



# PRO *MUSA*

A Global Programme for  
*Musa* Improvement



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# PROMUSA

## A Global Programme for *Musa* Improvement

Proceedings of a meeting held in Gosier, Guadeloupe, March 5 and 9, 1997

Edited by E. A. Frison  
G. Orjeda  
and S. L. Sharrock



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# Foreword

The first steps towards the establishment of the Global Programme for *Musa* Improvement (PROMUSA) were taken in 1996, with the aim to bring together, at the global level, all the major efforts in the area of banana and plantain improvement.

PROMUSA has been developed jointly by INIBAP and the World Bank through a process of extensive consultation with the various partners and stakeholders and has built upon experience gained from on-going activities such as the INIBAP Breeders' Network initiative and the World Bank Banana Improvement Project (BIP) funded by the Common Fund for Commodities.

As a result of these initial consultations, INIBAP prepared a draft proposal for the programme which was widely circulated as a means to stimulate discussion and to elicit further input into the proposal. This proved to be an extremely fruitful exercise and numerous positive and constructive comments were received. These comments were used to prepare a second draft of the document, which was again distributed by INIBAP to a wide audience. During this period of consultation, the document was distributed to over 50 individuals and institutes, the majority of whom provided feedback for incorporation into the document.

This participative approach led to the production of a final proposal which represented the common views of the interested parties. This proposal was presented for discussion at a joint World Bank/INIBAP meeting which was held in Guadeloupe in March 1997.

The Guadeloupe meeting brought together some 70 of the most prominent researchers involved in *Musa* improvement world-wide. Discussions of the proposed global programme took place in both plenary and working group meetings. Five Working Groups were formed to address the major research areas, while one group discussed the structure and *modus operandi* of the programme. At the end of the meeting the creation of a Global Programme for *Musa* Improvement was strongly endorsed by all participants and a programme structure, *modus operandi* and medium-term plan were agreed upon.

This document provides details of the strategy and medium term plan for PROMUSA. These include specific objectives for the programme as a whole, for which an initial 10-year period is envisaged, as well as outputs expected by the mid-term point (5 years) and by the end of the 10 year-period.

A more detailed strategy and medium term for each Working Group has also been elaborated, in which priority research activities and major constraints are identified and which also includes information on inputs required, expected outputs, indicators and timetables for the achievement of objectives. The Working Group reports also give information on existing facilities and expertise and provide an inventory of on-going research in each specific area.

Finally, proposals for an expanded, more flexible International *Musa* Testing Programme (IMTP) which has a greater regional focus and which could take on the role of a global and regional evaluation programme, are described.

Emile Frison  
INIBAP

Michel Petit  
The World Bank



## Guadeloupe Declaration

Bananas and plantains are one of the world's most important yet poorly studied crops. They are grown almost exclusively by smallholder producers, and play an important socio-economic role in many developing countries of the tropics. They are of major importance to food security as well as providing a valued source of income through local and international trade. In terms of gross value of production bananas and plantains are the fourth most important global food crop. Export bananas are the fourth most important commodity and as a fruit rank first.

Banana and plantain producers world-wide are facing ever increasing pest and disease problems, which in the absence of locally adapted resistant varieties, can only be controlled by the use of pesticides. The costs of such pesticides put them beyond the reach of many small farmers who are consequently suffering major production losses. For commercial banana producers, disease and pest control represents a considerable economic and environmental cost. The need for a wide diversity of genetically improved

Declaration of a meeting attended by 70 of the most prominent researchers involved in *Musa* improvement world-wide.

varieties with increased productivity, suitable for the range of growing conditions under which bananas and plantains are produced, and able to meet the differing local consumer demands, is recognised. It is mainly through genetic improvement that sustainable, environmentally sound, improved production can be achieved.

Advances in banana improvement that have been made in recent years indicate that a high return may now be expected on investment in *Musa* research. Although the research priorities for commercial and local consumption banana production may differ, there are considerable mutually beneficial "spill-over effects" from research carried out within each sector.

In order to foster close international cooperation and to facilitate the creation of synergies between ongoing research efforts, a global-level initiative is required. Such an initiative should provide the mechanisms through which individual efforts can be globally coordinated as a coherent and prioritized set of activities to more efficiently address critical research needs.

In recognition of the above statement, the creation of a Global Programme for *Musa* Improvement is strongly endorsed and it is recommended that significantly more resources be directed to *Musa* research, more particularly in *Musa* improvement.

Guadeloupe, March 1997





# PROMUSA – A Global Programme for *Musa* Improvement

## Introduction

Bananas and plantains are one of the world's most important yet poorly studied crops. They are grown almost exclusively by smallholder producers, and play an important socio-economic role in many developing countries of the tropics. They are of major importance to food security as well as providing a valued source of income through local and international trade. In terms of gross value of production, bananas and plantains are the fourth most important global food crop. Export bananas are the fourth most important commodity and as a fruit rank first.

The growing recognition over the last 10-15 years of the importance of bananas and plantains has coincided with recent advances in breeding techniques which have made it possible to overcome many of the barriers to genetic improvement of this crop. The number of *Musa* research and improvement programmes has thus increased considerably in recent years and the first disease-resistant bred hybrids are now being cultivated commercially. However the genetic improvement of bananas and plantains remains an expensive and slow task, and, considering the scale and diversity of the problems facing banana and plantain growers worldwide, these programmes are still too few in number. Many gaps remain in the knowledge of the major pests and diseases affecting *Musa* and there is a continuing need to conduct basic plant pathological research. Sources of resistance available to breeding programmes are also limited and these should be widened to avoid the dangers of genetic vulnerability. It is clear that there is still much work to be done before a range of pest and disease resistant varieties, suitable for the varying regional needs and conditions, will be widely available. It is only through close international collaboration, drawing together and building on the limited number of on-going initiatives in *Musa* improvement, that a significant impact will be made in years to come.

## Global Programme for *Musa* Improvement

Considerable progress has been made in recent years by *Musa* breeding programmes. The first hybrids to be released for general cultivation through the International Network for the Improvement of Bananas and Plantains/United Nations Development Programme (INIBAP/UNDP) International *Musa* Testing Programme (IMTP) were produced by the *Fundación Hondureña de Investigación Agrícola* (FHIA) and these are now being tested in national evaluation programmes in more than 50 countries. In some countries these hybrids are already being cultivated on a wider scale by farmers. Similarly, black Sigatoka-resistant plantain hybrids have been developed by the International Institute of Tropical Agriculture (IITA) and these have been widely distributed for evaluation by national programmes in Africa. Further improved hybrids

from several breeding programmes are presently being evaluated worldwide as part of the second phase of the IMTP and hybrids from other breeding programmes, including those of IITA, *Centre de recherches régionales sur bananiers et plantains* (CRBP) and *Empresa Brasileira de Pesquisa Agropecuária* (EMBRAPA), are ready for inclusion in a third phase. INIBAP has also established an informal *Musa* 'breeders' network', specifically to stimulate cooperation in such breeding efforts.

In parallel, a Banana Improvement Project (BIP), co-sponsored by the Common Fund for Commodities (CFC), the World Bank and the Food and Agriculture Organization of the United Nations (FAO), was set up in 1993 with funding for five years. This project aims to increase the productivity of export bananas, through the development of higher yielding, disease resistant varieties and by reducing the costs of production, especially the cost of pesticide applications. The potential for "spill-over effects" from this project to benefit smallholder producers is great.

The Global Programme for *Musa* Improvement (PROMUSA) has thus been developed as a means to bring together all the major efforts in the area of *Musa* improvement. It is a broad based programme which links the work carried out towards addressing the problems of export banana producers, including that of the BIP, with those initiatives directed towards improving banana and plantain production at the subsistence and smallholder level. The global programme builds upon existing achievements and is based upon ongoing research initiatives. PROMUSA is therefore a mechanism to further maximize the outputs and accelerate the impact of the overall *Musa* improvement effort. The programme is an innovative mechanism to bring together research carried out both within and outside the Consultative Group on International Agricultural Research (CGIAR), creating new partnerships between National Agricultural Research Systems (NARS) and research institutes in both developing and developed countries. The formation of such partnerships will also contribute to strengthening the capacity of NARS to conduct *Musa*-related research.

Recent biotechnological breakthroughs are now allowing rapid progress in *Musa* improvement and real impact can be expected in the near future. PROMUSA therefore, in the initial stage, is focusing specifically on research directly related to *Musa* improvement. Wide participation in the programme ensures that PROMUSA has a global perspective and the structure is such that ownership of research is broadly based. A global and regional evaluation programme operates in parallel to the improvement research activities allowing NARS, and subsequently farmers, early access to improved material as it is produced. The evaluation programme not only provides a mechanism for the rapid dissemination of research results to farmers, but is also a channel through which information from farmers is fed back into the programme.

It is recognised that issues related to intellectual property rights and biosafety regulations have profound implications for programme implementation and these will need to be addressed within the framework of PROMUSA.

Funding which becomes available to the programme will be channelled to priority activities through a number of mechanisms including competitive grants and specific contract research grants.

## Guiding Principles

- **The Global Programme for *Musa* Improvement will focus specifically on genetic improvement and supportive research and priority will be given to research which has a global or regional significance.**
- **PROMUSA will operate as a consortium and will rely on a range of funding mechanisms.**

Partners in the programme are expected to contribute in-kind their own research and, in addition, the programme seeks further resources in order to address priority research needs, as identified by the programme partners.
- **PROMUSA's organizational structure will be simple and efficient in order to ensure that maximum support is maintained for research activities.**

Programme activities take place in a series of thematic working groups, which allow continual interaction between group members. Interdisciplinary contact also occurs at regular intervals through meetings at the programme level and on a continuing basis through the programme secretariat.

Thematic working groups operate as networks, thus the formation of collaborative projects between group members, resulting in a division of labour and the creation of synergies is facilitated. Networking as a *modus operandi* not only fosters collaboration between network partners but also provides an efficient mechanism for priority setting and facilitates the regular flow of information between network members.
- **Participation in PROMUSA will be based on the capacity to contribute through a high scientific capability in *Musa* research and on comparative advantage and will be on a voluntary basis.**
- **Decision making within PROMUSA will follow a 'bottom-up' approach and participating scientists will be fully involved in this process.**

Decisions are based on scientific priorities identified by programme participants and based on users needs. The global and regional *Musa* evaluation programme plays a major role in this regard, providing a mechanism for the two-way exchange of information between NARS and research and breeding programmes. The provision of feedback information regarding farmers' needs is of particular importance in setting research priorities. The existing regional banana research networks also provide a useful channel through which information from national programmes is fed back to the global programme.
- **Partners in PROMUSA will benefit from:**
  - global prioritization of research needs;
  - improved possibilities for funding for programme participants due to recognition of the programme by donor agencies;
  - close interactions with, and knowledge of, other research teams within their area of specialization;
  - opportunities for interdependent research projects (i.e. projects requiring interdisciplinary and complementary partnerships);
  - improved access to information and resources;
  - participation in programme meetings and conferences.

## Programme Structure

The programme operates as a series of interlinked thematic working groups coordinated by an Executive Secretariat. The programme is directed by a Steering Committee and operates under a Programme Support Group. Further details of the programme structure are given below.

**Programme Support Group:** This is composed of major donors and stakeholders and thus comprises representatives from donor agencies (e.g. countries, International Fund for Agricultural Development (IFAD), CFC, UNDP, World Bank, Foundations, private sector); other relevant organizations (e.g. FAO and the Inter-governmental Group on Bananas); representatives of Advanced Research Institutes (ARIs), International Agricultural Research Centers (IARCs) and NARS. Membership is also open to other interested parties. The Programme Support Group provides visibility, guidance and support to the programme. It endorses the overall direction and strategy of the programme and contributes to identifying and providing additional funding and other resources as necessary.

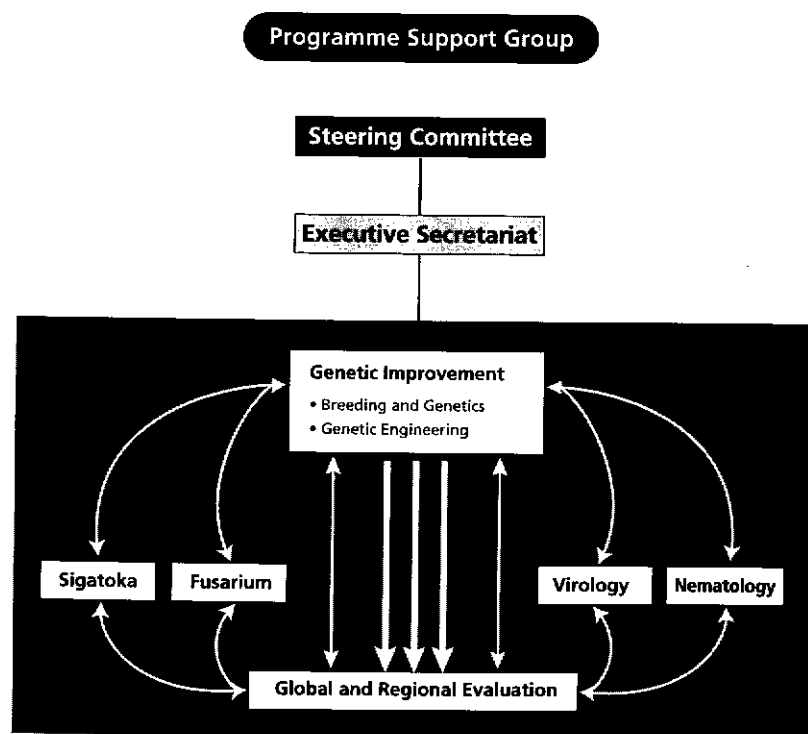
**Steering Committee:** The Steering Committee comprises representatives from NARS, ARIs and IARCs. In addition, the Chair of the Programme Support Group attends the Steering Committee meetings as an observer. This committee is responsible for proposing direction and providing oversight to the programme. It sets priorities based on technical advice from the working groups compiled by the Executive Secretariat and advises donors on the allocation of resources to the programme. The Steering Committee also approves the programme strategy, medium term plan and annual workplan. It commissions reviews of the programme, advocates on behalf of the programme and seeks external technical advice as appropriate.

**Executive Secretariat:** The Executive Secretariat is provided by INIBAP. It serves as the programme coordinator and is responsible for ensuring the smooth running of the programme as well as providing a programme secretariat. It also facilitates the organization of technical meetings, both thematic and interdisciplinary, and disseminates information to programme partners. It prepares reports and compiles lists of priorities, based on technical advice provided by the thematic working groups. Internal communication is a particularly important aspect of the programme, and the Executive Secretariat plays a critical role in stimulating contacts between groups. Regular inter-group information exchange is ensured through a programme newsletter, which is either published separately or included as a "PROMUSA" section in an existing newsletter. The Executive Secretariat also has an important role to play in providing feedback to the programme and ensuring a link with the end-users. The Executive Secretariat can also, if required, play the role of executing agency for funding provided to the programme.

**Thematic working groups:** The working groups are the heart of the programme. The members of these groups implement the programme workplan through a project portfolio which includes projects carried out by individual participants as well as collaborative projects involving a number of participants funded through various

mechanisms. Participation in the working groups is based primarily on capacity to contribute and comparative advantage, but will also depend on the priority research needs identified by each group. As the programme evolves, research priorities will change and this will necessarily result in changes in the make up of each working group. Working groups cover the major research needs, which at this stage include Genetic Improvement, Fusarium wilt, Sigatoka diseases, Nematodes and Viruses. This complement of working groups is however dynamic in nature and new groups may evolve during the lifetime of the programme.

Operating in small subject-specific working groups means that frequent interactions can occur between group members and flexibility is possible in the organization of group meetings. Each working group operates as a network within which research results, information, germplasm etc. are exchanged. In this way, each working group is able to ensure that it makes the best use of the resources available to the group as a whole and duplication of effort is minimized. Through the creation of synergies, the total output of the working group is greater than the sum of its component parts. Each working group develops a mechanism for ensuring regular contacts between group members and for facilitating information and material exchange between group members. Interdisciplinary contacts between the various working groups are also essential to address many of the major research needs and such links are therefore encouraged and facilitated.



## Programme Strategy and Medium Term Plan

### General Objectives

1. To increase the productivity of bananas and plantains produced for home consumption and local and export markets in an environmentally sustainable manner.
2. To foster the development of improved *Musa* varieties with a wide genetic base, and consumer acceptability and to disseminate these varieties to farmers through participating NARS.
3. To facilitate and stimulate partnerships among NARS, advanced research institutes, and IARCs to increase the efficiency and cost-effectiveness of global *Musa* improvement efforts.

### Specific Objectives

- To obtain the necessary basic scientific information to enable the production of a wide range of genotypes resistant to the major nematode pest species and to Sigatoka and Fusarium diseases.  
This will include:
  - Identification of sources of resistance - nematodes, Sigatoka, Fusarium;
  - Better knowledge of the types of resistance to nematodes, Sigatoka and Fusarium and an understanding of the inheritance of these traits;
  - Information on pathogenic variability and geographic distribution of major nematode pest species and of the Sigatoka and Fusarium fungi.
- Development of efficient breeding methodologies.  
This will include:
  - Broadened genetic base of material used by breeding programmes;
  - Identification of molecular markers and their use in marker-assisted breeding;
  - Development of biotechnological tools;
  - Integration of conventional breeding and biotechnology methodologies.
- Control of viruses in *Musa*.  
This is through:
  - Development of robust diagnostic systems for the major viruses affecting *Musa* in order to facilitate germplasm movement;
  - Production of transgenic virus-resistant clones.
- Evaluation and dissemination of improved varieties through a global and regional evaluation programme.

### Strategy

The production of improved, farmer accepted, *Musa* varieties through the development and application of conventional and biotechnological breeding approaches, incor-



porating resistance to pests and diseases to increase productivity and reduce pesticide use, operating in an environment in which collaborative partnerships and close interactions are fostered.

### Expected Outputs (5 years)

- New sources of resistance/tolerance to nematodes, Fusarium and Sigatoka identified and being used by breeding programmes;
- Fusarium and Sigatoka resistant varieties produced by conventional breeding approaches;
- Disease resistant hybrids of various types, including export bananas, plantains, Silk-type, Pome-type, cooking bananas, etc. being evaluated in multi- locational trials;
- Knowledge on the extent of adaptation of varieties that have passed through the evaluation programme;
- Database of agronomic and resistance/tolerance characteristics of the main varieties;
- Improved diploids available to breeding programmes;
- Molecular markers available for marker-assisted breeding schemes;
- Resistance constructs for banana bract mosaic virus (BBRMV), banana bunchy top virus (BBTV) and cucumber mosaic virus (CMV) developed;
- Diagnostic tests available for most major viruses including latent infections of BBTV;
- Basic information on banana streak virus (BSV) system for use in developing transgenic resistant plants;
- Efficient genetic transformation methods;
- Better understanding of host/pathogen relationships and mechanisms of resistance to nematodes, Fusarium and Sigatoka;
- Information on relationship between *Musa* varieties and various components of yield loss caused by nematodes;
- Fusarium and Sigatoka pathogenic diversity clarified.

### Expected Outputs (10 years)

- Fusarium/Sigatoka resistant clones developed through conventional and transgenic approaches;
- Nematode resistant clones developed through conventional and transgenic approaches;
- Virus resistant clones developed through transgenic approaches;
- Range of improved germplasm, with broad genetic base, available to NARS and farmers;
- Early screening tests developed;
- Resistance genes identified;
- More molecular markers available for marker-assisted breeding schemes.

## Working Group Reports

### Genetic Improvement Working Group<sup>1</sup>

#### 1. Scope of Work, Priority Research Needs and Major Constraints

It was noted that recent advances in the genetic improvement of *Musa* have been significant, with several hybrids of dessert banana, plantain and cooking banana being evaluated in many countries. Some of these hybrids have recently reached the stage of commercial production and marketing in a few countries.

Additional funding for on-going improvement efforts has therefore the potential to greatly enhance the production and release of better cultivars, both for the world export market and for the local consumption and market scenario.

##### 1.1 Scope of work

To produce improved *Musa* varieties through the integrated use of classical and biotechnological breeding approaches. In relation to classical breeding, the emphasis will be on research to improve the efficiency of breeding, including the development of marker-assisted breeding schemes, as well as on the enhanced utilization of plant genetic resources by breeders and on the greater exchange of breeding materials between breeders. In relation to genetic engineering, the scope of work is to develop efficient and transferable or generally accessible molecular biology and plant gene transfer tools for generating genetically modified bananas. Feasible methods identified are: transformation techniques through particle bombardment, *Agrobacterium tumefaciens* or protoplast electroporation as well as protoplast fusion techniques.

##### 1.2 Priority research needs

For the identification of priority research needs the Genetic Improvement Working Group divided into two sub-groups: the Breeding and Genetics sub-group and the Genetic Engineering sub-group.

(1) This report was prepared as a result of meetings held in Guadeloupe in which the following participants were involved: F. Bakry (CIRAD), F. Carreel (CIRAD), S. Clendennen (BTI), F. Côte (CATIE/CIRAD), J. Dale (QDPI), Do Nang Vinh (AGI), J-V. Escalant (CRBP/CIRAD), J- P. Horry (INIBAP), C. Jenny (CIRAD), D. Krigsvold (FHIA), N. Li (HKUST), K. Nair (KAU), S. de Olivera e Silva (EMBRAPA), G. Orjeda (INIBAP), A. Pires de Matos (EMBRAPA), P. Rowe (FHIA), L. Sagi (KUL), M. Smith (QDPI), R. Swennen (KUL), K. Tomekpe (CRBP), D. Vuylsteke (IITA). In addition, the following scientists were consulted subsequent to the Guadeloupe meeting: R. Gonsalves (Banana Board of Jamaica), S.C. Hwang (TBRI), P. Lagoda (CIRAD), M. Maluszynski (IAEA), G. May (BTI), H.P. Singh (NRCB), H. Tézenas du Montcel (CIRAD), A. Visser (ARC-ITSC), C. Vuillaume (CIRAD) and J. Zapata (IAEA).

### **Breeding and genetics sub-group**

An integrated approach to genetic improvement requires that all activities in the scope of the work should be undertaken. The highest priorities have been identified as follows:

- Enhanced utilization of plant genetic resources by breeding programmes;
  - Exchange of breeding materials and information;
  - Research into efficient breeding methodologies;
  - Marker assisted selection.
- **Enhanced utilization of plant genetic resources by breeding programmes**  
 Breeders need better access to existing *Musa* collections for increased availability of natural germplasm. Where gaps are identified in these collections, targeted exploration and collection of germplasm should be considered, with a particular focus on wild species. Such exploration should be coordinated by INIBAP/IPGRI. To improve access to existing collections, more effort should be put into good characterization of the germplasm and the ensuing databases/ information should be exchanged among collections and breeders. Existing working collections should also be supported in their germplasm conservation efforts.  
 This activity will result in a broader germplasm base in breeding programmes.
  - **Facilitation of the exchange of breeding materials**  
 Breeders' materials at various stages of improvement and of varying ploidy should be exchanged more vigorously among breeders. This can be facilitated by a better characterization of such materials and the exchange of information (including breeders' notes on combining abilities). This activity also requires faster and easier, but reliable virus diagnostics, which should be achieved both by increasing the INIBAP/Virus Indexing Centers (VICs) capacity and by decentralizing pathogen testing for regional germplasm movement.
  - **Research into efficient breeding methodologies**  
 Various schemes and approaches to genetic improvement should be tested to make banana breeding more efficient and to strengthen overall breeding efforts in order to better address national and regional needs.
    - Investigations into the genetics of important traits, e.g. disease/pest resistance and quantitative traits.
    - Marker assisted selection, particularly for screening of traits that are difficult or slow to evaluate, as such increasing the efficiency of breeding and selection.
    - Multilocational evaluation of germplasm to investigate genotype-by-environment interaction and to determine the stability of important traits.
    - Research into somaclonal variation and mutation induction as tools for genetic improvement, including the molecular analysis of the basis of somaclonal variation.
  - **Marker assisted selection**  
 Research related to molecular markers links with the work carried out by the genetic engineering working group. The following research areas are therefore applicable to both groups:

- Molecular mapping for marker assisted selection to identify single genes and genome segments for use in transformation of cultivars and breeding materials.
- Development of efficient recombinant DNA techniques for isolation and introgression into *Musa* of genes covering a wide array of desirable traits.

### **Genetic engineering sub-group**

- **Transformation protocol** (development of efficient transformation systems, including upstream and downstream tissue culture activities)  
 Development of pre-transformation tissue culture systems, which do not cause somaclonal variation, for the different varieties of interest. This would include investigations into genotype effect on the development of cell suspensions and the development of meristem cultures with a high capacity for regeneration.  
 Development of reliable constructs for transformation.  
 Development of a molecular tool box for controlled gene expression.  
 Priority ranking: High
  - **Availability of promoters**  
 There is a need to identify all kinds of strong promoters for transgene expression, including promoters that can be obtained from banana plants.  
 Priority ranking: High
  - **Molecular genetics**  
 Gene mapping;  
 Library construction (Bi-BAC);  
 Comparison of sequence homology between genotypes;  
 Cloning genes of important traits;  
 Transposon-tagging and T-DNA-tagging;  
 Priority ranking: High
  - **Selection markers, alternatives to antibiotics**  
 Priority ranking: Medium
  - **Development of early *in vitro* selection methods/mediums for identifying somaclonal variants with interesting traits.**  
 Priority ranking: Medium
  - **Development of *in vitro* early mutagenesis for producing plants with interesting traits.**  
 Priority ranking: Low.
- 1.3 Major constraints
- Short-term nature of funding - this is of particular relevance to genetic improvement programmes, which are expensive and time-consuming and require continuous support on a long-term basis.
  - Inadequate linkages between genetic engineering and conventional breeding programmes: these links need to be improved and facilitated especially for:
    - a. evaluation of transgenic products in the field
    - b. transformation of breeding lines

- c. evaluation of map-based cloning of genes of important agronomic traits
- Delayed access to virus indexed material due to time consuming virus indexing procedures.
- Intellectual property rights: Patents on constructs, products, etc. may prove to be a constraint in certain circumstances but this will vary depending on the type of product, the destination of the product (export, local, consumption) etc.
- Inconsistent national biosafety regulations: The fact that these regulations vary from one country to another is a limiting factor and has an influence on several activities including the use of particular constructs and selection markers and field evaluations.
- Access to national plant genetic resources.
- Lack of marketing information.
- Lack of acceptance of new export bananas into the established marketing channels.
- Lack of good characterization of host plant/pathogen interaction.

## 2. Research Strategy

The working group recognises that there are a number of approaches for the improvement of *Musa*:

- Breeding
- Genetic engineering
- Mutation.

The group recognises that the best strategy to strengthen *Musa* improvement is through the close integration of classical and genetic engineering breeding approaches and through the sharing of breeding material and knowledge among group members.

- The Executive Secretariat should facilitate the exchange of material and information;
- The different groups need to improve their tools, particularly in relation to the further development of marker assisted selection;
- PROMUSA should establish "resource centres" from which material could be obtained for genetic improvement research (e.g. cell cultures, libraries, vectors, Agrostrains etc.).

## 3. Areas for Collaboration

- Germplasm evaluation, including multilocational trials, such as IMTP.
- Marker assisted selection, exchange of markers.
- Integration of biotechnology and conventional breeding programmes, including testing of transgenics.

### 3.1 New areas for collaborative research

- Integration of somaclonal variation and conventional breeding.
- Information on available germplasm and breeding materials.

- Scientist exchange.
- Development of a breeding programme in Southeast Asia.

### 3.2 Identification of research needs requiring inter-group collaboration:

#### *Genetic engineering - Virology groups*

- Introduction of resistance to virus is thought to be feasible only by genetic transformation.
- A possible risk of activating BSV integration sequences through transformation is recognized and requires a collaborative approach.

#### *Genetic improvement - Nematode, Fusarium, Sigatoka groups*

- Isolation/introduction of mapped resistance genes.

## 4. Inventory of Existing Facilities, Expertise and On-going Research

### 4.1 Existing facilities and expertise

#### *Breeding and genetics sub-group*

|                          |  |
|--------------------------|--|
| CRBP<br>Cameroon         | Focus on breeding of plantains and other locally important varieties; large collection; diploid segregating populations; availability of different agroecologies; regional approach; contacts with NARS. |
| FHIA<br>Honduras         | Currently oldest active breeding programme with large collection of improved diploids; core funding; focus on dessert banana, plantain and cooking banana.   |
| CIRAD<br>France          | Focus on dessert banana breeding; different strategies; support from molecular biology and cell biotechnologies; large collection of natural and improved diploids.                                      |
| EMBRAPA<br>Brazil        | Focus on Silk and Pome bananas; availability of different agroecologies; can screen for resistance to yellow Sigatoka, Fusarium and <i>Pseudomonas solanacearum</i> race 2.                              |
| VASI<br>Vietnam          | Good collection, possibility of testing for Fusarium wilt; improvement of Cavendish.   |
| IITA<br>Nigeria          | Focus on plantain and cooking banana breeding; support from molecular biology; broad regional approach and good contacts with NARS; core funding.  |
| ARC-ITSC<br>South Africa | Nursery evaluation for Fusarium resistance.  |
| Banana Board<br>Jamaica  | Oldest collection in Latin America and the Caribbean; 4X x 2X breeding; synthetic 2X breeding; 4X resistant to Sigatoka and Fusarium   |

A list of acronyms and abbreviations is provided at the end of the document.



**Genetic engineering sub-group**

| Research area          | Transformation protocol |               | Promotors | Mapping | Gene cloning |
|------------------------|-------------------------|---------------|-----------|---------|--------------|
|                        | Tissue culture activity | Gene transfer |           |         |              |
| Institute              |                         |               |           |         |              |
| BTI, USA               | XX                      | XX            | X         | -       | X            |
| CATIE, Costa Rica      | XX                      | X             | -         | -       | -            |
| CICY, Mexico           | XX                      | ?             | ?         | ?       | ?            |
| CIRAD, France          | XX                      | XX            | X         | XX      | -            |
| CRBP, Cameroon         | X                       | -             | -         | -       | -            |
| Cuba                   | XX                      | X             | ?         | ?       | ?            |
| HKUST, Hong Kong       | X                       | -             | XX        | -       | XX           |
| KUL, Belgium           | XX                      | XX            | X         | -       | -            |
| QUT, Australia         | XX                      | XX            | XX        | -       | -            |
| Univ. Hawaii, USA      | X                       | X             | -         | -       | -            |
| Univ. Paris XI, France | XX                      | X             | -         | -       | -            |
| EMBRAPA/CNPMPF, Brazil | XX                      | X             | -         | X       | X            |
| ARC-ITSC, South Africa | XX                      | X             | -         | -       | -            |

A list of acronyms and abbreviations is provided at the end of the document.

X: have the relevant expertise in their domain XX: have the relevant expertise and have already obtained positive results.

## 4.2 Inventory of on-going research

| Institute     | On-going activity  | Expected output   | Time frame |
|---------------|--|---|------------|
| FHIA Honduras | Breeding for replacement for the Cavendish export banana | One, or more, black Sigatoka-resistant, dwarf hybrids with export qualities. This is being done by crossing disease-resistant, agronomically-improved bred diploids onto the Lowgate dwarf mutant of Gros Michel. Development of the FHIA-01 banana has shown that this 3X x 2X approach to genetic improvement is effective. | 5 years    |
|               | Breeding for dwarf, disease-resistant plantains          | One or more hybrids with the desired characteristics. Development of the FHIA-21 plantain hybrid has validated the approach being employed in plantain improvement.   | 5 years    |
|               | Breeding for dwarf, disease-resistant cooking bananas    | One or more hybrids for evaluation in East Africa. The FHIA-03 cooking banana already developed in this breeding objective is being cultivated commercially in Cuba.  | 5 years    |

| Institute      | On-going activity  | Expected output   | Time frame |
|----------------|--|---|------------|
| ITA Nigeria    | Enhanced <i>Musa</i> germplasm utilization   | Germplasm with desirable alleles available and used. Main focus is on ideotype breeding and multi-trait selection, and population improvement based on combining abilities. Desirable traits include Sigatoka, virus, nematode, Fusarium, weevil resistance, better root systems and good fruit quality.  | 3 years    |
|                | Improved plantain and banana genotypes and populations                                       | At least 10 improved genotypes tested in multilocational trials in at least 8 African countries. The breeding approach includes conventional and non-conventional cross-breeding, polycross breeding, varietal mixtures, and international dissemination of improved genotypes. Hybrids should include high and stable yield, resistance/tolerance to biotic and abiotic stresses, desirable plant habit, and good fruit quality. | 4 years    |
| EMBRAPA Brazil | Breeding for dwarf, disease and pest resistant bananas (including Moko resistance)           | One or more hybrid (Pome-type) resistant to black and yellow Sigatoka. One or more Fusarium resistant Silk hybrids. One or more hybrids resistant to Moko. All hybrids tested under farmers field conditions.   | 5 years    |
| CRBP Cameroon  | Plantain improvement by 3X x 2X → 4X   | Dwarf plantain hybrids with resistance to black Sigatoka and <i>R. similis</i> , virus indexed and evaluated in multi-locational trials in West and Central Africa. Improved plantain-like male parental lines developed and tested.  | 5 years    |
|                | Production of Plantain hybrids by 2X → colchicine → 4X x 2X → 3X. (Collaboration with CIRAD) | Improved 3X plantain hybrids tested in multilocation trials in West and Central Africa. New crosses ABAB x AA or BB.  | 5 years    |
|                | Diploid improvement for improved plantain-like parental breeding lines                       | Creation of disease and pest resistant plantain-like AB hybrids (BB x AA cv) for the triploid breeding scheme ABAB x AA cv.   |            |
|                | Germplasm characterization   | About 400 natural accessions evaluated for agro-morphologic traits, fruit quality and resistance to disease and pests.  | 3 years    |
|                | Plantain improvement by tetraploid breeding  | Selection of 10 or more tetraploid plantain hybrids resistant to BLS and other parasites for global and regional evaluation.  | 3 years    |

| Institute               | On-going activity   | Expected output  | Time frame |
|-------------------------|---|--|------------|
| CRBP<br>Cameroon        | Diploid improvement   | Twenty or more resistant plantain and cooking-type AA and AB for tetraploid and triploid breeding.   | 3 years    |
| (cont'd)                | AAB triploid breeding   | 10 or more plantain and cooking-type hybrids resistant to disease and pests available for global and regional evaluation.  | 5 years    |
|                         | Formal genetics   | Four or more diploid segregating populations. Better knowledge of the genetics of resistance and major agronomic traits.   | 3 years    |
| CIRAD<br>France         | Triploid breeding: production of AAA and AAB dessert bananas  | Creation of triploid hybrids resistant to BLS, YS, <i>R. similis</i> and Foc for exportation and local market in different countries.  | 5 years    |
|                         | Enhanced <i>Musa</i> germplasm characterization and utilization   | Development of a database. Precise identification of the relationships between cultivated triploids and related diploids.  | 5 years    |
|                         | Genetic mapping   | Research of QTL. Identification of genes of agronomic interest coming from banana genomes.   | 5 years    |
| Banana Board<br>Jamaica | Breeding varieties suitable for food and commercial production. Tetraploids and triploids resistant to black Sigatoka, Foc and nematodes. | Varieties (tetraploid) resistant to black Sigatoka are being crossed by diploids 4X x 2X and 4X x 2X => 3X. One very promising triploid has been produced so far. Continuation of this approach seems to be on a progressive path. | 5 years    |
| CATIE<br>Costa Rica     | <i>In vitro</i> regeneration methods  | <i>In vitro</i> cryopreserved cell suspension from different cultivars.  | 3 years    |
|                         | Methods of transformation   | Efficient methods of transformation.   | 3 years    |
| HKUST<br>Hong Kong      |   | Completion of sequencing two ACO and two ACS genomic DNA clones.   | 2 years    |
|                         |   | Study of the differential regulation of the gene expression of ACO and ACS.  | 2 years    |
|                         |   | Subcloning of the promoters that confer fruit or leaf specificity.   | 2 years    |

| Institute                   | On-going activity  | Expected output   | Time frame         |
|-----------------------------|--|---|--------------------|
| University of Hawaii<br>USA | <i>Agrobacterium</i> /meristem transformation system<br><br>Gene gun/cell suspension transformation system<br><br>Embryogenic callus transformation system | Efficient and reliable systems.   |                    |
| CIRAD<br>France             | <i>In vitro</i> regeneration and gene transfer method<br><br>Genetic engineering/mapping/gene cloning  | Efficient methods of transformation.<br><br>Better knowledge of the origin and genetic bases of the resistances/genes of interest.          |                    |
|                             | Genetic engineering (particle bombardment, <i>Agrobacterium</i> transformation)  | Transformed plants with exogenous genes expressed with improved agronomic/pest-disease resistance traits.                                   | 1997 onwards       |
| BTI<br>USA                  | Structural genes from banana. Promoter characterization  | Constructs for tissue-specific expression in banana. Bi-Bac library construction.   | 2 years            |
| ARC-ITSC<br>South Africa    | <i>In vitro</i> regeneration methods<br><br>Method of transformation   | Efficient and reliable systems for regeneration.<br><br>Gene gun.   | 2 years<br>2 years |
|                             | <i>In vitro</i> evaluation for cold tolerance  |   | 3 years            |
|                             | Nursery evaluation for Fusarium resistance   |   | On going           |
| John Innes Centre<br>UK     | AFLP mapping   | Technique is currently up and going in many crops, e.g. cereals, yams, cowpeas, other legumes etc. and is being applied to BSV integration. |                    |

| Institute                     | On-going activity                               | Expected output  | Time frame |
|-------------------------------|---|--|------------|
| John Innes Centre UK (cont'd) | Fluorescent <i>in situ</i> hybridization (FISH) | This is also being used for the BSV work and on A & B genotypes. For banana, it is in the early stages but is being widely used for other systems. Sensitivity is currently at low copy number genes but progressing towards single copy. Also being used for mapping retrotransposons.  |            |
|                               | Comparative mapping                             | Much work in comparative mapping of cereals (demonstration of colinearity) is now being extended to other monocots. There would be an interest at the JIC in involving <i>Musa</i> in this. With cereals it is proving very useful in identifying (and isolating) genes especially those for disease resistance.   |            |
| KUL Belgium                   | <i>In vitro</i> culture techniques              | New embryogenic cell suspensions. Embryogenic cultures from a wide-range of cultivars.   | 3 years    |
|                               | Genetic transformation                          | Large number of independent transgenic plants with useful gene constructs.   | 3 years    |
|                               | Molecular genetics                              | Characterization of novel promoters in banana. Molecular characterization of fungus/host interactions.   | 3-5 years  |
| IEB Czech Republic            | Flow cytometry                                  | Flow cytometry is used to screen ploidy levels and to determine nuclear genome size. The technique is being improved to detect aneuploids and to determine nuclear genome composition in triploids and tetraploids.  |            |
|                               | Molecular cytogenetics                          | Fluorescence <i>in situ</i> hybridization is used to localize repetitive DNA sequences on <i>Musa</i> chromosomes and is being improved to localize low and single copy sequences. Genomic <i>in situ</i> hybridization is used to characterize nuclear genome composition. Primed <i>in situ</i> DNA labelling is being tested for localization of microsatellites. |            |
|                               | Molecular genetics                              | Considering extensive experience in other crops, a possibility to construct chromosome specific DNA libraries in <i>Musa</i> is being considered.  |            |

A list of acronyms and abbreviations is provided at the end of the document.

## Medium-Term Plan

| Goal  | Inputs   | Outputs   | Indicators  |
|---|--|---|---|
| Broaden the genetic base of breeding programmes       | Targeted collecting. Germplasm collections. Characterization information.                          | Breeding lines with broad genetic base.   | New breeding lines being used in the development of pest and disease resistant hybrids. |
| Production of pest and disease resistant hybrids      | Germplasm. Equipment and expertise. Evaluation information on populations and hybrids.             | Segregating populations. Improved hybrids.  | Improved hybrids available to farmers.  |
| Increased knowledge of <i>Musa</i> molecular genetics | Equipment and expertise.   | Gene maps. Library construction (Bi-BAC). Information on sequence homology between genotypes. Cloned genes of important traits. Transposon-tagging and T-DNA tagging.   |   |
| Identification of selection markers                   | Equipment and expertise.   | Identification of selection markers as alternatives to antibiotics.   |   |
| Identification of molecular markers                   | Equipment and expertise.   | Molecular markers for identifying traits of interest.   | Molecular markers used in breeding.   |
| Development of transformation protocol                | Upstream and downstream tissue culture activities. Equipment and expertise. Biosafety information. | Transformed plants. Efficient system for obtaining cell suspensions or meristem culture. Reliable constructs for transgene formation. Strong promoters for transgene expression. Molecular tool box for controlled gene expression. | Transgenic plants being tested in field.  |
| Production of somaclonal variants/mutants of interest | Germplasm. Equipment and expertise.  | Method for the early <i>in vitro</i> screening of somaclonal variants for traits of interest. Method for early <i>in vitro</i> mutagenesis. Improved varieties.   | Availability of plants with traits of agronomic interest available for farmers.         |



## Sigatoka Disease Working Group<sup>1</sup>

### 1. Scope of Work, Priority Research Needs and Major Constraints

#### 1.1 Scope of work

Sigatoka leaf spot diseases of bananas involve two related pathogenic ascomycete fungi: *Mycosphaerella fijiensis* Morelet causing black leaf streak disease (BLS) and *Mycosphaerella musicola* Leach ex. Mulder, causing Sigatoka disease (SD). *M. fijiensis* is characterized by its stronger pathogenicity on a broader range of hosts, making BLS the most destructive leaf disease of bananas (AAA), plantains (AAB) and other cooking bananas.

Research on BLS is required in order to develop effective strategies to maintain the production of bananas and plantains and is of the highest urgency. Integrated solutions are needed, including the selection of resistant clones, cultural practices and rational chemical control. Because of the world-wide nature of the problem, it is necessary to focus on research in the following areas in close collaboration with breeding programmes.

- Expansion of knowledge on the types of resistance expression (phenotypes);
- Assessment of pathogenic variability and distribution of pathogenicity of *M. fijiensis* and *M. musicola*;
- Identification of new sources of resistance;
- Development of screening methodologies for resistance to black leaf streak and Sigatoka disease (inoculation under controlled conditions, biochemical markers, screening using toxins);
- Determination of the status of Sigatoka diseases in some areas, mainly in Asia;
- Identification of the different gene interactions during the polycyclic disease development (components of resistance) through epidemiological studies;
- Improvement of disease evaluation methods to improve the accuracy of the assessment of resistance phenotypes.

#### 1.2 Priority research needs

##### **Pathogenic variability of the pathogens**

Since BLS has a perfect stage, recombination of genes can occur inducing the appearance of new pathotypes. The geographical genetic diversity of *M. fijiensis* at the global level has already been investigated. However, there is some evidence for the existence of pathogenic variability as well. It is therefore essential to have a better understanding of the structure of pathogen populations in order to make the genetic

(1) This report was prepared as a result of meetings held in Guadeloupe in which the following participants were involved: H. Fagan (WIBDECO), E. Fouré (CIRAD), X. Mourichon (CIRAD), A. Pires de Matos (EMBRAPA), R. Romero (CORBANA), S. Tripon (INIBAP), P. Vi Nai (MAF). In addition, the following scientists were consulted subsequent to the Guadeloupe meeting: R. Fullerton (Hort Research), A. Johanson (NRI), R. Peterson (QDPI), and W. Tushmehirwe (NARO).

improvement of bananas more efficient and for the resistance to be durable. Research in the following areas is necessary:

- estimation of the extent and distribution of genetic variability within *M. fijiensis* and *M. musicola* populations using molecular markers (neutral markers) and to relate this information to pathogenic variability;
- quantification at the greenhouse level of the extent of pathogenic variability of the two pathogens by using a differential set of *Musa* cultivars (pathotypic markers). A set of pathotypes that represent the full pathogenic diversity of the pathogen populations will be selected in terms of "virulence and aggressiveness";
- detection at the field level in several areas the presence of pathogenic strains by planting large plots of selected or reference materials used as a source of resistance to maintain a greater selection pressure.

##### **Development of early screening methods**

The following strategies are suggested for research:

- Artificial inoculation under controlled conditions**

Inoculation procedures must be available for assessing qualitative (highly resistant) as well as quantitative (partially resistant) expressions of resistance using a set of pathotypes that represent the full diversity of the pathogen (pathogenicity/aggressiveness in populations).

Inoculation procedures to assess qualitative (vertical) resistance are simple and do not require rigorous control of inoculum and incubation conditions. On the other hand, for quantitative (horizontal) resistance, inoculum level, host age, and incubation and post-inoculation conditions, must be strictly controlled.

Artificial inoculation should not be so severe as to overestimate the capacity of an agent to cause disease or to under-evaluate the resistance of the host. Factors to be assessed will include:

- qualitative and quantitative differences in susceptibility,
- quality of inoculum (type, age, concentration of conidia, etc.)
- physiological age of acclimatized vitroplantlets,
- environmental conditions prior to, and especially after inoculation.

The correlation between the behaviour of young material in controlled conditions (growth cabinet/glasshouse) and the behaviour of mature plants in the field will be evaluated in using a set of reference banana cultivars already characterized for the field reaction to BLS and SD.

- Use of toxins produced by *M. fijiensis***

The role of toxins in pathogenicity remains unclear, therefore, research aimed at clarifying this relationship is important before toxins can be used for screening germplasm. Research is required in order to:

- investigate the relationship between the level of the resistance to the infection (under natural field inoculation) and the sensitivity to the toxic compound produced *in vitro* by *M. fijiensis*.

- compare of the behaviour of intact plants towards the toxins (detached leaf assays) and the sensitivity of banana tissue expressed *in vitro* (callus and cell suspensions).

The results will be used to develop an appropriate screening technique. The toxins can be applied to the detached leaves of plantlets or tissue cultured *in vitro* (callus, cell suspensions, protoplasts), if it is confirmed that toxin tolerance will be expressed in the regenerated plants.

### New sources of resistance

It appears to be necessary to look for new sources of resistance in areas where host and pathogen co-evolution have been demonstrated (Southeast Asia).

### Epidemiological studies

Sigatoka diseases are characterized by being polycyclic. Resistance in the host results from the effect on different components of the disease. It is essential to assess the weight of each epidemiological sequence (each gene interaction) in the resistant phenotype. These elements will be very useful for molecular mapping (QTL approach).

#### 1.3 Major constraints

- Lack of expertise in many areas (relatively few institutes working on this disease)
- Insufficient infrastructure
- Need for integration of plant pathology with breeding programmes.

## 2. Research Strategy

(a) Select a coordinator to identify possible collaborators for research in the following three main areas:

- mechanisms of resistance
- pathogen variability
- epidemiological studies.

(b) Bring the collaborators together to identify their potential contributions in the collaboration and develop research proposals in each area. The coordinator will identify which aspects of the proposals would require funding. The group should be open to additional collaborators with expertise as needed.

## 3. Areas for Collaboration

For the evaluation of pathogenic variability, collaboration is sought between banana producing countries (Costa Rica, Cameroon, Philippines, Tonga) and banana non-producing countries where this is possible because of quarantine regulations. To obtain a large collection of *M. fijiensis* and *M. musicola* isolates with good coverage of the different production zones (Southeast Asia, Pacific, Africa and Latin America), the working group propose:

- Close collaboration among all interested partners in supplying isolates, that will be further characterized for pathogenicity virulence or aggressiveness.

- Collaboration is also sought to conduct field trials in the four main areas, where genetic structure of *M. fijiensis* is already demonstrated (Southeast Asia, Africa, Latin America, Pacific) to detect pathogenic strains and host-parasite specific interactions (using reference banana clones). At this level, some areas of collaboration between Bureau of Plant Industry (BPI, Philippines), CRBP (Cameroon), *Corporación Bananera Nacional* (CORBANA, Costa Rica) and Ministry of Agriculture and Forestry (MAF, Tonga) can be expected.

The collaboration between CIRAD, University of Gembloux and CRBP should continue and be strengthened in order to increase knowledge on resistance mechanisms.

Similar studies on pathogenic variability and resistance mechanisms can be conducted for *M. musicola* through collaboration between Australia (QDPI), Brazil (EMBRAPA), Caribbean (WIBDECO, St Lucia).

#### 3.1 New areas for collaborative research

- Role of toxins in the pathogenicity of both pathogens: *Mycosphaerella fijiensis* and *Mycosphaerella musicola*
- Mechanisms of host resistance.

#### 3.2 Identification of research needs requiring inter-group collaboration

Stronger and continuous collaboration is needed between the Sigatoka working group and the Genetic Improvement group. The main areas of collaboration could be as follows:

- Identification of new sources of resistance, mainly at the diploid level. Screening in controlled conditions will allow the selection of lines for resistance by using pathotypes which represent the full diversity of pathogen populations.
- Improved accuracy of assessment methods for the inheritance of BLSA thus allowing each component of the disease cycle, i.e. gene interaction between the host and the parasite, to be distinguished. Such information is essential to map the genes involved in partial resistance (QTL work).

Interaction with the IMTP group is necessary to provide useful indications for a more appropriate selection of IMTP sites (according the pathogenic structure).

## 4. Inventory of Existing Facilities, Expertise and On-going Research

### 4.1 Existing facilities and expertise

|                            | Expertise | Lab. | Staff | Field | Greenhouse |
|----------------------------|-----------|------|-------|-------|------------|
| CIRAD, France              | yes       | yes  | yes   | no    | yes        |
| CORBANA, Costa Rica        | yes       | yes  | yes   | yes   | yes        |
| CRBP, Cameroon             | yes       | yes  | yes   | yes   | yes        |
| EMBRAPA/CNPMF Brazil       | yes       | yes  | yes   | yes   | yes        |
| MAF, Tonga                 | yes       | yes  | yes   | yes   | yes        |
| WIBDECO, Windwards Islands | yes       | no   | yes   | yes   | no         |

A list of acronyms and abbreviations is provided at the end of the document.

## 4.2 Inventory of on-going research

| Institute                                     | On-going research  |
|---|--|
| CIRAD<br>France                               | Host pathogen interaction (s).<br>Genetic diversity within <i>M. fijiensis</i> - Assessment of pathogenicity.<br>Early screening methods.<br>Diagnostic of BLSD and SD.  |
| CORBANA<br>Costa Rica                         | Epidemiological studies.<br>Pathogen variability and fitness related to fungicide resistance.<br>Field resistance screening.   |
| CRBP<br>Cameroon                              | Epidemiological studies.<br>Phenotype expression of resistance.<br>Field resistance screening.   |
| EMBRAPA/CNPMF<br>Brazil                       | Field resistance screening.<br>Variability within <i>Mycosphaerella musicola</i> .   |
| Hort. Research<br>New Zealand                 | Variability of pathogenic populations.   |
| IITA, Nigeria                                 | Epidemiological studies.   |
| NRI<br>UK                                     | Study of the distribution of <i>Mycosphaerella</i> species in East Africa;<br>large collection of isolates of <i>Mycosphaerella</i> species from around the world; capability in molecular diagnostics; analyses of molecular variability;<br>greenhouse pathogenicity testing; work on epidemiological aspects of the disease, in particular spore dispersal in the field in Central America and East Africa. |
| QDPI/University<br>of Queensland<br>Australia | Genetic diversity with <i>Mycosphaerella musicola</i> .  |
| University of<br>Gembloux<br>Belgium          | Characterization of resistance mechanisms.<br>Role of toxins in pathogenicity.   |

A list of acronyms and abbreviations is provided at the end of the document.

## Medium-Term Plan

| Goal                                | Inputs  | Outputs   | Indicators  | Time Frame |
|-------------------------------------|---|---|---|------------|
| Pathogenic variability of pathogens | Expertise and equipment.<br>Molecular markers.<br>Equipment and land for greenhouse and field screening.  | Provision of criteria for selection of appropriate field testing sites for resistance. Basic information on the pathogen.<br>A set of pathotypes that represent the full pathogenic diversity of the pathogen populations | Field sites being used for screening.<br>Availability of molecular markers.<br>Set of pathotypes available. | 5 years    |
| Early screening                     | Equipment and expertise.<br>Source of high quality plantlets. A set of pathotypes that represent the full pathogenic diversity of the pathogen. | A methodology for early screening of resistance useful to breeding programmes.<br>Correlation between greenhouse behaviour and field behaviour.<br>Clarification of role of toxins in pathogenicity.                      | Sources of resistance being used by breeders.   | 5-10 years |
| New sources of resistance           | Collecting missions in Southeast Asia.  | New sources of resistance available to breeders.<br>Widened genetic base of resistance.   | Sources of resistance being used by breeders.   | 5 years    |
| Epidemiological studies             | Expertise and equipment.  | Understanding of host-pathogen relationships.   |   | 5-10 years |
| Mechanisms of resistance            | Expertise and equipment.  | Identification of mechanism of resistance for use by breeders.<br>Identification of resistance genes.<br>Information for use in molecular mapping.  | Availability of molecular markers.  | 5-10 years |
| Status of Sigatoka disease in Asia  | Surveys.<br>Collecting missions.  | Information on the relative importance of black and yellow Sigatoka throughout Southeast Asia.  |   |            |



## Fusarium Wilt Working Group<sup>1</sup>

### 1. Scope of Work, Priority Research Needs and Major Constraints

#### 1.1 Scope of work

The Fusarium Wilt Working Group will continue to concentrate on the assessment of genetic and pathogenic diversity in *Fusarium oxysporum* f. sp. *cubense* (Foc) and on surveying the geographic distribution of genetic and pathogenic variants of the pathogen. Efforts will be made to identify additional parents which resist Fusarium wilt, especially tropical race 4 (VCG 01213-01216), as well as advanced hybrids of the different types (e.g. sweet-acid dessert, export dessert, etc.). Where appropriate segregating populations of the host and sufficient resources exist, marker-assisted selection will be investigated. Work will also be carried out to investigate why tissue cultured plantlets are more susceptible to this and other diseases and the means by which their vigour can be enhanced.

#### 1.2 Priority research needs

##### **Early screening test**

The ability to score Foc isolates for virulence on different banana clones and different clones for resistance to pathotypes of Foc would assist the breeding programmes and allow the informed deployment of germplasm to various producing regions. Expansion of existing facilities and resources would allow significant research in this area to be conducted.

##### **Field screening of parents and clones**

Field screening has been and will likely continue to be the primary means by which resistance is identified for the near future. Moreover, it will remain the only way in which the agronomic performance of clones can be assessed for the foreseeable future. Thus, significant on-going activities in this area should continue.

##### **Genetic variation in Foc**

To date, considerable progress has been made in characterizing the extent and geographic distribution of genetic variation in this pathogen. However, large and important production areas have not been assessed in Asia, Africa and the Americas. Continued work on the genetic diversity and phylogeny of populations of Foc is needed, especially in the non-explored areas.

(1) This report was prepared as a result of meetings held in Guadeloupe in which the following participants were involved: Z. de Beer (ARC-ITSC), K. Pegg (QDPI), A. Pires de Matos (EMBRAPA), R. Ploetz (Univ. of Florida), M. Smith (QDPI), R. Valmayor (INIBAP-ASPNET). In addition, the following scientists were consulted subsequent to the Guadeloupe meeting: J. Hernández (ICIA), A. Kangire (NARO), N. Moore (QDPI), M. Rivera (FHIA) and M. Rutherford (CAB-IMI).

#### **Marker-assisted selection**

Very little has been done in this area to date. Two F<sub>2</sub> populations have been produced for use in such work (see constraint #5), but it is not clear at this time whether better or different populations are needed. Despite the very great expense of such research, work in this area is warranted.

#### **Plantlet vigour**

Recent research in Australia has clearly demonstrated the greater susceptibility of tissue culture plantlets to Fusarium wilt when compared to conventional seed pieces. Research to elucidate the cause(s) for this phenomenon and means by which plantlet vigour could be increased and susceptibility decreased is needed.

#### 1.3 Major constraints

- Quarantine regulations restrict the use of nonendemic isolates of the pathogen in virtually all producing areas (the plantlet test described for UF-Homestead are conducted under APHIS-USDA permit). These restrictions constrain the scope of work that can be conducted in this area.
- Expertise and infrastructure are limited in the key areas in which tropical race 4 is found (Halmahera, Java, peninsular Malaysia, Sulawesi, Sumatra and Taiwan). In the two best locations, MARDI-Malaysia and Solok-Sumatra, routine screening of parents and hybrids for resistance should be possible but training of national scientists and the assistance of pathologists from other locations may be required.
- Lack of facilities world-wide constrain the use of the plantlet screening test developed at UF-Homestead. Considering the need for replication (at least six) and for internal controls of the race differentials, it is necessary to have increased incubator capacity in order to screen several clones or pathogen strains at the same time.
- Although the free exchange of improved parental lines among all interested parties is desirable, this is seldom the case. The delayed access to improved hybrids due to slow virus indexing techniques is also a constraint to *Musa* improvement research.
- Breeding programmes may be encouraged by collaborating organizations to provide advanced hybrids in exchange for information on the performance of parents and hybrids.
- It is not clear whether good segregating populations exist for determining the inheritance of resistance to diverse pathotypes of Foc or for developing molecular markers for use in breeding programs. Two F<sub>2</sub> populations have been created for such work. One (CIRAD-FLHOR) utilizes relatively homozygous parents of unknown, but assumed resistance to Fusarium wilt, whereas the other (FHIA) utilizes heterozygous parents of partially characterized resistance.

## 2. Research Strategy

Early screening tests have been developed but these require strengthening in order to produce significant information on pathogenic diversity in Foc and on resistance in the host. On-going field screening efforts will continue to provide much needed information

on the resistance of parents and progeny to the important pathotypes of Foc. As breeding (conventional and biotechnology) and the recognition of variants of Foc continues, this effort will need to be expanded. In addition, the results obtained from early screening will have to be verified in the field. Studies on the variation in Foc will continue with further collections being made in important production areas in Asia, Africa and the Americas. The efficiency of breeding efforts will be increased through the identification of markers which are linked to resistance and efforts will be made to enhance the hardiness of tissue culture plantlets.

### 3. Areas for Collaboration

Informal links established between University of Florida, QDPI and ARC-ITSC and breeding programmes;

University of Florida with FHIA and IITA;

ARC-ITSC with FHIA and CIRAD (in future);

QDPI with FHIA and CIRAD;

Internal link between University of Florida, Homestead and University of Florida, Gainesville;

Internal link between QDPI and CRCTPP and QDPI and QUT;

External link between ARC-ITSC and TBRI (Taiwan);

Need to develop close links with NARS on 01213/01216 areas e.g. MARDI, CRIH, synergies created where mutual benefit;

EMBRAPA/CNPMF with QDPI.

#### 3.1 New areas for collaborative research

##### *Early screening test - Fundamental to success of PROMUSA*

Requires a 3-way collaboration between a tissue culture laboratory supplying good quality plantlets, the early screening centre, and a field site to verify results.

##### *Genetic variation*

Inability to collect in some areas due to funding constraints.

##### *Field screening*

No site for detailed studies with 01213/01216 - sites available in Malaysia and Indonesia but clear mutual benefit must be provided.

##### *Molecular markers*

Need to identify suitable F<sub>2</sub> segregating populations. Ideally there should be a black and white situation for resistance/susceptibility - also need to study inheritance of resistance.

#### 3.2 Identification of research needs requiring inter-group collaboration

- Molecular Markers - need to collaborate with genetic improvement group.
- Evaluation of diploids and progeny - need to collaborate with genetic improvement group and IMTP.
- Hardiness of tissue culture plants - need to collaborate with nematology group.
- For pathogen diversity studies - need to collaborate with IMTP.

## 4. Inventory of Existing Facilities, Expertise and On-going Research

### 4.1 Existing facilities and expertise

| Organization                       | STRENGTHS  |  |   |   |  |
|------------------------------------|--|--|---|---|--|
|                                    | Early screening test   | Pathogen characterization  | Field evaluation  | Marker-assisted selection   | Plantlet vigour  |
| QDPI Australia                     | No, cannot use non-endemic isolates.   | VCGs, volatiles.   | Major strength fields with i. 0120 and 0129 and ii. 0124/0125.              | No, but will assist CRCTPP's efforts.                                 | Group has conducted the initial research on the topic.                 |
| CRCTPP Australia                   | No   | RAPDs and more recently sequence analysis of the IGS.  | No  | CRCTPP plan to analyze the FHIA and CIRAD F <sub>2</sub> populations. | Student is working to identify endophytes that may increase hardiness. |
| Univ. Western Australia            | No   | No   | No  | No  | Stress physiology is a strength of the group.                          |
| Univ. of Florida, Homestead, USA   | Major strength Permitted lab. contains world collection of the pathogen and have increased the reliability of the test via refinement. | VCGs, RFLPs, RAPDs depending upon the objectives of the research.  | 0120 and 01210 are present, but major potential is for work with 0124/0125. | Interested, but has conducted no work.                                | Interested, but has conducted no work.                                 |
| Univ. of Florida, Gainesville, USA | No   | Major strength Kistler and O'Donnell are the world authorities in this area and are at the forefront of virtually all research here. | No  | No, but could provide invaluable assistance in this area.             | No   |

| STRENGTHS                     |                                   |                           |   |   |                                    |
|-------------------------------|-----------------------------------|---------------------------|---|---|------------------------------------|
| Organization                  | Early screening test              | Pathogen characterization | Field evaluation  | Marker-assisted selection                                   | Plantlet vigour                    |
| TBRI<br>Taiwan                | Interested                        | No                        | Large field screening scheme in place. The VCGs at this location are unknown. Subtropical race 4 is present here. | No  | Interested                         |
| Taiwan National University    | No                                | VCGs.                     | No  | No  | No                                 |
| CITA<br>Canary Islands        | Interested                        | VCGs and RAPDs.           | 0120/01215 site for IMTP II.  | No  | No                                 |
| MARDI<br>Malaysia             | No                                | No                        | Good site with 01213/01216.   | No  | No                                 |
| CRIH/Solok<br>Sumatra         | No                                | No                        | Also has 01213/01216 site.  | No  | No                                 |
| University Sains<br>Malaysia  | No                                | VCG.                      | In cooperation with Mak Chai (Kuala Lumpur) has conducted experiments in another 01213/01216 site.                | No  | No                                 |
| University of Bonn<br>Germany | Interested                        | No                        | No  | No  | Ph.D. student works on endophytes. |
| Philippines                   | Interested                        | No                        | 0122 here.  | No  | No                                 |
| EMBRAPA/CNPMF<br>Brazil       | Preliminary work being conducted. | No                        | Major strength Fields infested, VCGs under characterization at QDPI.  | Interested; no work conducted but facilities are available. | No                                 |

| STRENGTHS   |   |  |   |                           |                 |
|---|---|--|---|---------------------------|-----------------|
| Organization  | Early screening test                                  | Pathogen characterization  | Field evaluation                                  | Marker-assisted selection | Plantlet vigour |
| FHIA<br>Honduras  | No  | No   | Yes to Race 1 and 2.                              | No                        | No              |
| IMI (CAB International)<br>UK (also networked with other UK and East Africa-based groups) | No  | Extensive experience - mitochondrial RFLPs and probes; PCR methods - RAPD, SSR, rDNA ITS & IGS, introns of other loci; extra/intracellular isoenzymes; metabolite and enzyme production/ activity; VCGs Permit held for importing and working with non-indigenous isolates. Comprehensive range of isolates held in internationally recognised Genetic Resources collection. | Undertaken by collaborators in East Africa.       | No                        | No              |
| IITA<br>Uganda  | No  | No   | Plans exists for work on putative 0124/0125 site. | No                        | No              |
| ARC-ITSC<br>South Africa  | Have developed screening test but needs modification. | RAPDs  | Major strength with 0120/01215 fields.            | Interested                | Interested      |

A list of acronyms and abbreviations is provided at the end of the document.

## Medium-Term Plan

| Goals                                      | Inputs   | Outputs   | Indicators   | Time frame   |
|--|--|---|--|--|
| Early Screening                            | Equipment.<br>Source of high quality plantlets (i.e. INIBAP, QDPI).<br>Financial Support.  | Characterization of pathogenic diversity. Clarification of race structure in Foc. Initial and rapid indication of resistance / susceptibility in parents and hybrids.                   | Availability of resistant germplasm. Early elimination of highly susceptible genotypes.                                | Diversity<br>5 years<br>Resistance<br>10 years   |
| Field Screening                            | Identify fields where cooperative work can be conducted on major pathogen groups. Financial support.<br>Source of high quality plantlets (i.e. INIBAP, QDPI).  | Identification of resistant parents and hybrids.  | Availability of superior parents and potentially useful clones.  | 10 years   |
| Identification of genetic variation in Foc | Collecting missions. Financial support needs to be substantial as many of the methods are expensive and time-consuming.  | Basic understanding of pathogen increased. Know where to deploy specific host genotypes.<br>Specific molecular probes.  |  | Collecting missions<br>5 years. Variation - major information in 5 years but ongoing for 10 years. |
| Marker-Assisted Selection                  | Populations with different combinations of susceptible and resistant parents. Large fields for resistance / susceptibility evaluation - each individual needs to be replicated many times.<br>Laboratory equipped to carry out molecular analysis. Tissue-culture laboratory to supply plants. | Enhanced capability of breeding programme to identify resistance among progeny. Identification of resistance genes for transformation.<br>Understanding of how resistance is inherited. | Molecular marker technology used by breeding programmes.<br>The development of resistant clones using these techniques | 10 years   |
| Plantlet Vigour                            | Equipment for histological and physiological studies. Glasshouse facilities and access to field sites to establish correlations.   | Physiological, morphological or biological reasons for greater susceptibility identified. More vigorous plantlets better able to combat pathogens.                                      | Improved plant performance in diverse situations.  | 5 years  |

## Nematology Working Group<sup>1</sup>

### 1. Scope of Work, Priority Research Needs and Major Constraints

#### 1.1 Scope of work

The scope of the nematology group is to concentrate on the identification of sources of nematode resistance and tolerance, their underlying mechanisms and the pathogenic variability between and within the major nematode pest species of *Musa*. These activities will also require an improved knowledge of the species composition of nematode populations in major *Musa* production areas.

#### 1.2 Priority research needs

Although the following five research needs are ranked, their interdependence precludes their strict prioritization.

##### **Screening for resistance**

Identification of useable sources of resistance and tolerance to the major nematode pest species, including the further development and refinement of early and rapid screening techniques. This research is needed because there is a severe shortage of identified sources of transferable resistance and/or tolerance to the major nematode pest species.

##### **Relationship studies**

Studies on the relationships between *Musa* genotypes, nematode reproduction, root damage, plant growth and other components of yield losses in *Musa*. This basic research is needed because it will facilitate further research into the genetic improvement of a range of *Musa* genotypes, including those traditionally grown for domestic consumption.

##### **Pathogenicity studies**

Studies on the variability in pathogenicity between and within different nematode pest species in different geographic regions and different *Musa* production systems. This research is needed due to the recognised pathogenic variability of *Radopholus similis* and its potential existence in other nematode pest species, as well as to counter the potential development of resistance-breaking nematode pathotypes which has occurred in other crop-nematode associations.

##### **Mechanisms of resistance**

Studies on the mechanisms and inheritance of resistance and tolerance to the major nematode pest species. This research is needed because basic knowledge on mechanisms

(1) This report was prepared as a result of meetings held in Guadeloupe in which the following participants were involved: D. De Waele (KUL), R. Fogain (CRBP), N. Price (CAB-IIP), J-L Sarah (CIRAD), N. Viaene (INIBAP/FHIA). In addition, the following scientists were consulted subsequent to the Guadeloupe meeting: J. Bridge (CAB-IMD), S. Gowen (Univ. of Reading), I. Kashajja (NARO), J. Pinochet (IRTA), P. Speijer (IITA) and J. Stanton (QDPI).

of resistance and tolerance will assist breeding programmes in genetic improvement, for example, by the development of molecular markers for use in classical breeding.

### **Species profiles**

Studies on the occurrence of the major nematode pest species within different geographic regions and different *Musa* production systems. These activities are needed because the relative importance of the major nematode pest species in the different geographic regions and production systems is poorly understood. In addition and directly related, is a lack of knowledge of the potential interactions between different nematode pest species and their possible influence on any resistance or tolerance identified in *Musa*. This knowledge will be needed to enable the future prioritization of research needs in the various geographic regions and production systems.

#### 1.3 Major constraints

- Lack of awareness of the importance of plant-parasitic nematodes as potential production constraints to *Musa*, largely due to a lack of comprehensive empirically derived data demonstrating yield losses caused by the most important nematode pest species to the major *Musa* genotypes.
- Shortage of personnel trained in *Musa* nematology, especially in national programmes.
- Severe financial constraints contributing to institutional instability of research efforts.
- Shortage of available services for specialised nematode identification to provide back-up to field scientists.

## 2. Research Strategy

- Conduct pot and field experiments for identification of resistant and/or tolerant *Musa* genotypes.
- Identify suitable locations with different species compositions for the conduct of field experiments for the evaluation of relationships between genotypes, nematode reproduction, root damage and other components of yield loss, and the performance of experiments.
- Further collect and identify pathogenic variability in nematode species, particularly species other than *Radopholus similis*.
- Perform detailed studies of the nematode population dynamics, morphological, histological and biochemical studies of the genotypes to elucidate mechanism of resistance once resistant and tolerant genotypes are identified.
- Sample an appropriate number of fields in representative *Musa* production systems and determine the nematode species composition of the area, to help prioritise the major nematode species.

## 3. Areas for Collaboration

### **Screening**

FHIA, CRBP, KUL, IITA, IITA-ESARC, EMBRAPA, CIRAD, University of Reading, QDPI, IRTA/ICIA, CAB/IIP, NARO. Output could be increased through an exchange of methodology, exchange of data on relative resistance of tissue-culture plants versus conventional plant material, exchange of data on the genotypes that are screened and their reactions. Early screening of promising or scarce genotypes in the screen-house can then be followed up by more extensive field trials. This would improve and enhance the screening processes at the various institutions.

### **Pathogenic variability of nematodes**

KUL, IITA Uganda, CAB/IIP, CIRAD, USDA, QDPI, CRBP, IRTA-ICIA. Output could be increased by exchange of methodology, results and nematode cultures. Replication or division of research would enhance the validity of research findings.

### **Mechanisms of resistance**

CIRAD, ORSTOM, University of Reading, IITA-ESARC, CRBP. Output could be increased by avoiding unnecessary duplication of research activities and findings thus generated would be more comprehensive.

#### 3.1 New areas for collaborative research

Studies of yield losses caused by nematodes in semi-intensive and small-holder production systems in different agro-ecological regions. Other areas of research that need expansion and collaboration between institutions are the study of mechanisms of resistance and the pathogenic variability. An expansion of screening sites (e.g. in Southeast Asia) will enable the assessment of locally important *Musa* genotypes and will accelerate the production of relevant research findings.

The adoption of common or mutually compatible research methodologies between different research groups should be encouraged. A good example is the recently prepared 'Technical Guidelines for the Screening of *Musa* Germplasm for Resistance and Tolerance to Nematodes'.

#### 3.2 Identification of research needs requiring inter-group collaboration

Studies on the relationships between *Musa* genotypes, nematode reproduction, root damage, plant growth and other components of yield losses in *Musa* will benefit from collaborative activities with both the Fusarium and Sigatoka working groups. In particular, this may help to establish the significance (or otherwise) of nematode-pathogen interactions. A further aspect which merits consideration is the basis of the apparent multi-pathogen resistance in some genotypes e.g. Yangambi Km 5. This would also require collaboration with the Genetic Improvement Group, for example in the development of a gene-mapping programme and on the determination of the inheritance of resistance.



## 4. Inventory of Existing Facilities, Expertise and On-going Research

### 4.1 Existing facilities and expertise

| Laboratory                        | Areas of expertise  | Particular characteristics  |
|-----------------------------------|---|---|
| QDPI<br>Australia                 | Field screening, yield loss assessments.  | Regional <i>Musa</i> collection.  |
| KUL<br>Belgium                    | Glasshouse screening, pathogenicity studies, mechanisms of resistance.                    | Molecular techniques INIBAP Transit Centre; nematode culture collection (about 15 accessions).                      |
| Univ. of Gent<br>Belgium          | Taxonomy.   |   |
| EMBRAPA/CNPMF<br>Brazil           | Screening, yield loss assessments, pathogenic diversity of nematodes, ecological studies. | Existing breeding programme, presence of most major nematode spp., agro-ecological diversity, molecular techniques. |
| CRBP<br>Cameroon                  | Field and screen-house screening, mechanisms of resistance.                               | Agro-ecological diversity. Presence of most major nematode spp. Existing breeding programme.                        |
| University of Dschang<br>Cameroon | Nematode taxonomy.  | Classical taxonomy.   |
| CORPOICA<br>Colombia              | Screening, yield loss assessments.  |   |
| CORBANA<br>Costa Rica             | Screening, yield loss assessments, pathogenic diversity of nematodes, ecological studies. |   |
| IDEFOR<br>Côte d'Ivoire           | Field screening, yield loss assessments.  |   |
| Various<br>Cuba                   | Screening, yield loss assessments.  | Strong potential but lack of international exposure and linkages.   |
| INIAP<br>Ecuador                  | Screening, yield loss assessments.  |   |
| USDA, Florida<br>USA              | Pathogenic diversity of nematodes.  | Molecular techniques.   |
| CIRAD/ORSTOM<br>France            | Glasshouse screening, pathogenicity studies, mechanisms of resistance.                    | Molecular techniques. Nematode culture collection (approx. 50 accessions).  |
| Univ. of Bonn<br>Germany          | Pathogen interactions.  |   |
| Univ. of Ghana                    | Field screening, yield loss assessments.  |   |
| CRI<br>Ghana                      | Field screening.  |   |
| CIRAD / ORSTOM<br>Guadeloupe      | Screening, yield loss assessments.  | Existing breeding programmes.   |

| Laboratory                            | Areas of expertise   | Particular characteristics   |
|---------------------------------------|--|--|
| FHIA<br>Honduras                      | Screening, yield loss assessments.   | Presence of several important nematode spp. Existing breeding programme.                                       |
| NRCB<br>India                         | Field screening, yield loss assessments.   | Regional <i>Musa</i> collection.   |
| RIF<br>Indonesia                      | Field screening, yield loss assessments.   | Extensive regional <i>Musa</i> evaluation.   |
| Volcani Research<br>Center<br>Israel  | Screening, yield loss assessments.   | Sub-tropical. Importance of <i>Helicotylenchus multincinctus</i> .   |
| MARDI<br>Malaysia                     | Field screening, yield loss assessments.   |  |
| CIRAD /ORSTOM<br>Martinique           | Yield loss assessments.  |  |
| Institut Hassan II<br>Morocco         | Yield loss assessments.  | Importance of <i>Meloidogyne incognita</i> . Sub-tropical.   |
| IITA<br>Nigeria                       | Field screening, screen-house screening, yield loss assessments, resistance mechanisms.          | Root system investigations. Existing breeding programme.   |
| Univ. of Los Baños<br>Philippines     | Field screening, yield loss assessments.   |  |
| ARC-ITSC<br>South Africa              | Field, screen-house screening, yield loss assessments.   | Sub-tropical.  |
| PPRI<br>South Africa                  |  | Availability of identification services (classical).   |
| IRTA/ICIA<br>Spain,<br>Canary Islands | Field, screen-house screening, pathogenicity studies.  | Molecular techniques. Sub-tropical. Nematode culture collection (approx. 30 accessions).                       |
| National<br>Programme<br>Tanzania     | Field screening, yield loss assessments.   |  |
| HRI<br>Thailand                       | Yield loss assessments.  |  |
| IITA/ESARC<br>Uganda                  | Field, screen-house screening, yield loss assessments, resistance mechanisms.                    | Existing breeding programme. Agro-ecological diversity.  |
| NARO<br>Uganda                        | Field screening, yield loss assessments  | Agro-ecological diversity, presence of most of the important nematode species and of <i>H. multincinctus</i> . |
| CAB-IIP<br>UK                         | Glasshouse screening, pathogenicity, mechanisms of resistance, taxonomy (classical & molecular). | Molecular techniques Nematode culture collection (>70 accessions).   |
| Univ. of Reading<br>UK                | Glasshouse screening. Pathogenicity, mechanisms of resistance.                                   | Nematode culture collection.   |

| Laboratory         | Areas of expertise                        | Particular characteristics  |
|--------------------|---|---|
| IACR Rothamsted UK | Molecular taxonomy                        |   |
| Univ. of Leeds UK  | Use of transgenes for nematode resistance | In potato, tomato and rice but technology should be transferable to <i>Musa</i> |

A list of acronyms and abbreviations is provided at the end of the document.

#### 4.2 Inventory of on-going research

| Institute                    | On-going research  | Expected outputs   | Expected time-frame |
|------------------------------|--|--|---------------------|
| FHIA Honduras                | Field and screen-house screening.  | Identification of resistant genotypes.   | 5 years             |
| EMBRAPA/CNPMP Brazil         | Screening. Survey in infected areas. Foc x <i>mycorrhizae</i> x nematode interactions. | Identification of resistant genotypes.   |                     |
| IITA Nigeria                 | Screening. Species composition. Yield loss studies                                     | Identification of resistant genotypes. Identification of yield loss for <i>Musa</i> AAB. | 3 to 5 years        |
| CRBP Cameroon                | Screening. Yield loss studies.   | Identification of resistant genotypes.   | 3 to 5 years        |
| IITA-ESARC Uganda            | Species composition. Screening. Yield loss studies.                                    | Identification of resistant genotypes.   | 3 to 5 years        |
| IRTA/ICR Spain               | Screening interactions. Pathogen diversity studies.                                    |  | ?                   |
| CIRAD France                 | Pathogenicity diversity and resistance mechanisms.                                     |  | 3 to 5 years        |
| CIRAD Guadeloupe, Martinique | Screening and damage studies of <i>Meloidogyne</i> .                                   | Identification of resistant genotypes.   | 3 years             |
| KUL Belgium                  | Resistance screening. Pathogenicity testing.   | Identification of resistant genotypes.   | 5 years             |
| CAB-IIP UK                   | Pathogenicity testing.   | Identification of molecular markers for pathogenicity.                                   | 3 years             |
| Univ. of Reading UK          | Screening and mechanisms of resistance.  |  | 3 years             |
| QDPI Australia               | Screening.   |  | ?                   |

A list of acronyms and abbreviations is provided at the end of the document.

#### Medium-Term Plan

| Goal                     | Inputs  | Outputs   | Indicators   | Time frame  |
|--------------------------|---|---|--|---|
| Screening for resistance | Collecting missions or existing germplasm collections. Equipment and expertise. | Sources of useable resistance/ tolerance following screening of part of available germplasm collection. Sources of useable resistance/tolerance following screening of an additional part of the germplasm collection and incorporated in agronomically useful germplasm. | Nematode resistance incorporated into new hybrids. | 5 years<br>10 years                                     |
| Relationship studies     | Equipment and expertise. Suitable evaluation sites.                             | Increased knowledge within the major <i>Musa</i> genotypes, of the relationships between nematode populations and various components of yield loss.   |  | 6 years   |
| Pathogenicity studies    | Equipment and expertise. Collections of nematodes from different regions.       | Nematode pathotypes will be grouped and ranked based on their pathogenicity.  |  | 5 years   |
| Mechanisms of resistance | Information from basic research activities. Equipment and expertise.            | Understanding of mechanisms of resistance/ tolerance at the morphological, cellular genetic and molecular level. Molecular markers identified. Resistance genes identified.   | Use of molecular markers by breeding programmes.   | 7 years   |
| Species profiles         | Surveys.  | Identification of global and regional significance of the major nematode pest species.  | Priority list of major nematode species.           | Several short term surveys during a period of 10 years. |

## Virology Working Group<sup>1</sup>

### 1. Scope of Work, Priority Research Needs and Major Constraints

#### 1.1 Scope of work

Viruses are recognised as a significant problem leading to constraints in *Musa* improvement on two grounds: yield losses which can be up to 100% and a constraint to germplasm movement. Four viruses are currently recognised as being significant: banana bunchy top virus (BBTV), banana streak virus (BSV), banana bract mosaic virus (BBrMV), and cucumber mosaic virus (CMV); there are reports of other viruses but these have not yet been characterised. As virtually nothing is known about these other viruses, it is suggested that a watching brief is kept on them. The main demands are for:

- Detection and diagnostic systems for breeding, tissue culture and germplasm movement (quarantine).
- Resistance to the viruses.

#### 1.2 Priority research needs

##### *Virus detection and therapy*

In relation to virus detection, there is a need for the development of improved, robust, simple and accessible diagnostic systems.

The importance of the simplicity and robustness would depend upon who was carrying out the diagnosis - international centres, NARS, propagation centres, users (farmers, industry, etc.) BSV presents a unique case as there is a strong probability that the three forms of the virus (encapsidated episomal, unencapsidated episomal and integrated) would require different diagnostic techniques. Further research is also needed in the areas of: virus variability, geographical distribution of the different viruses, cultivar reaction to virus infection and the molecular biology of the viruses themselves. In addition, research is required on the development of a technique to eliminate viruses from infected plants. Table A lists these four major topics on which virus detection research is needed (+) together with prioritization (#) both for the topic and virus.

The prioritization is based on the perceived importance for an efficient diagnostic system.

##### *Virus resistance*

There is a lack of resistance in *Musa* to the major viruses but there is the possibility that resistance is present in related genera and families which could be introgressed into *Musa*.

(1) This report was prepared as a result of meetings held in Guadeloupe in which the following participants were involved: G. Dahal (IITA), J. Dale (QUT), J. Hu (USA), R. Hull (John Innes Centre), M-L Iskra-Caruana (CIRAD), B. Lockhart (Univ. of Minnesota), S. Sharrock (INIBAP). In addition, the following scientists were consulted subsequent to the Guadeloupe meeting: H. Muyle (Gembloux) and J. Thomas (QDPI).

Table A - Research needs for virus detection

|                         | (1) | (2)  | (3)   | (4)    | #   |
|-------------------------|-----|------|-------|--------|-----|
|                         | BSV | BBTV | BBrMV | CMV    |     |
| Variability             | +   | +    | +     | + Sat* | (1) |
| Viral molecular biology | +   | +    | -     | -      | (2) |
| Distribution            | +   | +    | +     | -      | (3) |
| Cultivar reaction       | +   | +    | +     | +      | (3) |

\* It is not known if the variability of the reaction of CMV in banana is due to the virus or possibly associated Satellite RNA.

The major constraints and research needs on virus resistance are:

- Information on resistance in closely related genera and families;
- Methods for introducing such resistance into *Musa* by, for example, wide crosses.

The most promising approach to obtaining resistance in *Musa* is the introduction of viral or virus related sequences into the host genome. A major emphasis is required on transformation technologies, with the development of strategies to obtain combined resistance to several viral and fungal diseases and to important pests.

An understanding of the BSV system is required in order that strategies can be developed to overcome this problem.

Latent strains of BBTV need to be further studied.

The major research needs in relation to virus resistance are given in Table B. In this table (+) indicates lack of information and the need for more research. (H) is high priority with (H)\* being the highest; (M) is medium and (L) low priority.

Table B: Research needs for virus resistance

|                           | (H)   | (H)  | (H) | (M) |      |
|---------------------------|-------|------|-----|-----|------|
|                           | BBrMV | BBTV | BSV | CMV |      |
| Molecular biology         | -     | +    | +   | -   | (H)  |
| Variability               | +     | +    | +   | -   | (H)  |
| Resistance mechanisms     | -     | +    | +   | -   | (H)  |
| Transformation techniques | +     | +    | +   | +   | (H)* |
| Vector constructs         | +     | +    | +   | +   | (M)  |
| Selection markers         | +     | +    | +   | +   | (M)  |
| Transgenic stability      | +     | +    | +   | +   | (L)  |
| BSV                       | +     | +    | -   | +   | (H)  |
| Resistance testing        | +     | +    | +   | +   | (L)  |
| IP                        | +     | +    | +   | +   | (L)  |
| Biosafety                 | +     | +    | +   | +   | (M)  |

The first three topics relate to the viruses themselves and the design of transgenes. The next five topics are associated with transformation procedure, that of BSV being the potential problem of transformation stresses activating integrated viral sequences. The

last two topics are constraints which would become relevant on field release of transformed plants but for which consideration has to be given at early stages of the project. Selection markers, e.g. use of antibiotic genes, should also be considered at this stage.

### 1.3 Major constraints

- a lack of known sources of genetic resistance in *Musa* species.
- the small number of specialists working in this area.
- a need for further attention to be given to biosafety and IPR considerations.

## 2. Research Strategy

The goal is the control of viruses in *Musa*. This will be by transgenic approaches to conferring resistance in *Musa* and development of diagnostic systems to support this approach and to produce virus-tested germplasm and planting material.

## 3. Areas for Collaboration

### Transformation

Transformation is essential for progress in the development of transgenic resistance and in many cases requires collaboration between scientists working on molecular aspects of the viruses and those with expertise in transformation.

### Molecular markers

Collaboration in this area will have some application in use of transgenics for breeding but especially for resolving problems relating to BSV integration. It could also be of use in IP issues.

### Virus detection

Collaboration is required in this area in developing diagnostic systems suitable for the end user.

### 3.1 New areas for collaborative research

International screening of transgenic plants. The field release of transgenic *Musa* lines will require collaboration between the biotechnologists, biosafety officials, breeders, agronomists and others. Studies have to be made as to how to effect this as efficiently as possible.

Stresses which induce activation of integrated BSV. An understanding of the factors leading to the activation of integrated BSV is of major importance in the application of new technologies to *Musa* improvement. This will involve collaboration between molecular biologists, tissue culture experts, breeders and transformers.

### 3.2 Identification of research needs requiring inter-group collaboration

- Transformation with the Genetic Engineering Sub-group of the Genetic Improvement Group.
- Molecular markers with the Breeding and Genetics Sub-group.

- Breeding with the Breeding and Genetics Sub-group of the Genetic Improvement Group.
- Transformation strategies to obtain resistance to a range of pathogen and pests with the Sigatoka, Fusarium and Nematode groups.

## 4. Inventory of Existing Facilities, Expertise and On-going Research

### 4.1 Existing facilities and expertise

|                              | BBrMV | CMV | BBTV | BSV | Others |
|------------------------------|-------|-----|------|-----|--------|
| University of Southern China | -     | +   | +    | -   | -      |
| CIRAD, France                | +     | +d  | +    | +d  | +      |
| Gembloux/KUL Belgium         | +t    | +t  | +t   | +t  | +t     |
| IITA, Nigeria                | -     | +d  | -    | +   | +      |
| JIC, UK                      | -     | -   | -    | +   | +      |
| NUT, Taiwan                  | -     | +d  | +    | -   | -      |
| QDPI, Australia              | +     | +d  | +    | +d  | +      |
| QUT, Australia               | +     | -   | +    | -   | -      |
| UOH, Hawaii USA              | -     | +   | +    | +d  | -      |
| UOM, USA                     | -     | +d  | -    | +   | +      |
| EMBRAPA/CNPMF, Brazil        | -     | dt  | -    | -   | -      |

A list of acronyms and abbreviations is provided at the end of the document.

+ Indicates relevant work being done on virus

d Indicates work in diagnosis

t Indicates work on therapy to cure tissue culture lines

Most, if not all the work being done in these organizations is complementary and there is no obvious comparative advantage of any laboratory for any virus.

### 4.2 Inventory of on-going research

|                   | BBrMV | CMV | BBTV | BSV |
|-------------------|-------|-----|------|-----|
| Variation         | +S    | +M  | +M   | +M  |
| Distribution      | -     | -   | +M   | +L  |
| Molecular biology | +S    | +M  | +M   | +S  |
| Transformation    | +S    | +M  | +M   | +L  |

S = Short term (1 - 2 years)

M = Medium term (up to 5 years)

L = Long term (more than 5 years)

+ = Indicates research going on in that area for that virus

The outputs are that the research is directed towards gaining an understanding of the topic for that virus relevant to the aims of diagnostics and/or resistance by the transgenic approach.

### Medium-Term Plan

| Goal  | Inputs   | Outputs   | Indicators  | Time frame                    |
|---|--|---|---|-------------------------------|
| Development and demonstration of efficiency of a cassette for transgenic resistance to BBTv, BBrMV and CMV                          | Basic research on BBTv.<br>Efficient transformation system.<br>Strategies to address IPR considerations.                                       | Mechanisms for transgenic resistance to BBTv.<br>Information on genome variability within CMV isolates infecting <i>Musa</i> .<br>Resistance constructs for each of BBrMV, BBTv and CMV.  |   | 3 years<br>3 years<br>5 years |
| Development of robust diagnostic systems for BBTv, BBrMV and CMV  | Basic information on BBTv variation.   | Understanding of the molecular basis of latent BBTv strains.<br>Determination of the distribution of latent BBTv strains.<br>Development of diagnostics for the detection of latent BBTv strains.   | Availability of diagnostic systems suitable for use by a wide range of users. | 3 years<br>3 years<br>3 years |
| Genetically modified plants protected against viruses   | IP and biosafety considerations.<br>Transgenic and conventional approaches.  | Varieties transgenically protected against viruses, field tested and farmer released.<br>Varieties with integrated protection against other pathogens and pests and linked with other agronomic characters.   | Resistant varieties available to farmers.                                     | 10 years                      |
| Obtaining basic information on the BSV system which can be used for the development of transgenic strategies and detection systems. | Basic research on BSV system.<br>Information from breeders to indicate which parental lines produce highest proportion of BSV infected plants. | Understanding of the integration:episomal system. Identification of active integrated form(s) of BSV. Understanding of factors which activate integrated form(s).<br>Understanding of encapsidated/non-encapsidated episomal forms.<br>Development of diagnostics for the different forms.<br>Characterization of the status of integrated forms in cultivars of interest (parental lines, tissue culture stocks).<br>Understanding of variation of active integrated and episomal forms. |   | 3 years<br>5 years<br>5 years |



| Goal  | Inputs   | Outputs   | Indicators  | Time frame |
|---|--|---|---|------------|
| Genetically modified plants protected against BSV | Basic information.<br>Advice from breeders and tissue culturists as to important cultivars for transformation.<br>IP and biosafety consideration.      | <i>Musa</i> varieties transformed with constructs designed to protect against BSV.<br>Promising lines undergoing initial field testing. | BSV resistant varieties included in field trials. | 10 years   |
| Development of virus eradication methodologies    | Basic information on virus multiplication in the plant.<br>Understanding of the significance of integration of BSV sequence in the <i>Musa</i> genome. | Plants freed from virus infection.<br>Virus therapy methodology available.  | Availability of plants freed from virus.          | 5 years    |

## Global and Regional Evaluation<sup>1</sup>

### 1. Introduction

An efficient mechanism for evaluating germplasm is an essential component of PROMUSA. In relation to this, an International *Musa* Testing Programme (IMTP) has been co-ordinated by INIBAP since 1989. A global IMTP meeting was held in Guadeloupe on March 3<sup>rd</sup> and 4<sup>th</sup> to evaluate the results of the second phase of the programme and to discuss future proposals for this programme. In view of the increasing number of breeding programmes wishing to contribute hybrids and NARS wishing to evaluate these new hybrids, the participants at the meeting agreed to make some major modifications to IMTP to enable it to better respond to the different needs of programme partners. The new structure of IMTP should allow it to play an important role in the global and regional evaluation of germplasm in the framework of PROMUSA.

### 2. Scope of the Work

The International *Musa* Testing Programme (IMTP) is a global initiative in which improved varieties, breeding materials, accessions with possible sources of resistance and standard checks are tested at the global and regional level with the following objectives:

- To obtain pathological information and agronomic evaluation data to feedback to breeding programmes.
- To provide NARS with improved varieties.

### 3. Strategy

The identification of germplasm with disease resistance/tolerance, with desirable agronomic characteristics and with local adaptation by evaluation in different locations world-wide. The participation of NARS ensures that improved hybrids are made available to them at an early stage and through the creation of linkages, the bi-directional flow of information between breeding programmes and evaluation sites is facilitated.

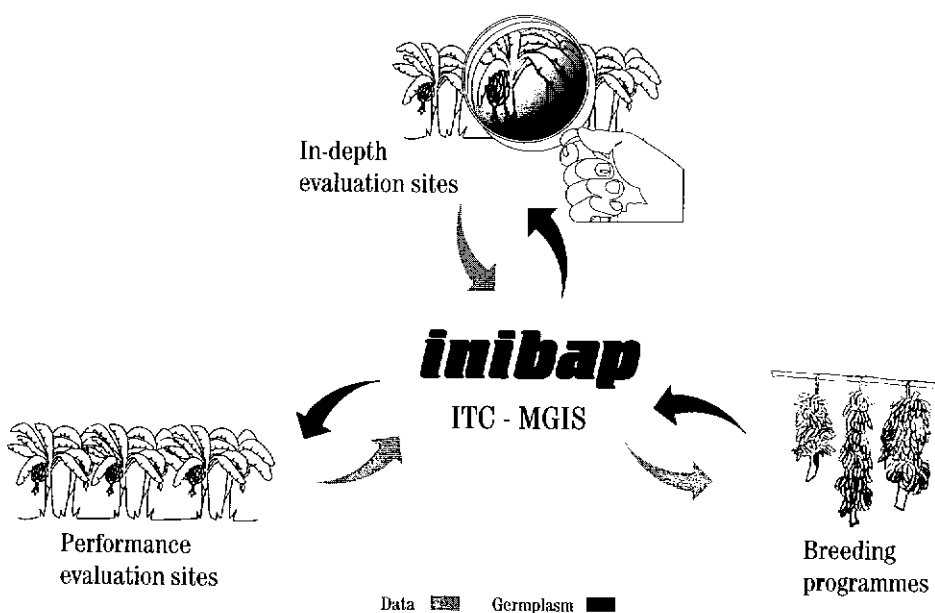
(1) This report was prepared as a result of meetings held in Guadeloupe in which the following participants were involved: F. Bakry (CIRAD), W. Collins (World Bank), J. Dale (QUT), Z. De Beer (ARC-ITSC), E.A.L. De Langhe, D. De Waele (KUL), J-V. Escalant (CRBP), H. Fagan (WIBDECO), E. Fouré (CIRAD), E. Frison (INIBAP), P. George (World Bank), J. Hernandez (ICIA), J-P. Horry (INIBAP), S.H. Jamaluddin (MARDI), C. Jenny (CIRAD), B. E. L. Lockhart (Univ. Minnesota), D. Mackenzie (USDA), L. Magnaye (BPI), N. Mirza (IDRC), X. Mourichon (CIRAD), S. Nanthachai (HRI), G. Orjeda (INIBAP), K. Pegg (QDPI), A. Pires De Matos (EMBRAPA/CNPMPF), R. C. Ploetz (Univ. Florida), R. Romero Calderon (CORBANA), P. Rowe (FHIA), J-L. Sarah (CIRAD), L. Sas (BADC), L. Setyobudi (RIF), H.P. Singh (NRCB), M. Smith (QDPI), R. Swennen (KUL), H. Tézenas Du Montcel (CIRAD), K. Tomekpe (CRBP), S. Tripon (INIBAP), R. Valmayor (INIBAP), P. Vi Nai (MAFF), N. Viaene (FHIA) and D. Vuylsteke (IITA).

#### 4. Modus Operandi

Promising new material is identified by breeding programmes and sent to the INIBAP Transit Centre (ITC) where it is introduced *in vitro*, virus indexed, and multiplied. This material is then made available to interested NARS for screening for disease resistance using standard guidelines previously agreed upon by the participants in IMTP.

In the first two phases, all material donated to the IMTP was evaluated at all sites using the same protocol, and this has yielded important information on disease resistance/tolerance. As a result, several improved varieties have been recommended for further distribution and are now being commercially cultivated in a number of countries. However, as the number of improved hybrids becoming available for testing increases, and with an expanding number of national programmes interested in evaluating such improved material, a new more flexible approach for IMTP has been developed (Figure).

Within the new structure of IMTP two different evaluation protocols will be used to address the two main objectives :



#### Revised structure of IMTP

ITC: INIBAP Transit Center

MGIS: *Musa* Germplasm Information System

#### *In-depth evaluation sites*

Very detailed studies will be carried out at the in-depth evaluation sites. The evaluations at these sites will include not only disease and pest resistance/tolerance screening but can also be combined with epidemiological studies on pathogen populations, studies on

host/pathogen relationships for different races of the pathogen and adaptability and yield studies. The evaluation protocols at these sites are elaborate, requiring time and expertise.

The in-depth evaluation sites will also be used for screening potential breeding parents for resistance to pathogens not present at the breeding sites. The information obtained will be primarily useful to breeders and pathologists.

#### *Performance evaluation sites*

At the performance evaluation sites the collaborators will assess the disease resistance, agronomic adaptation and stability of the improved hybrids under their particular conditions. The evaluation protocols to be used at these sites are simple requiring less time and expertise than those used at the in-depth evaluation sites. Moreover collaborators at these sites will select the clones they wish to evaluate, based on local needs and conditions. Such a programme has a strong regional focus, with varieties being selected for evaluation according to national/regional needs. The information obtained will be primarily useful to extension agents and farmers.

Both types of sites will provide feedback information on the agronomy, pathology and adaptation of improved varieties tested in the International *Musa* Testing Programme. This information will be compiled in a database which has been designed for this purpose and which in the future will be linked to the *Musa* Germplasm Information System (MGIS). The information is then fed back to the breeding programmes and is also available for other NARS to assist in their choice of appropriate well characterised varieties.

#### 5. Activities

- Identification of improved hybrids;
- Introduction *in vitro*, virus indexing, propagation and dissemination of material;
- Elaboration of the structure of a database;
- Gathering basic agronomic and disease resistance data from breeding programmes;
- Elaboration of the actual database in the framework of MGIS;
- Provision of evaluation training to IMTP site managers;
- Establishment of trials and carrying out evaluations;
- Gathering data from sites and feeding the database;
- Make the database available to collaborators - both breeding programmes and NARS.

## Acronyms and Abbreviations

|            |   |
|------------|---|
| ACO        | 1-aminocyclopropane-1-carboxylic acid oxidase   |
| ACS        | 1-aminocyclopropane-1-carboxylic acid synthase  |
| APHIS-USDA | Animal and Plant Health Inspection Service - United States - Department of Agriculture      |
| ARI        | Advanced Research Institute   |
| ARC-ITSC   | Agricultural Research Council, Institute for Tropical and Subtropical Crops, South Africa   |
| ASPNET     | Asia and Pacific Regional Network, INIBAP, Philippines                                      |
| BADC       | Belgian Administration for Development Co-operation   |
| BBrMV      | banana bract mosaic virus   |
| BBTV       | banana bunchy top virus   |
| BIP        | Banana Improvement Project  |
| BLSD       | black leaf streak disease   |
| BPI        | Bureau of Plant Industry, Philippines   |
| BSV        | banana streak virus   |
| BTI        | Boyce Thompson Institute, USA   |
| CABI       | CAB International, UK   |
| CATIE      | Centro Agronómico Tropical de Investigación y Enseñanza, Costa Rica                         |
| CFC        | Common Fund for Commodities, the Netherlands  |
| CGIAR      | Consultative Group on International Agricultural Research                                   |
| CICY       | Centro de Investigaciones Científicas de Yucatán, Mexico                                    |
| CIRAD      | Centre de coopération internationale en recherche agronomique pour le développement, France |
| CITA       | Centro de Investigación y Tecnología Agrarias, Canary Islands                               |
| CMV        | cucumber mosaic virus   |
| CNPMP      | Centro Nacional de Pesquisa de Mandioca e Fruticultura, EMBRAPA, Brazil                     |
| CORBANA    | Corporación Bananera Nacional, Costa Rica   |
| CORPOICA   | Corporación Colombiana de Investigación Agropecuaria, Colombia                              |
| CRBP       | Centre de Recherches Régionales sur Bananiers et Plantains, Cameroon                        |
| CRCTPP     | Cooperative Research Centre for Tropical Plant Pathology, Australia                         |
| CRI        | Crop Research Institute, Ghana  |
| CRIH       | Central Research Institute for Horticulture, Indonesia                                      |
| DNA        | deoxyribonucleic acid   |
| EMBRAPA    | Empresa Brasileira de Pesquisa Agropecuaria, Brazil   |
| ESARC      | East and Southern Africa Regional Centre, IITA, Uganda                                      |
| FAO        | Food and Agriculture Organization of the United Nations                                     |
| FHIA       | Fundación Hondureña de Investigación Agrícola, Honduras                                     |
| FLHOR      | Département des productions fruitières et horticoles, CIRAD, France                         |
| Foc        | <i>Fusarium oxysporum</i> f. sp. <i>cubense</i>   |
| HKUST      | Hong Kong University of Science and Technology  |
| HRI        | Horticulture Research Institute, Thailand   |
| IACR       | Integrated Approach to Crop Research, UK  |
| IARC       | International Agricultural Research Centre  |
| ICIA       | Instituto Canario de Investigaciones Agrarias, Spain  |
| IDEFOR     | Institut des Forêts, Côte d'Ivoire  |
| IDRC       | International Development Research Centre, Canada   |

|          |  |
|----------|--|
| IEB      | Institute of Experimental Botany, Czech Republic   |
| IFAD     | International Fund for Agricultural Development, Italy                                   |
| IGS      | intergenic spacer of the ribosomal DNA gene  |
| IIP      | International Institute of Parasitology, UK  |
| IITA     | International Institute of Tropical Agriculture, Nigeria                                 |
| IMI      | International Mycological Institute, UK  |
| IMTP     | International Musa Testing Programme, INIBAP   |
| INIAP    | Instituto Nacional de Investigaciones Agropecuarias, Ecuador                             |
| INIBAP   | International Network for the Improvement of Banana and Plantain, France                 |
| IP       | intellectual property  |
| IPGRI    | International Plant Genetic Resources Institute, Italy                                   |
| IRTA     | Instituto de Recerca i Tecnologia Agroalimentàries, Spain                                |
| ITC      | INIBAP Transit Centre, Belgium   |
| ITS      | internal transcribed spacer of ribosomal RNA gene  |
| JIC      | John Innes Centre, UK  |
| KARI     | Kawanda Agricultural Research Institute, NARO, Uganda                                    |
| KAU      | Kerala Agriculture University, India   |
| KUL      | Katholieke Universiteit Leuven, Belgium  |
| MAF      | Ministry of Agriculture and Forestry, Tonga  |
| MARDI    | Malaysian Agricultural Research and Development Institute, Malaysia                      |
| MGIS     | Musa Germplasm Information System  |
| NARO     | National Agricultural Research Organization, Uganda                                      |
| NARS     | National Agricultural Research System  |
| NRCB     | National Research Centre on Banana, India  |
| NRI      | Natural Resources International, UK  |
| NUT      | National University of Taiwan  |
| ORSTOM   | Institut français de recherche scientifique pour le développement en coopération, France |
| PCR      | polymerase chain reaction  |
| PPRI     | Plant Protection Research Institute, South Africa  |
| QDPI     | Queensland Department of Primary Industries, Australia                                   |
| QTL      | quantitative trait loci  |
| QUT      | Queensland University of Technology, Australia   |
| RAPD     | random amplified polymorphic DNA   |
| rDNA     | ribosomal DNA  |
| RFLP     | restriction fragment length polymorphism   |
| RIF      | Research Institute of Fruits, Indonesia  |
| SD       | Sigatoka disease   |
| TBRI     | Taiwan Banana Research Institute   |
| UF       | University of Florida, USA   |
| UNDP     | United Nations Development Programme   |
| UOH      | University of Hawaii, USA  |
| UOM      | University of Minnesota, USA   |
| USDA-ARS | United States Department of Agriculture-Agricultural Research Service                    |
| VASI     | Vietnam Agricultural Science Institute   |
| VCG      | vegetative compatibility group   |
| VIC      | Virus Indexing Centre  |
| WIBDECO  | Windward Islands Banana Development and Exporting Company                                |
| YS       | yellow Sigatoka  |

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