 3 µl of the PCR reaction was electrophoresed on 5% ethidium bromide stained Metaphor agarose gels or on 6% polyacrylamide sequencing gels for 2 h at 100W, and DNA visualized by silver staining according to the Manufacturer's guide (Promega Inc., USA).

SSR markers having a unique allele in either or both parents was analyzed in the entire F_1 progeny of 150 individuals. SSR markers that segregated in the expected ratio of 1:1, presence: absence of the unique parental allele in the F_1 progeny were placed onto the existing map of cassava using the linkage analysis computer package MAPMAKER 2.0 (Lander et. al.1987), as described earlier (Fregene et. al. 1997). The "group" command, with a LOD threshold of 4.0, and a recombination fraction of 0.3, was used to assign SSR markers to existing linkage groups, and the "try" command was used to find the most likely interval to place the new marker on the linkage groups. In a few cases the SSR led to a new linkage group being formed, or two smaller groups joining together, the order was ascertained using the "compare" function. Maximum likelihood orders of linkage groups with newly added markers was verified by the ripple function, and only orders ≥ LOD2.0 were accepted for the new frame work map of cassava. MAPMAKER analysis was done on a Macintosh G3 computer.

From "Enrichment A", a total of 148 positive clones, or <1%, was obtained from the SSR oligo screen of the libraries. Plasmid DNA was prepared from the 148 putative clones and a PCR pre-screen, to determine the proximity of the SSR repeat to the end of the clone sequence, was done before the clones were sequenced. The average size of the clones was 200bp. Of these, 66 clones, or 45%, contained the relevant SSR repeat sequences. Primers were designed for 35 unique clones; 4 were duplicates, while the other 17 were clones with the SSR too close to the end of the DNA clone to permit primer design. Enrichment B had 1400 positive clones, or >60% enrichment. Plasmid miniprep and DNA sequencing was performed for 544 clones, from which 479 clones had SSR repeat sequences, 30 clones had no SSR loci while 35 had sequences that needed repeating. No PCR pre-screen f the clones was performed. Of these 479 positive clones, 229 clones had the SSR repeat too close to the end of the sequence while 113 clones were duplicates. Primers could be designed for 137 clones.

One hundred and sixty four, or 95%, of the 172 SSR containing clones for which primers were designed, were dinucleotide repeats while the balance were trinucleotide repeats save for one tetranucleotide repeat. Table 7.2 shows the breakdown of the clones into nucleotide repeat classes. Thirty-seven, or 21%, of the loci were found to contain more than one kind of repeat -compound repeats. Approximately 20% of the SSR clones from both enrichment were duplicated sequences, while 45% had the SSR loci too close to the cloning site to permit primer design from the flanking regions. On a whole, 35% of sequenced positive clones were unique sequences with SSR loci well situated for primer design.

Table 7.2. Number, percentage and kind of SSR repeat sequences for which primers were designed.

Type of SSR	Number	Percentage (%)	Type of SSR	Number	Percentage (%)
	Enrichment A			**Enrichment B**	
GA/CT	12	34	GA/CT	80	58
CA/GT	5	14	CA/GT	30	22
(CA)(GA)	2	6	(CT)(CA)	15	11
ATT/TAA	5	14	(CA)(GA)	6	4
Others	11	31	Others	6	4
Total	35		**Total**	137	

A total of 6000 cassava insert, of average size 200bp, "was screened for Enrichment A" and 36 GA containing clones, or an average of approximately 1 GA marker every 34kb, was found. Assuming an average of one GA repeat every 225kb found for higher plants (Maroof et. al. 1994), this is a 6 fold enrichment. The Enrichment B, on the other hand obtained 875 GA clones from 2,300 clones of average size 250bp, or 1 GA marker every 700bp, or >300 fold enrichment.

SSR Parental Survey:

All 172 primer pairs successfully amplified the corresponding SSR loci in the parents of the cassava mapping progeny; but with different $MgCl_2$ concentrations, and 2 annealing temperatures, 55°C and 45°C respectively. The primer pair sequences, annealing temperatures, product sizes, and $MgCl_2$ concentrations are presented as an appendix at the end of this paper. One hundred and thirteen SSR loci, or 66 % of all SSR markers tested in the parents, revealed a unique allele in at least one of the parents; 45 SSR markers, or 26% showed a unique allele for both parents.

Genome location of SSR markers:

Twenty two SSR markers that were polymorphic in the parent on ethidium bromide stained 5% Metaphor agarose gels were scored in the 150 F_1 mapping progeny, along with a group of 14 SSR markers polymorphic in the parents only on PAGE gels. A total of 81 SSR loci from the 172 SSR markers analyzed to date have been mapped on the male- and female-derived molecular genetic map. The 81 SSR markers reveal a fairly

even spread over the cassava genome – sixteen of the eighteen linkage groups have at least 1 SSR marker, with an exception of 9 SSR markers clustered on linkage groups C, D, and J. An unusual observation is the complete lack of duplication of the SSR markers mapped so far. It is not clear at this stage why the SSR enrichments yielded no clones with duplicated loci.

Current efforts are geared to mapping the remaining SSR markers, and continued sequencing of the more clones to reach the objective of a few hundred mapped SSR markers. Further development of SSR markers will be in to search the 3' and 5" untranslated regions of expressed sequence tags (ESTs), expected from ongoing cassava EST projects, for SSR repeats.

Achievements:

- 172 new SSR markers developed for the molecular genetic map of cassava.
- 82 SSR markers placed on the molecular genetic map of cassava
- Training in SSR marker technology for visiting post-doc scientist from IITA.

OUTPUT 8: Maintenance of the germplasm bank collection.

The overall objective of this project is to maintain, study and whenever possible, to expand the germplasm bank collection including *Manihot esculenta* and wild *Manihot* relatives.

Activity 8.1. ***To carry out field operations, in coordination with other projects working on cassava,in such a way as to guarantee the rapid cleaning of our germplasm from the* 'frog skin disease'.**

Summary: Because of changes in CIAT's experimental station management, resulting in a concentration of plantings, and the recent increase in whiteflies populations, there has been a steady increase in the incidence of frog skin disease (viral origin and transmitted by whiteflies). The frequency of contaminated stakes has reached unacceptable levels. Therefore, a plan was devised for a drastic reduction of the occurrence of frog skin disease to manageable frequencies. It is accepted that it will be very difficult or impossible to completely erradicate the disease because it is endemic in Colombia.

The strategy consists in:

a) to plant all sexual seeds (seeds are disease free) in isolated environments outside the experimental station. The location for this planting must be in areas were there is no cassava grown nearby. Indexed vegetative stocks that are proven to be disease free are also planted in this "clean locations". A plot kindly offered by CENICAÑA, has been utilized for this purpose.

b) Regenerate from the in vitro collection, materials that have already been indexed and certified to be disease free. Results from these regenerations will also go to the "clean location". There are about 1300 accessions in this situation and they are currently being regenerated.

c) Maintain the current "contaminated" collection in the field until June, 2000. In the meantime there will be an accelerated process for indexing plants from the in vitro collection. Hopefully about half of the collection will be planted through the middle of next year.

d) By the second semester of 2000, all material certified to be disease free will be planted in plots located in the western extreme of the station. All new cassava plantings during next year, if not certified to be disease free, will be planted in the eastern side of the station. As we proceed this way gradually there will be less cassava planted on the east and more clean materials on the west. It is expected that in a matter of a year incidence of frog skin will be drastically reduced.

Collaborating institutions:

Agropecuaria Mandioca. Venezuela.
Agroselect Snacks, S.A.Venezuela.
Agrovelez. Colombia.
Almidones Nacionales S.A. Colombia.
American Soybean Association. Venezuela.
Asociación Colombiana de Porcicultores (ACOPOR). Colombia.
Brawijaya University. Indonesia.
CDA, Vaupés (Ms R. Peña). Colombia.
Centro para la Investigación en Sistemas Sostenibles de Producción Agropecuaria (CIPAV). Colombia.
Chinese Academy of Tropical Agriculture Science. CATAS, Hainan, China.
Comercial "Manihot". Venezuela.
Concentrados del Norte S.A.. Colombia.
Congelados Agrícolas S.A. (CONGELAGRO). Colombia.
Cooperativa Agroindustrial del Tolima (COOPALTOL). Colombia.
Corporación Colombiana de Investigación Agropecuaria (CORPOICA). Colombia.
Derivados de la Yuca y el Maíz. Colombia.
Empresa Brasileira de Pesquisa Agropecuaria (EMBRAPA). Brasil.
Empresa Pernambucana de Pesquiza Agropecuaria. Brazil
Federación Nacional de Avicultores. Colombia.
Federación Nacional de Avicultores .Venezuela.
Federación Venezolana de Porcicultura. Venezuela.
Fondo Nacional de Investigaciones Agropecuarias FONAIAP. Venezuela.
Fundación para la Investigación y el Desarrollo Agrícola (FIDAR). Colombia
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International Food Policy Research Institute (IFPRI). USA.
International Institute of Tropical Agriculture (IITA). Nigeria.
Institute of Agricultural Sciences (IAS), Ho Chi Minh City. Vietnam.
Instituto Agronómico Campinas (IAC). Brazil.
Instituto de Estudios Avanzados (IDEA). Venezuela.
Instituto Interamericano de Cooperación Agrícola (IICA). Colombia.
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Ministerio de Agricultura. Paraguay.
National Starch and Chemical Company. USA.
Rayong Field Crops Research Center. Thailand.
Red de Solidaridad (Presidencia de la República), Mitú (Mr. J. M. Girón). Colombia.
Secretaría de Agricultura, Mitú (Dr. Leticia Guerrero). Colombia.
Thay Nguyen University, Vietnam.
Universidad Central de Venezuela – Maracay. Venezuela.
Universidad Nacional de Colombia – Palmira Campus. Colombia.
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Collaborating institutions *(cont.):*

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UMATAs (O. Holguin, L.M. Giraldo, W. Ospinal Muñoz, M. T. Aristizábal). Quindío, Colombia.

Vietnam Agric. Sciences Institute (VASI), Hanoi, Vietnam.

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***Meetings, workshops and Field Days*:**

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- Alvarez, E., G. Llano, J.A. Restrepo, O.J. Ríos, R, Madriñán. Four meetings held with the Nodo, Vaupés Departament, in Mitú to report advances in the project *Desarrollo agrícola de la población indígena de la zona de influencia de Mitú-Monfort (Vaupés, Colombia): Control de pudriciones en yuca, mediante investigación participativa.*

- Alvarez, E., G. Llano, J.A. Restrepo, L.M. Panche. A field day in the Cucura Indian settlement, with the participation of 85 natives from eight communities close to Mitú.

- Alvarez, E., G. Llano, J.A. Restrepo, O.J. Ríos, L.M. Panche Five meetings organized in different communities to encourage women indigenous to disseminate the results of their trials and to highlight the importance of applying the participatory research method.

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***Theses in Progress*:**

Bedoya, J.M. Determinación del potencial genético con respecto al contenido de provitamina A y vitamina C en la colección núcleo de yuca (*M. esculenta* Crantz) del CIAT. A B.S. thesis at the Universidad Nacional de Colombia.

Dorn, Brigitte. Ecological cultivationof cassava to render natural enemies of sucking pests more effective and reliable. A Ph.D. thesis at ETH Institute, Switzerland.

Echeverri, J. A. Evaluación de 500 genotipos en cuanto al contenido de vitaminas A y C de la coleeción central de yuca del CIAT. A B.S. thesis at the Universidad Nacional de Colombia.

Gonzalez, J.E. Indirect measurement of cassava total mass root development by a capacitance meter. A B.S. thesis at the Universidad Nacional de Colombia.

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Llano G.A. Identificación de genes de resistencia a *Phytophthora* spp. en yuca, mediante el uso de sondas heterólogas. A M.Sc. thesis at Universidad Nacional de Colombia, Palmira.

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Perez Porras, G. A simulation of the environmental impact of a herbicide-resistant cassava variety. A B.S. thesis at the Universidad Nacional de Colombia.

Donors:

American Soybean Association. Venezuela.
Asociación Colombiana de Porciculturoes (ACOPOR), Colombia.
COLCIENCIAS, Colombia.
CORPOICA – Nataima. Colombia.
Department for International Development (DfID). England.
Federación Nacional de Avicultores (FENAVI), Colombia.
International Fund for Agricultural Development. Italy.
International Food Policy Research Institute. USA.
Ministerio de Agricultura y Desarrollo Rural, Colombia.
New Zealand Ministry of Foreign Affairs and Trade. New Zealand.
Nippon Foundation, Japan
PRONATTA (Programa Nacional de Transferencia de Tecnología), Colombia.
Rockefeller Foundation. USA.
USDA (United States Department of Agriculture). USA.

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