CBN-VI

CIAT

Embrapa

Sixth International Scientific Meeting of the Cassava Biotechnology Network

8-14 March 2004 CIAT, Cali, Colombia

Adding Value to a Small-Farmer Crop

Abstracts

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International Scientific Meeting of the Cassava Biotechnology Network (6, 2004, Cali, Colombia). Abstacts. Cali, CO: Centro Internacional de Agricultura Tropical (CIAT). 2004. p. 208

Descriptores AGROVOC:

1. Biotecnología vegetal. 2. Redes de investigación. 3. Subproductos. 4. Mercadeo. 5. Análisis de costos y beneficios. 6. Recursos genéticos vegetales. 7. Fitomejoramiento. 8. Variación genética. 9. Resistencia genética. 10. Conservación del germoplasma. 11. Cultivos alimenticios. 12. Estrés. 13. Tecnología postcosecha. 14. Tapioca. 15. Cultivo de tejidos. 16. Transformación genética. 17. Marcadores genéticos. 18. Transferencia de tecnología. 19. Países en desarrollo. 20. Africa. 21. Asia. 22. Colombia. 23. América Latina.

Descriptores Locales
1. Agrobiodiversidad. 2. Investigación participativa. 3. Técnicas moleculares.

Categoría de Materia AGRIS:F30 Genética vegetal y Fitomejoramiento.
A50 Investigación Agrícola

AGROVOC Descriptors:
1. Plant biotechnology. 2. Research networks. 3. Byproducts. 4. Marketing. 5. Cost benefit analysis. 6. Plant genetic resources. 7. Plant breeding. 8. Genetic variation. 9. Genetic resistance. 10. Germplasm conservation. 11. Food crops. 12. Stress. 13. Postharvest technology. 14. Tapioca. 15. Tissue culture. 16. Genetic transformation. 17. Genetic markers. 18. Technology transfer. 19. Developing countries. 20. Africa. 21. Asia. 22. Colombia. 23. Latin America.

Local Descriptors
1. Agrobiodiversity. 2. Participatory research. 3. Molecular techniques.

AGRIS Subject Categories: F30 Plant genetics and Breeding.
A50 Agricultural research.

Clasificación LC.: SB 211 .C3 I58

Compiled by: Claudia Stella Zuñiga
Agrobiodiversity and Biotechnology Project

Dear CBN-VI participant,

At the end of our CBN-V, in November 2001 in St. Louis (USA), the CIAT's group proposed that the next CBN meeting would return to Colombia. The suggestion was accepted since 10 years had passed from the first meeting in Cartagena de Indias, Colombia (1992) and to reinforce the regional framework of the CBN for Latin America and the Caribbean. Two months later, Chusa Ginés and Veronica Mera, the Coordinator and Social Scientist, respectively of CBN-LAC, lost their lives in a tragic airplane accident. After this tragedy staff at CIAT joined forces to endure this situation and develop initiatives to maintain the CBN-VI commitment and to honour the memory of Chusa and Veronica.

Considering their interest to the sustainable use of agricultural biodiversity, the idea of a biodiversity scholarship fund was supported by IDRC and the "Ginés-Mera Memorial Fellowship Fund for Postgraduate Studies" was launched in early 2003 to aid young students from Developing Countries to complete masters-level and higher theses in molecular biology, social and rights-related aspects of agrobiodiversity and its conservation. The theme of this meeting "**Adding Value to a Small-Farmer Crop**" highlights the main concern related to the sustainable use of the cassava crop and the principal CBN-LAC's objective: integrate the needs of small-scale cassava farmers, processors and consumers into biotechnology research planning.

This year, the meeting program, besides the special session for celebrating Ginés and Mera, will give emphasis to Plenary Sessions to discuss relevant themes and Posters Sessions with more time to discuss the papers. We have received around 160 papers from 30 countries in all three continents.

I would like to take this opportunity to thank Dr. Joachim Voss, Director General of CIAT and Dr. Joe Tohme, Manager of the Agrobiodiversity and Biotechnology Project at CIAT, who always maintained his support and encouragement that CBN-VI should be at CIAT. Likewise, the members of the Scientific and Local Committees and other CIAT's staff, who strongly supported the organization of CBN-VI. My thanks also go to the representatives of the Institutions that sponsored this event: Rockfeller Foundation, USAID, Syngenta Foundation, Embrapa, FAO, and AVEBE.

Welcome to Cali and enjoy CBN-VI at CIAT.

Dr. Alfredo A. C. Alves
CIAT, CBN Coordinator

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General Information

Sixth International Scientific Meeting of the Cassava Biotechnology Network (CBN-VI)

8-14 March 2004
CIAT, Cali, Colombia

Theme: Adding Value to a Small-Farmer Crop

Summary

CIAT will host the CBN's Sixth International Meeting on the use of biotechnology tools to add value to cassava, a small-farmer crop of central importance to food security in the tropics. Scientists from advanced labs and NARS from Africa, Asia, South America and Central America will attend the meeting to discuss how biotechnology can assist cassava farmers by developing, for example, more suitable varieties, disease-free planting materials, and better ways to conserve and process cassava after harvesting. This conference will help determine the future of cassava research and will permit cassava scientists from all three continents to share ideas and discuss the implementation of capacity building in developing countries for a range of technologies to be used with cassava.

Goal

To strengthen and speed efforts to maximize the contribution of modern biotechnology tools to the agronomic improvement of cassava and thereby contribute to improved food security in the tropics.

Objectives

- To facilitate dissemination of current information concerning cassava biotechnology, most importantly recent scientific and technical advances in this area.
- To promote the transfer of scientific information to cassava researchers through state of the art presentations and exchange sessions.
- To inform and educate biotech company representatives as to the importance of the crop in human development and the role of modern biotechnologies in cassava improvement.
- To publicize cassava and raise its profile in the world as a primary food crop for the 21st century.

Background on CBN

The Cassava Biotechnology Network (CBN) was founded in 1988 through the initiative of several individual scientists and the CGIAR-sponsored center CIAT (Centro Internacional de Agricultura Tropical, Colombia). CBN's mission is to maximise the contribution of modern biology to the agronomic improvement of cassava, a crop of central importance to food security in the tropics. Initial funding was supplied by CIAT, after which a 5 year project to support the CBN was secured from the Dutch government development agency (DGIS). An extension of this funding allowed the CBN to operate fully until the end of 1998. CBN was revived in 2001 under joint sponsorship of the Canadian International Development Research Center (IDRC) and DGIS. Although operating on a smaller scale than in the 1990s, CBN continues to sponsor both worldwide and regional activities dedicated to cassava improvement.

The CBN aims to provide a network for enhanced communication between scientists through the organization of meetings, newsletters, reference databases, a small grants program for scientists from developing countries and as an advisor for the preparation of cassava research and training proposals to a variety of agencies. CBN has been internationally acclaimed as one of the most successful organizations of its kind.

One of the most important activities of the CBN is the organization of scientific meetings to exchange state-of-the-art knowledge. These CBN International Scientific Meetings are held every other year in a different cassava producing country, and include field trips to cassava fields, processing facilities, factories and farmer cooperatives. Past scientific meetings included:

- CBN-I 1992: Cartagena, Colombia
- CBN-II 1994: Bogor, Indonesia
- CBN-III 1996: Kampala, Uganda
- CBN-IV 1998: Salvador, Brazil
- CBN-V 2001: St. Louis, USA

Participants

CBN-VI is expected to bring together 120 scientists from around the world, including 45 Latin Americans, 30 Africans, 15 Asians, 15 Europeans, and 15 North Americans

Subjects to be explored

CBN-VI's program will consist of keynote addresses, plenary sessions, panel and poster sessions covering the following subjects:

- The future of biotechnology in developing countries
- How to add value to cassava
- Innovative approaches in R&D for cassava
- Biodiversity and IPR/Biosafety
- Cassava nutritional value
- Abiotic and biotic stresses
- Transformation and transgenics products
- Small farmers and research planning
- Tools for cassava breeding
- Toward cassava biotechnology's next phase

In addition, there will be visits to cassava fields and CIAT's laboratory and processing facilities.

Project beneficiaries

- Output from this meeting will benefit plant biotechnologists and cassva breeders of NARS from Latin American, African and Asian countries where cassava is a staple food. The scientists will, in turn, transfer the information and technologies to cassava farmers, processors and consumers.
- NARS scientists will establish stronger ties to the network, in which they can benefit from the cassava genotypes, available for sharing and adapted to different environmental constraints that limit cassava production in many producing countries.
- The ultimate beneficiaries will be the small-scale farmers who can improve their income, and thus their standard of living, adding value to cassava through biotech tools explored in this meeting.

Continuing network activities

Planning for the future of cassava research is an important focus of this meeting. CBN will be able to outline its next phase toward the invigoration of its activities in both regional and global frameworks, articulating a common vision to move the research forward. All the outputs from this meeting will help the network to progress, for the next 2 years, in its three main complementary thrusts:

1. Priority setting and evaluation through the strategic use of social science to ensure that the end-users have a real voice in decision-making in the development and implementation of biotechnologies;
2. Technology diffusion by further adapting key biotechnologies together with small farmers by public sector research;
3. Information to promote awareness building/dialogue among scientists and end-users of the opportunities and constraints inherent to biotechnology.

Organizers

Centro Internacional de Agricultura Tropical (CIAT) / Cassava Biotechnology Network (CBN)
Alfredo A. C. Alves, *CBN Coordinator*
Joe Tohme, *Manager of Agrobiodiversity and Biotech Project, CIAT*

General Secretariat
Claudia Zuñiga, CIAT/Biotechnology Research Unit
Address: Km 17, Recta Cali-Palmira, AA 6713, Cali, Colombia
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Website: http://www.ciat.cgiar.org/biotechnology/cbn/index.htm

Donald Danforth Plant Science Center/ILTAB

Brazilian Agricultural Research Corporation (Embrapa)

International Institute for Tropical Agriculture (IITA)

Donors

- The Rockefeller Foundation
- USAID
- Syngenta Foundation
- FAO
- Avebe

Committees

International Committee

Role:

- Ensure the highest visibility possible for the crop itself and for the institutions participating, and to foster relations between private companies and cassava research.
- Raise sponsorship for the organization of the conference and invite high profile speakers and participants to the meeting.

Alfred Dixon, Coordinator of Global Cassava Development Strategy (GCDS), Nigeria

Alfredo Alves, Embrapa's Research Scientist and Coordinator of CBN, CIAT, Colombia

C. P. E. Omaliko, Director, National Biotechnology Development Agency, Nigeria

George Otim Nape, Deputy Director General, NARO, Uganda

Ivan Ingelbrecht, Head of Biotechnology Unit, IITA, Nigeria

Joachim Voss, Director General, CIAT, Colombia

Joe Tohme, Manager of Agrobiodiversity and Biotech Project, CIAT, Colombia

Juan Lucas Restrepo, Vice-Minister of Agriculture and Rural Development, Colombia

Klaas J. Tamminga, Research and Comunication Division of the DGIS, Netherlands

Klanarong Sriroth, Professor, Dept. of Biotechnology, Faculty of Agro-Industry, Kasetsart University, Thailand

Marc Van Montagu, Professor, Laboratory of Genetics, University of Ghent, Belgium

Marcio Porto, Deputy Regional Representative for Latin America and the Caribbean, FAO, Chile

Osei Safo-Kantaka, National Coordinator for Root and Tuber Programme, Kname Nkrumah University of Science and Technology, Ghana

Peter H. Raven, Director, Missouri Botanical Garden, USA

Roger Beachy, President of the Donald Danforth Plant Science Center, USA

S. Edison, Director, Central Tuber Crops Research Institute, India

Wardie Leppan, Team Leader, Sustainable Use of Biodiversity, IDRC, Canada

William Roca, Plant Cell Physiologist, Leader Project, CIP, Perú

Scientific Coordination Committee

Role: fund raising, scientific program, organize posters and plenary sessions, publications etc

Alfredo Alves, CIAT, Colombia
Ann Marie Thro, Consultant, USA
Anthony Bellotti, CIAT, Colombia
Chikelu Mba, IAEA, Austria
Claude Fauquet, Danforth Center, USA
Daniel Debouck, CIAT, USA
Eric Kueneman, FAO, Italy
Guy Henry, CIRAD, Argentina
Hernan Ceballos, CIAT, Colombia
Jan Salick, Missouri Botanical Garden, USA
Joe Tohme, CIAT, Colombia
John Beeching, University of Bath, UK
Larry Beach, USAID, USA
Martin Fregene, CIAT, Colombia
Nigel Taylor, Danforth Center, USA
Reinhardt Howeler, CIAT, Thailand
Richard Sayre, Ohio State University, USA
Richard Visser, Wageningen University
Valerie Verdier, Univ. de Perpinang, France
Wilhelm Gruissem, ETHZ, Switzerland

Local Organization Committee

Role: day to day organization of the meeting, travel, accomodation, proceedings, posters etc.

Alfredo Alves
Anna Knox
Anthony Bellotti
Bernardo Ospina
Claudia Stella Zúñiga
Elizabeth Alvarez
Elizabeth Caicedo
Hernan Ceballos
Jim McMillan
Joe Tohme
Martin Fregene
Nathan Russel
Patricia Fajardo
Paul Chavarriaga
Roosevelt Escobar
Sibel González

CBN-VI PROGRAM

Open Day: March 8, 2004 (Mon)

08:00-09:45: **Registration**

09:45-10:00: **Break**

10:00-12:00**: Open Ceremony**

- **Joachim Voss**, Director General, CIAT
- **Orlando Meneses**, Representative of the Ministry of Agriculture and Rural Development, Colombia

Keynote Speakers:

- **Deborah Delmer**, Rockefeller Foundation
- **Joe Tohme,** Agrobiodiversity and Biotechnology Project, CIAT

12:00-13:30: **Lunch**

13:30-15:30: **Celebrating Ginés-Mera**

Chair: **Ann Marie Thro**

Future of Biotech in Developing Countries

- **Jacqueline Ashby**
- **José Restrepo**

Deployment of transgenic cassava products

- **Marilia Nutti / Joe Tohme**
- **Lawrence Kent / Claude Fauquet**

15:30-16:00: **Break**

16:00-17:30: **Panel: How to Add Value to Cassava?**

Chair: **W. Gruissem** Reporter: **B. Ospina**

- **Guy Henry:** Products and Markets
- **Luiz Carvalho:** Biodiversity
- **Richard Sayre:** Transgenics

17:30-18:30: **Welcome Reception**

19:00: **Dinner**

Day Two: March 9, 2004 (Tue)

08:00-10:00: **Plenary Session 1**

Innovative approaches in R&D for cassava

Chair: **Reinhardt Howeler** Reporter: **Hernán Ceballos**

- **Bernardo Ospina:** Latin American and Caribbean consortium to support cassava R&D (Clayuca)
- **Andrew Westby:** Post harvest in Africa
- **Klanarong Sriroth:** New outlook for cassava in Thailand

10:00-10:30: **Break, Group Photograph**

10:30-12:30: **Plenary Session 2**

Towards Cassava Biotechnology Next Phase

Chair: **Joachim Voss** Reporter: **Chikelu Mba**

- **Alfredo Alves:** CBN Overview
- **Nebambi Lutaladio:** Global cassava development strategy
- **Claude Fauquet /Joe Tohme:** Global cassava partnership for genetic improvement

12:30-14:00: **Lunch**

14:00-16:00: **Plenary Session 3**

Cassava Nutrition Value

Chair: **Joe Tohme** Reporter: **Richard Sayre**

- **Delia Amaya:** Increasing micronutrient quality in cassava
- **Clara Brandão:** Cassava leaf flour
- **Larry Beach:** Perspectives for enhancing protein content in cassava

16:00-16:30: **Break**

16:30-18:30: **Poster Sessions**

PS1: Products and Market

PS2: Biodiversity and Genetic Resources

PS3: Nutrition Value

19:00: **Dinner**

Day Three: March 10, 2004 (Wed)

08:00-10:00: **Plenary Session 4**

Abiotic Stresses

Chair: **Morag Ferguson** Reporter: **Martin Fregene**

- **Alfredo Alves:** Physiological responses of cassava to water deficit
- **Elizabeth Bray:** Using Arabidopsis to identify genes in the drought-response pathways
- **Manabu Ishitanu:** Applying genomics tools to understand drought tolerance mechanisms
- **John Beeching:** Reducing post harvest physiological deterioration in cassava

10:00-10:30: **Break**

10:30-12:30: **Poster Sessions**

PS4: Abiotic and Biotic Stresses

PS5: Post harvest and Starch Quality

12:30-14:00: **Lunch**

14:00-18:30: **Lab and Field Visits**

Three Themes/Sections:

- ***Biotech Tools***
- ***Added Value***
- ***Field Work***

All participants divided in 3 groups: each group will visit the 3 sections (lab and field day)

19:00: **Dinner in Cali**

Day Four: March 11, 2004 (Thu)

08:00-10:00: **Plenary Session 5**

Transformation and Transgenic Products

Chair: **Omaliko** Reporter: **Krit Raemakers**

- **Paul Olson:** Seven habits of effective transgene product development
- **Paul Chavarriaga:** Progress in cassava transformation
- **Henry Daniell:** Chloroplast genetic engineering for agronomic trait improvement and molecular farming

10:00-10:30: **Break**

10:30-12:30: **Plenary Session 6**

Small Farmers and Research Planning

Chair: **Otim-Nape** Reporter: **Roosevelt Escobar**

- **Santiago Perry:** Innovating with small cassava farmers in Colombia
- **Reinhardt Howeler:** Working with farmers in Thailand: Spreading new varieties, improved practices and…new hope
- **Jacqueline Ashby:** Reaching end-users

12:30-14:00: **Lunch**

14:00-16:00: **Plenary Session 7**

Biotic Stresses

Chair: **John Colvin** Reporter: **Elizabeth Alvarez**

- **James Legg:** Epidemics of viruses on cassava in Africa: lessons learned
- **Valerie Verdier:** Expressed genes for CBB resistance in cassava
- **Anthony Bellotti:** Integrated management of cassava arthropod pests
- **Claude Fauquet:** The known and the unknown viruses and satellites infecting cassava in Africa

16:00-16:30: **Break**

16:30-18:30: **Poster Sessions**

PS6: Gene Discovery

PS7: Tissue Culture and Transformation

19:00: **Dinner**

Day Five: March 12, 2004 (Fri)

08:00-10:00: **Plenary Session 8**

Tools for Cassava Breeding

Chair: **Richard Visser** Reporter: **Yona Baguma**

- **Hernán Ceballos:** Doubled haploids
- **Chikelu Mba:** Induced mutagenesis and crop improvement
- **Alfred Dixon:** Conventional breeding: lessons learned
- **Martin Fregene:** MAS in cassava breeding

10:00-10:30: **Break**

10:30-12:30: **Poster Sessions**

PS8: Genomics and Markers

PS9: Participatory Research and Technology Transfer

12:30-14:00: **Lunch**

14:00-16:00: **Plenary Session 9**

Biodiversity and IPR

Chair: **Edison S.** Reporter: **Paul Chavarriaga**

- **Daniel Debouck:** Studies on cassava biodiversity
- **Safo-Kantaka:** Explotation of natural diversity
- **Carol Nottenburg:** IPR/Biotech and Genetic Resources

16:00-16:30: **Break**

16:30-18:00: **Closing Ceremony**

- **Richard Sayre:** Scientific Summary
- **Alfredo Alves:** Concluding Remarks
- **Joachim Voss:** Closing

19:00: **Dinner**

Day Six: March 13, 2004 (Sab)

10:00-12:00: **Meetings**

1) CBN / GCDS / GCP-GI

2) Advanced Cassava Transformation Group

Obs*: Other groups can be organized to discuss some hot topic, according their convenience. These meetings can be scheduled for the sixth day or any other day during extra times.*

Sixth International Scientific Meeting of the Cassava Biotechnology Network

CBN-VI

8-14 March 2004, CIAT, Cali, Colombia

PROGRAM OUTLINE

Time	08-Mar (Mon)	09-Mar (Tue)	10-Mar (Wed)	11-Mar (Thu)	12-Mar (Fri)	13-Mar (Sab)
08:00 - 09:45	Registration	**Plenary Session 1** Innovative approaches in R&D for cassava (3 lectures)	**Plenary Session 4** Abiotic Stresses (4 lectures)	**Plenary Session 5** Transformation and Transgenic Products (3 lectures)	**Plenary Session 8** Tools for Cassava Breeding (4 lectures)	
09:45 - 10:00	Break					
10:00 - 10:30	**Open Ceremony**	Break, group photograph	Break	Break	Break	**Meetings:**
10:30 - 12:00	Joachim Voss and Orlando Meneses **Keynote speakers** : Deborah Delmer Joe Tohme	**Plenary Session 2** Towards Cassava Biotech Next Phase (3 lectures)	**Poster Sessions** 4) Abiotic and Biotic Stresses; 5) Postharvest and Starch Modification	**Plenary Session 6** Small Farmers and Research Planning (3 lectures)	**Poster Sessions** 8) Genomics; 9) Partic. Res. and Tech. Transfer	**1)** CBN / GCDS / GCP-GI **2)** Advanced cassava Transformation Group
12:00 - 12:30	Lunch					
12:30 - 13:30		Lunch	Lunch	Lunch	Lunch	Other groups can be organized to discuss some hot topic, according their convenience. These meetings can be also scheduled for any day in extra times
13:30 - 14:00	**Celebrating Chusa-Ginés** : Future of Biotechnology in Developing Countries					
14:00 - 15:00	(2 lectures) Deployment of Transgenic Cassava Products (2 lectures)	**Plenary Session 3** Cassava Nutrition Value (3 lectures)	**Lab and Field Visits** 3 themes **Biotech Tools** **Added Value** **Field Work**	**Plenary Session 7** Biotic Stresses (4 lectures)	**Plenary Session 9** Biodiversity and IPR (3 lectures)	
15:00 - 15:30						
15:30 - 16:00	Break					
16:00 - 16:30	**Panel: How to Add Value to Cassava?**	Break	Break	Break	Break	
16:30 - 17:30	3 Lectures	**Poster Sessions** 1) Products and Market; 2) Biodiv. & Gen. Resources 3) Nutrition	**Lab and Field Visits** Continuation	**Poster Sessions** 6) Gene Discovery; 7) Tissue Culture and Transformation	Closing Ceremony	
17:30 - 18:30	Welcome Reception (Cocktail)					
19:00	Dinner	Dinner	Dinner in Cali	Dinner	Dinner	

PS1: Products and Market

Fufu yield of three improved cassava (*Manihot esculenta* Crantz) cultivars and their physico-chemical and sensory properties

S.C. Achinewhu; L. Barber; P.U. Umunna

Dept. of Food Science & Technology, Rivers State University of Science & Technology, Port Harcourt, Nigeria

Dried and roasted fufu flour produced from three improved cassava cultivars, TMS 3575, TMS 82/00058 and TMS 3044, were subjected to physical, chemical and sensory analyses. Physical analyses showed fufu yield of 32.6%, 30.8% and 28% swelling index of 355%, 362% (roasted flour), 295%, 285%, 298% (oven dried flour) water absorption capacity of 1.6, 1.8, 1.8ml/g (roasted), 0.8, 1.0, 1.0 (dried) ml/g, relative bulk density of 0.27, 0.26, 0.24g/cm3 (roasted) 0.48, 0.46, 0.48g/cm3 (dried) and starch yield of 56.6, 54.8% and 57.9% for TMS 3575, 82/0058 and 3044 respectively. The dried and roasted fufu flour showed pH values of 3.7, 3.9, 3.9 (roasted) 3.9, 3.9, 4.0 (dried) titratable acidity of 0.9, 0.9, 0.9 (roasted) 0.7, 0.8, 0.9% (fired) lactic acid for TMS 3573, 82/00058 and 3044 respectively. Seventy-two hours of submerged fermentation and subsequent roasting or oven drying reduced the HCN content from 8.6, 6.2, 7.2mg/100g in the raw cultivars to 0.79, 0.90, 1.0mg/100g (roasted), 0.9, 1.1, 1.0mg/100g (dried) in the fufu for TMS 3575, 82/00058 and 3044 respectively, a safe level for human consumption. All three cultivars have good sensory attributes. They could be recommended as good cultivars to processors and consumers of fufu.

Issues in the administration of micro credit in the commercialization of a shelf stable fermented cassava product in Southwest Nigeria

K. Adebayo[1]; A.O. Dipeolu[1]; I.A. Ayinde[1]; A. Jumah[2]; L.O. Sanni[1]; O.B. Oyewole[1]; A.Westby[3]

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2. Institute of Advanced Studies, Viena, Austria
3. Natural Resources Institute, University of Greenwich, Chatham, Kent, United Kingdom

Credit is an important input in all production systems in the real sector. The administration of micro credit has however been regarded as unattractive to the forma credit institutions because of high administrative costs and high default rate among beneficiaries. This on-going study on the development of the small and medium scale enterprise sector producing cassava based products to meet emerging urban demand is making counter-intuitive finding which challenge earlier assumptions in the administration of micro credit. Among these are the high repayment rate and low administrative costs associated with the approach adopted in the study. These finding have brought to the fore functional and policy related issues which are discussed in this paper.

Target varieties for managing cassava commodity chains in Africa

M.O. Akoroda

Department of Agronomy, University of Ibadan. Ibadan. Nigeria. (m.akoroda@cgiar.org)

Target varieties for the 44 cassava producing countries in Africa are not necessary those with high yields of fresh roots. The region is experiencing a cassava postharvest revolution. however, this process is a complex of production, transportation on-farm and off-farm, coupled with a strong diverse product processing and marketing and consumption sector. Targeting end-uses even in the breeders selection strategy is critical for isolating the elites that meet varied purposes of consumers both domestic and industrial. In all these steps, use of the right or appropriate varieties is the most important input. Plant breeders have already amassed a list of suitable varieties are that are yet to be satisfactorily deployed into the farming systems. Stem production is the major problem. The use of varieties that demand high fertilizers to give high starch yields are inappropriate. Dry matter of 25-45 percent are available but the key factor is how to combine the soil fertility restrictions and skilled labor use in locally economic ways to cut down the cost of production and processing. Price of final product is often high because the method, machine and manpower in use are traditional and needs optimizing. The 55 percent of world fresh root output of about 175 million tonnes is produced in Africa in 17 million hectares with hundreds of varieties that are poor yielding (7-9 tonnes per hectare in sole crop farms after 12 months of growth). It is thus clear that no sustained revolution can be made to alleviate poverty if the higher yielding varieties are not used in an increasing ratio to traditional varieties. The need for varieties that use less fertilizers or more efficiently exploit nutrients from the soil is evident.

A cost-benefit analysis of the processing of a shelf stable cassava fufu in Nigeria

I.A. Ayinde[1]; A.O. Dipeolu[1]; K. Adebayo[1]; O.B. Oyewole[1]; L.O. Sanni[1]; J. Adusei[2]; A. Westby[3]

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2. Institute for Advanced Studies, Stumpergasse, Viena A-1060
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Cassava (*Manihot esculenta*) is an important tropical staple crop; which when processed, produces different types of carbohydrate-rich foods which are consumed at least once in a day in Nigeria. Fufu is one of such processed foods and it is usually consumed in the cooked 'ready-to-eat' form, which has a low shelf life. A novel source of increasing the shelf life and reducing other problems associated with the consumption of this form of fufu was recently identified through the development of a new product obtained through the drying of a fermented product of cassava. The popularization of the production and transfer of this technology to the local cassava processors entails the determination of its economic viability and its project-worthiness as a potential small-medium enterprise needed to improve the livelihood systems of the potential beneficiaries. Input-output data were collected at the pilot level production stage and these were subjected to analysis of costs and returns as web as cost-benefit analysis (CBA); projected over a five year period. Results indicate that adoption of this technology would be a worthwhile intervention in the cassava processing systems in terms of value addition and enhancement of livelihood systems of processors through the generation of higher profit.

Development of different functional variants of banana yogurt-like cassava starch beverage

F.V. Corte[1]; S. Tchakirian[2]; S.V. de Fabrizio[2]

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Lactic fermented cassava starch products of different kinds has been successfully presented by our group since 1990. More recently, during CBN V, we presented an example of a tropical fruit (banana), together with cassava starch, as substrates to obtain a yoghurt-like fermented beverage. All these products, elaborated by a simple process, were intended to create new opportunities for farmers and processors as well as new markets for different regions. The global phenomena, lead people to acquire unexpected knowledge and raising interest on healthy foods and nutritional-biosafety aspects. Moreover, new functional foods are claimed by pharmaceutical industries and medical environments of many countries, including developing regions. This work provides the basis for the development of such functional foods. Food safety, is accomplished by lactic fermentation and previous thermal treatment. In case of children or populations with deficit of protein, the desired nutritional balance is obtain by the addition of protein. Vegetarian requirements can be fulfilled by a strictly vegetable formulation, while diets with low fat and glucose contents could be also followed by a vegetable formulation, but sweetened by edulcorants. To cover all these expectation, we present two variants to the original banana yogurt-like beverage poposed before (cassava starch 4%, fresh blended banana 6%). The basic formula is nutritionally improved with 3% milk, obtaining the first variant: more rich in high quality proteins, calcium and vitamins. For most diets, the desired effect is accomplished by using 6% starch, 1,5% soy protein plus sweetener and only banana flavour. Fermentation was followed by pH, acidity and lactic acid bacteria (LAB) enumeration. as well as sensorial and rapid tests of consistency. In all cases, pH and acidity reached safe levels. LAB population remained slightly higher at the end of the fermentation, while texture was significantly improved. In this manner were developed three different purpose functional yogurt-like drink with the probiotic properties of a real yogurt, being an alternative for the diet of different cases including those people with casein intolerance and celiacs.

Effect of cassava based diets on cholesterol content of table eggs

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This paper gives a review of two studies conducted using cassava based diets for laying hens. Study one included unpeeled cassava root meal (UCRM) at levels of up to 20%. In study two, cassava root sievate (CRS) was included at varying levels from 0 to 20%. Both studies involved hens 30 to 40 weeks in lay. Eggs were collected and analyzed for cholesterol content while blood samples were collected for estimation of the lipid profile in blood. Results showed a clear reduction in egg yolk cholesterol and other lipid fractions in the blood. Both studies clearly indicated that feeding cassava based diets resulted in reduced cholesterol content of eggs up to 12.31% in UCRM based diets and 55.45% in CRS based diets. A clear indication of lowered cholesterol and other lipid fractions in the blood was also recorded.

Affordable mechanical efforts for adding value to cassava product in Malawi. A case study with peeling methods and size reduction

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Four improved peeling knives were tested to evaluate their performance compared to four local knives. Four common cassava varieties in Malawi (Nyasungwi, Gomani, Chitembwele and Mbundumali) were used during the tests which took place at Chitedze Agricultural Research Station - Farm Machinery Unit and Nkhotakota. Karonga and Mulanje. A total of 355 farmers participated in the tests which involved them using the knives. On average the improved peeling knives were inferior to the local knives in terms of peeling rate, achieving 29 kg per hour compared to 32 kg per hour with the local knives. However, the improved peeling knives resulted in 10% percent loss compared to 12% using the local knives. A higher peeling efficiency (86%) was achieved from improved knives than that of common knives (85%). Two cassava mechanical chippers (wooden and metallic) were also tested at the same station and locations mentioned above. They were tested for performance in terms of chipping rate, chipping efficiency and percent loss in comparison with hand chipping. On average, the mechanical chippers were superior to the hand chipping in terms of chipping rate and chipping efficiency achieving 91 kg per hour compared to 20 kg per hour and 86% compared to 15% respectively, It was also shown that hand chipping resulted in higher percent loss than the mechanized chipping (25%, compared to 14%). In terms of ease of drying, the mechanized cassava chips dried faster (on wet basis) with a relative drying rate of 98 g (mc) per hour than the hand made ones that achieved 34 g (mc) per hour.

Cassava utilisation and marketing in coastal Kenya

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Cassava is the second most important staple food crop in the food deficit Coast province of Kenya. It is also becoming a major cashcrop in the region. Its yields are low (5-10t/ha) due to the low yielding potential of the preferred local variety (Kibandameno) but which is early maturing and has good taste. However, some varieties in the region yield as much as 32t/ha. The successful use of cassava as a cash crop requires it to be processed into quality products that increase its shelf life, facilitate marketing, and improve palatability. Attributes that would affect adoption by farmers need to be known. To understand the prevailing status of cassava enterprise at the Coast, a questionnaire survey was conducted covering 179 farmers and 44 small scale cassava processors in 4 districts of Coast province, in 2003. Multi-stage sampling technique for selecting farmer sites, farmers, and processors was used. The objectives were to document the existing processing technologies, to identify areas of training on processing and post-harvest handling, to identify desirable cassava attributes during variety selection, and to assess the level of use of cassava as a livestock feed. Results showed that early maturity and good taste were the most desired cassava attributes. Training on cassava processing at household level was needed to increase the shelf life of the fresh cassava sold to off-farm markets by 25.4% of the farmers, and to increase quality of the chopped dry cassava pieces (makopa) sold to factories. Small scale processors needed training on processing high value products to capture wider markets and afford hired labour, on better product packaging, and on better processing equipment that are less tedious and safe. Cassava was also fed to cattle, goats and chicken by 60.4% of the farmers, therefore economic returns of feeding it to livestock needs to be compared with alternative cassava uses.

Cassava leaf production research in Thailand

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The unlignified upper part of the cassava plant is potentially a good source of protein for animal feed rations because of its high yield and nutritive value. Factors affecting yield and protein content of cassava foliage are presently being researched to determine the most suitable practices for cassava foliage production in different parts of the country. Experiments on varieties, plant spacing, fertilizer application and cutting height and frequency were conducted in 2001/02 and 2002/03, and are being repeated this year again at Rayong, Nakhon Ratchasima, and Khon Kaen Field Crops Research Centers. The following results were obtained from these trials: Most of the recommended varieties for root production such as Rayong 5, Kasetsart 50, Rayong 72 and Rayong 90 produced 11-16 t/ha of dry leaves from 4-5 cuts during a 12 month growth cycle, in addition to 10-35 t/ha of fresh roots. The most appropriate plant spacing for leaf production tends to be location specific. At Rayong planting at 30x30 cm gave higher leaf yields than at wider spacings. However, when both leaf and root yields are considered, a spacing of 60x60 cm produced a higher net income. -For high leaf production, the application of at least 300 kg N, 75-150 kg P_2O_5 and 150 kg K_2O/ha is required. -Cutting as often as every 1.5-2 months seems suitable when regrowth is rapid during periods of adequate rainfall, while cutting may be delayed for up to 3 months when regrowth is slow during the dry season.
-Cutting height is probably location specific. A low cutting height of 15 cm above the ground produced good leaf and root yields at Rayong, while at Khon Kaen 25 cm was better. Though we have already obtained good information about cassava leaf production from these trials, it is not yet possible to draw definite conclusions. We need to select those practices that optimize both leaf and root yields and minimize costs in order to increase farmers' net income.

Increased cassava trade through increased production, utilization and commercialization

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Cassava (*Manihot esculenta* Grantz) is an important food securuty as well as income-generating crop for small-scale farmers in the sub-sahara Africa. Livelihood of over 500 million people depend on cassava. Production of the crop is however, constrained by;

- Cultivation of low yielding varieties
- Pests and diseases
- Limited processing and utilization options
- Socio-economic factors (poor mans crop)
- Limited commercialization and marketing

Due to the above constraints, farmers in the developing countries cultivate cassava on small pieces of land just enough for family food. Nevertheless, cassava yields are several folds more per unit area as compared to important staple cereal crops in similar environments including stress of poor soil fertility and low moisture regime. Cassava therefore possesses great potential to spur greater development in the sub-sahara Africa amongst the small-scale farmers. Better still, biological scientists have now developed clones which are high yielding and are resistant to major cassava pests and diseases as well. Analysis of cassava roots and leaves indicate that they are rich in carbohydrates and digestable protein respectively. Nutritionists have developed large number of recipes of cassava for human food. Animal nutritionists have also formulated animal feeds from cassava. Countries such as, Thailand, Asia, Latin America and South Africa have exploited these potentials. As a result productivity and production in such countries has increased. On going work in Kenya is introduction of CMD and CGM resistant cassava clones which are also high yielding. Multiplication and distribution of planting materials of improved cassava varieties. Pilot village processing units have also been introduced to enhance production of high quality chips which attract market from baking firms in Kenya. There is evidence that productivity and production is on the increase. The scope of utilization and commercialization is also on increase. This has led to creation of job opportunities and contributed to poverty alleviation in the country.

Improving the sensory quality and detoxification of two commonly consumed cassava products (flour & gari) in Nigeria using palm wine yeast-solid substrate fermentation techniques

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Pure strain of Palm-wine yeast was used in the fermentation of cassava pulp for 72hrs aerobically, the mash obtained was subsequently processed to flour and gari, the forms in which cassava is popularly consumed in Nigeria. The sensory quality, antinutrient (tannin, phytate and cyanide) content and *in vitro* multi-enzyme protein digestibility for both the fermented and unfermented cassava products were subsequently analyzed. The result of the study revealed that Palm-wine yeast solid substrate fermentation of the cassava pulp prior to processing to cassava flour and gari caused a significant increase ($P<0.05$) in the sensory quality of the gari as typified by significant improvement in its taste, colour, aroma, texture and generally acceptability, when compared to the unfermented cassava product. Furthermore, it caused a significant increase ($P<0.05$) in the digestibility [flour (79.1%); gari (67.3%)] of the cassava products. Conversely, it causes a significant decrease ($P<0.05$) in the cyanide [fermented {flour (14.2mg/kg), gari (6.4mg/kg)}; unfermented {flour (21.3mg/kg), gari (14.6mg/kg)}], tannin [fermented{flour (0.16%), gari (0.10%)} unfermented {flour (0.20%), gari (0.20%)}]and phytate [fermented {flour (510.7mg/100g), gari (484.0mg/100g)}; unfermented {flour (874.4mg/100g), gari (662.8mg/100g)}] content when compared to the unfermented cassava products, while Aflatoxin B1 was not detected in the products. This study therefore revealed that Palm wine yeast, a cheap and abundant fungi could be use to improve the sensory quality, protein digestibility and detoxify two popularly consumed cassava products in Nigeria flour and gari.

Design and performance evaluation of a hydrocyclone system for cassava starch milk concentration

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Hydrocyclone has been used in the starch processing industries since long. But it is generally selected without giving any consideration to the properties of the fluid and solid particles to be separated from the suspension. An attempt was made to design a hydrocyclone for concentration of cassava starch milk taking into account of the physical properties of starch and water. Diameter of hydrocyclone was designed based on the d_{T50} particle size (ie. particle diameter for which the hydrocyclone has 50% efficiency of separation) and the other dimensions were fixed as per Rietma's correlation. A 30 mm hydrocyclone so designed for cassava starch milk concentration had an overall length-150mm, cylinder length-25mm, cone length-125mm, vortex finder length-12mm, diameter of inlet-8.4mm, overflow-10.2mm and under flow-2.6mm. It was tested for its performance *viz.,* total efficiency, reduced efficiency, underflow volume split, increase in under flow concentration and recycling efficiency for different operating parameters (0.6-3 kg/cm^2) and feed concentrations (2-8%). Underflow concentration, total and reduced efficiency were enhanced with increasing pressure and decreasing concentration, while underflow volume split was not affected by the operating parameters. Five units of hydrocyclones with equal inlet and overflow diameter connected in series operating at 3 kg/cm^2 gave about 1.56% overflow concentration and this overflow stream when recycled, provided about 0.40% and 0.46% starch concentration in the overflow and underflow stream after 3 and 4 passes , respectively. A model system of six such batteries, if operated simultaneously was effective in reducing the starch in the overflow to 0.46% resulting a net saving of 43% water, which could be reused for rasping operation. The use of hydrocyclone system thus could reduce the water consumption and thereby the effluent generation. Thus the system can be useful in place of the conventional method of starch settling prevailing in India in which larger detention time often leads to deterioration of starch quality and high BOD and COD in the effluent released from the starch factories.

Studies into the production and qualities of cassava grits (Tapioca) in Nigeria

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Tapioca is a roasted granular product from cassava starch. It has a status of smuggled product in Nigeria, hence, the need to study its production processes with a view of transferring its methodology to local processors. As part of the INCO-DEV sponsored research on SME in West Africa, tapioca from major markets in SouthWest Nigeria and those produced from four varieties of cassava (TMS 30572, TMS 30351, Idileru and Odongbo) in the laboratory using same methods of processing were analysed for chemical, pasting, functional, mineral and sensory qualities. Effects of roasting methods (traditional, manual and electrically rotated rotary driers) on the qualities of tapioca were also investigated. For varietal influences, there were no significant differences in some properties expect for moisture content (8.1-11.7 % db), absorption capacity (231-610%), solubility index (18.0-42.2%) and swelling power (10.3-36.5%). Mineral contents ranged between Ca (0.67-1.43 mg/l), Cu (0.02-0.07 mg/l), Fe (0.13-.089 mg/l), K (5.66-13.07 mg/l), Mg (0.33-5.15 mg/l), Mn (0.01-0.09 mg/l), Na (0.06-0.59 mg/l) and Pb (0.56-1.87 mg/l); pasting properties had peak viscosity of 487.4-684.4 RVU), peak time (3.18-4.94 min) and pasting temperature of 74.6-74.9oC). There were significant differences ($p<0.05$) in the sensory qualities of tapioca samples except for colour and taste. For the roasting methods, there were significant differences ($p<0.05$) in dispersibility, colour L*, water absorption capacity, pH, final viscosity, mineral contents and sensory qualities of tapioca samples.

Influence of extrusion conditions on the functional characteristics of cassava flour extrudates

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A single screw extruder was developed incorporating the provisions to control and measure the processing parameters, such as, barrel temperature, screw speed, pressure and energy. The effect of processing variables, viz., moisture content of flour (14-18%, d.b), barrel temperature (100-120°C) and screw speed (100-200 rpm) on functional properties such as water absorption index and water solubility index of the cassava flour extrudates. The overall water absorption index increased from 2.47 to 3.90 g of gel/g of dry samples and water solubility index decreased from 46.57 to 39.94% for the increase of moisture content from 14 to 18%. As the temperature incresased from 100 to 120°C water absorption index decreased from 3.3189 to 3.0467 g of gel/g and water solubility index increased from 42.53 to 44.16%. Increased in screw speed from 100 to 200rpm lowered the overall water absorption index from 3.5178 to 3.0250 g of gel/g of sample and increased the water solubility index from 42.58 to 45.81%. The water absorption index was negatively related to water solubility index with a correlation coefficient of –0.7896 ($p < 0.01$).

Demand assessment for cassava in industrial sector of India

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Cassava, introduced as food security crop in India, has attained the status of commercial crop generating higher net income. This crop is grown for using as industrial raw material especially in Tamil Nadu and Andhra Pradesh states. Industrially cassava is finding diverse uses to produce many value added products viz., starch, sago, wafers, chips, flour etc. A study conducted by the author on assessing the industrial demand for cassava in India revealed that the industrial sector driven demand is boon for future of cassava in India. Among the many value added products, demand for cassava starch (both native and modified) is derived demand from a wide range of industries viz., textile industry, corrugation box industry, paper conversion industry, adhesives, paper industry besides food industry to produce sago and wafers. Projected cassava starch demand in textile, corrugation box and paper industry were worked out to be 0.87, 3.0, 2.0 lakh tonnes respectively by 2020 in India. Encouraging demand for cassava chips and flour exists in animal feed, textile and adhesive industries. The study also indicated that more than half of the cassava starch produced is being used in India for the production of Sago and this demand is growing continuously. This whopping industrial demand for cassava can only be met through increased cassava production. Modern biotechnological tools like tissue culture, genetic engineering can be handy for producing high starchy, high yielding cassava varieties so as to meet the projected demand in future.

Production of cassava wine by biotechnological method

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The production of cassava wine, an alcoholic beverage from cassava was developed from the current process of making traditional rice alcoholic beverage by collecting the effective microbes from 3 sources of Look-pang, the herb-containing rice cake inoculum and from the application of biotechnology by using the commercial starch-degrading enzymes. Initially, the starch in cassava roots, after peeled, washed and ground, was hydrolyzed by α-amylase enzyme (Termamyl® 120L, Novo Nordisk, 100°C for 2 hrs) and the liquefied starch was further hydrolyzed to glucose by glucoamylase enzyme (AMG 300L, Novo Nordisk, 70°C for 12 hrs). The syrup was then fermented by mixed pure culture isolated from Look-pang A, B and C for 4 weeks. The mash was clarified by sedimentation process and kept for quality evaluation. All isolated mixed culture provided the products with good alcohol content (10 to 11%v/v) comparable to commercial yeasts (11%v/v). The sensory attributes, when evaluated by 25 trained panels, of cassava wine produced by mixed culture isolated from Look-pang A and B, were favorable. The wine did not contain any harmful chemicals and hazardous metals. By using the commercial enzymes and pure culture, the process of making cassava wine could be readily controlled and the products had more consistent quality.

Cassava or rice starchy fermented products as functional products for different intended purposes

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At present, there is general agreement concerning the important role of the gastrointestinal microflora, particularly lactic acid bacteria (LAB) in health preservation and diarrhea treatments. The intestinal microflora is susceptible to be altered by several factors, including food composition. The LAB are able to accomplish starch and milk biotransformation to obtain functional foods for different intended purposes, depending on the starchy substrate and the strain used. Formulations using cassava 6% starch or rice flour water suspensions (S6) plus 3% milk powder (M3), were thermally treated and fermented with three different kind of *Lactobacillus* and *Streptococcus thermophilus* (Zuleta et al, in press) *. Studies of LAB growth kinetic, fermentation properties and utilisation of starch, were addressed on the formulated products. Moreover, LAB survival and permanence in the gastrointestinal tract was assessed using an experimental model in young adult rats, based on the principles of a method suitable for humans. In this study was observed that all fermented products showed LAB counts of 8.0 log CFU/mL or higher. LAB strains were all recovered from faeces at significant levels, indicating a substantial microflora improvement. The survival of *Lactobacillus* in the gut mainly depended upon the bacterial characteristics and it was also affected by the physicochemical properties of the food.The different behavior of strains could be linked to the starchy vehicle food physico-chemical characteristics such as the gelified starch-protein net. The nature of this net and concomitant water retention could explain the higher volume generally observed in feces from cassava starchy products, useful to improve intestinal tract regularity, and the contrast with the drying effect of rice starchy products, which are recommended by WHO for diarrhea treatment. (UBACYT, B0065).

PS2: Biodiversity and Genetic Resources

Breeding system of cassava (*Manihot esculenta* Crantz) ethnovarieties from different regions of Brazil

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Cassava is one of the most important sources of carbohydrates in tropical regions. The objetive of this study was to estimate the multiloci and uniloci outcrossing rates of eight cassava ethnovarieties from different regions of Brazil, in order to provide more information on the breeding system of these varieties. Open pollinated seeds from eight progenies, with eight to 20 plants per progeny, in a total of 122 plants, were evaluated with microssatelite markers. DNA quantification was made in 4% polyacrilamide gel stained with silver nitrate. The amplification was conducted in 6% polyacrilamide gel, also stained with silver nitrate, using nine pre-selected primers. Data was analyzed with PopGen32 and MLTR programs, considering 1.000 bootstraps for the MLTR analyses. The number of alleles per locus varied from 2 to 5, and the percentage of polymorphic loci from 77.78% to 100.0% for the progenies evaluated. The multilocus outcrossing rate was estimated as 0.96 ± 0.03, indicating that the cassava ethnovarieties studied are considered outbreeding plants. The difference between the multilocus and unilocus rates (0.156 ± 0.04), significantly different from zero, indicated the occurrence of 15.6% of biparental inbreeding rate. The estimate of the paternity correlation (0.38 ± 0.07), also significantly different from zero, indicated that part of the offspring was derived from related crosses, sharing the same paternal and maternal genitor.

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Chromoplasts associated proteins from color storage root diversity in cassava (*Manihot esculenta* Crantz)[1]

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Accumulation of β-carotene in the cassava (*Manihot esculenta* Crantz) storage root might result of a variety of regulatory mechanism, including steps in the syntheses pathway and its end product sequestration as well. Such mechanisms may be especially important in non-green tissues performing massive carotenoids biosynthesis, such as in cassava storage roots. Recently, in our laboratory, carotenoids accumulation in cassava was found to be strongly associated with the occurrence of carotene-sequestering proteins isolated from root chromoplasts. Surprisingly, the protein content correlated with high amount of a particular form of carotenoid presence as well as tissue age. These observations are further reported in the present communication. Results so far indicated that soluble protein content in the storage root of color cassava can reach values 2 to 5 times the amount commonly reported for white cassava. Based on size exclusion chromatography we were able to identify three groups of proteins-carotenoids complex coming in association with three distinct pattern of carotenoids accumulation in traditional clone of cassava identified in the Amazon. SDS-PAGE analytical gel indicated a group of small proteins (8 to 14 kDa) in early separated group, a wide size range (14 to 150 kDa) protein group in intermediate separation group, and finally a group of protein size of 40 to 68 kDa. These complex sets of protein groups are under current investigation for further purification in HPLC, full size characterization and sequence.

Celebrating diversity in storage root of cassava (*Manihot esculenta* Crantz)[1]

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Cassava (*Manihot esculenta* Crantz) belongs to the Euphorbiaceae plant family and is the only cultivated species of the genus *Manihot*. Eighty, out of 98 species of the genus, occurs in Brazil with distribution from the high lands of the central plateau of Brazil down to low lands in Amazon Basis and Northeast cost. This pool of genes from the diversity of the genus and the cultivated species is under intense studies in our laboratory to gain knowledge in the storage root potential for improving storage root quality associated to human health. Herbarium database, field trip expedition, DNA molecular markers technology, organization of a GENEBANK of clones, functional genomics technology, and development of new commercial products from the diversity has been performed. In the present document we are reporting the compiled information generated to illustrate our concept working-model to improve cassava. Results, so far, indicate that: 1. The Central plateau of Brazil presents the largest diversity in terms of number of species of the genus. 2. The low lands of Brazil, mainly Amazon Basis, present the largest natural populations within the cultivated species. 3. The largest diversity within the cultivated species occurs in Amazon and Northeast of Brazil. 4. Unusual storage root traits have been identified in traditional clones with high carotenoids, protein and free sugar content and diverse starch type. 5. Natural mutations have been described for the first time in cassava storage root. 6. Processing technologies have been approached to add value to the new cassava clones, which includes powder TUCUPI, vitamin supplement capsule, cassava pickles and natural glucose concentrate.

A novel sugary cassava (*Manihot esculenta* Crantz) accumulating phyto-glycogen in the storage root may be mutated in the gene coding for the branching enzyme[1]

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A new class of cassava (*Manihot esculenta* Crantz) storage root, named sugary cassava, has been described with accumulation of a diverse starch type. A gene expression analyses with genes coding for the major enzymes in the starch pathway is presented in the present report for the clone CAS36.1 that accumulate glycogen like starch. cDNA clones for the genes coding for sucrose synthase, phosphoglucomutase plastidial isoform phosphoglucomutase cytosolic isoform, ADPG pyrophosphorilase, starch synthase, branching enzyme and debranching enzyme (isoamilase) were used for their expression analyzed in the storage root of the sugary clone (CAS36.1) and a farina type (IAC12-829) of cassava. All the genes were expressed in both types of cassava except the gene coding for branching enzyme that was not expressed in the sugary clone CAS36.1. This result strongly suggest that this gene is mutated, some how, altering the expression of the protein enzyme responsible for the formation of amylopectin in the sugary cassava CAS36.1. Sequence homology analyses of a cDNA fragment of this gene was carried out indicating that the non expressed branching enzyme gene in the storage root of the clone CAS36.1 could be the gene coding for isoform II described for corn endosperm. Further research is underway in our lab to analyze the molecular genetics mechanism responsible for the differential expression of this gene.

Inheritance of agronomically relevant traits in cassava

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Very little is known about the inheritance of relevant traits in cassava. Three diallel studies were conducted with 9 or 10 parental clones adapted to the sub-humid, acid soil savannas or mid-altitude valleys, respectively. Each F1 cross was represented by 30 clones. Evaluation trials for each diallel set were grown in two representative locations with three replications. Genotypic and environmental effects were considered fixed and random, respectively. Analysis of variance (combined across the two locations used for each type of environment) showed highly significant genotype x environment interaction for most variables studied. There was a clear consistency in the relative proportion of the sum of squares due to genetic effects explained by general (GCA) and specific (SCA) combining ability effects across the three diallel studies. On average, about 49% of the cross sum of squares for fresh root yield, was explained by GCA. For harvest index, GCA explained about 60% of the cross sum of squares. For dry matter content, height of first branching and plant type score GCA effects were much more important, explaining from 70-75 % of the crosses sum of squares. Plant health variables (reaction to super-elongation disease, white flies, thrips and mites) showed the highest impact of GCA effects explaining from 82 to 86% of the cross sum of squares.

Alternative for estimating general combining ability in cassava breeding

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Cassava breeding is difficult and, compared with other crops, inefficient. Because of the low multiplication rate each cycle of selection takes about six years for completion. Initially a large number of genotypes are evaluated and visual selection is performed on single plant plots. The second and third stage of evaluation and selection involve non-replicated plots generally with 8 and 20 plants, respectively. Selection is usually based on a visual inspection of the plots with few or no data taken. Because no data is taken, a good opportunity for determining the general combining ability of the parental lines whose progenies are evaluated is missed. A modification of the traditional evaluation scheme has been introduced at CIAT's cassava breeding project for an estimation of the general combining ability or breeding value of the parental lines on which each cycle of selection is based. The procedure has provided valuable information for the identification of elite cassava germplasm with the capacity of producing outstanding progenies. Selection of parental lines, therefore, is not based on the performance of the clones themselves, but on the quality of the progenies they produce. The variables recorded for the identification of elite clones whose progenies are superior involve not only yield performance, but also insect and disease resistance, dry matter content, harvest index and plant architecture.

Identification of naturally occurring and irradiation-induced mutant GBSSI alleles of cassava in a heterozygous genetic Background

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Waxy starch or high amylopectin (95-100%) starch is valued in the food, paper, adhesive, and animal feed industries because of the stability of its native or modified gels under a wide ranges of temperature and pH. More than 3 million tons are produced every year from a single source, the special corn hybrid - ZPSC-704 WX. Corn waxy starch commands a premium price, twice that of native starch. There is a growing interest from both the private and public sectors in a waxy starch phenotype from cassava. Three approaches have been embarked upon namely, genetic transformation (anti-sense and sense silencing), irradiation-induced mutation, and screening of the germplasm bank for waxy starch phenotypes. The heterozygous nature of cassava complicates the identification of the waxy phenotype via the latter 2 methods as the waxy gene (GBSSI) is recessive and cassava is a highly heterozygous crop. However the identification of a mutant waxy gene by molecular methods.

Agro-biodiversity of cassava in different three agroecological zones of Suriname— A preliminary analysis

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Cassava (*Manihot esculenta* Crantz) is a staple food crop among the Ameri-Indian and Maroon ethnic groups of Suriname where it is cultivated under mainly subsistence agricultural systems while it is an important commercial crop in the Javanese group. Productivity of the cassava crop as well as the agricultural system is this context is low. Rural migration and introduction of processed foods in their diets may lead to the erosion of both the biodiversity as well as the knowledge base. Therefore a rapid rural appraisal survey was done in various agroecological zones representing each of these groups over a period of 15 months to better understand the agronomic and biological diversity of the cultivated genotypes. Preliminary data will be presented to illustrate the large diversity that exist in terms of cropping systems, land preparation and intensity of cultivation, as well as differences in planting methods and the use of planting materials. Indigenous knowledge base identification in terms of cultural habits and food needs and preferences was also done. The constraints to productivity improvements in such systems are also identified with the purpose of justifying a larger study to address these issues.

Assessment of cassava diversity in Uganda using SSR markers

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The genetic diversity and differentiation of 272 cassava landrace accessions collected in Uganda was assessed in this study using 35 simple sequence repeat (SSR) markers. Twenty accessions each from previous studies of diversity in Tanzania, Ghana and Guatemala, and 18 from holdings at CIAT and IITA that represent the core collection from Latin America were also included in the study to give a total of 9 samples, based on country of origin. From the raw SSR marker data, allelic variation at all loci was used to estimate the parameters of genetic diversity and differentiation, and to estimate the strengths of various forces shaping them. SSR data was analyzed by GENSURVEY (Vekemans & Lefebvre,1997), FSTAT 2.9 (Goudet, 1995) and NTSYS-PC(Rohlf, 1993). The number of alleles observed at each locus in the data set ranged from 2 to 12 alleles per locus (over the 35 loci. The average gene diversity, H_e, that estimates the probability that two randomly selected alleles in a given accession are different, was more than half for all samples (0.576 ± 0.0519). The least values for average gene diversity were observed in Lira and Luweero districts while the highest was found in Kasese. This affirms earlier findings of higher varietal diversity in the western and southwestern districts of Uganda as opposed to those in the eastern districts (Otim-Nape et al, 2001). It is worth noting that these values are lowest for the districts that were worst hit by CMD. Genetic differentiation averaged over all loci was 0.103 ± 0.009 (jackknifing) and 0.082 ± 0.126 calculated by bootstrapping at 99% confidence interval (data not shown), as estimated by F_{st} (theta). This concurs with previous diversity studies in Tanzania confirming low differentiation between country samples. In conclusion, results affirm genetic divergence between African and Latin American accessions as found earlier and . reveals high genetic diversity and a low differentiation in the Ugandan accession. There is a substantial role played by the cassava mosaic disease (CMD) on cassava genetic constitution in respective districts.

Phylogeography and the origin of domestication of cassava: Insights from G3*pdh* sequence data from cassava and wild relatives in the Guianas

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Many aspects of the origin of domestication of cassava are still unresolved. Among the controversial points are the nature of the domesticate (a "compilospecies" resulting from complex interspecific hybridizations, or derived from a single wild ancestor), the number of domestication events, and the place(s) of origin of domestication. Olsen & Schaal (*PNAS*, 1999) studied these questions using sequences of the nuclear gene G3*pdh* and concluded that cassava was probably domesticated once, along the southern rim of Amazonia. However, their sample of the domesticate was limited to 20 individuals from the CIAT core collection, and their sample of wild *Manihot* did not include populations from the Guianas that are currently treated by Allem as belonging to *M. esculenta* subsp. *flabellifolia*, considered by Allem and by Olsen & Schaal as the sole ancestor of cassava. We re-examined the conclusions of their study, by sequencing haplotypes of this gene from a large sample of local Amerindian varieties of cassava from the Guianas and from wild populations of this region, and re-analyzing the entire data set. Our results largely support the conclusions of Olsen & Schaal of a single origin of domestication of cassava along the southern rim of Amazonia, followed by the (probably rapid) diffusion of domesticated cassava across Amazonia. Post-domestication introgression from populations of related wild has increased the haplotype diversity of domesticated cassava. We obtained evidence for natural hybridization in French Guiana between cassava and a wild relative. This wild relative, currently included in *M. esculenta* subsp. *flabellifolia*, appears to be closer to *M. pruinosa*, considered up to now to be endemic to forest-savanna ecotone along the eastern rim of Brazilian Amazonia.

Mining the primary gene pool of cassava: Introgression of resistance to the cassava green mite and high root protein from accessions of *Manihot esculenta* sub spp *Fabellifolia* and *Manihot tristis* into cassava

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High levels of resistance to the cassava green mites were found in 4 inter-specific hybrid families, CW68, CW65, CW67, and CW66, from the wild *Manihot esculenta* sub spp *flabellifolia* accession MFLA 437- 007 following a very dry spell in January 2002 at CIAT Palmira and a subsequent heavy incidence of the green mites. Selected genotypes showing the highest levels of resistance to mites were crossed extensively to elite parents of CIAT's cassava gene pools and a total of 45 BC_1 families were developed. The back cross progenies were planted in a seedling trial, and evaluation for mites resistance revealed a large proportion of genotypes were resistant. The BC_1 progenies were cloned and re-evaluated for to green mites during the dry spell of the current season. Bulk segregant analysis (BSA) of the F_1 and BC_1 families using SSR and RAPD markers have identified putative markers associated with resistance to green mites. These markers are being confirmed for their utility for MAS of resistance to green mites. Similarly high protein content, 10 to 18%, was observed in the storage roots of several accessions of *Manihot esculenta* sub spp *flabellifolia* and *M. tristis*. These wild species were crossed extensively to elite parents of cassava gene pools and more than 30 F_1 families were obtained. Protein content in these progenies ranged from 5 to 10% and root sizes also ranged from very thin roots to commercial size storage root. Back crosses to elite cassava parents will be conducted this season and BSA will be performed on the BC_1 families to identify markers and introgressions that bear the high protein genes. The amino acid profile was evaluated for 2 wild accessions and two inter-specific hybrids and they revealed a very high amount of free arginine, aspartate, and glutamate amino acids, but very low levels of free cysteine and methionine, both sulphur containing amino acids. SDS-PAGE analysis of crude protein extract, using several extraction methods, did not produce a clear-cut band to allow for further analysis of the protein composition of these high protein accessions. Several more extraction methods are being tested. This is the first examples of the implementation of an advanced back cross QTL (ABC-QTL) for the introgression of genes, in this case high levels of resistance to green mites and high protein content, from wild relatives into cassava gene pools.

Cassava genetic diversity in Brazil

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The understanding of the genetic organization of cassava germplasm in Brazil is important for many reasons: Brazil is the site of one or more domestication centers of the species; is the primary origin of most cassava varieties growing in Africa and Asia; is one of the biggest producers of cassava in the world; have a huge area that includes several different agroecosistems were cassava have been planted for centuries. To access this organization 10 microsatellite markers were used and PCoA and cluster analysis were performed involving 623 bitter ("mandiocas bravas") and sweet ("macaxeiras") cassava varieties. Three broad ecological regions were sampled: Amazonian, Sub-Tropical Continental, Sub-Tropical Littoral. Some sub-regions were also defined for more detailed analysis. The results suggest the existence of three general groups of varieties: Group I that includes mostly "mandiocas bravas" predominantly originated from Amazon, especially from the High Negro River basin, but also "mandiocas bravas" from all over the country. Group II that includes mostly "macaxeiras": practically all "macaxeiras" and some "mandiocas bravas" from Amazon and Sub-Tropical Littoral regions belongs to this Group. Group III composed basically by "macaxeiras" includes predominantly varieties from the Sub-Tropical Continental region. A good sample from the Northeast Semiarid Region is still missing in order to complete the picture of the current genetic diversity organization of cassava in Brazil.

Evaluation of genetic relationships among *Manihot* species and determination of possible hybrid or introgressed plants between *M. leptophylla* and cassava

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Allem proposes that *Manihot leptophylla*, a species whose distribution range includes southern Colombia, coastal Ecuador, Perú and reaches to Belém in Brazil, could be part of the synonomous complex of sub-species which includes *M. esculenta* ssp. *esculenta, M. esculenta* ssp. *flabellifolia*, and *M. esculenta* ssp. *Peruviana*. In Ecuador *M. leptophylla* is found as small populations only in the western provinces of Guayas, Manabí, Esmeraldas and Los Ríos. Our studies have led to the identification of at least three distinct ecotypes of *M. leptophylla* in Ecuador, one of these ecotypes (Guayas Province) is morphologically more similar to cassava and molecular analyses also indicate that it is genetically closer to cassava while differentiating more from other *M. leptophylla*. Although the hypothesis of a hybrid origin of cassava has been discarded by the molecular evidence of Olsen & Schaal which affirms the ancestry of cassava to *M. flabellifolia*, the occurrence of natural hybrids has not been discarded and current research carried out with *M. glaziovii* points to the contribution of this species to the genetic constitution of cassava. As we have determined that *M. leptophylla* is genetically distant from cassava our present work is focused on evaluating the relationship of *Manihot leptophylla* to other species of the genus, especially regarding its systematic standing among the central and south American species. Additionally, there is the need to provide proof of the contribution of *M. leptophylla* into the genetic constitution of cassava. Given that there is evidence of the occurrence of natural hybridization and introgression processes, the question remains regarding the genetic flux between the wild and the domesticated species (uni or bi-directional exchange). At the same time, it is also necessary to gather data that will confirm the presumption based on markers evidence that this species may have participated in the genetic differentiation of sweet and bitter varieties.
[Results from 15 SSR loci that were used to evaluate the genetic relationships among *M. leptophylla*, *M. esculenta* (sweet and bitter varieties*), M. flabellifolia, M. baccata*, *M. aesculifolia*, *M. rubricaulis*, and *M. chlorostica* and to determine hybrid or introgressed plants are presented].

Plant breeding prospects of polyploidizing cassava interspecific hybrids

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The wild species; *Manihot glaziovii, M.pseudoglaziovii, M. oligantha M.caerulescens, M.dichotoma, M.pilosa, M.neusana, M.pohlii,M.reptans, M.aesculifolia, M.grahamii*, and *M. anomala* were hybridized with cassava. Interspecific barriers were broken using different methods and techiques. Morphological markers were identified and enabled confirming the hybridization between the different taxa. Polyploid types of these interspecific hybrids were obtaibed by applying 0.2% aquous colchicine solution to lateral buds for a period of 24 hrs. In addition to total tetraploids, sectorial and periclinal chimeras were identified with different frequencies. The polyploidization has resulted in restoring interspecific hybrids fertility leading to formation of new gene pools from which may evolve new synthetic species. Description and photos of these doubled chromosome types are being presented. The study of apomixis frequency in polyploidized types shoed it has increased significantly reaching 51% compared to 1-2% in diploid types.

Keywords: Wild *Manihot*, polyploidy, chimera, geen pool

Genetic diversity of cassava in Ethiopia: Its implication for food security and the need for biotechnology research

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Thirty-six germplasm accessions of cassava representing IITA gene pool and Ethiopian land races were studied with respect to different qualitative and quantitative characters contributing to characterization and genetic divergence at Jima Agricultural Research Center during 2002-2003 cropping season. The minimum descriptor lists of the International Cassava Germplasm Network and International Plant Genetic Resources Institute were adopted in the study. Analysis of variance for quantitative characters indicated significant ($p<0.05$) variations among the accessions for majority of the characters except plant height, number of main stems, stem girth and storage root length. Cluster and distance analyses of quantitative characters based on multivariate analysis pointed out to the existence of six divergent groups. The clustering pattern of germplasm accessions was found not necessarily related to geographical origin and genetic diversity. The maximum distance was observed between clusters V and VI ($D^2 = 722.93$) while the minimum was between clusters II and I ($D^2 = 33.41$). The present study indicated a considerable amount of variability for majority of the characters of interest in cassava for exploitation towards strengthening the food security system of the country. Nevertheless, the need for confirmation the conventional characterization and diversity analysis approaches through advanced tools of biochemical and molecular approaches and widening of the genetic base for strengthening cassava improvement strategy are suggested.

Genetic diversity of cassava (*Manihot esculenta* crantz) landraces in Ghana using SSR markers

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Cassava a major staple in Ghana that ranks first in area under cultivation and utilization has been characterized morphologically as a sep towards improvement. Morphological markers are highly subjective and environmentally influenced hence show little polymorphism. In this study a total of 36 Simple Sequence Repeat (SSR) loci markers were used to assess the genetic diversity and differentiation in 320 cassava landraces collected from all cassava growing regions of Ghana. An average number of 5 alleles for each locus and 0.5245 + - 0.0045 gene diversity were observed. The total heretozygosity (Ht) and genetic differentiation (Gst) ranged widely across the markers. Genetic differentiation estimated by Fst (theta) had an overall value of 0.04 and a characteristic very low value for samples between regions, with the exception of some accessions from Northern Ghana the semi arid and arid ecological zones with moderate to high genetic differentiation. This is in consonance with other finding that agricultural practices and the allogamous nature of cassava produces a large pool of volunteer seedlings that natural and human selection acts upon to maintain a high level of diversity and low differentiation. The first two PCA access accounted for 42% of the total variation in the genetic distances. The PCA showed loose clustering of the landraces by regions except for the genotypes from Northern region which had a sub-structure being the most differentiated.

Phenotypic and genetic correlations among agronomically relevant traits in cassava

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There is a limited knowledge about the inheritance of agronomically relevant traits and their relationship. Thirty-eight elite clones were evaluated in a uniform regional trial across thirteen locations in the northern coast of Colombia. Phenotypic and genetic correlations (within parenthesis) were measured in individual location data as well as in the analysis combined across locations. In the analysis combined across the thirteen locations, fresh root yield showed the following correlations: fresh foliage productivity –0.237 (-0.375); harvest index 0.735 (0.789); root dry matter content –0.268 (-0.323). Fresh foliage productivity had the following coefficients with other traits: harvest index -0.786 (-0.833) and dry root matter content 0.031 (0.031). Phenotypic and genetic correlations between harvest index and root dry matter content were –0.110 and –0.124, respectively. In every case genetic correlations were higher than phenotypic ones. Harvest index showed the highest correlation with fresh root productivity, supporting the findings previously reported at CIAT and highlighting the importance of harvest index as one of the variables that should accompany root productivity in the selection indexes used for improving cassava productivity. As expected, harvest index was negatively associated with fresh foliage productivity. These results are useful for an improved implementation of selection indices in cassava breeding.

Stability and genotype by environment analysis in cassava

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Cassava is recognized by its capacity to grow and produce competitively under environmental constraints where few other crops can compete. However, the crop is also distinctive for its remarkable magnitude of genotype by environment interactions. Thirty-eight elite clones were evaluated in a uniform regional trial across thirteen locations in the northern coast of Colombia. The stability of performance was analyzed following the methodology proposed by Eberhardt and Russell. Large variation of mean productivity was observed ranging from 36.2 (SM 1565-17) to 20.9 (SM 1657-14) t/ha of fresh roots. The highest yielding clone (SM 1565-17) also showed an excellent stability with the regression coefficient not different from zero (b=1.052). Clone CM 4843-1 showed an intermediate yield potential with average fresh root yield of 28.9 t/ha, but an excellent adaptation to low-productivity environments (regression coefficient 0.344). On the other hand, clones SM 1973-23, SGB 765-2 and SGB 765-4, which also had intermediate fresh root productivity (ranging from 25.6 to 28.3 t/ha), showed a particular adaptation to high-productivity environments (regression coefficients > 1.30). The two SGB clones were derived from a farmers' participatory breeding scheme in that region of Colombia. It was unexpected to conclude that they are particularly adapted to high-productivity conditions. Results suggest that it is possible to identify cassava germplasm with high yield potential and stability of production. Other variables analyzed were harvest index and dry matter content in the roots.

Analysis of genotype by environment interactions in cassava using the AMMI model

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The additive main effects and multiplicative interaction analysis (AMMI) has been used in the analysis of genotype by environment interaction. Thirty-eight cassava clones were evaluated in uniform trials across thirteen environments with three replications in each trial. All locations were in the northern coast of Colombia from Pitalito in the Atlántico Department to Mutatá in the Urabá region (Antioquia Department). Trials at two locations (Carepa and Necoclí), both in the Urabá region were repeated for two consecutive years in 2001 and 2002. The AMMI analysis was very efficient in distinguishing different locations and separating those genotypes that did not significantly interact with the environment from those that contributed significantly to the genotype by environment interaction sum of squares. Furthermore, for those clones exposing high interaction with the environment, particular locations in which these clones had an outstanding performance could be identified. The AMMI approach, therefore, was useful in identifying clones with high productivity and wide adaptation (low genotype by environment interaction); identifying clones that were particularly adapted to certain locations; and in organizing the information so that adequate selection of key evaluation locations could be accomplished.

Heritability of agronomically relevant traits in cassava

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A molecular genetic map of cassava has been developed. However, little is known about the inheritance of agronomically relevant traits for this crop. Thirty-eight elite clones were evaluated in a uniform regional trial across thirteen locations in the northern coast of Colombia. Broad sense heritability coefficients were estimated for individual locations and across location data. Mean fresh root productivity ranged from 53.0 (Carepa, Antioquia Department) down to 18.3 t/ha (Caracolí, Atlántico Department). Heritability coefficients for this trait, based on individual location data ranged from 0.876 (Corozal, Sucre Department) down to 0.304 (Necoclí, Antioquia Department) with a mean value of 0.560. In the analysis combined across the 13 locations, heritability coefficient (which is already corrected by the effect of genotype by environment interaction) was 0.789. Heritability coefficients in the combined analysis across the thirteen locations for dry matter content in the roots, harvest index and fresh foliage production were 0.947, 0.929 and 0.840 respectively. Coefficients of variability for fresh root yield, based on individual location data, ranged from 30.9 down to 19.4% with a mean of 24.5%. In the analysis combined across the thirteen locations, the coefficient of variation was 24.8%. Results indicate the high potential of this crop as a source of raw material for different purposes (mean fresh root productivity of 33.8 t/ha) but also the possibilities of a genetic improvement of the crop based on these relatively high heritability estimates.

Molecular diversity in the land races of cassava in India

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Cassava is an introduced crop in India. Still, over the years, a number of local varieties have evolved. They are mostly natural hybrids, selected by farmers for cooking quality, chip making and early maturity. These land races were screened for morphological as well as yield characters. The data showed high variability for height of plants, branching habit, number of tubers, weight of tubers and colour of the stem, petiole, emerging leaf, tuber skin, rind as well as flesh. However, there was not mach variability for reaction to Cassava Mosaic Disease and most of the varieties were susceptible. Duplicates among the land races were identified by morphological, biochemical and DNA analysis. Some of the distinct popular varieties were subjected to RAPD analysis using a number of random primers. The banding pattern showed very high polymorphism. Similarity Index varied from 90 to 25. This hints at the existence of a secondary centre of diversity for cassava in India, as in Africa. The soil and climatic conditions in India, especially in the tropical region, are ideally suited for crop establishment as well as for flowering and natural hybridization. Details are given in the paper.

Comparative reproductive ecology of wild and domesticated cassava gives evidence of selection for rapid growth in traditional agroecosystems

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Although cassava is vegetatively propagated by farmers, sexual reproduction continues to play important roles in both traditional management and in scientific breeding programs. We compared the seed and seedling ecology of domesticated cassava managed by Amerindian farmers in Guyana and French Guiana with that of a close wild relative of cassava found in savanna-forest ecotones in this region. In both wild and domesticated cassava, physiological seed dormancy, mediated by soil temperature, ensures that seeds in the soil seed bank do not germinate under a vegetation cover. Germination is triggered by high soil temperatures that signal removal of covering vegetation by disturbance (fire, mowing, ploughing, etc., in wild cassava; field clearing in domesticated cassava). Seedling functional morphology presented strong contrasts between wild and domesticated cassava. Seedlings of the two taxa appear to have evolved divergent adaptations to contrasting environments. Traits of wild cassava are those expected in environments where rapid initial growth is less important than protection from risks to above-ground parts from fire, herbivores, and drought. Traits of domesticated cassava are those expected in environments where human management minimises such risks and favours rapid initial growth. These contrasting strategies are also reflected in the ecophysiology of photosynthesis. When grown under identical greenhouse conditions, domesticated cassava was characterised by higher CO_2 exchange rates (expressed per unit mass), and higher specific leaf area, nitrogen content, and nitrogen use efficiency than wild cassava. Wild cassava had higher leaf dry matter content and C/N ratios. We propose that all these differences constitute part of the domestication syndrome of cassava and result from natural and human selection for rapid growth in agricultural environments, richer in resources and more protected from risks than the environments of wild ancestors.

Sugary cassava (*Manihot esculenta* Crantz): Preliminary morphological characterization and agronomic evaluation[1]

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In addition to the farina and table type of cassava, a new kind of cassava (sugary cassava) was, recently, identified and isolated under cultivation in Amazon. Biochemical and molecular characterization of this sugary storage root phenotype indicates unusual storage root traits, including high free sugar content (mainly glucose) and distinct starch molecule structure in comparison to the conventional cassava cultivated for farina and table harvesting propose. Since the scientific community did not know the sugary cassava type, there is no morphological and agronomic research applied to them. In complementation to our biochemical and molecular characterization research applied to the sugary cassava, here we are present our preliminary morphological characterization and agronomic evaluation for our GENEBANK collection of this type of cassava grown in Amazon region. Fifteen sugary clones were grown in field plots at EMBRAPA-Amazonia Oriental in the Amazon for field evaluation in comparison with farina and table type of cassava. Morphological characterization showed large diversity in leaf, petiole, stem, and storage root as usually occurs in the other types of cassava. No particular morphological characteristic could be used to distinguish sugary cassava from farina or table type of cassava, except the size, weight and density in water of the fresh storage root. Agronomic evaluation indicated a production of storage root fresh weight per plant varying from 2 to 10 kg with 10-month growth, and dry weight of 9 to 22%, while the table and farina cassava produced 2-4 kgFWt./plant and 30 to 40% dry weight.

Manihot caerulescence: A new source of resistance to cassava mosaic disease (CMD)

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Cassava is a subsistence crop, which provides high calorific value throughout the developing world. In India cassava occupies about 0.26 million hectares producing 5.868 million tons annually. Cassava mosaic disease (CMD) is the most important problem of this crop in India leading to 16-80 per cent yield loss. Wild *Manihot* species has been used as source of many useful traits in cassava. At the Central Tuber Crops Research Institute, (CTCRI) thirty seven accessions of wild *Manihot* species comprising of *M. glaziovii, M.pseudoglaziovii M. caerulescence, M. tristis, M. peruviana* and *M. flabellifolia* were sceened for resistance to cassava mosaic disease (CMD) through wedge grafting All the accessions of *Manihot caerulescence* exhibited high level of resistance and were used as donor parents for transferring resistance to elite Indian cultivars. Resistant interspecific crosses were also graft inoculated reciprocally with highly susceptible cultivars to confirm resistance. This study point towards the need for characterization and utilization of this novel source of CMD host resistance through biotechnological tools.

Evaluation of genetic diversity of traditional varieties and spontaneous sexual plants by microsatellites

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Cassava (*Manihot esculenta* Crantz), is a tropical crop and one of the most important sources of carbohydrates in human diet. In traditional agrosystems cassava is a subsistence crop characterized by an important diversity. Fifteen pairs of microsatellite primers were used to assess the genetic variability of 22 varieties of cassava traditionally grown by quichuas and 44 volonteer seddlings growing simultaneously in a quichua traditional field in Napo. Fourteen loci were polymorphic. 44 alleles were found in varieties and 45 alleles in volunteer seedlings populations. Given the vegetative multiplication of cassava, landraces would be spected to be clonal but identical genotypes (clones) were not observed. Some volunteer seedlings show a low differentiation with cassava varieties; this could by explained by the cross polinization between varieties that gave way to the seeds. An important intervarietal and intravarietal diversity was detected; the 22 landraces were different; diversity inside varieties could generate confusions in the local classification; varieties morphologically similar were classified under the same name being genetically differents. The incorporation of volunteer seedlings in this study allowed that they could be evaluated with the rest of traditional varieties with the chance that if they present interesting characteristics they could be selected by the farmers and increased the crop genetic variability in account that they were originated from cross polinization. The important genetic variability found in a single farm shows a dynamic management of the crop by the quichuas farmers as is found in other traditional groups who show a big interest in acquiring new varieties and actively exchange cuttings with other farmers; they also take in to consideration volunteer seedlings that could by part of the stock of stems cuttings by farmers for the next cassava crop.

Assessment of genetic diversity and improvement on cassava germplasm collected in China using molecular markers

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Cassava (*Manihot esculenta* Crantz) cultivation and revolution in China pass over two hundred years. Several elite cultivars such as HN 205 have been bred and disseminated. Presently, cassava is an important energy sources crop in south China, higher starch content in storage roots, resistance to Brown leaf spot (*Cercosporidium henningsii*) and to abiotic stresses will be the key breeding objectives. In the study, genetic diversity within 102 accessions collected in nursery of Chinese Academy of Tropical Agricultural Sciences has been investigated using SSR and AFLP markers. The genetic relationship of all accessions were shown in single tree by neighbor-joing analysis. Many of the duplicated collections were identified. The results supply benefit information for genetic improvement of these materials. Meanwhile, two F1 experimental populations could be used in molecular marker of starch content, waxy and yield characters have been accomplished. Investigation of quality and yield characters of F1 individuals also has been finished. How to develop a new approach for cassava breeding integrating biotechnology tools was discussed in the paper.

Progress in cassava core germplasm conservation in Thailand

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The systematic collection of locally grown material in all over the cassava growing areas of Thailand started in 1993. Formerly, maintaining in the field only and to ensure its long-term availability with low risk of loss contamination by pests or pathogens. In 2000, CIAT and DOA of Thailand have agreed on the establishment a program of cassava germplasm conservation for the another safe duplication of cassava genetic resources. In 2001,the Rayong Field Crops Research Center (RYFCRC) of Thailand have been received a duplicate set of the CIAT cassava core collection as *in vitro* plants with 2 tissue culture tubes for each clone. At present RYFCRC have been received 608 accessions for 2 years. The plant has been subcultured and preserved at RYFCRC. The *in vitro* collection, the cultures are maintained in under slow growth condition : 20 $\pm$ 1 ^{O}C constant temperature with 1000 – 3000 lux illumination during 16 hours a day provided by cool white fluorescent lamps, at 70-90% relative humidity. Three plants are grown in each 4.5x12 cm. glass tube containing a modified Morishige and skoog mediums developed at CIAT and 10 plants of each clone are routinely maintained. After multiplication the material have been transfer to greenhouse and then to the field. In the future, to evaluated for yield, starch content, morphological and physiological character, molecular , a high resistant to diseases and pests and the data that will be share with CIAT for the cassava germplasm database. Finally, we can assume that virtually all the additional economic effect generated by the higher fresh root yield of new cultivars and entering direct to the better living of small farmer.

PS 3: Nutrition

Cassava – based nutritionally balanced low cost weaning foods for children of the developing world

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Nutritional inadequacy at the weaning stage has been highlighted as one of the main reasons for malnutrition in infants. Most of the popular brands of weaning foods are beyond the reach of majority of the population in the developing world due to the high cost factor. Being an energy rich root crop of tropical countries, cassava has ample scope as the base raw material for making low cost weaning foods, aimed at the nutritionally and socially impoverished segments of the society. Nevertheless, cassava starch / flour is characterized by high viscosity leading to a thick consistency of the weaning foods such that they fall short of the adequate nutrient density. Attempts were made to reduce the bulk and thick consistency of cassava-based weaning food formulations using pre-treatments like dry heat treatment, microwave exposure, roasting, malted cereal or germinated legume incorporation in cassava flour etc. Enhancement in protein content up to 10-12% could be achieved through fortification with cereal flours (wheat, rice and finger millet) and green gram flour. Physical properties of the weaning food formulations like water holding capacity and dispersibility, nutritional evaluation as well as the viscosity characteristics were studied. Considerable reduction in viscosity leading to a thin gruel consistency could be achieved through the addition of malted cereals and germinated legumes prior to processing the cassava flour-based formulations. The cost economics of the acceptable formulations were worked out against the prevailing market prices of popular brands.

Production of safer cassava food products having accelerated cyanogenesis

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Cassava leaves and roots contain potentially toxic levels of the cyanogenic glycoside, linamarin. Upon tissue damage linamarin is released from the vacuole and deglycosylated by the cell wall localized enzyme linamarase to produce acetone cyanohydrin. Acetone cyanohydrin is then broken down either spontaneously (pH > 5.0 or temperature > 35 ^{o}C) or by hydroxynitrile lyase (HNL) to produce acetone and hydrogen cyanide. During food processing essentially all free cyanide is removed by water extraction or volatilization. Consumption of residual cyanogens (linamarin or acetone cyanohydrin) in incompletely processed cassava roots, however, can result in cyanide poisoning due to conversion of the cyanogens to cyanide in the body. Significantly HNL is not expressed in cassava roots unlike leaves. Since acetone cyanohydrin may be stabilized by the low pHs often encountered during root processing we hypothesized that the low levels of HNL in roots may effectively reduce the efficiency of cyanogenesis and cyanide removal. To test this hypothesis we have over-expressed HNL in transgenic cassava plants under the control of a double 35S CaMV promoter. We show that HNL activity increased more than 2-fold in leaves and 13-fold in roots of transgenic plants relative to wild-type plants. Elevated HNL levels were correlated with substantially reduced acetone cyanohydrin levels and increased cyanide volatilization in processed or homogenized roots. These results demonstrate that acetone cyanohydrin can be effectively eliminated in processed roots of plants over-expressing HNL. Importantly, intact roots of transgenic plants have normal levels of linamarin and linamarase. Thus, transgenic plants with elevated HNL levels retain the herbivore deterrent attributes of cyanogens that may be of importance to subsistence farmers while providing a safer food product to the consumer.

Cyanogenic glycoside transport in cassava: Implications for the generation of a cyanogen-free cassava plant

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The evidence for long-distance transport of cyanogenic glucosides in plants is not well documented although it has been demonstrated that cyanogenic glycosides are transported during early stages of development in sorghum and rubber tree. Here, we present evidence in support of the long-distance transport of cyanogenic glucosides in cassava. Cassava accumulates potentially toxic levels of cyanogenic glycosides (linamarin, 95% and lotaustralin 5%) in all parts of the plant. The first dedicated step in linamarin synthesis is catalyzed by two similar cytochrome P450s encoded by the *CYP79D1* and *CYP79D2* genes. Using tissue-specific promoters to drive the expression of CYP79D1/D2 antisense constructs, we have successfully generated transgenic cassava plants in which the steady-state levels of the CYP79D1 and CYP79D2 transcripts have been selectively reduced or eliminated in leaves and roots. Importantly, root linamarin content was unaltered in transformants in which CYP79D1/D2 transcripts steady-state levels were reduced to non-detectable levels in roots. In contrast, the root linamarin content of transformants having substantially reduced CYP79D1/D2 transcripts levels in leaves was reduced by 99%. These results suggest that linamarin made in the leaves is transported to the roots. Further analysis of the growth of transgenic cassava in media lacking ammonia suggests that cyanogenic glucosides may function as an important mobile nitrogen source in young plants in addition to their proven ability to deter herbivory.

ICP-MS screening of mineral elements from cassava—An emerging industrial crop in South India

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Manihot esculenta Crantz (Euphorbiaceae) has had many folk medicine uses in tropical and subtropical countries where it has been a staple nutraceutical food for millions of peoples. Leaves and roots have been a folk remedy for tumors and cancers. Cassava is currently cultivated on 242,000 hectares (ha) in India - an annual production of 5.9 million tonnes (t). The states of Kerala and Tamil Nadu cultivate 213,000 ha (88 percent of the country's total area) and have average productivities of 19.8 and 37.7 t/ha, respectively. Earlier, this crop was grown mainly as a staple food but recent area expansion is attributed to its industrial value. Leaf-nutrient analysis is the best method for diagnosing cassava plant nutritional status, and represents an important tool for determining future multiplication requirements. The present study performed to the possible relationship between macro- as well as micro-elements in the leaves of young-stem and in the tuber at different stages of development, were carried south Indian soil grown cassava by following a special leaf-sampling technique. The data have revealed accumulation of N, P, K, Mg, Zn and Cu, a decrease of Zn, Ca, Fe and B, and no change in Mn content in the leaves as the tuber development progressed. Clonal variation influenced the level of N, K, Mg, Mn and B in the leaves. The metabolism of each element has been discussed in the light of various agro-climatic conditions prevailing in this area.

Burkina faso: Stake of the production of cassava as alternative to cereales deficit under sahelian climate

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Agriculture in Burkina Faso is an agriculture of subsistence dominated by cereales which occupy more than 88% of the annual cultivated surfaces. The rainy caracter of the agricultural production exposes this agriculture to climatic effect (dryness, temporal and space variation of the rainfall) and to the continual environment and soils degradation. For these recurrent reasons the cereal deficit more or less important year to year train an importation of more than 4 billion of F.CFA (approximately 6850000 US Dollars) of foodstuffs each year by burkinabè state. It is for it that cassava has been retained for its great adaptation to the climatic variations and its weak requirement of soil nutrients. On the basis of agronomic test in farmers fields in different parts of the country done on cassava new varieties introduced from IITA-Ibadan (Nigeria) near of local varieties six varieties have retained for their adaptability and for their high yield. It is: TMS 91/02312; TMS 92/0067; TMS 94/0270; TMS 92/0325; TMS 92/0427 and TMS 4(2)1425. The different pathogenics and ravagers of cassava in Burkina Faso were listed and the suitable methods of fight are in study. In the country the cassava's cycle of production is eleven to twelve months. Indeed, cassava spends several months (6 to 9 months) in dry condition. Harvest generally intervenes at the beginning of rainfalling in June generally marked by a food shortage especially in rural zone; the use of these high-yield varieties of cassava and their popularization could constitute an alternative to the resolution of the frequent problem of cereal deficit in the country at this period of the year.

Assessment of genetic improved cassava plants expressing a nutritious storage protein (ASP1) gene

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Protein malnutrition is a severe problem in sub-Saharan Africa as well as other poor regions where many people are dependent on cassava as a staple diet. Although cassava is an excellent carbohydrate source that contributes to food security significantly, its storage roots are deficient in proteins. The nutritional quality of cassava may be improved by genetic breeding. Here we present our further results of genetic improved cassava lines expressing an essential amino acid-rich storage protein (ASP1) gene, which is driven by the CaMV 35S promoter, in their leaves and storage roots. The expression of ASP1 has been found to correlate with the copy numbers of the transgene. The ASP1 tetramer could be detected in leaves as well as in primary roots of cultured transgenic plants by Western analysis. Analysis of one-year-old plants from second vegetative generation in the greenhouse demonstrated the stable expression of ASP1 both in the leaves and storage roots. Increase of protein content and essential amino acid composition in storage roots was achieved in several transgenic lines. These results indicate that the nutritional improvement of cassava storage roots can be achieved by constitutive expression of ASP1 in transgenic plants. Currently we study the possibility of improving the levels of expression and accumulation of ASP1 using cassava storage root specific promoters and signal sequences that can target the protein to storage organelle plastid and vacuole.

PS4: Abiotic and Biotic Stresses

The case for an integrated approach to the management of cassava mosaic disease in Uganda

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Among the possible control measures for controlling cassava mosaic disease (CMD), the use of resistant varieties is the most effective and has been used in Africa for several decades. Recent studies in Uganda indicated progress of CMD to be slow in resistant varieties compared to susceptible cultivars ($P<0.001$), leading to low cumulative and final CMD incidences. Transient symptoms (often leading to '*recovery*' from CMD) and '*reversion*' (production of CMD-free plants by cuttings from diseased plants) were observed. In field experiments, up to 90% recovery and 60% reversion in CMD-resistant varieties were recorded. However, in another study, ratooning of CMD-resistant plants resulted in more severe symptoms on re-growth of previously diseased plants and an increase in incidence of CMD-affected plants compared to disease in the previous generation. Besides apparently exerting pressure for selection of more virulent virus types, this has the effect of increasing the amount of virus in area and an increased risk of further spread, especially to susceptible cultivars. Cultivar differences have been observed in the proportion of mild and severe CMD. Where the highly resistant variety Nase 3 is widely cultivated, the severe 'yellow mosaic' was more common than mild phenotype. The latter was predominant in areas mainly growing moderately resistant varieties. It is therefore noted that there is potential for recovery, reversion, selection of CMD-free planting material and rouging to generate beneficial interactive effects, especially under conditions of low CMD spread, resulting in better control of CMD rather than using each practice singly.

Nataima-31, a cassava (*Manihot esculenta*) variety resistant to the whitefly, *Aleurotrachelus socialis*

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The whitefly, *Aleurotrachelus socialis*, is one of the major cassava pests, causing considerable yield loss in Northern South America. Yield losses of 5 to 79 percent have been reported from Colombia. Due to the whiteflies' short development cycle (30 to 35 days) and rapid population increases, the use of chemical pesticides is uneconomical for the small cassava farmer and pesticide application is not considered as a long-term viable solution to whitefly damage. Host plant resistance offers the farmer a low cost, sustainable, long- term alternative solution. The cassava genotype MEcu 72 was identified as highly resistant to *A. socialis*. A cross with MBra 12, a high yielding, good quality genotype, resulted in numerous progeny with moderate levels of whitefly resistance, high yield and good cooking quality. CORPOICA, the Colombian Institute of Agronomy, evaluated four progeny over a four-year period at three localities in the Tolima Valley and compared these to local farmer varieties. It chose the genotype CG 489-31, subsequently named Nataima-31, to be officially released to producers in the Tolima region. A field day attended by about 200 producers and technicians, was organized in March 2003 to release the variety. Stem cuttings of Nataima-31 were distributed to farmers participating in the field day activities.

Resistance screening to whitefly infestation in a range of cassava genotypes established in multilocational trials of IITA in Nigeria

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Host plant resistance to whiteflies is not common in cultivated crops. In cassava, although resistance breeding efforts have been recently geared towards host plant resistance mechanism against *Bemisia tabaci* Gennadius, large scale screening of a range of cassava genotypes is still limited. In this study, twenty-two new cassava elite clones and eight other genotypes, as checks, were assessed for whitefly infestation in 1999/2000 and 2000/2001 cropping seasons in experimental fields of the International Institute of Tropical Agriculture (IITA), Nigeria. On each scoring day, the vector count was done between 6 and 8 AM, when the whiteflies were relatively immobile, by counting adult whitefly populations on the five top-most expanded leaves of the selected cassava cultivars.All through the 6-month scoring period, there was a highly significant difference (p<0.01) in whitefly infestation among the newly developed cassava elite clones. Whiteflies were observed on the underside of the five top-most leaves of cassava plants and the infestation pattern varied significantly between the cassava genotypes. Vector population build-up was observed in Ibadan (Forest-savannah transition zone) and Mokwa (Southern guinea savannah) 2 months after planting (MAP), while the build-up was delayed to 4 MAP in Zaria (Northern guinea savannah zone). Mean whitefly infestation across cassava genotypes was significantly highest (16.6 whiteflies/plant) in Ibadan, as revealed by Duncan multiple range test. The lowest mean infestation per plant was observed in Zaria (0.2). Moreover, the two locations, Ibadan and Onne (humid forest) had the highest numbers of adult whiteflies and also showed a relatively high incidence of Cassava mosaic disease (CMD). In general, whitefly infestation was very low in all locations at 5 and 6 MAP. During this period, five cassava genotypes (96/1439, 91/02324, 91/02324, 95/0166 and 96/1708) significantly supported higher infestation compared to other genotypes (p< 0.01), whereas plants of clones 96/1089A and 96/1800 supported significantly lower whitefly infestations across genotypes in all locations. Invariably, percentage disease incidence and symptom severity scores obtained for the latter two genotypes were remarkably lower compared to other genotypes. The preferential whitefly visitation, the differences between locations in relation to whitefly population and the relationship between CMD incidence and whitefly populations are discussed.

Identification of a reolike virus infecting *Manihot esculenta* and associated with cassava frog skin disease

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The causal agent of cassava frogskin disease (CFSD) has remained unknown since the disorder was first identified in 1971. Although a viral agent has been suspected, it has been difficult to develop conclusive evidence. Virus like particles of 45 and 70 nm in diameter were found in partially purified preparation. Nine or more species of dsRNA were associated with cassava frogskin disease. Through cloning, it was found that these extractions contained ribosomal RNAs which were the dominant cDNA clones produced. The putative protein of one cDNA clone (S5) had homology with the P5 protein of rice ragged stunt virus (RRSV). The technique of reverse transcriptase AFLP was used to associate markers with CFSD plants. One AFLP product (clone S1) was consistently associated with affected plants and it was cloned and sequenced. This putative protein of this product has homology with the p 1 protein of RRSV. Northern blot analyses were made with dsRNA obtained from healthy and CFSD affected varieties. The probes were derived from the clones obtained previously. When hybridizing with the S5 probe, a positive reaction that corresponds to a dsRNA molecule of approximately 3000pb was found only in the CFSD affected materials. There was no reaction with the S1 probe. These different experiments lead to the conclusion that a virus in the family reoviridae is infecting cassava and is associated with cassava frogskin disease.

Gradual adaptation of biotype B of *Bemisia tabaci* (Gennadius) (Homoptera: Aleyrodidae) on Cassava (*Manihot esculenta* Crantz)

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The biotype B of *Bemisia tabaci*, a well-known pest on cassava (*M. esculenta*) in Asia and Africa, is associated with African Cassava Mosaic Disease (ACMD) that causes high economic losses in Africa. At present, there is no register of *B. tabaci* biotype B on cassava crops in the neotropics and it has been speculated that the absence of ACMD in the Neotropics may be related to the inability of *B. tabaci* to colonize cassava. It is also considered that its potential adaptation represents a threat for the production of cassava in the neotropics. With the purpose of verifying if biotype B has the potential capacity to adapt to cassava, experiments were carried out in the growth chamber (25±2 °C, 70±5 HR. 12 L:12 D). The development of *B. tabaci* population were evaluated on a legume (*Phaseolus vulgaris*), followed in two Euphorbiaceae, (*Euphorbia pulcherrima* and *Jatropha gossypiifolia*) and finally on cassava. The average longevity of the original populations on *P. vulgaris*, *E. pulcherrima* and *J. gossypiifolia* was of 3,1, 5,6 and 3,25 days, respectively; the highest rate oviposition (average 2,64 eggs/female/2days), the lowest development time (44,41 days) and the highest value of r_m (0,48 day^{-1}) were those of populations originating of *J. gossypiifolia*. It was verified that the 27.5% of the original population from *J. gossypiifolia* fed and reproduced on cassava. This study represents a successful attempt to adapt the B biotype of *B. tabaci* on cassava, verifying the possible influence of close phylogenetic hosts as gradual steps in the adaptive capacity of B-biotype on cassava.

Screening transgenics unveils apparent resistance to hornworm (*E. ello*) in the non-transgenic, African cassava clone 60444

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In Colombia like in most cassava-growing countries in Latin America, one of the major pests is the cassava hornworm. Yield and planting material losses can reach up to 64 and 72%, respectively, depending on the severity of the attack. Resistance is still elusive, although, more recently, the attention is being placed on the African cultivar 60444 (Nig11), which seems to be protected from attacks of the cassava hornworm. It was detected when transgenic plants of cassava, of the same cultivar, carrying one cry1Ab gene, were challenged with larvae of first instars. The variables being measured were daily weight increase and mortality. Cultivars 60444 (non transgenic) and CMC-40 were run as controls. The former is a very susceptible one and produced a mortality around 20%, while it was 83% for 60444. Most larvae fed with transgenic and non-transgenic lines derived from 60444 died by day five or six, and their average weigh did not exceed 0,5 g; none reached the pupa stage. Contrastingly, those fed on CMC-40 lasted until day twelve; reached their highest weight at day eleven (average beyond 3 g) and most became pupa. There was only one larva, fed on a transgenic line, whose weight was close to 2 g by day eight, did not gain extra weight afterwards and did not become pupa. These results indicate that cultivar 60444 may contain genes for resistance to the hornworm, an observation that needs to be confirmed in the field, in cassava growing areas where the pest is endemic, like in Tolima or the Caribbean Coast of Colombia. It is also worth noting that 60444 derives from crosses with the wild species *Manihot. Glaziovii*, a species know to be the source to resistance to ACMD as well.

Eco-physiological knowledge system of cassava: Deficit- and excess-moisture stress tolerance at whole plant level

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A large body of evidence on the anatomy, morphology and behavioral physiology regarding the water deficit and excess moisture stress tolerance of cassava (*Manihot esculenta Crantz*) in various agroecological zones of Nigeria has been collected in the last decade and a half. In the mean time much advances has taken place also in informatics and diagnostics and crop modeling tools that have not been efficiently applied to address the issue of mechanisms of tolerance in different stress ecologies. The available knowledge system used to identify known and relevant indicators of drought tolerance in cassava at the whole plant level is described in this paper. This information is expected to complement functional genomics knowledge of cassava to overcome yield reducing abiotic stress conditions in the field. It will also delineate major gaps in the knowledge base of whole plant physiology of cassava in relation to drought tolerance where future research efforts ought to concentrate. Work is ongoing to collate and strengthen the ecological knowledge system to test and validate performance in situ.

Potential characteristics of Nigerian landrace cultivars of cassava for crop improvement and biodiversity conservation

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The stay –green ability (SGA), photosynthetic efficiency (PE), camopy development (CD) , fresh root yield, dry matter, flowering , pest and disease status and recovery rate from past and diseases, of 10 newly collected landrace cultivars of cassava were assessed in the gene bank of the National Root Crops Research Institute, Umudike, Nigeria. Significant variations in SGA. PE, fresh root yield, dry matter, flowering, pest and disease recovery rates occurred among the cultivars . Cultivars Durungwo sustained the highest SGA (80%) followed by Aburuasua (65%), Nwocha and Dimugbo (60%). Photosynthetic efficiency was highest in Durungwo (.0.924) followed by Otupam (0. 876). The highest root yield was obtained by Durungwo (20.3t/ha) followed by otupam (18.6/ha) and Aburu asua. Dry matter yield followed the same trend. Three cultivars –Iwa Bende, Nwocha and Dan –warri flowered. The cultivars were moderately susceptible to major disease except for Durungwo the highest yielder exhibited the highest SGA, PE,DM, moderate resistance to disease and the interdependence of these phenomena is exploitatble for use and for rapid screening in the development of improved cassava lines and for bildiversity conservation.

Use of isolated protoplasts/cells in studies on cassava mosaic disease

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Cassava mosaic disease is a major constraint for production in many cassava growing countries. Distinct begomoviruses have been reported to cause mosaic diseases of varying intensities in different cassava growing areas. Environmental conditions and genotypical variations influence symptom severity. The cellular location of the infectious agents and sites of their accumulation are important in further understanding of the host-pathogen interrelationships. A number of commercial pectinolytic and cellulolytic enzymes were utilized to separate host tissue components of *Manihot esculenta* Crantz. cultivars popularly grown in India. Maceration of adjacent cells, cemented by pectin rich matrix and the cellulosic cell wall in healthy and mosaic symptomatic cassava leaves were attempted. Effects of enzyme concentration, nature of osmotica, pH and physical conditions and yield of isolated protoplasts/cells are investigated. Advantages of such preparations in virus detection tests involving electronmicroscopy and microscopy with fluorochromes like DAPI, Hoechst 33258 as well as responses in media specially formulated for protoplast cultures will be presented.

Investigations on antiviral plant extracts for management of cassava mosaic disease in India

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Cassava mosaic caused by Indian cassava mosaic virus (ICMV, *Begomovirus*, *Geminiviridae*) is a severe, widespread and economically important disease of cassava (*Manihot esculenta*) in India. An environment friendly integrated management approach is essential to manage the disease. Several plant extracts are known to have inhibitory effect on viruses. Some extracts are known to inhibit the virus directly while others are known to induce antiviral proteins in plants. In the present study 20 known antiviral plant extracts were screened against ICMV by inoculating tobacco (*Nicotiana benthamiana*) under glass house conditions. Aqueous leaf extracts (5 per cent) of *Mirabilis jalapa* and *Boerhavia diffusa* were found to inhibit ICMV infection on tobacco when applied 1 to 12 hour before inoculation. Mixing of these plant extracts with ICMV inoculum at 1:1 ratio was also found to inhibit the virus. Various methods and combinations of these extracts are being tested to utilize these extracts for the management of the disease. Use of these locally available plant extracts seems to be one of the best components of biologically based environment friendly integrated disease management system.

Toxicity of cassava based bioinsecticide on certain pests of stored products

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Large number of seeds is produced by certain popular varieties of cassava, but due to the lack of economic importance seeds are thrown as waste. An attempt was made to prepare biopesticide from cassava seed using polar and non-polar solvent extraction meted, and its insecticidal properties were evaluated against *Araecerus fasciculantus*, *Sitophilus oryzae* and *Lasioderma serricorne*, which are categorized as the major pest of many stored products including plain dried and parboiled cassava chips. Cotton ball (1 cm dia) socked with 0.5 ml of petroleum ether extract of cassava seed extract (CSE) at different concentrations (10,20,40 and 60%) was exposed to *S. Oryzae* and its fumigant actino was compared with control (Solvent alone). A positive correlation between the mortality of the weevils due to the treatment with 60% concentration was 30.6 76.0 and 100% respectively, on 1,3 and 7 days after treatment (DAT) as against 0.6 to 2.0% in the control. The extract in different quantities (0.1, 0.3 and 0.5 ml) transferred into cotton balls (1 cm dia) was exposed to batches of *S.oryzae* and thus the minimum quantity required to kill the insect was standadized. The mortality was 96% on 1 DAT in the treatment with 0.5 ml exposure, but it was only 1.3% in the control. Fumigant action was also noticed in the ethyl acetate and methanol extracts of cassava seed on all the test insects, however, extraction with petroleum ether extract at a dose of 5% showed that mortality on 1 DAT was 97.2% in *L. Serricorne,* 94.8% in *S. Oryzae* and 82.8% in *A. Fasciculatus* as against 9.2, 5.2 and 1.2% respectively in the control. Compared to the three test insects, *L. Serricorne* was highly susceptible to the extract. Treatment of *L. Serrocrne* with three doses (1,3,5%) showed high mortality (97.2%) on the 1 DAT in 5% concentration, and mortality was 89.2, 53.2 and 9.2% in the treatments with 3%, 1% and control respectively. This study reveals that cassava based biopesticide is a promising eco-friendly insecticide and potential alternative to synthetic insecticides.

Cassava leaf—A potential source of bioinsecticide against *Sitophilus oryzae* (L.) (*Curculionidae*: Coleoptera)

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Cassava, a staple and subsidiary food for millions of people, is extensively cultivated in tropical and subtropical countries. Large quantity of leaves, which is estimated to be 5 tones / hectare is often discarded as waste at the time of harvest. A preliminary investigation was carried out with leaves of important tuber crops viz. *Manihot esculenta, Ipomoea batatas, Solenostemon rotundifolius. Maranta arundinacea, Xanthosoma* sp., *Dioscorea esculenta ,D. alata, Amorphophallus paeniifolius, Colocasia esculenta, Pachyrrhizus erosus* to test the insecticidal properties. The leaves were distilled in aqueous medium and bioassay was done on *Sitophilus oryzae*, a major pest on stored cassava chips and other stored product, and proved that distillate from cassava leaves only caused high toxicity to the test insect. Screening with nine locally available cassava varieties proved that all the selected varieties contained toxic principles to kill *S. oryzae* and variety H226 showed maximum activity. Aqueous extracts collected from the leaves of upper, middle and lower parts of 10 month old cassava variety, H226 when tested against *S. oryzae*, high mortality was observed in all the extracts. When extracts collected from each part was fractionated separately into five fractions (10 mL each,) and tested on *S. oryzae*, there was cent percent mortality in the first two fractions of all the leaves collected from three parts. But leaves of upper portion showed further activity even with 3rd fraction. Mortality due to the application of 3rd fraction of upper leaves was 79.2%, whereas it was negligible in the other two fractions. The cassava leaf based biopesticide (CLBP) at different quantities (0.1, 0.25, 0.5, and 1.0 mL) were transferred into cotton balls taken in small plastic vials (1 cm long and 0.5 cm dia.) and exposed to batches of *S. oryzae* for the study of fumigant action. When fumigation was done with the CLBP in grain filled containers with *S.oryzae*, the infestation rate was found to be negligible.

Yield maximization in cassava (*Manihot esculenta* Crantz) through 'Systematic Approach' in fertilizer use

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Maximum Yield Research (MYR) and Maximum Economic Yield (MEY)systems are important under the present situations of increased demographic changes which have reduced the per capita availability of both land and food. Among the root and tubers , cassava is found in a variety of production systems and perform well under various levels of management from low input to high input systems. The 'Systematic Approach' involves the determination and elimination of soil nutrient constraints for balanced supply of all potentially deficient essential nutrients for sustainable high yield. In the 'Systematic Approach' of evolving fertilizer optima for a Typic Kandiustult soil, studies on critical level of nutrients , original nutritional status of the soil, its sorption capacity, and green house/screen house nutrient survey were carried out. Based on this concept the safe level of any one of the nutrients, is to ensure its maintenance in the available pool of the soil to a level which is neither deficient nor toxic to the crops grown and this corresponds to three times the critical level of the nutrient and the critical levels of P and Kwere arrived as 8.23 μg g^{-1} and 43.5 μg g^{-1}respectively. The above basic studies revealed N,P,K and Ca as the limiting nutrients for this particular soil and the optimum treatment for cassava was fixed as NPK @100:300:300 kg ha^{-1}. Field experiments were conducted for two seasons with different levels of the optimum treatment fixed in a short duration (6-7 months) cassava variety ' Sree Vijaya'.The optimum treatment gave an yield of 43.41 t ha^{-1}whereas its average yield under the existing package of practices recommendations is 20-24 t ha^{-1}. The quality parameters viz., starch and cyanogenic glucosides were also satisfactory. Economic analysis showed this optimum treatment as the best in terms of highest gross return (Rs 1,31,125/-),net return (Rs.83,150/-),added return (Rs.70,178/-),added profit (Rs. 56,142/-)and BCR (2.73).

Bemisia whiteflies cause physical damage and yield losses to cassava in Africa

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Cassava production in large parts of East and Central Africa has been devastated by a pandemic of cassava mosaic virus disease (CMD) from the late 1980s to the present day. A characteristic feature of this pandemic has been the co-occurrence of superabundant populations of *Bemisia tabaci*, the whitefly vector of the viruses causing CMD. In addition to their vectoring activity, *B. tabaci* populations are also now so great in some epidemic-affected areas that they are causing physical damage to the cassava crop. Reports of such physical damage have been made from Uganda, Tanzania, Rwanda, Burundi and both eastern and western Democratic Republic of Congo. Symptoms include mottled chlorosis on upper leaves, leaf deformation, sooty mould on lower leaves and general plant stunting. Experiments to investigate yield effects have shown that losses range from > 50% in the worst affected cultivars to almost nothing in the least affected. Of particular concern is the fact that some of the worst affected cultivars have recently been released on the basis of their high levels of resistance to CMD. However, the variability in response of the cultivars to *B. tabaci* populations does suggest that sources of resistance to whiteflies exist within cassava germplasm available in East Africa. Both host plant effects and *B. tabaci* biotypes have been proposed as possible reasons behind the increase in cassava whitefly populations. Conventional ecological as well as molecular study approaches are currently being used to evaluate these two hypotheses in order to identify whether either one or both are key determinants of *B. tabaci* population increase. This knowledge is considered to be an essential pre-requisite for the development of effective and sustainable management approaches for cassava whiteflies in East and Central Africa.

Preliminary evidence of correlation between foliar and root resistance to root rot caused by *Phytophthora tropicalis* in cassava

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The disease Phytophthora Root Rot affects cassava (*Manihot esculenta* Crantz) in different production areas and attacks all the plant's major organs, that is, roots, stems, and, sometimes, leaves. Farmers use several methods to control this economically significant disease, but ecological management by host-plant resistance is the preferred method. Root and leaf resistance to *Phytophthora tropicalis* was determined in 22 cassava clones, both during penetration of the pathogen through the peel (infection phase) and after infection in the parenchyma. Effective methodologies for preparing inoculum, inoculating, and evaluating resistance types were developed for both roots and leaves (lobules) obtained from field plants. We observed significant ($P < 0.05$) clonal differences for root and leaf resistance at the two stages of infection. The results indicate that *P. tropicalis* is able to infect unwound roots. A correlation of +0.28 ($r^2 = 0.08$) between resistance during (in the peel) and after infection (in the parenchyma) was observed for the roots, indicating that the association between these two types of resistance is low, and therefore should be determined separately. The correlation between leaf resistance (percentage of lesions 72 h after inoculation) and root resistance (percentage of lesions 10 days after inoculation) of the 22 clones was intermediately positive for the pathogen penetration ($r = +0.37$, $r^2 = 0.14$). The correlation between leaves and roots suggest that leaves can be used to predict the resistance to root rot in cassava populations.

Transmission of cassava brown streak virus by whiteflies

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Two viral diseases, the cassava mosaic disease and cassava brown streak disease (CBSD) affect cassava in eastern Africa. CBSD is most important in the coastal districts of Kenya, Tanzania, Mozambique and the lake shore area of Malawi. Typical CBSD symptoms include various patterns of foliar chlorosis especially on older leaves and purple brown lesions on green stems. Major yield losses (up to 70%) occur in the form of brown corky necrosis of tuberous roots, which makes them inedible. CBSD is a poorly studied disease and only recently shown to be caused by *Cassava brown streak virus* (CBSV), a member of the genus *Ipomovirus* of the family *Potyviridae*. The vector of CBSV was unknown, although whiteflies (*Bemisia tabaci* and/or *B. afer*) have been suspected candidates strongly because the spread of CBSD coincided with high whitefly populations in field conditions and some ipomoviruses are transmitted by whiteflies. Vector transmission of CBSV was carried out using *B. tabaci* and *B. afer*, and other insects such as beetles, thrips and spiralling whitefly. In addition, soil and seed transmission experiments were carried out by using soil from CBSD-affected and -unaffected cassava fields, and seeds from diseased and healthy plants. Plants inoculated using both *B. tabaci* and *B. afer* (~15-20 each, four inoculations) produced typical CBSV symptoms five weeks after inoculation under laboratory conditions. Infection of CBSV was confirmed by the reverse transcription polymerase chain reaction (RT-PCR) using virus-specific primers. This is the first conclusive evidence for the vector transmission of CBSV by whiteflies. Plants of soil and seed transmission experiments produced no symptoms after six months and CBSV was not detected by RT-PCR. Further studies are initiated, first, to understand if both the whiteflies species are required for CBSV transmission and to study virus-vector relationships. Results of these experiments will be discussed in relation to CBSD management.

Assessment of *Xanthomonas axonopodis* pv. *manihotis* genes expressed during plant infection

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Cassava bacterial blight (CBB) caused by *Xanthomonas axonopodis* pv. *manihotis* (*Xam*) is a major disease, endemic in Latin America and Africa. Recent analysis in our laboratory showed the high potential for *Xam* populations to change. The pathotypic composition of *Xam* evolved rapidly and may lead to overcome the deployed cassava resistance. It is therefore important to determine more accurately the molecular basis of pathogenicity of *Xam* by developing a large-scale analysis of expression profiling in *Xam*. A genomic library of *Xam* strain CIO46 was constructed and 3450 clones were amplified by PCR and printed on glass slides. A set of spot controls from tomato, potato and cassava housekeeping genes, human genes and spiked controls were also included in the *Xam* array. The resulting microarrays contained 16896 elements. Microarray hybridization was performed using RNA obtained from bacteria extracted from cassava stems, at different times after inoculation, and RNA from bacteria grown in culture media. This study allowed the identification of 93 differential expressed *Xam* clones. Among those 77 were up regulated and 16 were down regulated. The nucleotide sequence of 77 up regulated clones was analyzed and some clones showed homology to known genes involved in bacterial infection processes like: cell wall modification, type II site-specific deoxyribonuclease, putative ClpA/B-type proteinase, DAPA aminotransferase. Sequences related to permease activity, like ABC-2 and ABC transporters were also characterized. We also found genes of unknown function or showing no homology with sequences in the databases (41 clones). They may represent new genes putatively involved in the *Xam* infection process. Further sequences analyses are in progress. The differential expression analysis of selected *Xam* clones is being confirmed by quantitative real-time PCR. DNA microarray proved to be a useful tool that allows gene expression analysis even in non sequenced genomes such as *Xam*.

Adaptation of cassava genotypes to low soil fertility

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In India cassava is cultivated mainly in the southern states viz. Kerala, Tamil Nadu and Andhra Pradesh. Wide variation exists with in major cassava growing soil types as well as in soil fertility. In Kerala the crop is largely grown on ultisols (lateritic soils), alfisols (red soils) and entisols (alluvial soils and sandy loam). The former soils are very poor in organic matter, available N, P, K and generally deficient in Ca, Mg and Zn. In Tamil Nadu cassava is grown on vertisols (black soils) and alfisols (red loam). These soils are low in organic matter and available N but medium with respect to P and K and deficient in Zn, Cu and Fe. In Andhra Pradesh also the cassava growing soils (entisols and alfisols) are low in organic matter and plant nutrient elements. Due to low fertility of soils, large dosages of organic manure (12.5 t ha^{-1}) and fertilizers to provide NPK @ 100:50:100 kg $ha^{-1}$are recommended for cassava. Cassava, essentially being the crop of small and marginal farmers, very few have the financial resources to follow the above recommendation. So identification of cassava genotypes adapted to low soil fertility conditions and popularizing them is considered as an appropriate, eco-friendly and sustainable technology. With this objective field experiments were conducted at Central Tuber Crops Research Institute, Thiruvananthapuram to select cassava genotypes adapted to low soil fertility management. The genotypes included in the study were evaluated on the basis of fresh root yield, total biomass production, harvest index and P adaptation index. The results showed that the land race Mankuzhanthan was adapted to low soil fertility management. It was also found that the varieties Sree Prabha, Mankuzhanthan and Malayan-4 were capable of giving satisfactory root yields even in the absence of applied P . Monitoring VAMF association on cassava roots indicated that in the absence of applied P the fungal colonization was higher. Experiments at CTCRI also indicated that variation exists among cultivars in sensitivity to nutrient deficiencies. The variety Sree Visakham was found to be very sensitive to Mg deficiency, whereas the triploids were sensitive to Ca deficiency.

Host plant resistance to cassava green mite in Africa and its interaction with biological control

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The leaf feeding cassava green mite (CGM), *Mononychellus tanajoa* (Bondar), is a very serious pest of cassava in Africa. Host plant resistance and biological control with exotic phytoseiid predators were recommended in the 80s as major components of the integrated pest management strategy for the pest. Progress has been made with both control strategies, but the successful management of CGM seem to be a matter for the future. In the 90s, cultivars with moderate to high levels of resistance to CGM were developed at the International Institute of Tropical Agriculture, Ibadan, Nigeria. Three of such genotypes include TMS 920427, TMS 920342 and TMS 920342, which combine high levels of resistance and good storage root yields. The physical, physiological and chemical characteristics of cassava affect CGM. Denser and longer leaf trichomes, the retention of many green leaves during the dry season, and low levels of leaf nitrogen, potassium, phosphorus, tyrosine, phenylalanine and isoleucine as well as high levels of leaf calcium and fatty acids were positively correlated with the mite resistance. CGM aggregated on the top leaves of cassava at low levels of resistance as compared with a more even within plant distribution at high levels of resistance, suggesting that the phytoseiid predators may need a higher searching efficiency on resistant cassava genotypes. The negative interaction recorded between CGM and local phytoseiid predators was either weak or not detectable at higher levels of resistance. The promising exotic predator, *Typhlodromalus aripo*, which has spread to many countries in sub-Saharan Africa, showed better fitness on cassava with large pubescent apices, relative to the glabrous cultivars. For the successful management of CGM in Africa, interactions between host plant resistance and biological control should be either complementary or synergistic and not antagonistic. Entomologists, plant breeders and ecologists should collaborate with the aim of producing cassava cultivars with appreciable levels of resistance to CGM and able to encourage predator activity in Cassava-based ecosystems.

Host-plant resistance to African *Bemisia tabaci* in local landraces and improved cassava mosaic disease resistant genotypes in Uganda

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In the previous decade, a severe cassava mosaic virus disease (CMD) pandemic devastated cassava production in Uganda. The effects of the pandemic were mitigated effectively by the release and adoption of CMD-resistant cassava varieties. In the recent past, however, large *B. tabaci* populations have appeared on these varieties. Preliminary assessments have shown that this causes substantial yield loss. For instance, losses of about 40% were recorded in Nase 12 (Legg, unpublished data), a variety, which has gained wide-spread acceptability amongst farmers due to its good tuber quality and resistance to CMD. In response to this emerging threat, the National Cassava Programme of Uganda, in collaboration with NRI and IITA-ESARC, has embarked on a search for host-plant resistance to African *B. tabaci* and currently over 450 cassava clones are being evaluated. Furthermore, data on whitefly populations for over 280 local landraces have been reviewed. Initial results have revealed some promising genotypes, which show considerable *B. tabaci* resistance, amongst both the local landraces and the new introductions. We consider that identifying the cassava genes conferring resistance to African *B. tabaci* is now of great importance, given that this biotype now causes considerable direct damage. We consider that priority should also be given to developing hybrids with resistance to African *B. tabaci*, as the benefits will be considerable.

Tracking the spread of the 'Uganda variant' *East Africa cassava mosaic virus* – Uganda (EACMV-UG2) in East and Central Africa using nucleic acid-based diagnostic techniques

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Cassava mosaic virus disease (CMD) has been known on the African continent for more than a century although its impact on cassava production is currently greater than ever. In the 1980s/90s, an epidemic associated with a recombinant virus, *East Africa cassava mosaic virus* – Uganda (EACMV-UG2), designated the 'Uganda variant', was reported from central Uganda and has subsequently spread to neighboring countries. Urgent measures were required to address this regional threat, and a key component of the management programme developed was a regime of virus/disease monitoring and diagnostics. Through regular and extensive surveys, spread of the 'Uganda variant' has been monitored, allowing the targeting of control measures to affected areas and the identification of threatened zones. During surveys, fresh samples are collected from the top-most tender leaves showing CMD symptoms, and total DNA is extracted *in situ* using a portable kit. Polymerase chain reaction (PCR) is then performed in the IITA-Uganda laboratory using the universal primers (UNIF and UNIR). This is then followed by RFLP, which uses the endonucleases *Mlu*I and *Eco*RV to restrict purified DNA recovered from the PCR products. Based on the lengths of restriction fragments generated, it is possible to detect the presence of EACMV-UG2 and virus mixtures. It has been demonstrated that whilst there is a rapid spread of EACMV-UG2 from Uganda into northwestern Tanzania (>400 km in five years), there is much slower spread of the Uganda variant through western Kenya (<100 km in five years). New spread of the Uganda variant into north-eastern Burundi and eastern Gabon has also been demonstrated, with most EACMV-UG2 diagnoses associated with whitefly-borne infections. Vital information has been generated in this way, aiding the targeting of control measures and thereby reducing the impact of further spread of severe disease to important cassava-growing areas of sub-Saharan Africa.

Cassava macro- and micro-nutrient uptake and partitioning in alley cropping as influenced by *Glomus spp* in sub-humid tropics and its impact on productivity

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The beneficial effects of arbuscular mycorrhizae (AM) under alley cropping on the macro- and micro-nutrient uptake and their partitioning in cassava (*Manihot esculenta* Crantz) were investigated under field and potted conditions in two sites in the derived savanna belt of Nigeria. Cassava was grown either between multipurpose trees or as a sole crop. Multipurpose tree species *Leauceana leucocephala* and *Senna siamea* in alley farming system were used. *Glomus mosseae* and *G. fasciculatum* were used as the inoculum. The uptakes of N and P by cassava plants were improved in non-sterilized controlled experiments. The macro-nutrients investigated were significantly partitioned to the cassava shoot than the root. This partitioning of macro-nutrients was also found to be influenced by VAM inoculation. The VAM fungi introduced were able to enhance the uptakes of some of the trace elements (Fe, Zn, and Cu) in cassava both on the field and semi-controlled experiments. Fe and Cu were significantly partitioned to the roots while the tissue concentration of Zn was significantly higher largely in the shoots than the roots. Cassava macro-nutrition was not significantly improved by VAM fungi inoculation under field or potted conditions. It was also concluded that introduction of exotic VAM fungi can improve cassava micro-nutrient uptake under alley cropping where competition for nutrient uptake is strong. AM fungi has an overall beneficial effect on productivity of cassava under low fertility conditions.

Whitefly *bemisia tabaci*: Biotypes, indian cassava mosaic virus and its biocontrol agents

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Investigations were undertaken under USDA/USIF funded collaborative research at CTCRI with an objective of exploring potential natural enemies of *B. tabaci* and identification of different Biotypes in India. Four biotypes were identified *viz.* cassava biotype and sweet potato biotype (both from Trivandrum), Kannur biotype and Bhubaneswar / Parbhani biotype; based on host transfer and ovipositional studies, cross breeding, vectoring efficiency, isozyme and PCR-RAPD studies. ELISA, dot-blot immunoassay, PCR and EM studies confirmed role of cassava biotype in ICMV transmission. New nested primer and few sequencing primers were designed from the highly conserved region of AV1 coat protein gene of ICMV. Nested primer found to be a very good diagnostic tool for rapid and sensitive detection of ICMV in plants as well as in *B. tabaci.* Using these primers, AV1 coat protein gene of ICMV-Tri was sequenced and it showed similarity with ICMV-Tri, ICMV-Adivaram, ICMV-Maharashtra and Sri Lankan Cassava Mosaic Virus (SLCMV); and is distantly related to African cassava mosaic virus (ACMV). Phylogenetic relationship of ICMV with SLCMV, ACMV, South African cassava mosaic virus, East African cassava mosaic virus were clearly depicted. Nine aphelinid parasitoids, thirteen predators and four pathogens were identified as potential biocontrol agents. Molecular markers were used to amplify the D2 region of 28S RNA of nine aphelinid parasitoids and got them identified, in which few of then are reported for first time from India. Their detailed biology, percent parasitism and mass rearing and their use in biological control are embodied. Biology, feeding potential and mass rearing methods of different predators are documented. Infectivity and pathogenicity of potential pathogens are reported.

Influence of relative humidity and temperature on infestation of cassava chips by *Sitophilus oryzae* (L.)

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Sitophilus oryzae (L.) is one of the major pests of dry cassava chips. It is a major pest of rice and other cereals causing heavy economic loss. Influence of r.h. (40, 50, 60, 70 and 80%) on population growth and damage caused to cassava chips by *S. oryzae* was studied at low temperature (20^{o}C) and also at ambient temperature (26-30^{o}C). A parallel study was also conducted at ambient temperature (26-30^{o}C) and r.h. (68-90%). Fifty adult insects were released in glass jars containing 100 g cassava. Five replicates (4+1 control) per treatment were placed in air tight plastic containers containing 0.51 glycerine / water solution to maintain the respective r.h. Boxes were aired every three weeks and specific gravity of the solution checked to maintain the desired humidity. After 5 months, weight loss, dust formed and population of adult insects were recorded. Both low temperature (20^{o}C) and low r.h. (<50%) were found unfavourable for the insect to multiply and cause damage. Weight loss caused to cassava chips at 80% r.h. at 20^{o}C was only 7.0% while at the same r.h. at ambient temperature the value was 19.1%, weight loss caused at ambient temperature and r.h. was 27.5%. Adult progeny developed were also more at ambient conditions (261), the values were 238 for samples kept at 80% r.h. at ambient temperature and only 37 at 20^{o}C. Weight loss per cent increased from 0.5 to 13.7 with the increase in r.h. 40 to 70% at ambient temperature while the value ranged from 0.2 to 5.1 at the same r.h. at low temperature (20^{o}C).

Breeding cassava mosaic disease (CMD) resistant progenies under open pollination using MNga-1 line as the resistant source

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The propagation of cassava through true seeds (sexual seeds) rather than by clones is a promising option due to its manifold advantages like enhancing the multiplication rate, longer seed viability, ease of storage and transport. The high genetic heterogeneity and consequent variation among seedlings is the major stumbling block in sexual propagation. In the present study, a promising cultivar "Ambakadan" with profuse fruit setting, seed output and male sterility was identified. Nursery technique and agrotechnique were standardized for the establishment of a seed crop. Open pollinated and hybrid progenies of "Ambakadan" were evaluated for important economic traits. Tuber yield at first clonal stage (C_1) was significantly superior to that of the seedlings. By providing higher plant population at seedling and first clonal stage, yield levels could be enhanced. The dry matter content and starch output of seedlings and first clones were comparable to that of the commercial varieties. Similarly, the HCN and cooking quality of seedlings and first clones were at acceptable levels. In the open pollinated (OP) and hybrid progenies of "Ambakadan" the CMD infection increased drastically in subsequent generations due to secondary spread of the pathogen. In order to breed more homogeneous, high yielding, high starch sexual progenies having high level of mosaic resistance, hybridization of "Ambakadan" with fourteen male parents were undertaken. A CMD resistant line MNga-1 obtained from CIAT Colombia was used as one of the male parents. Evaluation of the hybrid progenies at seedling and first clonal stage along with clones of high yielding varieties as control indicated the superiority of the cross involving MNga-1 over the others in respect of CMD infection. A breeding strategy has been evolved to generate more homogeneous CMD resistant sexual progenies by raising a pollination block in isolation involving the male sterile 'Ambakadan' as the female parent and the MNga-1 as the male parent. Every five rows of female parent are interspersed with one row of the male parent. The OP seeds are collected from the female parent. The sexual progenies need to be rouged systematically at least for two consecutive generations before carrying forward rest of the CMD resistant population for cultivation.

Genetic structure and population dynamics of *Xanthomonas axonopodis* pv. *Manihotis* in Colombia from 1995-1999

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Restriction fragment length polymorphisms (RFLPs) were used to study the population genetics and temporal dynamics of the cassava bacterial pathogen, *Xanthomonas axonopodis* pv. *Manihotis* (*Xam*). The population dynamics were addressed by comparing samples collected from 1995 to 1999 from six locations, spanning four different edaphoclimatic zones (ECZs). Forty-five different *Xam* RFLP types or haplotypes were identified between 1995 and 1999. High genetic diversity of the *Xam* strains was evident within most of the fields sampled. In all but one site, diversity decreased over time tithin fields. Haplotype frequencies significantly differed over the years in all but one location. Studies of the rate of change of Xam population during the cropping cycle in two sites showed significant changes in the haplotype frequencies but not composition. However, variations in pathotype were described after 1997 in these ECZs, tus revealing the dramatic change in the pathogen population structure of *Xam*. Disease incidence was used to show the progress of cassava bacterial blight in Colombia during the 5-year period in different ecosystems. Low disease incidence values were correlated with low rainfall in 1997 in ECZ1.

Light-induced regulation of gene expression during tuberisation in cassava (*Manihot esculenta*)

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Several genes have been identified as important for tuberisation in sweet potato roots and solanum tubers; however, to date there have been no reports on the genes that are expressed in tuberising cassava roots. Furthermore, it is possible to manipulate planting conditions such that cassava stems can be induced to form stem tubers. Total RNA was isolated from tuberising and non-tuberising stems and from tuberising roots and using selected primers, RTPCR's were conducted to identify those genes which were down-regulated by light exposure and those that were expressed during tuber-inducing (dark) conditions. Genes which are expressed in both stem and root tubers and those which are specific for each organ could be identified. These data would have implications for increasing the yield of all tuber crops including cassava.

Pathogenicity of infectious clones of cassava mosaic viruses and potential for their use to screen cassava genotypes for disease resistance

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Cassava mosaic virus disease (CMD) is considered the most important constraint to cassava production throughout Africa. Use of resistant varieties is the most favoured management strategy, although it is greatly limited by lack of empirical data on the causal viruses and their pathogenicity in different cassava genotypes. In Uganda, different strains occur and have varying effects on the productivity of cassava. In order to relate pathogenic variation to the breeding strategy, we have isolated and established the current distribution of the causal viruses, produced infectious clones and determined the complete sequences of representative isolates. The infectious clones have been used to demonstrate pathogenic differences in single and mixed infections in the laboratory host *Nicotiana benthamiana* and cassava. The clones are currently being used to screen a range of cassava genotypes for resistance to CMD. This paper presents the progress and discusses the potential for developing and use of pathogenicity-based tactics in rational breeding for resistance to CMD.

Biochemical alterations in cassava genotypes differing in the leaf retention capacity during drought

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Cassava (*Manihot esculenta*) is generally considered as a hardy crop resistant to drought stress conditions. However, drought significantly reduces the yield as well as the starch content of storage root of cassava. Cassava genotypes show wide variations in their response to drought stress but the biochemical response of cassava to drought stress is little understood. This paper reports the biochemical alterations in the leaves of four cassava genotypes, CE- 165and CI-301 having high leaf retention and CE-326 and CE-283 having low leaf retention under drought stress conditions. Under drought stress, the contents of free proline, total free amino acids and soluble proteins markedly increased in the leaves of four cassava genotypes as compared to the plants under irrigated conditions. The increase in free proline, total free amino acids and soluble proteins was greater in the leaves of CE-165 and CI-301 as compared to CE-326 and CE-283. Free proline content increased by 300-400% in the leaves of CE-165 and CI-301 as compared to 50—100% increase in CE-326 and CE-283. Drought stress decreased the free sugar content by 50% in the leaves of CE-165 and CI-301 while it caused no appreciable change in the leaves of CE-326 and CE-283. The possible role of free proline and proteins in relation to drought tolerance in cassava is discussed. Further studies on other stress metabolites and protein profile are suggested to understand the biochemical mechanism of drought tolerance in cassava.

Cassava and geminiviruses

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Cassava mosaic disease (CMD) is caused by several cassava mosaic geminiviruses (*Geminiviridae*: *Begomovirus*) and it is now recognized as the most important constraint to the production of cassava in Africa, and a growing problem in India. The disease was first recorded in 1894, becoming widespread throughout Africa by the mid- 20h century. A total of 8 distinct cassava-infecting geminivirus species have now been identified, with more likely to be discovered in the coming years. In the 1990s an unprecedented cassava epidemic occurred in Uganda allowing the first geminivirus recombinant to be identified. Inter-species recombination is now recognized as a major driving force to create biodiversity in geminiviruses. This epidemic has spread throughout East and Central Africa, but it remains unclear as to what exact biological conditions are required to initiate and promote such a pandemic. Synergism between two species of geminiviruses, namely African cassava mosaic virus (ACMV) and East African cassava virus (EACMV), is certainly an important factor, but such dual infections have been found in West and South Africa without causing the same degree of devastation. Recently, we have discovered new types of geminivirus satellites in cassava from East Africa which are associated with severe CMD. These DNA molecules have the capacity to enhance symptoms and even break natural resistance in otherwise highly resistant landraces. For many years we have been working towards the development of transgenic cassava plants with resistance to a range of cassava-infecting geminiviruses. Here we report the production of transgenic plants with transgenically imparted resistant to three distinct geminivirus species and to the synergistic dual infection with ACMV and EAVMV. Evidence will be presented that the mechanism of protection within the transgenic plants is via a natural gene silencing mechanisms. Progress towards testing these plants in East Africa against local geminiviruses via whitefly inoculation will be described.

Effects of cassava mosaic disease on cyanogen accumulation and starch yield in cassava (*Manihot esculenta* Crantz)

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Cyanogens are compounds that release toxic hydrogen cyanide. Starch is the primary form of carbohydrate energy storage in plants. CMD is the viral disease of the cassava (*Manihot esculenta* Crantz). Preferred cyanogens and starch management is by breeding and processing of low and high cyanogens and starch respectively. Our present understanding of cyanogens and starch in cassava emphasizes their role in plant defense signaling mechanisms and storage energy respectively. However, from the nutritional point of view, cyanogens and starch are toxic and lethal and nutritionally respectively. As a corollary, these divergent aspects have translated into conflicting priorities on cyanogens and starch research. Nonetheless, this study aimed to contribute to the gap in present knowledge on the effects of Cassava Mosaic Disease (CMD) status and cultivar resistance on cyanogenic potential (CNp) and starch yield in cassava. High yielding improved varieties and local landraces have been collected, characterized and evaluated through breeding in Uganda. Field and greenhouse studies were used to analyse the CNp and starch accumulation patterns in BAO and Tongolo, the local highly CMD susceptible landraces, and Nase 3, a moderate resistant. A total of 400 and 36 cassava plants were studied, divided into scores 1, 2,3,4,5 tags from healthy to highly susceptible genotypes in the field and greenhouse, respectively. Tagging was by scoring the genotypes. Analysis revealed increasing CNp yield ($P<0.001$), but inversely correlated with starch content. CNp accumulation strongly depended on variety and CMD status singly or combined, and varied with crop age and vigour. CMD modulates changes in CNp yield but does not affect CNp accumulation profile. In future studies on implicated genetic link between CNp and CMD (stress) vigour x variety and CMD status, effects of temperature and nutrition on CNp and starch yield are needed.

Progress towards incorporation of resistance to cassava mosaic disease from exotic germplasm to cultivated varieties in India

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MNga-1, an exotic line received from CIAT, Cali, Colombia was found as resistant to the cassava mosaic disease prevalent in the cultivated varieties in India through continuous field testing carried out for 8 years followed by graft testing with CMD infected susceptible stalk plant. This line showed 25-30t/ha tuber yield and 25-27% starch content. This line is presently being tested in different locations in India for assessing its potential as a variety for direct utilisation in disease affected areas. Intervarietal crosses were carried out with six cultivated varieties and MNga-1as it had shown stability to the resistant character. Seeds of selectively pollinated plants (full sib progeny) and open pollinated parents (half sib progeny) were collected and used in raising the seedling progeny. 1419 seedling plants (475 full sib and 944 half sibs) were screened in a heavily infested field.704 plants were selected based on symptom expression ranging from mild to no symptom. Some of the selections showed good plant type and tuber characters. Higher number of symptom-free plants were obtained (79.8%) from half sib progeny of MNga-1.Variation was observed among the different crosses in the expression of the resistant character ranging from 31-56% symptom-free plants. The selections are further being evaluated in clonal progenies for screening and conformation of results. The results are presented and discussed.

Studying begomoviruses infections in susceptible and resistant cassava breeding lines

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Screening cassava for resistance to cassava mosaic disease (CMD) relies on natural infections with particular virus(es) and their whitefly vectors present at a given time and location. Inoculation with defined viruses at an early stage in breeding for resistance can present a major improvement to the resistance development in cassava. Partial multimers of DNA A and DNA B components of several begomoviruses were constructed and introduced into cassava by microprojectile bombardment. Cloned DNA of East African cassava mosaic begomovirus isolates from Kenya (EACMV-UG2-[Ka]) and EACMV-UG2-[Si]) reflecting recombinant EACMV-UG2 viruses, was only infectious to susceptible cassava genotypes, TME 117, 96/0304 and 96/1039, while biolistic inoculations with chimeric clones, DNA A from EACMV-[KE-Kwale] and DNA B from EACMV-UG2-[Si]) forming a pseudorecombinant virus, was also infectious to the highly resistant genotypes, TME 3 and TME 4. Symptom expression and virus replication were recorded in 2-4 leaves following the bombarded ones. However, virus infection was not sustained and plants recovered from infections. A quantification of virus by real time PCR revealed remarkable variations of virus concentrations in non symptomatic leaves of resistant breeding lines. However, despite an initial symptom expression phase and virus replication in resistant cassava, virus infections were subsequently aborted. In conclusion, 96/1089A, 96/0160, TME 3 and TME 4 proved highly resistant to all begomoviruses tested. The usefulness of the methods applied for studying virus resistance and the description of virus replication in resistant and susceptible breeding lines are discussed.

Increased African cassava mosaic virus resistance in transgenic cassava plants expressing different viral antisense RNAs

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African cassava mosaic virus (ACMV) is an essential component contributing to the epidemics of cassava mosaic disease, the most important and devastating diseases of cassava in Africa. We developed cassava plants with increased ACMV resistance using improved antisense-RNA technology. Three antisense-RNA constructs were designed for the expression of viral antisense-RNAs as 3'-untranslated regions (3'UTR) of a selectable hygromycin phosphotransferase gene, which is driven by the CaMV 35S promoter. The targets for antisense-RNA interference are the viral mRNAs for *Rep (AC1)*, *TrAP (AC2)* and *REn (AC3)*, which play key roles in ACMV replication and transcriptional regulation. DNA and RNA blot analyses confirmed the integration and stable expression of the three viral antisense-RNA genes in corresponding transgenic plant lines. Results from transient viral replication assays in isolated leaves confirmed that replication of two ACMV isolates was strongly reduced or inhibited in transgenic lines. Compared to wildtype plants, delayed symptom developments and attenuated symptoms were observed in transgenic plants upon ACMV-N*Og* infection under our greenhouse conditions. Significantly reduced viral DNA accumulation was detected in the infected leaves of transgenic plants. Potential mechanisms of viral antisenses on ACMV replication and accumulation, e.g. post transcriptional gene silencing and transgene methylation, were studied. Our results demonstrate the feasibility on ACMV resistance by expressing viral antisense-RNAs against viral mRNAs specifying essential nonstructural proteins.

Extension of cassava leaf life by autoregulatory inhibition of senescence

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Prolonging the life of cassava leaves could result in higher root yield, improved root quality, and more frequent leaf harvesting. Similar to strategies first explored in tobacco, we expect that expression of the isopentenyl transferase (ipt) gene from *Agrobacterium tumefaciens* under control of the senescence-induced *SAG12* promoter from *Arabidopsis* should lead to delayed cassava leaf senescence via an autoregulatory senescence inhibition system. We have transformed cassava plants with the ipt gene under control of the *SAG12* promoter. Seven transformed plants regenerated under paramomycin selection were confirmed for the insertion of the *SAG12-ipt* cassette by PCR and Southern analyses. RT-PCR analysis revealed the expression of *ipt* in mature but not young leaves of the transgenic plants. After dark-induced senescence treatment of mature leaves from both *in vitro* and greenhouse-grown plants, significant stay-greenness and repressed chlorophyll degradation were observed in the transgenic line 529-28 compared to wildtype. The line also displayed resistant to leaf senescence after drought treatment. Only 10% leaves of 529-28 become senescent in comparison with 50% of wildtype and 20% of line 529-48 from 3 month old plants. The expression of *ipt* was increased in the old leaves of drought-treated 529-28 lines. During the development of transgenic plants, the decrease in chlorophyll, total protein, and Rubisco content in mature leaves was repressed. Interestingly, the transgenic plants also showed an early storage root bulking in comparison with wildtype plants. Evaluation of the yield of leaf and storage root will be carried out under greenhouse conditions. The “stay-green” cassava provides a new germplasm for subsistence farmers to allow frequent leaf harvesting with improved root production.

PS 5: Post Harverst and Starch Quality

Identifying the full set of genes involved in cassava post-harvest physiological deterioration

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Cassava roots suffer from a rapid post-harvest physiological deterioration (PPD) that can render the roots inedible and unmarketable within 24-72 hours of harvest. Increased urbanization and the changing nature of cassava use from a locally consumed crop to a commodity, has lengthened the distance and time between the farmers' fields, markets and processors, thereby incurring losses, wastage and discounting of poor quality cassava. PPD is thus a major constraint to the development of cassava for farmers, processors and consumers alike, and the successful application of strategic research is necessary to understand and solve this problem. With a view to fully understanding PPD and to ultimately producing the tools to control this problem, we have identified some of the key genes that play major roles during PPD and mapped these on the molecular genetic map of cassava. Recently, we have embarked on a programme employing massively parallel methods of gene analysis (cDNA microarrays) to identify the full set of genes involved in PPD. Screening over 11,000 cDNA clones from early and late PPD-related libraries using a series of labelled probes from a range of time points during the time course of deterioration has led to the identification of genes whose expression is either significantly increased or decreased during PPD. The putative identity and function of many of these genes has been established.

Native starch quality of Malawian cassava genotypes in different environments

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The starchy tuberous roots of cassava (*Manihot esculenta* Crantz) provide more than half of the calories consumed by more than 800 million people in the Sub-Saharan Africa. Cassava is a staple for more than 30% of the Malawi population, while in the rest of the country it is grown for food security, as a snack, and a cash crop. Industries that use starch in Malawi have not been willing to use cassava starch because the powder sold by some suppliers as 'cassava starch' was inferior. Trials with 20 cassava genotypes were planted at Chitedze and Makoka in Malawi in November 2000, and were harvested in December 2001. The quality parameters measured included protein, moisture and ash content, pH and whiteness. The results show that all the cassava genotypes produced starch containing no detectable protein, similar to the starch used in the pharmaceutical industry. The moisture content ranged from 11.85 to 13.65%, which is lower than the recommended maximum of 14%. The recommended maximum of 0.5% for ash was much higher than the values of cassava starch, which ranged from 0.10 to 0.20%. The recommended pH for starch is between 4.5 and 7.0, and cassava starch was within these limits, ranging from 5.0 to 5.9. The cassava starch was as white as the corn starch currently being used. Trials have shown that cassava starch can successfully be used in the making of tablets, batteries, packaging material and textile manufacture. Thus, native cassava starch is suitable for use in various industries. This study has also dispelled fears that cassava starch is of low quality and that it is not effective in some industries. Hence, the ideal quality of starch was confirmed in this study.

Physicochemical characteristics of starch of commercial cassava varieties grown in Thailand

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Cassava starch samples extracted from the roots of several commercial varieties grown in Thailand, i.e. Rayong 5, Rayong 90, Kasetsart 50 and Rayong 72, were evalulated in terms of their physicochemical properties using a Brabender Amylograph (BU). These roots were obtained from cassava planted both in the early and late rainy season, on Houypong soil series in Rayong Field Crops Research Center, and harvested at monthly intervals from 6-24 months after planting. The analyses indicate that the starch samples (at 5% starch) had pasting temperatures, peak viscosity, final viscosity, setback and breakdown ranging from 65.1 to 67.8 ^{0}C , 345 to 505 BU, 264 to 350 BU, 109 to 180 BU, and 169 to 296 BU, respectively. Cassava starch obtained by planting on different soil series in Nakhon Ratchasima Farm had the highest starch viscosity on Chokchai soil series followed by Stuk , Warin, Sikhiu, Yasothon, Surin, and Korat series, respectively.

Biochemical composition in cassava root and physico-chemical properties of starch

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Six commercial cassava varieties of Thailand were evaluated for biochemical composition and physio-chemical properties of their tapioca starch under 7 different soil series, Satuk Walin Korat Chokchai Surin Yasotorn and Srikewt, of Nakhornrachasima province, a major cassava planting area. Split plot in randomized complete block design was employed with the main plot consisted of 6 cassava varieties and the sub-plot were 4 harvesting times, 6, 8, 10 and 12 months. Amount and size of amylose were determined by the High Performance Size Exclusion Chromatography (HPSEC) technique. Biochemical compositions of starch from 6 cassava varieties (when planting in early rainy season and harvesting at 4 different harvesting times) showed that fatty content have ranging from 0.095-0.329%, phenolic ranging from 1.507-2.816%, HCN ranging from 4.0-378 ppm, fiber have ranging from 1.234-2.325%, amylose have ranging from 17.3-23.5%, and amylose size have ranging from 2042-3762 DPn. However, planting in late rainy season found that fatty content have ranging from 0.065-0.294%, phenolic ranging from 0.0827-0.1490%, HCN ranging from 70-217 ppm, fiber have ranging from 1.573-3.492%, amylose 17.1-22.6%, and amylose size have ranging from 2763-4420 DPn. Physio-chemical properties from their flour, when planting in early rainy season informed that peak viscosity have ranging from 381-451 RVA, trough viscosity have ranging from 114-169 RVA, final viscosity have ranging from 193-254 RVA, breakdown have ranging from 235-312 BU, and setback have ranging from 81-119 BU. In addition, planting in late rainy season showed that peak viscosity have ranging from 317-369 RVA, trough viscosity have ranging from 116-154 RVA, final viscosity have ranging from 183-260 RVA, breakdown have ranging from 167-233 BU, and setback have ranging from 64-108 BU. Finally, the results indicated that Rayong 5, when planting in the early rainy season and harvesting at 8 months after planting, was the best variety for cassava starch industry, due to low hydrocyanic acid, phenolic acid, fat, protein and fiber content. All soil series provided good starch quality, however, planting Rayong 5 in Chokchai soil gave the best cassava starch viscosity.

On the lactic acid fermentation of cassava

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The perishable nature and bulk of cassava left it aside in the rat race of urbanisation and presently in globalisation restricting its use in the production centres. The overall trend in developing countries is toward consumption of processed food products. In this background the involvement of lactic acid fermentation as an efficient means of post harvest processing in cassava is discussed. The latin American technology takes 30 to 40 days to offer puffing-up trait to fermented cassava starch. The low cost biotechnique developed in India (*Govt. of India Patent No.187378*) to extract cassava flour having puffing-up qualities takes only 48h. and as such will have an edge over the artisanal processes. The feasibility of adopting the Indian technique to global arena in the current realm of Intellectual Property Rights is worthy of discussion and prove as a tool for adding value to a small farmers crop.

Use of modulated DSC for rapid and accurate determination of amylose content in different starches

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Recently there has been a concerted effort to breed low- and high amylose cassava starches using genetic modification. So it is imperative that there should be methods to determine the amylose content in starches rapidly and accurately. The most common procedure used is iodimetric determination, while other techniques used include potentimetric titration and Gel Permeation chromatography. But these methods are either not sensitive or are very slow. Differentail Scanning Calorimetry is being increasingly used to estimate the amylose content in starches. The method is based on the enthalpy of gelatinisation of amylose-surfactant complex and is rapid and very accuarate. The conventional DSC has been found to be not very accurate when the amylose content is very less. In addition, the procedure requires use of lysolecethin which is a very costly chemical. We have standardised a method using Modulated DSC in which the starch slurry is subjected to repeated cycles of heating and cooling which will be useful in formation of amylose-surfactant complex. Cetyl trimethyl ammonium bromide a very common surfactant was used to complex with starch. The heating cycle used was 30-120°C at 3°C /min., cooling to 30°C at 30°C/min, reheating to 100 °C at 3°C /min and fast cooling. The ΔH(Gelatinisation Enthalpy) was calculated from the graph and by comparison with the ΔH value for standard potato amylose, the amylose content in the starch sample was calculated. Various cereal, tuber, natural high amylose and genetically modified potato starches were tried and stastical analysis of the results showed that the method can be used for rapid and reliable determination of amylose content in starches even in low-amylose starches. The significance of modulated DSC and advantages of the methods are discussed.

Use of microwave heating in physical, chemical and enzymatic modification of cassva starch

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Modification of cassava starch for imparting specific characteristics for special applications is becoming more and more common to meet the demand of consumers for novel and safer products. The modifications carried out include physical, chemical and biotechnological processes. Heat-moisture treatment, steam pressure treatment and radiation constitute physical methods, oxidation, esterification, etherification and crosslinking are chemical processes, while treatment with enzymes form biotechnological procedures used for modification of starches. Use of microwave heating has recently assumed lot of interest in view of shorter reaction times and cleaner products. Microwave energy is more effective since it acts by dielectric polarisation of molecules and hence accelearates organic reactions. Reports on use of this technology in modification of starch are scanty and hence attempts were made to produce modified cassava starch using this technique. The three processes tried were 1. physical modification: production of pregelatinised starch, 2. chemical modification: starch phosphate and 3. enzymatic modification.: amylase thinning of starch. For pregelatinised starch, 50g starch slurried in 100ml water was subjected to microwave heating in two stages and the product was obtained within 40 minutes which on powdering yielded white granular powder having cohesiveness and required solubility in cold water suitable in textile applications. Starch phosphate was obtained in good yield (90%) and quality (high viscosity and solubility) by heating starch-phosphate-urea mixture (100:5:2) for 45 minutes at controlled power (compared to conventional method which requires a heating time of 150 minutes at 145-165 degrees Celsius). For enzymatic treatment, 2 g starch suspended in 10 ml water was treated with 2.4 mg Termamyl at low power for 1, 2 and 5 minutes. The progress of thinning was monitored by reducing value determination. It was found that the reducing value increased from 1.5 to 26.5 in 1 minute while total gelatinisation occured within 5 minutes. Details of the processes and product properties are presented in the paper and the results clearly show that microwave heating can bring about starch modification much faster.

Development of waxy starch cassava varieties via the anti-sense mediated silencing of the granule bound starch synthase I (GBSS I)

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Higher incomes from cassava in the developing world where the crop is generally found will require the industrialization of the crop and the development of novel industrial products from cassava. There are several novel products that can be produced from cassava, including modified starches such as 100% amylopectin or 100% amylose starches, from the down regulation of the granule-bound starch synthethase (GBSS) gene or the starch-branching enzyme (SBE) gene. Industrial application of either pure amylopectin or pure amylose starches, such as the production of high-value biodegradable polymers from pure amylose starches or the use of 100% amylopectin in thickeners, pastes and glues, have a market with unlimited growth potential. With funds from the Colombian Ministry or Agriculture and Rural Development, a project was been initiated to genetically engineer industrial cassava varieties for the production of waxy starch using an anti-sense and sense construct of the GBSSI gene. Three constructs were made for genetic transformation. They include the full length GBSSI gene in the sense and anti-sense orientation in the binary vector pCAMBIA 1305.2, and the gene in the anti-sense orientation in the binary vector pBIG101. Friable Embryogenic Callus (FECS) of the cassava genotype TMS60444, MCol2215 and CM 3306-4 were transformed via *Agrobacterium tumefaciens* with the three constructs. Results of GUS transitory assay revealed a successful incorporation of the gene. Two lines from transformed FECs of the cassava genotype TMS60444 were successfully regenerated and 4 plants transferred to the screen house. Preliminary gus assays of leaf and stem revealed successful expression of the construct and a possible endogenous silencing. Transformation events from the other genotypes are being regenerated. Confirmation of the waxy phenotype awaits biochemical tests on roots of the transgenic plants.

Physico-chemical properties of root, flour and starch of bitter and sweet cassava varieties

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Cassava (*Manihot esculenta* Crantz) is typically classified by high and low cyanide content present in fresh roots as "bitter" and "sweet" type, respectively. Rather the cyanide content, this study aimed to evaluate physico-chemical properties of root, flour and starch between bitter and sweet types. Two commercially utilized varieties of the bitter type including Rayong 5 and Kasetsart 50 were planted in the same identical field as three varieties of the sweet type including Rayong 2, Hanatee and MKUL 36-Y002. Only the cyanide contents in root could be used to identify cassava into two groups as the high cyanide type (Rayong 5 and Kasetsart 50) and the low cyanide type (Rayong 2, Hanatee and MKUL 36-Y002). The starch content in roots of bitter cassava (83 to 84% dry basis) was slightly higher than the sweet one (79 to 82% dry basis). The chemical composition of flour and starch from bitter and sweet types was similar except that the fiber and ash content in sweet cassava flour were higher than the bitter one. The viscosity of flour and starch were depending on variety and did not relate to bitter-sweet type. The other properties of extracted starch including the amylose content (17 to 18%) and size ($DP_n \approx 2100$), paste clarity (64% Light transmittance of 1% solution), gel texture (Young Modulus of elasticity = 24 to 29 k.Pa), were not significantly ($p>0.5$) different.

Strategies for effective cassava post harvest system in Nigeria

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As part of efforts by all stakeholders to launch Nigeria into the world cassava trade, a National Survey was instituted by IITA to assess cassava sub sector as one of the components to Cassava Mosaic Disease (CMD) project. The main thrust of the survey was to provide comprehensive information on production, processing, marketing and distribution of the cassava chain. This paper presents an overview of the post harvest section of the cassava business. The survey indicates that quite a lot has been done in the area of traditional processing of cassava gari in South West, South East and South South of Nigeria. Starch and fufu are localized across regions with some few innovations at pilot level. There was little or no presence of industrial products like chips, pellets, ethanol etc. The paper highlights various constraints such as high cost of cassava production, lack of appropriate technology for processing cassava into high quality products, inadequate marketing infrastructure, lack of market information and inconsistent government policies. Pragmatic work is, however, required in products that will add value to cassava business such as processing of cassava into Chips, Starch and Pellets. Various Strategies of upgrading the current traditional processing methods and achieving high value products from cassava were discussed.

Fermentation in cassava, a tool in ensuring food security and safety in tropical African region

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The African continent has suffered a great deal from food security and safety problems for a long time. Natural climatic changes such as drought, economic state and limited assess to modern processing technology through information dissemination has been a major factor promoting this state. Many unique qualities has been identified in cassava plant such as high carbohydrate source and survival of varieties over several harsh conditions that makes it of good potential as a food crop. Cassava is a major crop of the tropical African communities. It has been a significant factor contributing to food security in the region. In this studies the role of fermentation at ensuring food security and safety, major fermentation techniques in use at the region for some cassava products and current trends aimed at improving product quality and presentation will be discussed. Underlining microbial potentials capable of conferring safety on fermented products will be discussed as well as the potential of starter cultures in solving processing and nutritional problems. In particular, traditional experiences of the local entrepreneurs and relevant academic institutions will be used to stress major points of the discussion.

Added value of amylopectin cassava starch

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Starch is the major storage carbohydrate in plants and one of the most important plant raw materials for food and industrial applications. Approximately 60% of the starch production is used for food purposes, whereas 40% is used as starch for paper and industrial specialties. The ratio of amylose to amylopectin has a great influence on the physicochemical properties of the starch. In most crops, starch contains between 20-30% amylose and 70-80% amylopectin. A major drawback of the use of this mixture of polymers in many commercial uses is its extreme high viscosity (thickness of the solution) when heated and its tendency to "retrograde" (recrystallization of amylose) at ambient or low temperatures. Retrogradation is a problem in many applications because the resulting gel becomes firm and tough and may become brittle as it loses moisture. To avoid problems with retrogradation, starch is subjected to common chemical treatments. To avoid the use of these environmentally unfriendly chemicals and to reduce production costs it is for many applications desirable to have a pure or enriched fraction of either amylopectin or amylose. Crops producing so-called amylose-free or high -amylopectin starches have been generated. For most cereals such as maize, sorghum, barley, rice and wheat amylose-free mutants have been generated. Via genetic modification (suppression of the Granule-bound Starch Synthase gene) an amylopectin potato and recently an amylopectin cassava was developed. Total production of the amylopectin starches, mainly waxy maize starch, is at this moment roughly 500,000 tonnes per year. Based on the final properties of the starches, waxy maize, amylopectin potato or cassava starch each have their benefits for different applications and market segments. Waxy maize for example has particular benefits in dairy products and sauces because of its short, smooth and shiny texture. On the other hand, amylopectin potato starch is particularly preferred in the paper industry because of its low protein and lipid content. Amylopectin cassava starch with its small granule size and low protein and lipid content could have a significant impact in many market segments.

Production and properties of amylose-free cassava starch

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We previously showed that by using particle bombardment we were able to obtain amylase-free cassava plants in the genotype TMS 60444. In a large scale greenhouse experiment over 1000 plants were set up for the production of starch. Sufficient starch was isolated for different analyses. In this presentation data will be presented on the biochemical, physico-chemical as well as rheological properties of this starch and it will be compared with amylose-containing starch of cassava.

PS 6: Gene Discovery

Recent advances in the identification of genes conveying whitefly (*Aleurotrachelus socialis* Bondar; Homoptera: *Aleyrodidae*) in cassava (*Manihot esculenta* Crantz)

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A complex of whitefly species cause considerable yield loss to the cassava crop in Latin America, Africa and Asia as direct feeding pests and virus vectors. The most economically important species in northern South America on cassava is *Aleurotrachelus socialis*. The combination of direct feeding damage and lower photosynthetic rate reduces yields as much as 79% when prolonged attacks occur. The cassava clone MEcu 72 has consistently expressed the highest levels of resistance to *A. socialis*. *A. socialis* feeding on this resistant clone had less oviposition, longer development periods, reduced size and higher mortality than those feeding on a susceptible clone. Owing to the importance of this pest, it is important to understand and characterize the genetics of resistance expressed in the MEcu 72 genotype; therefore a cross was made between MEcu 72 (Resistant) x MCol 2246 (Susceptible). Segregation of the F1 population of this cross was evaluated using molecular markers such as micro satellites and AFLPs. The use of micro satellite markers showed an association between molecular markers and resistance. A linkage map was constructed using a micro satellite data, an RGA and field phenotypic characterization. This will accelerate the selection of whitefly resistant germplasm and the identification of resistant genes.

A catalogue of 6000 expressed genes in cassava: Identification of genes implicated in cassava bacterial blight resistance and starch biosynthesis

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Cassava is a major staple food for more than 500 million people worldwide. Roots serve as a low cost carbohydrate source for human nutrition. Cassava bacterial blight is a major disease causing important yield losses. A number of tools are yet available for cassava functional genomics (genetic map, BAC librairies) together with transformation system. Expressed Sequence Tag (EST) provides an immediate and productive method of gene discovery. Fourteen cDNA libraries were obtained from root tissues and from stem tissues challenged with *Xanthomonas axonopodis* pv. *manihotis*. A random sequence analysis of cDNA was performed in order to identify potentially informative genes. A total of 14168 single-pass sequences were obtained from 5'ends of cDNA. Cluster analysis using Stack-pack system permitted the identification of 6046 unigene comprising 2032 contigs and 4014 singletons. About 18% of the cassava EST sequences show no similarity to previously described sequences in public databases and therefore can be considered as cassava specific genes. Sequence analyses permitted the assignment of a putative functional category for 32% of sequences on the basis of the gene ontology classification scheme. A group of genes belonging to a large multigene family was identified. We characterize a set of genes detected only in infected libraries putatively involved in the defense response to pathogen infection. By comparing two libraries obtained from cultivars contrasting in their starch content, a group of genes associated to starch biosynthesis and differentially expressed was identified. This is the first large cassava EST resource developed today and publicly available thus making a significant contribution to genomic knowledge of cassava.

Identification of resistance-gene analogs in cassava (*Manihot esculenta* Crantz), and their relationship to three *Phytophthora* species

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In an attempt to identify homologies between cassava genes and disease resistance gene analogs from different species, three species of *Phytophthora*, causal agent of cassava root rot, were evaluated in two families, for resistance. QTLs associated to resistance to *Phytophthora tropicalis, P. melonis* y *P. palmivora* were identified, based upon phenotypic and genotypic data from the cassava map. Two strategies were used to find resistance genes in cassava. The first approach was hybridizing heterologous probes from maize and rice, labeled with ^{32}P[dATP] to total cassava DNA, digested with six restriction enzymes (RFLP). The second strategy was amplifying conserved regions of DNA, using PCR with degenerated NBS and Pto kinase primers, in three cassava genotypes resistant to *Phytophthora tropicalis* and *P. palmivora*. Clones obtained were sequenced and compared with known resistance genes. Specific primers were designed from the sequences, allowing DNA regions of parental material and resistant and susceptible individuals, to be amplified. Bands were separated by denaturing polyacrylamide gel electrophoresis, and non-denaturing polyacrylamide gel. Two QTLs associated to *P. palmivora*, two associated to *P. melonis* and one associated to *P. tropicalis* resistance, were identified, which explain between 7.3% and 8.3% of phenotypic variance. By hybridization was concluded that cassava has a very low homology with the genes of the monocotyledons tested. A total of 28 NBS and 2 Pto kinase clones were obtained by PCR; of these, five showed homologous sequence with NBS-LRR resistance gene analogs (RGAs). Four of them showed open reading frames with conserved motifs of the NBS region, which means they were considered as RGAs. Three different RGAs classes were identified, which did not show association with resistance to *Phytophthora*.

Cloning carotene synthesis genes from cassava roots

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Our research seeks to increase the knowledge about genes that code for specific steps in the cassava's carotene metabolism to understand its regulation in roots. To achieve this goal, combinations of consensus primers were generated to PCR-amplify orthologous sequences of *Phytoene synthas*e and *Phytoene desaturas*e genes of the carotene pathway in cassava. Genomic DNA from leaves, and cDNA produced from fresh roots were used for PCR amplifications. The cultivars chosen for the study were CM 523-7, whit low root carotene content, and MPer 297 whit high root carotene. The consensus primers allowed the amplification of orthologous fragments, both in genomic DNA and cDNA with high homology, as compared to known clones, for *Phytoene synthase* and *Phytoene desaturas*e. Possible exonic and intronic sequences, as well as conserved domains belonging to families of these genes were identified and characterized. The next step comprises the cloning of genomic sequences to further characterize promoter and regulating sequences in high an low carotene content cultivars.

PS 7: Tissue Culture and Tranformation

Introduction of inbreeding in cassava through the production of doubled haploids

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Inbreeding in cassava has seldom been pursued. Several factors, particularly the time required to obtain high level of inbreeding (9-10 years), prevented this practice to be routinely used in the genetic improvement of this crop. Inbreeding in cassava is desirable because it: a) reduces genetic load; b) allows for the identification of useful recessive alleles (particularly relevant, those for starch quality traits); c) greatly facilitates traditional genetic and molecular studies; d) allows for the exchange and storage of germplasm using botanical seed; e) facilitates mutation breeding approaches; f) allows a more efficient exploitation of heterosis; and g) ultimately could allow for a more consistent, predictable and sustainable genetic improvement of the crop. A new initiative has been launched to introduce inbreeding in cassava genetic improvement. A tissue culture protocol for the production of doubled haploids (from cassava flowers) will reduce the time required for the production of homozygous lines from nine to, perhaps, 1-2 years. While the tissue culture protocol is being developed, elite cassava clones will be self-pollinated for two consecutive generations (to reach an average of 75% homozygosity). These partially inbred materials will be screened in search of previously undetected, useful recessive traits. If inbreeding depression is not a limiting factor, a third self-pollination will be made, or else lines derived from the same elite clone will be recombined to reconstitute a “full vigor” composite version of these elite clones (thus completing an S2 recurrent selection cycle for tolerance to inbreeding depression). Partially inbred germplasm has been planted in the cassava breeding nurseries at CIAT experimental station and could be visited by participants of the meeting.

Testing Brazilian cassava cultivars for FEC production and transformation capacity

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The economic importance of cassava in Northeast Brazil is based on its use for human consumption. Its importance also depends on its characteristics as a substitute for corn and wheat flour in poultry and baking industry. Genetic transformation is a reality for cassava and promises to speed up the improvement of the crop. However, the transformation technology is limited to few cultivars, due to genotype differences. It is then imperative to select cultivars from Northeast Brazil that are more suitable for genetic transformation, by testing their regeneration capacity in vitro after transformation. To accomplish this objective, CBN funded a small project to bring transformable tissues, mostly somatic embryos, from eight farmer-preferred cultivars from Brazil (Água Morna-BGM365, Amansa Burro-BGM549, Aparecida-BGM123, Bujá Preta-BGM1467, Milagrosa-BGM004, Rosa-BGM260, Rosinha-BGM394 e Tapicina-BGM1063), and test them in CIAT for transformation with the best *Agrobacteirum*-plasmid combinations available in the latter. Some embryogenic tissues were also induced to produce Friable Embryogenic Callus (FEC), the most commonly used cell system to transfer genes into cassava. Preliminary results showed that one cultivar produced FEC, and few plants have been regenerated from somatic embryos subject to infection with *Agrobacterium*. Molecular confirmation of transformation is awaited.

Update on cassava genetic transformation at CIAT

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Among the plasmid/*Agrobacterium*-strain combinations more recently tested are LBA4404-pGV1040 for herbicide tolerance, Agl1-pCAMBIA1305.2, Agl1-pCAMBIA1305.2 with GBSSI in sense and antisense orientation, and C58C1-pBIGCry for insect resistance. The cultivars transformed up to date are Per183, Nig11 (60444), Col2215 and CM3306, with an efficiency of transformation no greater than 7% (scored by the number of transgenic FEC lines that produce plants). In the case of insect resistance, bioassays were run in biosafety greenhouses to control the cassava hornworm (*Erinnyis ello*). Although the results were promising, they were preliminary and require further validation. A new source of resistance to the stem borer seems to have been unveiled during the testing of non-transgenic plants of cultivar 60444. Obtaining waxy cassava is another goal in our research program, and the recent achievements are summarized in another abstract. Transformation efficiency was recently improved by reducing coculture temperature and by bringing bacteria and cells in closer contact. This treatment produces roughly 50% transgenic FEC lines with cultivar 60444. Work is underway to improve the efficiency with more cultivars. New cultivars from North-East Brazil and Colombia are being tested in an attempt to shorten the protocol and reduce somaclonal variation. Also recently we started searching for cassava root-specific promoters, in a collaborative work with the University of Freiburg in Germany, to introduce genes that enhance Vitamin-A content in roots. We are also negotiating the Positech system to select transgenic tissues with mannose, for which we have established growth curves on mannose for cultivars 60444 and Col2215.Field-testing will be done for the first time this year. The permit to evaluate transgenics in the field is currently being analyzed by the Colombian Biosafety Technical Committee. An answer is expected quickly.

Effect of sucrose with or without abscissic acid on post-thaw viability, embryogenic competence and plant recovery from embryogenic calli of cassava

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The effect of sucrose with or without ABA in cryoprotective medium on post-thaw viability and/or embryogenic competence and subsequent plant recovery of embryogenic callus clumps of cassava was investigated. Post-thaw viability depended on duration of cryoprotection on the 0.3M sucrose cryoprotective medium. The presence of only ABA in the cryoprotective medium resulted in callus proliferation but loss of post-thaw viability. However, the inclusion of sucrose with low concentrations of ABA (5-10 mg/l) in the cryoprotective medium decreased callus proliferation; subsequent cryopreservation resulted in post-thaw viability and/or embryogenic competence, except at the higher concentration of ABA (20mg/l) where there was complete loss of post-thaw viability. Also, callus clumps pre-treated with low concentrations of ABA with sucrose prior to cryopreservation resulted in significantly higher plant recovery than only sucrose pre-treated callus clumps. Plants recovered from cryopreserved callus clumps were phenotypically similar to non-cryopreserved callus clumps. Cryopreservation of embryogenic calli of cassava has potential for long-term conservation of cassava.

Implementation of the encapsulation-dehydration cryopreservation method for the cassava core collection

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Cryopreservation techniques can be divided into classical and new. CIAT has developed alternatives for both kinds of techniques (Escobar et al., 1997, Escobar et al., 2000). New techniques based on encapsulation-dehydration, which are simple and rapid, would be useful for preserving large germplasm collections such as that of cassava. Not too many steps are involved as in the classic methods. Using these techniques with a wider number of clones will give a general idea about how safe this conservation method is in the long term with respect to repeatability and consistency after freezing. Actually, we are conserved under liquid nitrogen 348 clones (55.2% of the core collection). Of the total clones cryopreserved so far, 68% have surpassed 30% as plant formation at least (the threshold value). Material that did not complete the threshold value will be identified to initiate adjustment of the actual method. Some cryopreserved materials were regrown and planted in field; we did not observed any differences in root characteristics. It was possible recover plants from materials conserved for more than 1 year. We initiated to establish some logistic aspects in the management of cryo-bank, when it was important to consider number of beads and tubes per clone. In the same way, we maintain the research with wild materials.

Somatic embryogenesis and plant regeneration in cassava genotypes from the Northeast of Brazil

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The availability of protocols for the genetic transformation and regeneration of model cassava genotypes has prompted us to develop protocols for somatic embryogenesis (SE) and plant regeneration of major cassava genotypes from the North-East of Brazil. *In vitro* culture of the genotypes Água Morna and Rosinha were established by cultivating shoot apexes isolated from field grown plants, in MS medium, supplemented with 0.1 mg/L BA and solidified with 0.7% agar. SE induction was performed using isolated shoot apexes, containing up to three leaf primordia. These explants were incubated for three weeks, in the dark, in Petri dishes containing MS medium supplemented with picloram in the range from 1 to 16 mg/L. Somatic embryos were induced at each picloram level tested. The primary embryos were matured by transferring to MS medium supplemented with BA at 0.4 mg/L, and cultivated under light conditions. After embryo maturation, pieces of green cotyledons were transferred to Petri dishes containing picloram in the range from 1 to 16 mg/L, in order to induce secondary SE. Again, secondary SE could be observed at all levels of picloram tested. In order to assess whether these culture conditions could be used to other genotypes, isolated shoot apexes from the genotypes Amansa Burro, Aparecida, Mata Fome, Milagrosa, Rosa e Sacaí were incubated in MS medium supplemented with picloram 4.0 mg/L, for 3 weeks, in the dark. SE was observed in each genotype, with frequencies ranging from 50 to 80%. Green cotyledons of somatic embryos of each genotype were used as explants for inducing shoot organogenesis. The explants were transferred to MS medium supplemented with a factorial combination of BA and IBA. Shoot organogenesis frequencies ranging from 20 to 40% were obtaining using culture medium supplemented with 0.8 mg/L BA and 0.1 mg/L IBA. Rooted plants were obtained at a 70% frequency by transferring shoots to culture medium supplemented wit4 0.4 mg/L BA. We are now establishing the conditions for obtaining and transforming friable embryogenic calli of each of the genotypes. This is being presently done with the help of the Biotechnology Unit of CIAT.

Minisett multiplication of meristem culture derived cassava stems for quality planting material production

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Cassava is one of the most important tuber crops grown in India for food as well as for industrial purpose. However, major hindrance in the fast spread of cassava is its low multiplication rate coupled with the threatening menace of cassava mosaic disease (CMD) which tends to decline the tuber yield. Study conducted revealed that these twin problems could be effectively countered by meristem culture coupled with agrotechniques. In order to clean the planting material from virus, micro dissection of meristem from shoot buds was done followed by growing them on growth media. Subsequently they were hardened in a shade net house and transplanted in field. At harvest on maturity, the stems were made in to minsetts of single, two and three nodes. Raising minisetts in a shade net nursery was found to be useful to select healthy plants and thus obtain a uniform stand in the field. Comparative evaluation indicated that two node minisetts was ideal which projected a multiplication ratio of 1:50, as compared to the traditional ratio of 1:10. Transplanted minisetts gave 19 percent higher establishment than directly planted ones. However, root spread, yield components and tuber yield were significantly higher in directly planted minisetts. Under rainfed condition, transplanted minisetts gave better performance while under irrigated condition, direct planting was ideal. Among the various spacings studied, 60x45 cm was the optimum compared to other wider or closer spacings. Harvest was done at five month stage for further multiplication by minisetts. It was observed that for planting material production from 1.0 ha. of land by minisett technique, only 740 stems would be required while by the traditional system the requirement would be 2500 stems. The study further indicated that planting materials raised from 1.0 ha in the first year could be multiplied to 92.5 million stems by the third year by adopting this technique. The figure could still be manifold depending on the number of meristem culture materials generated and multiplied following the minisett technique.

Production of the first transgenic cassava plants in Africa: Progress and challenges

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Agricultural biotechnologies including plant tissue culture, molecular marker-assisted breeding and transgenic systems hold significant potential to resolve constraints affecting crop production in sub-Saharan Africa (SSA). Presently, biotechnology research for the improvement of staple foods in SSA is mostly carried out in the developed nations. In order to prevent an increasing gap in technological capacity between the industrialized North and developing South, biotechnology R&D should be supported and expanded in developing countries. For cassava (*Manihot esculenta*), a staple for more than 300 million people in SSA most research into it's the technologies required for its genetic transformation take place in European, North and South American laboratories. Impact of cassava transgenic technology for agricultural development in Africa will depend on how successful these capacities are transferred/adapted to the Africa environment. The International Institute for Tropical Agriculture (IITA) serves as a platform for biotechnology transfer between advanced laboratories and the NARS, and is therefore the ideal environment to initiate such work. Transfer of cassava biotechnologies to Africa was initiated in 1998 through a multinational collaboration between the Institute for Plant Sciences, Zurich, ILTAB/Danforth Plant Science Center, USA and IITA, Nigeria. This has resulted in establishing for the first time capacities for plant regeneration via somatic embryogenesis and organogenesis in more than 10 farmer-preferred genotypes. Most importantly, inoculation of friable embryogenic callus with *Agrobacterium tumefaciens* has resulted in transgenic cassava plants expressing the *uidA* and *hpt* marker genes. This represents the first confirmed success in obtaining transgenic cassava plants on the African continent and acts as an important beginning for the use of transgenic technologies to address cassava production constraints such as disease resistance, starch modification and nutritional enhancement. Establishing routine capacity for the production of transgenic cassava and transfer of this technology to the NARS remain as further challenges.

Transformation of cassava with a modified *e.coli glgc* gene for increased plant yield

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Cassava (*Manihot esculenta* Crantz), a member of the family Euphorbiaceae, is one of the most important food crops of sub-Saharan Africa. One of the constraints for cassava starch production is the long growing season. Cassava typically takes 9-12 months to yield a good harvest. This is longer than other major starch-producing crops such as corn and potatoes. We report here the generation of transgenic cassava with increased starch biosynthesis capacity. This was achieved by enhancing the activity of ADP-glucose pyrophosphorylase (AGPase), the rate-limiting enzyme in the starch biosynthesis, in transgenic cassava. To do this, we transformed cassava with a modified *E. coli glgC* gene encoding AGPase. The *glgC* gene was modified by site-directed mutagenesis (K296E/G336D) to remove the allosteric regulation (enhancement by fructose1,6-P and inhibition by AMP) sites and to increase the velocity of the enzyme. Root-specific expression of the *glgC* gene product was achieved using the tuber-specific patatin promoter of potato. We obtained antibiotic-resistant putative transformed plants which have been shown to have integrated and expressed the transgene by PCR, Southern blot, RT-PCR and enzyme activity analyses. AGPase enzyme activity in transformed plants was increased by more than 65%. Significantly, transgenic plants expressing the bacterial *glgC* gene had two-fold greater top (leaf and stem) and root biomass than wild-type plants grown in the greenhouse.

Efficacy of silver nitrate for slow-growth conservation of cassava (*Manihot esculenta* Krantz). Determination of viability and genetic stability

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In order to extend the subculturing period in *in vitro* conservation of cassava the addition of silver nitrate has been investigated as an option of slow-growth. After 20 months of storage, the viability and reculturing ability of the *in vitro* cultures was assessed. The cultures survived with 100% success in regrowing to their original growth form and on transfer to glasshouse conditions. A morphological, biochemical and molecular analysis was performed to assess the genetic stability of regenerated plants from six varieties of cassava maintained in a medium with 10 mg/lt silver nitrate. A total of 22 morphological descriptors, fifteen isozymes systems (at least twenty genes are involved in their genetic control) and AFLPs technique (three primer combinations) were tested. No morphological nor biochemical variation was observed. Similarly for the AFLPs fingerprints, our results indicate no variation for the evaluated regions of the genome (an average of 63 analyzable fragments by primer combination). A sample of 4,827 cassava varieties from 24 regions of origen has been subcultured in presence of silver nitrate, and slow-growth was observed for all varieties evaluated. We continue to evaluate the extent of conservation time for all accessions using this protocol.

Development of ICMV replicase gene construct in plant transformation vector for imparting resistance in cassava

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Cassava mosaic disease is a major constraint in all cassava growing areas in India. Using commercially synthesized AC1 (Replication associated protein) gene specific primer, AC1 gene was amplified from the ICMD infected plants and cloned in pGEM-T vector and named as pICMV-Rep. Sequencing of AC1 gene showed that it had 1042 bp which encoded for 347 amino acids. Comparison of this sequence with published sequences showed that it has high similarity with ICMV than ACMV sequences. Full length AC1 gene and C-terminal truncated version of AC1 gene was recloned in different plant transformation vectors viz., pBin19, pBin AR, pBI12, pCambia vector 1305.2 and recombinant colonies for both type were obtained in pBin19 and 1305.2 and named as pBin 19 R, pBin 19 R/E and pCambia R. These clones were mobilized to *Agrobacterium tumefaciens* strain LBA4404 through triparental mating procedure confirmed by amplification of AC1 gene in these clones. AC1 gene and its truncated version were introduced into tobacco (*Nicotiana tabacum*) explants through *Agrobacterium* mediated gene transfer method. Transformed explants were selected using Kanamycin resistance as marker for selection and incorporation of the gene was confirmed by PCR analysis.

Screening South African cassava (*Manihot esculenta* Crantz) cultivars for the production of embryogenic tissues

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The pre-requisite for any cassava (*Manihot esculenta* Crantz) transformation program that proposes to develop improved plants is the availability of a reliable regeneration system. Presently laboratories that prioritize cassava research are able to reliably regenerate plants from a range of cultivars. Unfortunately, some cultivars are still either recalcitrant or resisting attempts to induce useful levels of embryogenesis from their tissues. Here we report the production of organized embryogenic structures (OES) from five various southern African regionally important cvs. T200, T400, P4/4, P4/10 and TMS30333. A West African cv. TMS60444 was used as the positive control as it has proved to be a good model system for comparative purposes. By utilizing improved procedures developed at ILTAB for producing embryogenic tissues from various African cassava cultivars, OES were produced from leaf lobe explants of all the above cassava cultivars. South African (SA) cvs. T200 and T400 performed well (76 % and 57 % respectively), producing OES at a frequency and quality approaching that of the model cv. TMS60444 (76 %). Both were shown to be significantly superior for the production of embryogenic structures to the other two SA cvs. P4/4, P4/10 and a Zimbabwean cv. TMS30333. Further optimization of regeneration procedures for the two S.A. superior genotypes T200 and T400 will also be reported.

Induced mutagenesis accelerates crop varietal development and aids gene discovery

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Mutation, the alteration of the genetic make up of an organism, is a natural phenomenon in the evolutionary process that leads to new individuals, species and genera. The generation of variation can be artificially accelerated through induced mutagenesis with the attendant production of new and often times useful genetic variation. Exploitable mutations, though rare events, have contributed greatly to crop genetic improvement with more than 2,250 released crop mutant varieties, in over 170 crop species (including cassava). The impact has been worldwide with mutant varieties being cultivated in 59 countries. Since its inception in 1964, the Joint FAO/IAEA Division has promoted the use of induced mutagenesis to develop superior crop varieties. Currently, the use of induced mutations as a tool for gaining a better understanding of gene function and structure through the application of reverse genetics and gene discovery technologies is gaining momentum. Induced mutation has potential for aiding cassava genetic improvement as well as facilitating the discovery and exploitation of useful genes. Along with molecular genetic markers, cell, tissue and organ cultures and other complementary biotechnologies, induced mutation forms part of the plant breeder's tool kit in enhancing the efficiency of crop varietal development. This is especially important where the gene(s) for the desired trait is/are either not in the genepool accessible to the breeder, difficult to introgress or where genetic transformation is neither feasible nor acceptable. This paper highlights some of the achievements of induced mutagenesis in improving the levels of stress tolerance (drought, salinity, diseases) and quality traits (such as cooking quality in cassava). We conclude by highlighting how cassava genetics and improvement can benefit from the activities and facilities in the Plant Breeding and Genetics sub-Programme of the FAO/IAEA Joint Division, as has been the case with other plants.

Development of an improved system to transform cassava

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The development of a reliable methodology for genetic transformation of cassava is essential to the introduction of new traits into the crop. The current transformation protocols for cassava are long (about six months), amenable to somaclonal variation, and inefficient due to low integration of genes of interest into the genome. This study was undertaken to develop a somatic embyogenic-based system to transform cassava. Undifferentiated calli pieces and geminating somatic embryos for cassava cultivar M. col 2215 were inoculated with *Agrobacterium tumefaciens* plasmid carrying two genes targeted to suppress two genes of interest in the starch biosynthetic pathway. Explants and the bacterial plasmid were co-cultured for forty-eight hours under continuous light on a shaker, and in amended shoot regeneration medium comprising 50% Murashige and Skoog basal salts, yeast extract (0.4 gm/L), sodium chloride (0.1 gm/L), glucose (1%, w/v), galactose (1%w/v), benzylaminopurine (1 mg/L), gibberellic acid (10 mg/L), thiamine-HCl (10 mg/l) and myo-inositol (100 mg/L), and pH adjusted to 5.5. Following inoculation explants were incubated in the dark (1, 2, or 3 weeks) or transferred directly to a 12-hour light regime. Plants were regenerated following successive transfers of inoculated explants onto shoot induction, shoot regeneration, and shoot multiplication media. RT-PCR analyses of twenty-one putative transformants showed integration of the two genes of interest in all putants, but suppression of targeted genes in only one of the twenty-one putants. Results obtained are discussed with a view to developing a robust transformation system for cassava.

In vitro culture of isolated pollen, shoot tip and nodal explants of cassava: Assessment of growth response

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Growth and morphogenesis from different explants of cassava are prerequisite for its genetic manipulation. *In vitro* culture of isolated pollen, shoot tip and nodal segments of four cassava genotypes showed differential growth response in MS medium supplemented with various growth regulators. Of the different concentrations and combinations of 2,4-D (0.5 mg/l), NAA (0.25-0.5 mg/l), GA_3 (0.5-1 mg/l), BA (0.25-0.5 mg/l), zeatin (0.5 mg/l) and boric acid (50-100 mg/l) used for regeneration, supplementation of 0.5 mg/l 2,4-D in the medium produced callus invariably from all the explants and the genotypes tested. However, subsequent culture for growth and differentiation showed shoot organogenesis from the callus produced from shoot tip and nodal explants and embryogenesis in pollen cultures. Morphogenetic response was better from the callus induced from nodal explants as compared to shoot tip explants. Combination of NAA (0.25 mg/l), BA (0.5 mg/l) and GA_3 (0.5 mg/l) found to be quite productive for shoot organogenic response. In such combination the percentage of callus responded to shoot organogenesis was 40-60% and the mean number of shoot buds produced per 50 mg of callus was 6.5-7.2 among the genotypes. On the other hand use of boric acid and zeatin along with other growth regulators was found to be quite effective for pollen cultures in enhancing the frequency of proembryoids (1.5-2.8%). Results are encouraging for monitoring growth and morphogenesis of callus tissues induced from different explants of cassava.

Influence of border cell on growth and establishment of *Glomus microcarpum* var *microcarpum* in genetically transformed roots of cassava

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Cassava (*Manihot esculenta* Crantz) is highly susceptible and dependent on mycorrhizal fungi. Genetically modified adventitious hairy roots were induced in cassava, by infecting the plants with A4 wild type having agropine (vir) region, kanamycin resistant and hormone independent strain of *Agrobacterium rhizogens*. Based on the analysis of opines, and PCR analysis, the hairy roots were proved to be transformed root clones. A living root system is a prerequisite for germination, growth and establishment of AM fungi. Transformed hairy roots were co cultured with *Glomus microcarpum* var. *Microcarpum* at 25 +- 1C under a photoperiod of 14 hr duration. The growth and establishment of AM fungi, its mass production and AM fungal propensity for Border Cell (BC) production in transformed roots of cassava was found to be related phenomenon in the process of simbiosis. The pre-germinated spores of *Glomus microcarpum* var *microcarpun* infected the transformed roots in different phases consisting of 1) extension of germ tube, 2) attachment of germ tube, 3) formation of mycelial fan like structures, 4) formation of apprasoria, 5) formation of intercellular mycelium, formation of septa, arbuscules and vesicles 6) spread of infection in new roots and new regions of old roots and formation of extra cellular mycelium. The transformed root supported the growth of the AM fungi under initial growth phase, logarithmic growth phase, stationary phase where growth and infection of the AM fungus is equalized by the fast growing transformed roots and a phase of decline indicating the inadequate nutrient availability for both growing root as web as AM fungi. As BC production is related to distinct pattern of gene expression, AM Fungal propensity exhibited significant positive correlation. At 4-5 hr, light regime the AM Fungal Colonization spore production and Border cell production also showed a positive correlation in transformed roots of cassava.

Production of amylose free plants in a commercial genotype

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At the previous CBN meeting we have shown that down regulation of *gbssI* leads to the production of plants with amylose-free starch. The frequency of amylose-free plants was very low. Furthermore, the plants contain the luciferase gene and are produced via particle bombardment in the Nigerian genotype TMS60444. The past year amylose free plants were produced via Agrobacterium mediated transformation using only the pat gene as selectable marker in an Indonesian cassava genotype. About 1000 genetic modified plants were produced. These plants were analyzed for the starch composition, somaclonal variation and the presence of vector-DNA. Currently the amylose-free, vector DNA-free plants are analyzed for copy number.

Anther culture in cassava (*Manihot esculenta* Crantz)

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Cassava tissue culture technology has been used as a means of overcoming some problems associated with the traditional method of Cassava breeding. Although much research has been done on somatic embryogenesis and regeneration, investigations on androgenesis and gynogenesis are limited. Anther culture is commonly used in crop breeding as a tool for rapid generation of homozygous breeding lines from genetically diverse or heterozygous genotypes, and are obtained in one generation through spontaneous or induced doubling of the chromosomes of haploid microspores. The main objective of our work is to investigate the response of anthers of an adapted variety on different media combinations to produce haploid plants in Cassava. Unopened flower buds from terminal inflorescence of a locally adapted variety (Mulluvadi) were collected and pre-treated under 4^{o}c for 3days.They were sterilized with 0.1% mercuric chloride for 1 minute and then with 100% ethanol for a minute. A total of 240 anthers containing uninucleate to binucleate microspores were placed in a test tube containing callus induction media supplemented with 2,4-D (2,3,5,6 and 12 mg/l) + kinetin (0.5 mg/l) + sucrose (3%) and incubated at 25^{o}C for 4 to 6 weeks. Longitudinal ruptures in some anthers were seen after 20 days in two media combinations. In order to establish callus cultures, calli produced by responding anthers were subcultured to produce more callus. Callus clumps were transferred to medium containing MS+ 2,4-D (2mg/l) + Kinetin (0.5 mg/l) + BAP (1mg/l)+ Zeatin (0.5 mg/l) + GA (0.5 and 1 mg/l) + Sucrose (3%). Regular observations on callus colour, proliferation, nature of callus and differentiation were taken. The frequency of callus formation was 20% and two types of callus friable (embryogenic) and nonfriable were observed from the callus obtained on medium consisting of MS + 2,4-D (2mg/l) + Kinetin (0.5 mg/l). Culturing this type of callus on MS + BAP (1 mg/l) + GA (1 mg/l) resulted in obtaining embryoid like green structures. These calli were cultured on a suitable medium to obtain plantlets. Studies to determine origin of callus, protein profile of the calli derived from somatic tissues and reproductive tissues (microspores) are in the final stages and the details of the results will be presented.

Possibilities with gari as mycological culture medium

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The recovery of certain microorganisms from samples necessitates the use of expensive, selective media. Investigations into the possibility of using cheap, easily available local materials as media for the cultivation of a number of such microorganisms showed that gari - made from the tropical staple, cassava (*Manihot esculenta)*, was very effective in the isolation of several moulds from various sources. The moulds included *Aspergillus niger*, *Monilia sitophila*, *Penicillium* spp. and *Rhizopus stolonifer*, from soil and fruits and vegetables, and species of *Aspergillus, Cladosporium, Fusarium, Halophythophthora, Mucor, Nigrospora, Penicillium, and Rhizopus* from leaf litter of *Rhizophora racemosa*, *Avecennia africana* and *Laguncularia recemosa* – the red, white and black mangroves, respectively, from sites in Port Harcourt, Nigeria. No antibiotics or other selective agents were needed. Gari was as effective as the prohibitively expensive selective media (which are not readily available in many third world countries because of financial and technological constraints) in the recovery of the moulds from the various sources examined. Gari on its own forms an opaque gel with a rough surface, making it unsuitable for use in colony counts; but this was easily overcome by using a gari agar medium, with gari and water in a ratio of 1: 20 (w/v) rather than the ratio of (2:3, w/v) used in gari alone as the culture medium. On the whole, gari was found to be a cheap, highly effective medium that can be used for the recovery/isolation of moulds from various sources frequently investigated in third world countries.

Induced tetraploids and triploids in cassava: A comparative study with special reference to tuber yield and quality

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The role of cassava (*Manihot esculenta* Crantz) is rapidly changing from a traditional human food crop to an efficient agro industrial crop in many parts of Asia. Being so, there is great need for augmenting its production potential, especially starch yield. This crop has almost all the necessarry features required for successful polyploidy breeding especially because fertility is not a decisive factor as true seeds are not required for commercial cultivation. Research on genetic improvement of cassava at the Central Tuber Crops Research Institute (CTCRI), has revealed the potential of triploidy breeding as a novel, additional tool in cassava for producing high yielding hybrids having high starch content. In this programme autotetraploids were produced first in twelve elite cultivars by colchicine treatment and were screened for several characters. There was a drastic decline in pollen fertility and the tetraploids were found to be on par with diploids for yield and starch content. Triploid cassava produced by crossing diploids with induced tetraploids gave encouraging results. Out of 16,179 flowers crossed in twenty combinations of interploid crosses, 354 triploids were isolated from eight cross combinations. The comparative performance of tetraploids and triploids was assessed and the data revealed the superiority of the latter in most of the characters. The higher yield coupled with the higher starch content of the triploids compared to tetraploids was the most attractive attribute. Also, studies on the rheological properties of starch from triploid cassava revealed considerable starch modification which might be due to triploidy *per se*. This facilitates its direct application for industrial purposes without any chemical treatment. As artificial polyploidy has been used as a step in the process of transferring single valuable characters, attempts are underway to produce resistant tetraploids and triploids for Cassava Mosaic Disease by incorporating the CMD-resistant cultivar MNga-1 received from CIAT in the polyploidy breeding programme.

Virus resistance and production of transgenic cassava plants at ILTAB/Danforth Plant Science Center

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Particle bombardment of friable embryogenic callus with DNA sequences from African cassava mosaic virus (ACMV), resulted in the recovery of transgenic cassava plants with significantly enhanced resistance to cassava mosaic disease. Integration of a defective interfering sequence (DI) produced plants which displayed delayed onset of mosaic disease and reduced symptom severity after particle bombardment with infectious clones of ACMV. However, DI transgenic plants, failed to mobilize DI sequences and do not display resistance when challenged with East African cassava mosaic Cameroon virus (EACMCV). Integration of the replication associated protein (*AC1*) gene resulted in plants with resistance to ACMV, EACMCV, Sri Lankan mosaic virus (SLCMV) and dual inoculation with ACMV and EACMCV. In one transgenic plant line no symptoms were seen after challenge with any single geminivirus species, and only very mild disease observed after dual inoculation, a situation which can result in total yield loss in farmer's fields. Subsequent molecular analysis of the AC1 transgenic plants revealed that in the most resistant lines, AC1 derived mRNA and protein was expressed at only low levels. Probing with the AC1 sequence revealed presence of siRNAs in the non-inoculated plants, providing evidence that transgene-induced gene silencing was imparting resistance against the viral pathogen. The most resistant of these plants are now undergoing challenge with whitefly transmitted geminiviruses in East Africa. New transgenic plants are being generated at ILTAB using *Agrobacterium*-based technologies to investigate the phenomena of transgene-induced gene silencing and its use in controlling CMD in Africa and India. In a separate project, efficacy of a ss-DNA binding protein is being investigated as a mechanism to prevent geminivirus replication and movement in cassava. A 10-1000 times increase in transgene expression levels have been achieved in cassava tissues through manipulation of the expression cassette. More than 50 cassava plants transgenic for this gene are now undergoing analysis. Full efficacy of such strategies for controlling geminivirus-induced disease may require targeting of transgenic products to subcellular compartments. By expressing GFP with targeting sequences we have shown the ability to accumulate transgenically expressed proteins to both the ER and plastids with cassava leaves and root tissues.

In vitro production of certified cassava seed of industrial genotypes and for fresh consumption to improve the crop's competitiveness in the department of Casanare, Colombia

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In Casanare, farmer organizations have been advancing the cultivation of cassava, seeking alternative valid technologies that permit them true industrial development. At present, projects are being developed for the cassava crop's industrialization in the municipalities of Tauramena and Aguazul, where there are two processing plants. To guarantee competitiveness, income, and access to new markets, both for fresh and processed cassava, the need to start with seed of excellent quality is recognized, and can only be achieved through *in vitro* propagation of seed free of virus and other systemic diseases. To meet the expectations of increased cassava planting area in the department, and the supply of healthy seed, the above project is being developed in agreement with Colciencias, UNITROPICO, and the Microenterprise Center of Llano (CEMILLA). Achieving the project's planned objectives made possible the: 1) setting up of a suitable tissue culture laboratory; 2) adoption and application of *in vitro* techniques for the regeneration of complete plants; 3) construction of a greenhouse for material hardening, and 4) establishing of observation plots of mother plants and multiplication lots of certified good quality cassava seed, free of systemic pathogens such as virus, using *in vitro* techniques. This infrastructure and the development of production processes using tissue culture have permitted an important linkage of the technology to educational and production sectors of the region, representing great social and economic impact for the people of Casanares.

Influence of carbohydrate concentration on cassava growth *in vitro*

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Cassava line TMS 60444, which has been used in cassava transformation, was grown under varying sucrose concentrations in vitro. Nodal segments from the cassava were grown on agar gelled Murashige and Skoog salts and vitamins and supplemented with 0, 1, 2, 4 or 6% sucrose. Shoot growth and development was monitored over a five week period. Sucrose levels of 2% and 4% produced the most active growth and tallest plants. Plant internodal length and leaf size was reduced at the 6% level. This was attributed to the increased osmotic potential of the medium. The elevated sucrose concentration should allow more carbohydrates to be stored in the plant tissue for it to draw on as it becomes photoautotropic. Though the growth rate was reduced at the 6% sucrose level, the plants may be more adapted to acclimatization ex *vitro.*

PS 8: Genomics and Markers

An update on development of an EST-database for *Euphorbiaceae*

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Euphorbiaceae comprises a genetically diverse plant family that includes species having economic impacts on world economies. Apart from cassava, other globally important agricultural species of *Euphorbiaceae* include: castor bean (*Ricinus communis*), an important oil crop; rubber tree (*Hevea brasiliensis*), an important source of rubber; poinsettia (*Poinsettia pulcherrima*), an important horticultural crop; leafy spurge (*Euphorbia esula*) an important perennial pest weed of North American plains and prairies; and annual weeds such as hophornbeam copperleaf (*Acalypha ostryifolia*), and endangered species such as *Akoka* and telephus spurge. Several research groups have realized the potential of using a genomics-based approach to, for example, identify markers and increase our knowledge of plant genome structure, organization and gene function within the *Euphorbiaceae* family. As part of a genomics-based approach, an EST- (expressed sequence tag) database for *Euphorbiaceae* is being developed. To date, at least 25,000 ESTs have been identified from various tissues and genotypes of *Euphorbiaceae*. Preliminary data indicate that 6,000-7,000 unigenes have been identified within the *Euphorbiaceae*-specific EST-database. Two new EST projects have recently been started for cassava genotype TME 117 (drought-tolerant) and whole plant leafy spurge and should provide an additional 5,000-8,000 unigenes. These new ESTs will be added to the developing database and, taken together, these resources will provide a valuable resource for breeding programs working on improving genetic stocks of desirable species and for scientist involved in developing methods to control the growth of undesirable species.

Development of a diversity array technology (DArT) chip for cassava

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Genetic resources, mostly held by small farmers represent a critical resource for the future productivity and stability of production of the crop. How to evaluate and use in a systematic manner the vast amount of variability present in cassava is still a challenge to most cassava breeding programs. Genotyping micro-array technologies offer the highest throughput available up to date. One of them diversity array technology, DArT ™ (CAMBIA), is sequence-independent (low-input) and allows the fingerprint of an individual's genome based on a high number of polymorphic sites spread over the genome. These screening procedures should allow testing of thousands of individual samples in a speedy manner. Plant materials used for the generation of the DArT chip was chosen to represent a broad as possible diversity of the cultivar, a few genotype of its wild progenitors and 2 wild species were included to capture a large number of polymorphic fragments. They include 14 accessions from Brazil, 14 from Colombia, 4 from Guatemala, 2 each from Nigeria, Cuba, and Ecuador, Peru and Thailand respectively. Others include, one accession each from Argentina, Bolivia, Costa rica, Fiji islands, Indonesia, Mexico, Panama, Venezuela, and USA. Six and 2 improved varieties were included from CIAT and IITA respectively. Three enzyme combinations, /PstI/BstNI, PstI/ApoI, and PstI/TaqI) were tested in a preliminary experiment to determine the best enzyme combination,the library from PstI/BstNI gave the largest number of polymorphic clones (132), followed by *Taq*I (112) and ApoI (69). In total, 313 candidate polymorphic clones were obtained in the preliminary experiment. Libray expansion yielded 440 polymorphic clones (14.3%), for the PstI/TaqI array, and 554 polymorphic clones (18.0%) for the PstI/BstNI array, both consistent with the polymorphism frequency in the smaller in the preliminary experiment. A dendrogram was drawn for the 80 cassava samples based on analysis with the polymorphic clones fromm both arrays. There were differences between the two dendrograms obtained, inspection of the data suggests that the BstNI array contains a higher proportion of clones derived from repetitive sequences than PstI/TaqI array. Typing using repetitive sequences introduces a bias in genetic diversity analysis due to over-representation. The PstI/TaqI array does not show a high proportion of clones with repeated sequences and can be used as a routine genotyping tool for genetic diversity analysis.

Comparative gene expression study to identify genes possibly related to storage root formation in cassava (*Manihot esculenta* Crantz)[1]

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Cassava storage roots result from swelling of adventitious roots by secondary growth. Storage root formation involves continuous elongation (primary growth) and radial growths. The cells of vascular cambium, a meristematic tissue, are rapidly divided and expanded resulting in secondary growth that gives an increased diameter, rising secondary phloem to the outside and secondary xylem to the inside with parenchymatic cells packed with starch. In the present work we aimed to gain insight into molecular processes occurring during cassava storage root formation. Identifying genes expressed during secondary growth, will be possible to elucidate molecular processes related to parenchyma cells development (starch and carotenoid accumulation) and prospect specific promoter and enhancers able to drive efficient storage root specific expression. In this work, we report a comparative gene expression study by Northern blot analysis in adventitious and storage roots to identify such genes. cDNA clones derived from our cDNA subtractive library were sorted by cellular process possibly related to storage root formation and used as probes in this study. Results, so far, revealed five genes with higher expression level in the secondary xylem tissue than adventitious roots. Among them, the gene coding to Pt2L4 protein, a putative RING Zinc Finger and LEA genes were strongly induced in the secondary xylem tissue.

Reliability of constant traits in morphological characterization of cassava as compared to AFLP markers

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Cassava (*Manihot esculenta* Crantz) is the second most important staple food crop in Sub-Saharan Africa providing an average of 285 calories per person per day. It is also an important food and cash crop in Malawi. A prerequisite for any genetic improvement programme is knowledge of the extent of genetic variation present between cultivars and genetic distances between them, and closely related species. This can be achieved through characterization of germplasm either using morphological, biochemical or DNA markers. This study was therefore initiated with the aim of comparing morphological and DNA based molecular marker (AFLP) technologies in characterizing cassava genotypes. Trials with 16 cassava genotypes were conducted from 2000 to 2002. Field work was done at Chitedze and Makoka Research Station in Malawi, while the NDA finger printing was done at the University of the Free State in the Republic of South Africa. This study revealed that genetic distances determined by morphological characterization using mainly constant traits correlated to similar values using AFLP fingerprinting. It is however, a prerequisite that morphologic data into binary characters needs careful consideration to achieve meaningful results.

Subtractive cDNA library and macro-array applied to isolate specific genes related to the color diversity in the storage root of cassava (*Manihot esculenta* Crantz)[1]

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This study was performed to evaluate if cDNA subtractive library in combination with macro-array technology can identify genes using their transcriptionally defective mutant in the diversity of color in the storage root of cassava. Diversity of color in cassava storage root is due to the presence of a variety of carotenoids, including β-carotene, lycopene, phytoene and lutein that are synthesized and accumulated in chromoplasts of this type of root. Three putative mutants in our GENEBANK were identified to accumulate solely lycopene, β-carotene, or lutein. If this differential accumulation is in the synthesis pathway or in the process of accumulation is not yet known. Three cDNA subtractive libraries were prepared and the generated ESTs are being test for its tissue specificity, traditional clone specificity and for the tissue age effect in the storage root. Results, so far, indicate that the number of genes are differentially distributed among cell process including 20% coding for complex carbohydrate metabolism related genes, 16% coding for senescent cell process, 15% coding for protease as the major classes of genes. Secondary metabolism derived compounds genes code for 8% of the genes identified. Color phenotype and tissue age in the storage root are being tested for the differentially expressed EST using macro array and conventional mRNA blot as well as the eight gene coding for the carotenoid syntheses pathway.

Characterization of differentially expressed sequence tag (EST) from sugary phenotype of storage root of cassava (*Manihot esculenta* Crantz)[1]

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The sugary storage root phenotype was, recently identified from traditional clones cultivated in Amazon. At the level of above ground morphology there is no particular distinction of sugary cassava from the farina and table cassava. Major distinction among these types of cassava is observed only in the storage root, which shows profound alteration in storage root size and density in water, secondary xylem cells development, parenchyma cells size and turgidity, diverse starch structure presence, and osmotic parenchyma cells environment. Therefore it is expected to observe a profound effect on genes being expressed and regulated differentially in the sugary cassava in comparison with the farina type. We are using cDNA subtractive library to construct an EST database with silenced and expressed genes for this kind of storage root. Macro array and conventional mRNA blots are being used for differential gene expression analyzes. The BLAST and clustering analyses of 1200 high quality EST sequences gave 267 cDNA clones that are tested with macro array and conventional RNA blot analyzes. Preliminary gene sequence annotation for 953 cDNA clones indicated that 25% are NON HITS, 22% of the clones code for unknown protein, 17% codes for carbohydrate related metabolism, and other minors functions. cDNA clones are being tested for gene expression analyzes gain information on tissue specificity, tissue age effect, and phenotype specificity.

Molecular marker-assisted and farmer participatory improvement of cassava germplasm for farmer/market preferred traits in Tanzania

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Tanzania is the fourth largest producer of cassava in Africa with average yields of about 8 t/ha (FAO, 2001). This is well below the continent's average of 10tons/ha and the average yield of 14 tons/ha of Africa's (and the world's) largest producer, Nigeria. The low yield is caused by many factors including susceptibility of commonly grown varieties to major diseases and pests such as cassava mosaic diseases (CMD) and the cassava brown streak disease (CBSD). Cassava varieties grown by small farmers in Tanzania has however been shown to be very diverse and could be the basis of a successful breeding project. A farmer participatory, molecular marker-assisted, decentralized, breeding scheme was proposed as a means to speed up the process of improving local cassava germplasm for resistance to pests and diseases in Tanzania. The scheme was recently approved for funding by the Rockefeller Foundation. The proposed breeding project will take farmer preferred germplasm by agro-ecology and cross them to improved introductions that have resistance to Cassava Mosaic Disease (CMD), Cassva Green Mite (CGM), and Cassava Bacterial Blight (CBB). Given the fairly large number of parents that will be used, molecular markers associated with pest and disease resistance will be employed to reduce, in a logical manner, the number of progeny to a manageable number. The progeny selected by MAS will be evaluated in a single season in the corresponding agro-ecology and then evaluated over two cycles in collaboration with end-users (rural communities and cassava processors). The project will be carried out in a total of six years divided into 2 three-year phases. A principal objective of the project is the development of capacity for participatory plant breeding and marker-assisted breeding. This would be achieved by training 2 national program breeders at the MSc. and PhD level, and through 2 training workshops on participatory plant breeding and marker-assisted breeding.

QTL mapping for resistance to root rot caused by *Phytophthora tropicalis* in cassava

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Varietal resistance is one of the main tools for managing root rot in cassava (*Manihot esculenta* Crantz), caused by different *Phytophthora* species, and it is therefore necessary to understand the genetics of resistance of this important crop. The resistance of parents and progeny of cassava families K (M Nga 2 x CM 2177-2) and CM 9582 (M Bra 1045 x M Cr 81) to *P. tropicalis* were evaluated. Fresh roots of 92 individuals of family K and 43 of family CM 9582 were inoculated, in addition to the parental materials of each family. Based on the phenotypic evaluation and the molecular map of M Nga 2 (female parent of family K), QTLs associated to the resistance to *P. tropicalis* were identified and mapped, using simple marker analysis. Eight QTLs were defined by analyzing 92 individuals, two of which explained 9.0% and 8.6% of phenotypic variance. Cassava family K genotypes (69 individuals), evaluated during 2000 and 2001, showed a percentage of infected root area between 22% and 95%. The correlation between the evaluations of 2000 and 2001 was -0.15. The variability in the expression of resistance between years indicates that the environment affects the phenotypic expression, generating variation. Cassava family CM 9582 genotypes showed an infected area between 70% and 90%. The distribution of frequency of cassava family K genotypes, based on root area affected by *P. tropicalis*, corresponds to a normal distribution, with one genotype presenting moderate resistance in both years of evaluation, 51 genotypes susceptible, and 17 genotypes highly susceptible. Minor genes were found to control resistance in family K to *P. tropicalis*.

Characterization of a resistance gene cluster in cassava

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Oligonucleotide primers designed from conserved domains encoded by R-genes have been used to amplify NBS sequences from cassava leading to the identification of twelve classes of Resistance Gene Candidates (RGC). Mapping the RGC by RFLP was not very successful due to the low level of polymorphism observed. The genomic organization of these RGCs was explored by screening a cassava BAC library with the 12 RGC classes as probes. This allowed the identification of 42 BAC clones. Most of the RGCs detected one to six BACs suggesting the presence of a small gene family or single copy, except one that detected 26 BACs and represents a multigene family. Using fingerprinting analysis, BACs corresponding to each RGC class were assembled in 10 contigs and 19 singletons. Members of the two TIR and non-TIR NBS-LRR subfamilies occurred together within individual BAC clones. We obtained 67 BAC end sequences and primers were designed from 17 of them and used for mapping. Some BAC clones co-segregated and were localized in linkage group E and J. One BAC (11E5) located on group J contained five NBS sequences. Partial sequencing revealed the presence of two complete RGCs encoding two highly similar proteins. At least one is expressed constitutively in cassava tissues. Identification and sequence analysis of the RGCs provide new insights into the genome organization of cassava especially with regards to resistance genes.

RXam-1: A *Xa21* homologue associated to bacterial blight resistance in cassava

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The *Xa21* gene confers a broad spectrum of resistance to *Xanthomonas oryzae* pv. *oryzae* and encodes a receptor like-kinase carrying LRRs in the putative extracellular domain (Song *et al*., 1995). The structure of *Xa21* is unique in carrying both the receptor domain LRR and the kinase domain. By a PCR approach and using specific primers of *Xa21*, Bonierbale *et al*. (1997) isolated a 900pb fragment (named PCR250) in cassava cultivars showing high level of resistance to *Xanthomonas axonopodis* pv. *manihotis* (*Xam*). The PCR250 fragment showed 56 % similarity with the rice *Xa21* gene. PCR250 was localised in the linkage group X and associated to a QTL (XM5) explaining 13% of resistance to *Xam* strain Cio-136 (Jorge et al., 2000). In this study the PCR250 fragment was used to screen a BAC library (TMS3001, susceptible to *Xam*). Four BACs clones (BAC1-4) were identified, two of them were assembled in one contig containing two copies of the PCR250 fragment. Partial shotgun of one of the BAC clones (BAC2 from TMS3001) and a primer walking approach were used to sequence the complete cassava *Xa21* homolog gene. No ORF was detected in this BAC clone from TMS3001. Designing new specific primers and using a 5'RACE and 3'RACE RT-PCR approach, the genomic sequence and the complete cDNA of the *Xa21* homolog was obtained from the cassava resistant cultivar MBra685. The *Xa21* cassava gene homolog was named *RXam-1* and is ~3600 bp long containing an ORF of 1181 amino acids. The cassava RXam-1 protein shows 43% identity and 59% of similarity with the Xa21 protein of rice. RT-PCR experiment shows that the *RXam-1* gene is induced in the resistant variety 72 hours after infection by *Xam* strain CIO136. Studies on the *RXam-1* gene function are in progress and will be presented.

Molecular marker-assisted breeding for resistance to the cassava mosaic disease in Latin American cassava gene pools

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Molecular marker-assisted selection (MAS) for CMD resistance at CIAT is both a pre-emptive measure, should in case the disease is accidentally introduced in Latin America, and an effort to contribute the broad genetic variation of CIAT's elite germplasm to Africa. MAS for CMD resistance has been made possible by the discovery of several SSR and SCAR markers associated with the single dominant gene CMD2. The MAS scheme currently used at CIAT involves crossing CMD resistance donor parents, obtained from IITA, to CIAT's elite cassava parents (by agro-ecology) and germination of the resulting seeds as embryo axes followed by multiplication and molecular analysis, using leaf tissue from *in vitro* plants, with the SCAR marker RME1 and the SSR marker NS158, both closely linked to *CMD2*. Establishment as embryo axes is necessary to fulfill pyhto-sanitary conditions for the shipment of these germplasm to partners. Genotypes that carry the resistant allele are shipped to collaborators in Africa and India and also sent to the green house for hardening and transfer to the regular breeding scheme. Molecular analysis is by a rapid DNA miniprep isolation method using 96 well plates followed by PCR and eletrophoresis. An excel file format was developed for the collation and storage of the marker and other relevant information such as pedigree, phenotypic evaluation, number of plants available and where. This year, a total of 2315 seeds were harvested from more than 2000 controlled crosses between CMD resistant parents and elite parents of the 5 cassava gene pools by agro-ecology or backcross derivatives of *M. esculenta* sub spp flabellifolia resistant to the green mite. More than 1,500 genotypes have been processed by the MAS scheme described above. The cassava MAS lab currently has two persons and together they can process 192 genotypes in 2 days or 480 genotypes per week or over 24,000 samples in a year, molecular marker analysis alone. Work in ongoing to improve this by doing the grinding and DNA isolation in 96-well plates. Current costs of a single SSR marker data point analysis for cassava at CIAT is US$0.30, processing 24,000 samples in a year requires a budget of US$7,200. The bottle neck remains the tissue culture establishment step.

Simple sequence repeat (SSR) assessment of genetic diversity of local cassava varieties from Guatemala

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Although several studies have demonstrated a likely South American origin for the cultivar, the diversity of cassava and its wild relatives in Meso- America is great enough to suggest a second center in Meso-America. Besides, the potential of Meso-American diversity in cassava improvement has not been properly assessed. Three recent studies of genetic diversity in land races from South America and Meso-America have revealed unique alleles in land races from Guatemala at a frequency high enough to suggest a Meso American center of cassava diversity. The results of the three studies were based upon 6, 4, and 13 Guatemalan land races. The small sample size of the previous study could distort the allele frequencies and lead to wrong conclusions. A larger collection and SSR characterization of land races from Guatemala was therefore made to confirm preliminary data of a Meso-American center of diversity and to secure the largely untapped diversity in Guatemala before it becomes extinct. Collection of cassava land races was carried out all over Guatemala in May 2002, a total of 128 accessions were collected in the departments of Baja Verapaz, Quiche, Huehuetenango, Alta Verapaz, San Marcos, Escuintla y Santa in Guatemala, representatives samples from Africa and South America were included for comparison. A set of 36 SSR markers, carefully chosen to represent a broad coverage of the cassava genome with moderate to high polymorphism information content (PIC) and robust amplification, were used in this study. Statistical analysis on the raw SSR data include: genetic distance analysis using a of a distance matrix based upon 1-proportion of shared alleles (1-PSA), principal component analysis (PCA) and cluster analysis (UPGMA) of the distance matrix, and parameters of genetic diversity and differentiation. Genetic diversity, as assessed by the average gene diversity (H_E) was high in the accessions analyzed 0.5422 ±0.2468. Unique alleles were observed in the accessions from Guatemala for half of the markers used. Accessions from Guatemala form two groups, one that clusters along with land races from Brazil, Nigeria and Colombia in a broad group and a second group that is clusters separately. The results observed confirms previous observation of a high genetic differentiation of between certain groups of cassava land races from Guatemala and these from other parts of Latin America and Africa (Fregene et al. 2003). A UPGMA cluster analysis of the genetic distance data also produced 2 clusters of the Guatemalan accessions similar to that found with the PCA. The origins of highly differentiated samples of cassava germplasm from Guatemala can be explained by independent domestication events in populations of different *Manihot* species that yet exist or are now extinct or an introgression from *Manihot* species in certain regions that overlap in geographical spread with cassava. The highly differentiated landraces from Guatemala may represent heterotic pools, like those for maize.

Positional cloning of CMD2 the gene that confers high level of resistance to the cassava mosaic disease (CMD)

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Previous work revealed that the SSR markers SSRY 28 and NS158 are the closest markers to the gene *CMD2* that confers a high level of resistance to the cassava mosaic disease (CMD), and are located at distances of 9 and 3 cM respectively. High resolution of the *CMD2* region of the genome was initiated in this region of the genome to identify markers more closely linked to *CMD2*. The fine-mapping population was 1690 individuals from a cross between TME3, the source of CMD2 and the improved variety TMS30572. The cross was evaluated in the 2002 growing season for CMD resistance in the field at IITA, Ibadan, under heavy natural pressure of the disease. The population was evaluated with the 2 SSR markers according as described by Mba et al. 2001 and a total of 112 recombinants between the markers and *CMD2* identified. DNA from 10 resistant recombinants and 10 susceptible recombinants were combined to form 2 bulks which were then evaluated with several markers system including AFLPs, ISTRs, RAPDs and SSRs in a modified bulk segregant analysis (BSA) method.(Michelmore et al. (1991). Markers that were polymorphic in the recombinant bulks were then analyzed in individuals of the bulks. Two polymorphic RAPD markers were identified, one of them RME-1 is less than 1cM from the gene and it was cloned and converted into a SCAR marker. A bacterial artificial chromosome (BAC) library with more than 10X coverage for the cassava genome was constructed from the cassava variety TME3, donor parent of *CMD2*. The BAC library was screened with the SCAR marker RME-1 and 250 positive clones were identified, suggesting that this marker is from a repetitive sequence region. The BAC clones are being used to construct ends of the BAC clones that are at the extreme of the contigs will be sequenced and mapped in the recombinants to identify which contigs belong in the region of *CMD2*. Successive hybridizations of the BAC end markers to the BAC library will be carried out until markers that flank the gene on both sides are identified.

Genetic mapping of QTLs affecting productivity and architecture in a full-sib cross from non-inbred parents in cassava (*Manihot esculenta* Crantz)

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An attempt was made to identify quantitative trait loci (QTLs) fpr several productivity and architecture traits in a full-sib progeny of 144 individuals from two non-inbred parents in cassava. A molecular linkage map of this cross constructed previously with over 250 markers was the source of molecular markers. The progeny were grown under field conditions at two locations (Palmira and Quilichao) in Colombia and evaluated in two years (1998 and 1999) for architecture and productivity traits. Architecture traits evaluated were plant height (PH), branching height (BH), branching levels (BL), branching index (BI), stem portion with leaves (SPL) and LAI. Productivity traits were those related to total dry matter production and distribution, which were evaluated, were fresh root yield (FRY), fresh shoot yield (FSY), harvest index (HI) and number of storage roots (NR). Phenotypic evaluation of the traits in this population revealed continuous variation for all traits. Broad sense heritability estimates, ranged from 36% (for NR) to 94% (for BH). Several significant phenotypic correlations were observed between architecture and productivity traits. Primary QTLs, using single-QTL model, and secondary QTLs, by a primary QTL interaction model, were detected by interval mapping. A total of 30 primary QTLs and 84 secondary QTLs were detected. We identified 35% of detected QTLs in two or more trials, the other were environment specific. Several genomic segments affecting multiple traits were identified and were in agreement in correlation among traits. All QTLs identified for FRY were found associated with either component traits of productivity or architecture traits. This study suggests that QTLs for plant architecture can be used to improve productivity. However an exhaustive search and analysis of QTLs controlling architecture is required before marker-assisted selection (MAS) for increasing productivity can be initiated.

Assessment of simply sequence repeat (SSR) diversity of cassava land races in Nigeria

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"A study of the genetic diversity of 270 cassava land races in Nigeria was assessed by characterizing the accessions using Simply Sequence R."

Signifcant variations and diversities in total Heterozy gosity (HT) and Genetic differentiation (GST) occurred among the accessions. The mean gene dirersity recorded was 0.49-057. Cassava land races from the humid and sub-humid regions of Nigeria had higher mean gene diversity compared to those from the semi-arid region of Nigeria. There were strong differences in genetic distances between all pairs of individual accessions. The PCI AND PC2 values accounted for 26% and 16% of the total variouce respectively. The PCA further revealed a sub-structure in the accessions from the semi arid region of Nigeria. The differences in the gene diversity of accessions in the humid and semi-arid region suggest that the variations in agricultural practices of Cassava farmers coupled with the allogamous nature of Cassava , gave rise to a large pool of Cassava with diverse genes. This studey implies that there is need to harness the potential characteristics fo these land races for crop improvement.

Identification of defense-related cassava genes by subtractive hybridization and using a cassava cDNA microarray

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Cassava Bacterial Blight (CBB) caused by *Xanthomonas axonopodis* pv. *manihotis* (*Xam*) is a destructive disease in the South America and Africa and yield losses range between 12 and 100%. Cytochemistry and biochemistry of defense response to CBB have been well studied. However, the response of the plant to pathogen attack at the molecular and cellular level remains uncharacterized. Identification of genes associated with defense responses is one of most critical steps leading to the elucidation of disease resistance mechanisms in cassava. In this study, we identified differentially expressed genes during pathogen attack by subtractive hybridization, using the Differential Subtraction Chain method. A pool of cDNA obtained from infected plants was used as "tester" and a pool of cDNA obtained from healthy plants was used as "driver". 1536 clones were isolated from the resistant varieties (MBRA 685 and SG 107-35). Of these, 110 randomly selected clones were sequenced and a homology search was conducted. The sequence analysis showed that 16 cDNA clones shared homology with plant genes involved in defense responses, 70 clones were either homologous to plant genes of unknown function or showed no homology, representing new genes potentially involved in cassava defense responses. A cDNA microarray was constructed by spotting the clones identified from our subtractive libraries. Other clones potentially involved in cassava defense responses were also included. The cassava defense cDNA microarray was used to confirm the differential expression of the clones.

Global transcriptome analyses of cassava-*Xam* interaction using a cassava cDNA microarray

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Cassava is a staple crop for millions of people in the tropics. The application of molecular genetic analysis for cassava breeding has been limited compared to others crops. Recently progress have been made in the development of genomic and bioinformatics tools to increase our knowledge of cassava genome structure and cassava gene function. A large cassava EST database has been developed in our laboratory. This represents an important contribution to the genomic resource and permits the beginning of a large-scale analysis of expression profiling in cassava. A cassava cDNA microarray was constructed and used to study the interaction between cassava and *Xanthomonas axonopodis* pv. *manihotis (Xam)*, the causal agent of bacterial blight. For the microarray construction, 6040 clones from the cassava unigen set were amplified by PCR and printed on glass slides. A set of spot controls from tomato, potato and cassava housekeeping genes, human genes and spiked controls were also included in the cassava array. The resulting microarray contained 7059 elements. Microarray hybridization was performed using cDNA from cassava plants (resistant variety MBRA685) collected at 12, 24, 48 hours and 7 days post-infection as treatment and cDNA from healthy plants as control. Results obtained will be presented and discussed. Functional genomic tools such as the cassava microarray give a first comprehensive overview of the molecular basis of the cassava defense response to the bacterial blight pathogen and will help in the future in understanding the defense mechanisms to other important pests and diseases.

Knowledge from QTL mapping studies on the wild relatives of maize and its application on cassava

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During domestication strong selection on specific traits has gradually decreased the genetic variability in crops. This has lead to accumulation of some genotypes and traits, whereas others have been lost. Some of the lost traits (e.g. pathogen resistance) are of importance for cultivation of modern crop cultivars. These traits and genotypes are preserved in the wild relatives. Understanding the genetic basis of trait changes as a result of domestication is fundamental for breeders, other geneticists and evolutionary biologists. Two parameters of primary interest are the number of loci controlling a trait and the relative magnitudes of their effects. The most powerful means of identifying these loci is quantitative trait loci (QTL) mapping. QTL studies on maize and several other crops have demonstrated that numerous traits that distinguish crop plants from their wild relatives are often controlled by a relatively small number of loci, where some of these loci have a large effect. However, my own research on the wild relatives of maize, teosinte, showed that the evolution of traits under natural selection has, in contrast to crop domestication, only involved genes of small to moderate effects. Cassava is a contrast to maize in several respects. Cassava is grown for its starchy roots which are not used for propagation. Instead it is vegetatively propagated through stem cuttings. A major part of the cassava breeding takes place in the farmer's villages. This local breeding has resulted in a less directed selection in cassava than in maize and many crops grown in the industrial world. For future breeding it is of great importance to elucidate the genetic basis of trait differences between cassava and its wild relatives. Based on the knowledge from my work on teosinte I will use a QTL mapping approach to study the genetics of cassava domestication. I will analyze the segregation of molecular markers and traits in an F_2 population derived from a cross between cassava and its proposed ancestor *Manihot esculenta* spp. *flabellifolia*. The characterization of QTLs controlling trait differences will make it possible to reintroduce genetic variation from the wild ancestor in an effective and directed way using marker-assisted selection.

Two cassava promoters related to vascular expression and storage root formation

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Cassava (*Manihot esculenta* Crantz) storage roots, organs accumulating large amounts of starch, develop from primary roots via secondary growth. The availability of promoters related to storage-root formation is a prerequisite for engineering root traits in cassava. Two cDNAs, c15 and c54, were identified from a storage-root cDNA library of cassava MCol1505 via differential screening. The transcripts of c15 and c54 were detected in storage roots but not in leaves by Northern analysis. Homology analysis of the deduced amino acid sequences showed that C15 is likely to be related to cytochrome P450 proteins, which are involved in the oxidative degradation of various compounds, while C54 may be related to Pt2L4, a cassava glutamic acid-rich protein. The promoter regions of *c15* and *c54* were isolated from the corresponding clones in a cassava genomic library. A 1,465-bp promoter fragment (*p15/1.5*) of c15 and a 1,081-bp promoter region (*p54/1.0*) of c54 were translationally fused to the uidA reporter gene, and introduced into cassava and *Arabidopsis thaliana* (L.) Heynh. The expression patterns of *p15/1.5::uidA* and *p54/1.0::uidA* in transgenic plants showed that both promoters are predominantly active in phloem, cambium and xylem vessels of vascular tissues from leaves, stems, and root systems. More importantly, strong β-glucuronidase activity was also detected in the starch-rich parenchyma cells of transgenic storage roots. Our results demonstrate that the two promoters are related to vascular expression and secondary growth of storage roots in cassava.

Conserved expression of a root-hair specific promoter LeExt1.1 from *Lycopersicon esculentum* in cassava

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Using root hair specific promoter to target gene expression in trichoblasts has great potential to improve nutrient uptake and transformation from soil in cassava, a crop usually growing on acidic soils low in nutrients, particularly phosphorus. Here we report on characterization of a root hair-specific promoter LeExt1.1, which is isolated from tomato (*Lycopersicon esculentum*) and directs the expression of an extension-like protein LeExt1 in its root hair, in transgenic cassava (*Manihot esculenta* Crantz). A binary construct containing the β-glucuronidase under control of the LeExt1.1 promoter was used to produce transgenic cassava plants via *Agrobacterium*-mediated transformation. Of the 6 transgenic cassava plant lines, strong GUS expression was detectable only in the trichoblastic cells of rhizodermis within the differentiation zone of primary roots, but not root tip. Among the root hairs induced from primary roots, the predominant GUS expression was observed in the differentiation zone close to root cap. There is no or very weak GUS activity detectable in other tissues or organs, such as vascular tissue, leaf, stem, and storage roots. Similar expression pattern of the promoter had been found in both tomato and potato. Therefore, the promoter LeExt1.1 can be used to regulate genes of interest for improved nutrient mobilization in cassava.

PS 9: Participatory Research and Technology Transfer

New technologies: Factors influencing farmers decision to adopt and sustain use of improved technologies-experience from cassava research and development in Uganda

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Cassava mosaic disease (CMD) is the most important constraint to cassava production in Uganda. The epidemic was first reported in 1988. Since then, it has been affecting cassava production in East and Central Africa. To date, improved cassava varieties are the most effective means of controlling CMD. A study was conducted in Soroti, Kumi, Lira and Masindi districts of Uganda to investigate farmer's use of different cassava technologies in relation to the CMD constraint. The outcomes were to highlight issues for research for the Uganda National Cassava Programme. A qualitative research meted was adopted where individual farmer interviews were conducted and enriched with farmer-group discussions to understand transfer and adoption of CMD resistant technologies. The uses of improved and indigenous cassava varieties were ranked to obtain growing trends in variety use. Secondary data and interviews with key informants gave background information on actors and activities that influenced dissemination and adoption of improved cassava. The study revealed that farmers were eager to grow and adopt improved cassava irrespective of their age. It also showed that adoption of new technologies is positively correlated with availability of assets/capital, other inputs like labouring and access to extension services. From the study, the conclusion was drawn that interventions of government and non-government organizations positively contribute to adoption of improved cassava. UIT improved varieties, CMD severity was substantially reduced and farmers realized better yields and made innovations on processing techniques. The study result highlights the importance of a better understanding of the character and conditions of effective interventions by government and non-government actors. The implications for the design of decentralized participatory research are discussed.

Farmers participatory research and transfer on cassava technologies: Lessons for action from impact assessment in India

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Cassava in India is extending dual function of food security and raw materials to a range of industries and is cultivated in different types of production systems. Farmers Participatory Research and Transfer Programme (FPRTP) has become an integral part of cassava R & D sytem in India. Starting FPRTP as a semistructured extensive mode has now bloomed to a structured intensive method by passing through various phases of participatory programmes viz Lab to Land, participatory on farm evaluation, Institution Village Linkage Programme enabling the participation of more than 3000 farmers in 20 villages of three states covering eight cassava production systems. Impact assessment was done on all the programmes in terms of technology generation and transfer on the parameters of productivity, profitability, knowledge, adoption, spread of technologies and skills of onfarm experimentation. Impact study showed that FPRTP was instrumental in identifying production system specific varieties, varietal characters, agro-techniques amalgamating recommended and farmers practices, cropping systems and promoting farmers as experimenters .The impact parameters of technology transfer indicated that productivity and profitability increased to 20-60% in various systems and adoption rate to 20-70%. The FPRTP has helped in significantly improving the knowledge level and technology spread to 20-70%. A model of FPRTP was evolved for effective and organised farmers participatory research and transfer which depicts the actors, methods and linkages for future actions.

IFS: Building scientific capacity in developing countries

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The International Foundation for Science (IFS) supports promising young scientists from developing countries. They are identified through a careful selection process and they receive support in their early carrier to enable them become established and recognized nationally and internationally. IFS was founded in 1972 as an independent, non governmental organization with the mission to contribute to the strengthening of research capacity in developing countries. IFS has profiled itself in research on the sustainable management of biological resources. Since its creation IFS has supported some 3,500 scientists in 100 developing countries. Small grants (up to USD12,000) and supporting services are provided for projects in areas such as agriculture, forestry, aquaculture, food science and nutrition, natural products, and water resources as well as for research on the sustainable utilization and conservation of natural ecosystems. Projects may address biological, chemical, physical processes as well as socio-economic relationships important in the conservation, production and renewable utilization of the biological resource base. Grants can be renewed twice. Since cassava is not only important as a staple food but also in contributing to raised rural incomes, IFS supports innovative research on this crop and welcomes collaboration with other organizations involved with cassava. Since science and technology capacity has not evolved equally in all developing countries, the main focus of IFS is on countries with vulnerable research infrastructures, in cooperation with key organizations in the South.
Applications can be submitted in English or French all year round. Forms can be downloaded from www.ifs.se or write to the above address.

Community response to improved agricultural technologies: A case of Wakiso district in Uganda

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The concern of low technology adoption has become a subject of interest. Most notably, several agricultural technologies have been demonstrated to have impacted little or not at all at people level. In part, this has been in a consequence of poor technology delivery outlets, and has continuosly pondered the agricultural research scientists and their associates the technology providers. To gain insight into this cyclic cycle of misery, the Uganda National Cassava programme launched an outreach programme with the main goal to identify and integrate the most pertinent factors that would presumably acelérate ultimate technology adoption. In highlight, the process involved a needs assessment study to capture the crucial underlying factors to technology adoption; socio-economic, gender, community structural typologies, and general agro-ecosystem análisis. Preliminary results have consistently shown that existing farmer groups are more reliable to work with (>70%), support rapid technology adoption and propagate technology sustainability post intervention. During this study, female farmers demonstrated unrivalled memory and predicative capacity than males, although males often enroded this treasure by interfering with demales freedom of expresión. In addition, ideological differences and farmers'dynamics were identified as pertinent in the technology adoption process. Together, this study clearly demonstrated the need for constant redress of farmers changing priorities to match the dynamic farming systems. The implications and practice of this study are discussed.

Crops research institute of Ghana: Organization committed to developing affordable and desired food and agro-industrial products

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The CSIR-CRI is the largest of the 13 Institutes of the Council for Scientific and Industrial Research (CSIR), a statutory semi-autonomous corporation mandated to implement, coordinate and advise on scientific research and development policies in Ghana. Formed in 1964, CRI is currently committed to become a centre of reference and excellence in Africa, for quality agricultural crops research, technology transfer and dissemination that is impact oriented. Our research activities aim at developing and disseminating sustainable, appropriate and environmentally sound technologies. Such technologies include improved high yielding, pest and disease resistant and consumer acceptable varieties of food and industrial crops (e.g. cassava, yams, maize, cowpea, soybean, rice) and better crop management and post harvest practices. CRI has a multi-disciplinary team of 77 scientists (26 PhD, 36 MS, and 15 BS). We have a long history of agricultural research, and technology development and dissemination. Our recent research thrust is focused on agro- industrial rural development based on modernized agriculture in line with the governments vision. There are 12 programmes at CRI: Maize, Rice, Legumes, Roots & Tubers, Horticulture, Seed Technology, Resource and Crop Management, Crop Protection, Biochemistry/Biotechnology, Socio-Economics, Biometrics, Training and Communication. Our achievements include the development of high yielding improved varieties of cassava, maize, cowpea, soybean, rice, etc. that have been released and are widely grown by farmers in Ghana and other third world countries. One of these varieties is the highly acclaimed QPM maize variety Obatanpa, which is produced under different names in other countries. CRI is also proud to have supplied the cassava variety used for industrial starch production in the Presidential Special Initiative (PSI). Clients include farmers, agro-industrialist, food processors, vendors and exporters. As the foremost national scientific agricultural research and technology transfer institution, CRI is expected to impact positively and significantly on the livelihoods of rural dwellers, help reduce rural poverty, improve incomes and ensure food security. CRI must therefore be up to date in new and modern science and technology transfer efforts directed at crop production, productivity, yield and post harvest activities. Agro processing and adding value to essential and targeted agricultural products for domestic and export market is critical to our survival and relevance in the long term and would vigorously be pursued.

LE MANIOC (*Manihot esculenta*): Une culture rendue évidente pour tous

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Quatre clones améliorés de manioc d'origines IITA, préalablement sélectionnés en station (RT94/D70, RT94, RT95/23 et RT95/56) ont été testés en milieu paysan avec le cultivar local de 1998 à 2001 dans une approche participative décentralisée dans trois localités de la zone de savane du Tchad. Il s'agit de SIDO, KOUTOUTOU et KANA. Les écartements pratiqués par chaque paysan ont été évalués. Selon les caractéristiques des racines (qualité alimentaire, taux d'acide cyanhydrique etc.), ces clones à rendement élevé, résistants aux maladies/ravageurs et dotés de bonnes caractéristiques des racines (qualité alimentaire, taux d'acide cyanhydrique etc.) sont appréciées par les producteurs de manioc.
Il existe des perspectives d'avenir encourageantes en vue de la réalisation des objectifs du programme dans les années à venir, grâce à l'efficacité de la sélection et de l'amélioration végétale. Abstract : Four (4) new varieties of cassava issued from IITA (RT94/D70, RT94/D01, RT94/23 et RT95/56) have been first of all selected in an agricultural research station and then tested in 1998-2001 with a participative and decentralized approach in the savannah area in Chad with the local cultivars. The places where the test took place were SIDO, KOUTOUTOU and KANA. The row-spacing used by the farmers have been evaluated. According to the characteristics of the roots selected by the producers (feeding quality, rate of hydro cyanic acid etc…), those clones with a high level of efficiency, resistant to diseases/cassava pests and with very good characteristics of alimentary roots have been appreciated by the farmers (cassava producers). There are prospects that are encouraging for the achievement of the goals of the program in the years to come thanks to the efficiency of the selection and thanks to the improvement of plants.

Scaling-up biological and organic inputs with regional participation: Technologic and social development model for cassava production systems

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Fundamental changes in agricultural development involve rapid expansion of participatory approaches which are based on interactive learning between professionals and farmers. This project evaluates the potential use of biological inputs as mycorrhizal arbuscular (MA) fungi and organic inputs earthworm compost, in order to improve the productivity of under low fertility soil conditions. Moreover, farmers have to deal with the problem of low availability of high quality seeds and high dependence on chemical inputs. To contribute on the solution of these limiting factors , two pilot plants scale production for MA and earthworm compost were established at the regional centers of research Caribia (Santa Marta) and Turipaná (Monteria) with a capacity of 90 ton/year to MA and 35 ton/year to earthworm compost each on. These plants were designed to provide the initial biological and organic inputs to six local plants, placed strategically to have access to 54 participative groups from the Colombian Caribbean region. This products were evaluated under field conditions, resulting in increase of 38% in yield for a cassava production system. The activities developed until now have allowed design the of model for the scale-up of biological and organic inputs, the direct participation and involvement of small farmers throughout the process of research and technology transference is guarantee of the adoption of these technologies. These activities have an important impact in the production systems, in the community organization and generate new employment opportunity at the regional level without a negative impact on natural resources and environment.

Strategic research interventions on production and processing of cassava technologies in India

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Inadequate supply of food to overcome the hunger and combating malnutrition of myriad of weaker sections in India is a major concern. Demand projections for Cassava in India is estimated as 10 million tonnes by 2020 as against the present production of 5 millions tonnes, a huge task ahead. Added to this Cassava area trends over the past three decades is dismal. Present rate of productivity augmentation is too inadequate to meet the challenge. Cassaca mosaic disease (CMD) and lack of quality planting materials also pose great threats. Conventional crop improvement and protection could not handle the emerging demands from cassava. Identification of genes responsible for yield enhancement through marker assisted selection and introduction to cassava through gene transfer techniques is attempted. *Ama 1* gene enhancing protein level in tubers is being introduced into cassava. Genes responsible for ß-carotene enrichment and increasing the shelf life of tuber are also attempted. Similarly CMD resistant transgenic plant production are underway through pathogen derived resistance approach. Large scale multiplication of virus free planting materials are being produced through meristem tip culture techniques. Thrust in value addition research of late includes diversification of the cassava starch utilization for tablet and capsule making as well as ecofriendly detergents based on cassava starch. Nutritionally enriched food products like low cost health drinks and weaning food are also attempted, aiming at the socially and economically downtrodden segments of the population. To meet the challenge of production augmentation, nutrition enrichment and value addition, the R & D strategies warrants a shift in the approach in terms of networking and linkages nationally and internationally. The paper also discusses the structural and functional adjustments for networking and linkages for generating biotechnological driven cassava technologies.

Cassava propagation by small-scale farmers using a low-cost *in vitro* system

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Between 2002-2003 an interdisciplinary group composed by CIAT, FIDAR, CBN and a group of small-scale farmers established a low-cost, *in vitro*, cassava propagation system that allowed the latter to produce certified, planting material. This permitted to certify the quality of material each cycle, and support seed releases, or renewing materials in the Department of Cauca (Colombia). The methodology was tested in the filed with six clones that produced 6000 plants. The plants were harvested and certified as Frog Skin Disease-free by the Colombian Institute of Agriculture (ICA). Certified cuttings were then used as initial explants in a two-node, rapid propagation system. The purpose was to increase the number of plants for distribution among farmers. We believe that the system could be implemented in other cassava growing regions in Colombia, where there is need to renew planting material. This experience leads the incorporation of *in vitro* propagation (tissue culture) into farmer's routine agricultural practices. At CIAT, research will continue to incorporate other crops, with minimum investment, taking advantage of the low-cost facilities already in place. Other CBN-funded pilot sites, in Ecuador and Brazil, are scheduled to start similar cassava propagation schemes this year.

Use of *in vitro* technology by small farmers to clean and preserve native cassava varieties in Southern Colombians Andean region

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The Cauca region, Colombia is a zone of high cassava production, mainly to attend the starch markets. Nevertheless in the last years, diseases and insect attacks and low available of clean and certified material of better response clones have not permitted that be maintained large planting areas.
With farmers support, we collect 27 local materials and they were sown in two places (900-1300 msnm and 1400-1700 msnm) for evaluation of their characteristics. It was determined 21 morphological descriptors. Cuttings of these materials were carried to CIAT and carried out them thermotherapy and meristem culture to virus cleaning. This material will be use as source of clean material for local seed bank establishment. Initial plot were harvested and the local material named "Totoqueña" show better performance than other ones. The group, based on these data selected 12 materials for plants other plot with replications. Besides morphological descriptors, a molecular analysis using AFLP's was carried out.

Improvement of cassava productive system in the microregion of the plain of Cordoba and Sucre, Colombia

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In Colombian Atlantic coast, the cultivation of cassava is predominantly of rural economy, although in some isolated cases, private investors are beginning to develop process of Cassava drying in order to be used in the production of concentrate foods. This alliance that binds the private sector and the small producers tends to improve in a time period of five years, the productivity, the quality and the drying process of the cassava crop for the producers of the municipalities of the plain de Cordoba and Sucre and stimulate the associative production ways that enable the farmer economy to be integrated in the agrobusiness using the strategy of creating a cooperative (APROYSA).With this alliance we expect to cultivate 800 has, build 4 artificial drying plants each having an area for mixed drying.In addition, it will generate 37.600.00 daily wages per year in the agricultural production. Each producer will receive a net income of $ 416.000.00 per ha with a rentability of 24.16%.At the end of the alliance, at least 80% of the producers in the project will know and apply the multiplication technologies for clean and certified seeds, as well as the management and production of organic fertilizer and the agronomical management of the cultivation that permit to guarantee a constant production of raw materials for the private sector and with an excellent quality for concentrate food industry. These strategies of productive alliances should proliferate all around the country, allowing that the cultivation of Cassava generate better incomes to the small producers and supply the demand for concentrate food industry.

Regional progress in production and management of high quality seeds of cassava

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The production of clean seeds of cassava in projects of regional development have allowed us to give an answer to technologicals limitations, with high levels of adaptation to the local and regional conditions, that facilitate the development of productive center with an important participation of farmers and groups of women. These strategies have enabled the development and standardization of efficient laboratory protocols with average results of five knots in five weeks, survival percentages in greenhouse period greater than 80 %, multiplication of super elite seed (SSE) with production averages of 7x, management and multiplication of SSE seeds in breeding grounds with average rates of 3.5x with and efficient production of basic seed in a period 7 months with survival rates greater than 80 %. This seed can be multiplied in sandbanks for 15 days, with germination rates of up to 100% with a root development capacity of 500 seed per m2.These results in local seed scaling have enabled us to include nearly 5000 producers in the Atlantic coast region, Cundimarca, Tolima, Huila. Santander and the Llanos Orientales, increasing the productive yield from 12 to 22 Tn / ha in average.

Identifying cassava clones that are appreciated by smallholder farmers—A factor of importance to PPB and to maintenance of genetic resources

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Cassava (*Manihot esculenta Crantz*) clones, that is groups of plants with identical genotypes, which are appreciated by farmers, could be of great value to participatory breeding. It is shown that smallholder farmers, both in Africa and in Latin America, aim at having a single clone as a local variety although, mostly, a fraction of the plants that they classify to a variety do not belong to this clone. Evidence from both continents also suggest that many of the cassava clones kept by smallholder farmers are under evaluation for production capacity and could be looked upon as members of a breeding population. Consequently one could claim that farmers maintain both a production population, that is a set of clones appreciated for their production capacity, and a breeding population, that is clones used as options for the future. Examples showing the difference in distribution of a clone when using molecular markers compared to local variety names for identification will be presented. Criteria for classifying a clone as belonging to the production population and sampling strategies for identifying such clones will be discussed. The relevance of classifying the smallholder farmers' cassava plants into a breeding and a production population will also be discussed as well as the impact on the collection and maintenance of genetic resources of such a subdivision.

Biotechnology and rural development: An alternative for the integration of biotechnology in Cuban rural communities

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In Cuba, large investment has gone into the massive diffusion of biotechnology's advantages in the fast multiplication of plants, and big units of *in vitro* micropropagation (biofactories) have been developed in the last 15 years. Nevertheless, there are limitations to diversifying varieties and responding to market demands, and to farmers' biophysical and sociocultural demands. When the collapse of communism stopped the supply of inputs, the environmental and socioeconomic differences became more marked. The diverse and complex situation in Cuba demands strategies that facilitate the strengthening of local structures and of complementary methods of biotechnology application that are sustainable from the social, economic, and environmental point of view. The proposal attempts to offer evidence of the potential of *in vitro* multiplication with regard to local needs and the development of human potential in a bottom up sense. In the medium term, an increase in crop production will be possible through applying tissue culture techniques for the micropropagation of cassava cultivars, amplifying clonal diversity in this crop in two rural localities in the west of the country, using participative strategies for clone selection in each environment, and a strong component of training farmers in the advantages of these techniques and in developing abilities to put them into practice, This also will raise farmers' self-esteem in becoming a definitive part of variety and production policy, and management and maintenance of the seed in the cassava crop, which represents a staple in the diet of the Cuban small farmer. The present work reflects the advances achieved during a first stage in the Project's development in San Andrés and La Jocuma, municipality of La Palma, in the province of Pinar del Río, Cuba.

Working with farmers: Spreading new cassava varieties, improved practices ……. and new hope

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Cassava (*Manihot esculenta* Crantz) is the third most important food crop in southeast Asia and the most important upland crop in the northeast of Thailand. The crop is usually grown by small-holders in marginal areas of sloping or undulating land. Most farmers realize, however, that cassava production on slopes can cause severe erosion, while production without fertilizers will lead to a gradual decline in soil productivity. In order to enhance the adoption of new varieties and improve the sustainability of cassava production under a wide range of socio-economic and bio-physical conditions, a farmer participatory research (FPR) approach was used to evaluate promising cassava germplasm, and to develop effective soil conservation practices, balanced fertilization and cropping systems that tend to produce greater short-term benefits. This FPR project, funded by the Nippon Foundation from 1994 to 2003, started initially in only 2-3 sites (villages) each in China, Indonesia, Thailand and Vietnam, but later expanded to about 32 sites in Thailand, 35 in Vietnam and 33 in southern China. By the end of the project, new high yield and high starch varieties had been adopted in over 1 million ha (98% of the cassava area) in Thailand, 100,000 ha (40%) in Vietnam and 36,000 ha (10%) in China, benefiting at least 800,000 cassava farmers. The new varieties and improved agronomic practices adopted by farmers resulted in a gradual and substantial (2.6 t/ha) increase in cassava yields in Thailand as well as a significant increase of 4.2 t/ha in Vietnam from 1994 to 2002. In both countries, cassava is now an important vehicle for rural development and is given farmers new hope for a better future.

Results of applied biotechnology on cassava crop in Cuba

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A group of researches giving specific solutions to present difficulties in Cassava crop in Cuba is presented. Such results have contributed to develop cassava crop, as well as yield and quality of planting material. This work has been performed by the biotechnology lab. from INIVIT since 1994 to 2003. A general objetive was to establish an optimized methodology for an efficient somatic embryogenesis and genetically stable plants. The clons used were: 'CMC-76' and 'CEMSA 74-725'. As variants for field test, traditional planting materials (cuttings) and vitroplants from organogenesis and somatic embryogenesis were used. Evaluations were carried out 10 months after planting and variables were total plant height (cm), height of the first branch (cm), 'seed' number (cutting) per plant and yield (kg.plant-1). During five growing cycles, it was observed that for alll evaluated variantes and in both clons, the best responses were obtained during the second and third cycle, except for the variable related with the height of the first branch which resulted better in the first growing cycle. The durabillity of rejuvenation effect on cassava was determined and up to five cycles in field conditions were found to be workable with better results than plants from traditional planting materials. Besides, morphoagronomic and molecular markers were used to determine that plants regenerated from somatic embryos were genetically stable.

Farmer participatory varietal selection: The case of cassava in Ghana

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Conventional plant breeding (CPB) has made immense contribution to the improvement of cassava in Ghana. However, the adoption of new cassava varieties resulting from CPB has not been encouraging due to the fact that researchers normally involve farmers at the latter stages of varietal selection and with little or no regards to the multitudes of farmers' preferences which are often beyond yield and resistance to diseases and pests. Farmers on the other hand, place emphasizes on traits that are adaptable to the existing farming systems and post harvest characteristics that satisfy their long developed taste, flavour and cyanogenic potential requirements. As a step to improve the adoption of improved cassava varieties in Ghana, a study was initiated to ascertain the traits of interest to farmers in five different locations in Ghana and to take advantage of their knowledge of the crop. This paper reports on the criteria farmers use in selecting desirable cassava varieties both at the vegetative and the harvesting stages of the crop. Traits desired by farmers' in the various locations were also recorded.

Cassava nutritional studies in cropping systems involving coconut, banana and elephant foot yam using radio tracer techniques

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Interspecific root competitions in cassava + coconut, cassava+banana and cassava + elephant foot yam cropping systems were studied in field trials by comparing the absorption of applied ^{32}P in the sole and mixed systems.Absorption of ^{32}P in two crop cropping system was evaluated by quantifying the radioactivity absorbed during 15 days and 30 days after application by the crop to which the radiophosphorus was applied as well as by the neighbouring non- treated component crop. The results reveal that cassava can be raised as an inter crop in coconut without much competition for nutrients, when cassava is planted in two rows around the coconut palm at a spacing of 90x 90 cm leaving a distance of 2 m radius from the base of the palm. Cassava can also be grown in association with banana and elephant foot yam without any competition for nutrients.

Innovation in the Venezuelan Program of cassava improvement

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The Ministry of Science and Technology of Venezuela with the aim of contributing with a reduction in the food dependence in our country, it has been promoting the development of cassava crop through the definition of a working agenda of research and training, as well as promoting the integration process of the different people related to cassava crop. The general project aims are to evaluate cassava cultivars from different regions and countries in order to select the ones with better performance, higher quality and better yield in each production region, to establish cassava germoplasm banks, *in vitro* and in the field, and start with the establishment of regional centres of management and multiplication of high quality planting material (CEMMY). At the moment five field assays with 20 selected cassava CIAT elite cultivars it has been performed. The better local cultivars are also being introduced in germoplasm banks through meristems isolation and thermotherapy. A regional cassava germoplasm bank has been established at UDO. More germoplasm banks are expected to be developed in the near future. A program for training of cassava growers in an integrated management of the crop and production of planting material it's on the way. One of the most important results expected from the Project it is the transference of the agro technologies direct to the cassava growers, to set the minimum bases to produce high quality planting material, and to start with the selection and improvement of local cassava cultivars through a National Program of Cassava Germoplasm Improvement. The main result of the project will be the identification of genotypes with superior characteristics that can be used as parents in cassava breeding programs; identification of high yield cultivars showing resistance to pests and diseases and a better adaptation to different environmental conditions. The achievements of the objectives will allow an important development of cassava crop, the related farmers and all the cassava industry of our country.

Huay Bong 60: New developed Thai cassava (*Manihot esculenta* Crantz) variety with improved starch yield and quality

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Huay Bong 60 is new developed Thai cassava variety under the collaboration between Kasetsart University and the Thai Tapioca Development Institute. It was developed since 1991 by conventional breeding between Rayong 5, the latest developed variety by Rayong Field Crop Research Center and Kasetsart 50, the latest developed variety by Kasetsart University. After seedling selection, single row selection, preliminary yield trial and standard yield trial, it was selected for further evaluation in regional yield trial for at least 10 locations and had the average yield of 35.94 ton/ha with the average starch content of 25.4%. The average yield and starch content of Huay Bong 60 was slightly higher than those of Kasetsart 50, the most widely grown variety in Thailand. Extracted starch was white with high paste viscosity. This new developed variety also has good plant type, good stake with good rate of germination and can be another good variety for cassava industry.

Sampling variability in cassava roots for total carotene content

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As the interest for a precise determination of the provitamin A carotenoids content of foods is becoming more generalized, efforts had to be directed at improving the validity/reliability of the data. Because of the inherent difficulties in carotenoid analysis, which are sometimes not perceived by the analysts themselves, the reliability of a substantial portion of existing data may be questionable. It is recognized that the carotenoid composition varies as a function of several factors (e.g. stage of maturity, cultivar, handling, analysis method, etc.). However, little is known about the variability of the samples used for measuring carotene content in tissue like cassava (*Manihot Sculenta* Crantz) roots. The specific objective of this project was to measure sampling variation for β-carotene in cassava roots for a more uniform, comparable and reliable data. Although no position in the root resulted in a statistically significant difference for carotenoid content, there was a clear trend for the proximal section to have higher carotene contents that the distal section. Also the periphery of the root tended to have lower amounts of carotenoids that the more internal tissue.

The overall objective 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. This research is part of the HarvestPlus Initiative and was financed by DANIDA and USAID.

Effect of processing on B-carotene content of cassava roots

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Most of the available data on carotenoid contents refer to unprocessed food sources. However, the ultimate effect of those carotenoids in human health depends heavily on the way the foods are processed and consumed by the population. In addition to the original quality and quantity of carotenoids present in cassava, their bio-efficacy will depend on the amount lost upon processing the roots. The effects of processing on bio-availability of carotenes, therefore, needs to be determined. The influence of different preparation or processing methods on -carotene and total carotenoids contents of cassava roots was investigated. Retention of β-carotene upon different processing methods is variable and decreases in the following order: lyophilization, boiling, oven-drying, sun-drying and gari. Prolonged processing and cooking upon gari preparation results in substantial loss of β -carotene.

The purpose of this study is to determine losses of the vitamin A potency during processing of cassava roots. This research is part of the HarvestPlus Initiative and was financed by DANIDA and USAID.