

ALTERNATIVE *EX SITU* CONSERVATION TO ENSURE LONG-TERM VIRUS-FREE CONSERVATION OF *MUSA* GERmplasm

AB Molina¹, FS dela Cruz, Jr.², LS Gueco², OP Damasco², VC Huelgas², FM dela Cueva², TO Dizon², MLJ Sison², VGO Sinohin¹, MA Alforque³, and LE Heradura³

¹Biodiversity International - Commodities for Livelihoods, c/o IRRI, Los Baños, Laguna, Philippines
²Crop Science Cluster - Institute of Plant Breeding, College of Agriculture, University of the Philippines Los Baños, College, Laguna 4031, Philippines ³Bureau of Plant Industry-Davao National Crop Research and Development Center, Bago-Oshiro, Davao City, Philippines

Introduction

Current efforts to conserve *Musa* germplasm include maintaining the collections in the field genebank and backing up with *in vitro* conservation. While field conservation has the advantage of having the germplasm readily available for characterization, evaluation and distribution, it is exposed to natural calamities such as typhoons, floods, as well as pest and disease infestation. Field conservation also incurs high cost of maintenance while *in vitro* conservation may result to somaclonal variation. The limitations of current conservation strategies make long-term conservation difficult to achieve.

An alternative conservation strategy needs to be developed to offset the limitations of current conservation strategies and to ensure long-term conservation of these valuable genetic resources, especially where diseases are prevalent.

Methodology

- Complementary conservation of *Musa* in the Philippines is done through field gene banking at the Bureau of Plant Industry (BPI) and Institute of Plant Breeding (IPB), and *in vitro* at IPB.
- National Repository, Multiplication and Dissemination Centers (NRMDC) were also established at BPI and IPB to evaluate new cultivars and superior landraces, promote their utilization potential, and make them available to farmers and interested users.



- An alternative strategy using screenhouse conservation was assessed to offset the limitations of the conventional methods and evaluate its potential for long-term conservation.
- The health status of various *Musa* conservation methods were evaluated through symptoms and using antibody based detection technique Enzyme-Linked Immunosorbent Assay (ELISA).

Results

Field maintenance of *Musa* germplasm

The BPI-DNCRDC Banana Field Genebank was established in 1977 but after 10 years, rehabilitation was done because of rapid spread of diseases due to viruses. Re-establishment was also done three (3) more times due to poor growth and infection of viral and systemic diseases.

A. Historical rejuvenation of field genebank showing decline of accessions due to diseases* in BPI-DNCRDC

Country of Origin	Number of accessions rejuvenated				Accessions Lost
	1977	1987	1998	2006	
Philippines	102	51	81**	46	56
Malaysia	28	26	23	19	9
Thailand	33	30	16	13	20
Papua New Guinea	71	0	0	15***	71
Indonesia	15	11	11	1	14
TOTAL	249	118	131	94	170

* Mainly BBTv
 ** Additional accessions were collected from other germplasm centers or recollected from original sites
 *** New accessions introduced from *Musa* International Transit Centre

B. Field maintenance of wild *M. balbisiana* germplasm



- Wild *Musa balbisiana* were collected from 1997-1999 and were conserved in the field genebank of IPB in 2000.
- The health status of these collections were determined using ELISA in 2008 and results indicate that 91.67% of the collection were tested positive for banana bract mosaic virus (BBRMV) while 22.22% is positive for cucumber mosaic virus (CMV).

Evaluation of 108 wild *M. balbisiana* accessions maintained in the field genebank to banana bract mosaic virus (BBRMV), cucumber mosaic virus (CMV) and banana bunchy top virus (BBTV) using ELISA

Virus	Positive	Strong positive	Very strong positive	Negative
BBRMV	86 (79.63)	12 (11.11)	1 (0.93)	9 (8.33)
CMV	23 (21.29)	1 (0.93)	0	84 (77.78)
BBTV	0	0	0	0



Newly-established field genebank of introduced and local banana cultivars



Field genebank infected with BBTv starting at 5 months after planting

C. Percentage incidence of BBTv of NMRDC cultivars in 2 sites

1. Mainit, Bay, Laguna

Cultivar	Percent Incidence			
	17 MAP	20 MAP	23 MAP	30 MAP
19 Introduced	0 - 7	2 - 16	2 - 55	2 - 60
8 Local	0 - 9	7 - 19	10 - 68	18 - 88

2. CES, Pili Drive, UPLB

Cultivar	Percent Incidence				
	5 MAP	8 MAP	11 MAP	14 MAP	19 MAP
6 Introduced	0 - 0	0 - 2	0 - 7	0 - 12	0 - 33
6 Local	0 - 5	0 - 10	0 - 12	5 - 24	7 - 69

Screenhouse conservation



Pot maintenance in BPI



Pot maintenance in IPB

- Twenty-one (21) IMTP and seven (7) local cultivars were planted in 18" x 20" clay pots in BPI and 14" x 18" plastic pots in IPB and maintained inside an insect-proof screenhouse.
- Assessments were conducted every six (6) months and both collections remained uninfected even after 4-6 years of continued maintenance.

Health status of banana cultivars conserved inside the repository screenhouse in IPB and BPI

Cultivar	ELISA (BBTV and BBRMV)			
	2006	2007	2008	2009
IPB, UPLB				
• 20 Introduced cultivars	-	-	-	-
• 7 Local cultivars	-	-	-	-
	2003	2004	2005	2008
BPI-DNCRDC				
• 25 Introduced cultivars	-	-	-	-
• 4 Local cultivars	-	-	-	-

Conclusions

- Conservation of *Musa* germplasm inside an insect-proof screenhouse can be an effective alternative strategy for long-term conservation.
- Plants conserved inside the screenhouse are maintained virus-free and routine indexing can be done to ensure that the germplasm is virus-free. When infected plants are observed, they can be easily replaced.
- No variation in horticultural traits was observed after 5 years of continuous maintenance.