

IMTP Phase II final results: Response to *Fusarium* and stability of genotypes across environments

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Summary

We present the results from the *Fusarium* (FOC) sites of the International *Musa* Testing Programme. Twenty genotypes were evaluated in Australia, Brazil, Honduras, Indonesia, Malaysia, the Philippines, South Africa, Spain, Taiwan and Uganda using the same evaluation protocols. Information on the agronomic performance and response to disease is presented as well as an analysis of stability of the clones across sites.

Introduction

In 1991, INIBAP was requested to develop the second phase of the International *Musa* Testing Programme (IMTP). The proposal was approved for a total of three years. The proposal included support to breeding programmes, virus indexing of the germplasm donated by breeding programmes, support to research on viruses, publication of results and staff. No funds were allocated for the establishment and maintenance of trial sites. However, most National Agricultural Research Systems (NARS) decided, given the relevance of IMTP for their programmes, to finance the trials themselves.

As was the case with IMTP Phase I that began in 1989, the aim was to identify, in multilocational trials around the world, resistant banana and plantain hybrids which would meet local requirements and with which small-scale farmers could replace existing susceptible cultivars. Another objective of IMTP was to stimulate breeding by providing information to breeding programmes on the pathological response of their improved cultivars under different ecological conditions. As a spillover effect, IMTP also helped to increase the capacity of national organisations to carry out research on bananas and plantains.

IMTP Phase II evaluated germplasm for resistance to three diseases, namely: black Sigatoka (*Mycosphaerella fijiensis*), yellow Sigatoka (*M. musicola*) and Fusarium wilt (*Fusarium oxysporum* f. sp. *cubense*). Four breeding programmes contributed germplasm (INIBAP 1994); the material was propagated by the INIBAP Transit Center (ITC) and delivered as *in vitro* plantlets. Thirty-seven (37) sites asked for testing material. The majority of IMTP Phase II trials were planted during 1996 and 1997. This report includes results from the

Fusarium (FOC) sites in Australia, Brazil, Honduras, Indonesia, Malaysia, the Philippines, South Africa, Spain, Taiwan and Uganda.

Materials and methods

Plant materials

ITC #	Improved cultivars	ITC #	Landraces	ITC #	Reference clones	Response
ITC0504	FH1A-01	ITC 1259	Burro CEMSA	ITC 1122	Gros Michel	Race 1 susceptible
ITC0506	FH1A-03	ITC 0653	Pisang Mas	ITC 0643	Bluggoe	Race 2 susceptible
ITC1264	FH1A-17	ITC 1138	Saba	ITC 0570	Williams	Race 4 susceptible
ITC 1265	FH1A-23	ITC 1062	Pisang Nangka	ITC 0650	Pisang Creolan	Adult plant resistance
ITC 1262	EMB 402	ITC 0712	Cultivar Rose		Local cultivar	Varied
ITC 1261	EMB 404	ITC 1123	Yangambi Km 3			
ITC 1282	GCTCV 119	ITC 0312	Pisang Jari Buaya			
ITC 1271	GCTCV 215	ITC 0001	Pisang Lilin			
		ITC 1249	Calcutta IR 124			

Experimental design

A completely randomised design with 20 replicates per genotype was used for field design. Plant spacing was 3 x 2 m. No guard plants were used. Plant material was provided to each site as propagating tissue cultures or in vitro plants.

Evaluation

A common protocol was used for all evaluation sites. Reaction to fusarium wilt was evaluated using external and internal symptoms. Seven external symptoms were recorded every month for all plants in a 1-to-3 scale, where 1 was no symptoms and 3 was severe symptoms observed. Monthly recordings began three months after planting and were carried out until harvest. Internal symptoms were recorded at harvest. They were determined by the amount of corm discolouration. The corm was sliced horizontally and the cut area evaluated. Internal discolouration of the rhizome was rated from 1 to 6, where 1 indicated no discolouration; and 6 indicated total discolouration (INIBAP 1994). Agronomic characters were also recorded and infected tissue of the corm was sent to Plant Pathology Unit, DPI, Indooroopilly, Queensland, Australia for VCG identification.

Analysis of stability

Linear regressions of the environmental index on the varietal averages of various important traits were performed. The environmental index of each site was used as the independent variable and the varietal average was used as the dependent variable. The environmental index is a variable that

measures the difference between the average of a given environment with respect to the global average for a given trait. It is obtained by simply subtracting the site average for a given trait from the average for the same trait considering all sites. For example for bunch weight, a positive value of the environmental index indicates a better than average performance of a site while a negative value indicates the opposite. The regression equation of the environmental index on cultivars is a good method to estimate the response of a cultivar to a unit of improvement of the environment. For agronomic traits, a slope (β) of 1 is characteristic of stable cultivars because it indicates that the cultivar changes its average response in the same measure as the general performance of the site. A slope higher than 1 indicates a positive homeostatic response where the genotype improves at a higher rate than the environment average. It means that the cultivar responds better than expected under a positive environment.

On the other hand, the interpretation of the slope (β) for *Fusarium* resistance (expressed as internal symptoms) is quite different as the desired situation is that the average response of the cultivar changes to a lesser degree than the general infection score of the site. A slope (β) lower than 1 indicates a positive homeostatic response where the genotype shows lower infection scores than the site average. It means that the cultivar keeps its resistance/tolerance under higher pathogen pressure.

Results and discussion

Figure 1 provides an overview of the main effects of genotype and site on the average bunch weight. The most favourable site for yield was Taiwan, where management skills are highly developed. Second best yields were obtained from the Uganda site. It should be noted that this site was in a farmer's field, indicating the level of care the farmer devotes to get good production.

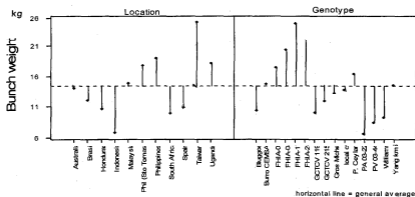


Figure 1. Main effects of location and genotype on the general average for bunch weight.

Internal discolouration of the rhizome is a good estimate of resistance to FOC. Also, this value combined with the agronomic performance is a measure of the tolerance of the accession. However, in the case of IMTP Phase II, it has been quite difficult to make an overall estimation of the resistance or tolerance of the plant genotype to FOC due to large differences on ratings across sites. It is believed that the differences were due to a subjective element of visual interpretation and not only to the differences in pathogen pressure. This is particularly true for discolouration averages across sites infested with FOC race 1 as there were only three sites infested with this race in the whole IMTP trial and one of them (Bago Oshiro, Davao, Philippines) had exceptionally high averages. The overall averages mentioned below should therefore be considered with caution. For comparisons with the reference clone, and resistance or tolerance ratings it is preferable to use the average scores of each genotype within site.

Agronomic performance and response to disease in sites infested with FOC race 1:

FOC race 1 was present in Brazil, Honduras and the Philippines (Bago Oshiro, Davao City) (Figure 2). For VCGs list see Orjeda *et al.* 1999. The average score considering all genotypes across sites infested with FOC race 1 was 2.4. The susceptible reference clone Gros Michel had an average score of 3.2 across sites. The improved genotype with the lowest discolouration score across sites was GCTCV 119, a Giant Cavendish (AAA) tissue culture variant developed by the Taiwan Banana Research Institute (TBRI). This clone had an overall score of 1.3, which reflects its resistance to FOC race 1.

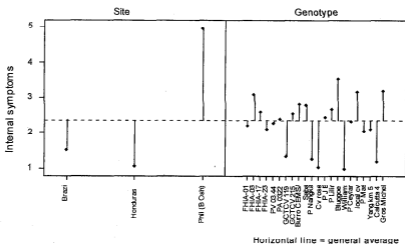


Figure 2. Main effects of location and genotype for the internal symptom average in sites with FOC race 1.

The genotype with the second lowest discolouration score was FHIA-23, an AAAA hybrid between Gros Michel Highgate x SH-3362. This hybrid had an average discolouration score of 2.1 on sites infested with FOC race 1. Although this score was below the overall average and it was the lowest for FOC race 1 of all FHIA hybrids, it already indicates infection in the corm. Given its excellent yield, this hybrid can be classified as very tolerant. It remains to be tested in the following generations to verify tolerance stability.

FHIA-01, an AAAB tetraploid hybrid obtained from crossing Prata Anã x (Prata Anã x SH 3142) showed an average score of 2.2. Although the scores of FHIA-01 already indicate more than isolated points of discolouration in the vascular tissue, they are still lower than the general average score. Despite its yield being the lowest of the FHIA hybrids, FHIA-01 showed the lowest variance across sites amongst the FHIA hybrids. These results suggest that FHIA-01 is tolerant to FOC race 1. This hybrid performs well under a variety of environments and responds positively to good management conditions.

PV 03.44, a Pome-type (AAAB) tetraploid hybrid was developed by EMBRAPA/CNPMP from a cross between Pacovan and Calcutta 4. It had an average score of 2.3 on sites infested with race 1, which was just under the general average for these sites. However, its yield was usually low with a general average of 8.4 kg and a maximum average of 11.3 kg in South Africa.

GCTCV 215 is also a tissue culture variant obtained from Giant Cavendish (AAA) by TBRI. It had an average score of 2.5 for race 1 sites, higher than that of GCTCV 119 and slightly higher than the overall average. However, it is to be noted that this average is substantially influenced by the exceptionally high scores (5.6) recorded in one site in the Philippines (Bago Oshiro, Davao). On the other two sites infested with race 1, this genotype had an average score of 1 (no discolouration). Since this genotype was always considered to be resistant to race 1 these contradictory results deserve re-evaluation.

FHIA-17, an AAAA hybrid obtained by crossing Gros Michel Highgate x SH-3362, had discolouration scores which were above average. Its average score was 2.5 across sites. However, as in the case of GCTCV 119, it is to be noted that this average is substantially influenced by the exceptional high scores (5.6) recorded in one site in the Philippines (Bago Oshiro). On the other two sites infested with race 1, this genotype had an average score of 1 (no discolouration) which is to be expected for Cavendish clones. FHIA-17 was the best yielding genotype of all across sites. It had an average weight of 25.2 kg. FHIA has reported this genotype to be resistant to fusarium wilt race 1. These findings indicate contradictory results among sites that merit further investigation.

FHIA-03, a tetraploid AABB hybrid, had an average vascular discolouration score of 3.08 across sites with FOC race 1, the highest score of all the improved genotypes and very similar to that of Gros Michel, the susceptible reference.

This score is above the total average score when all the test genotypes are considered and it certainly indicates susceptibility. Despite its high discolouration score, this hybrid had very good yield with an average of 20.4 kg across sites and maximum average of 29.8 kg. It seems that this genotype shows tolerance in the plant crop cycle.

Agronomic performance and response to disease in sites infested with FOC race 4:

FOC race 4 was present in Australia, Indonesia, the Philippines (Sto. Tomas), South Africa, Spain and Taiwan. Williams, the susceptible reference for sites infested with race 4, was a dwarf somaclonal variant that could not be used for comparisons. The average overall discolouration score for the race 4 sites was 2.4 considering all genotypes (Figure 3). GCTCV 119 had an average score of 1.3 across sites infested with race 4. As in sites infested with race 1, it was also the lowest discolouration score of all the improved cultivars across trial sites. Yet indications exist that this genotype can be quite susceptible to FOC race 4, although no infection had been recorded for this genotype in the Indonesian site, samples of this genotype sent previously to QDPI confirmed infection. Its average yield was lower than 10 kg bunch. This genotype was very variable with good average yields between 15 kg and 22 kg under some conditions and very low (3 kg) in others. It is believed that management practices have a strong influence on the agronomic performance of this genotype.

GCTCV 215 had an average score of 1.7 (data from Indonesia). However, this was not considered since only one plant was evaluated. This average was the second lowest of all improved genotypes. This somaclonal variant had an average yield of 11.9 kg across sites. However, this average increases to 12.7 kg if the data from Indonesia are not taken into consideration. Since this genotype was always considered to be resistant to race 1 and susceptible to race 4, these contradictory results deserve re-evaluation.

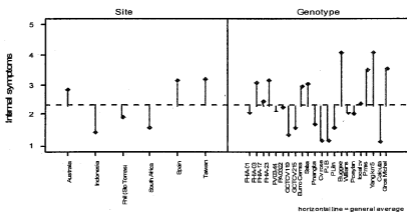


Figure 3. Main effects of location and genotype for the internal symptom average in sites with FOC race 4.

FHIA-01 showed an average discolouration of 2.04 across sites infested with FOC race 4. This score was similar to the average score of the susceptible clone Williams (2.05). However, Williams plants were dwarf somaclonal variants, thus their score is not reliable. FHIA-01 had an average bunch weight across sites of 17.6 kg and a maximum average of 24 kg with optimum management conditions. Given the good performance of FHIA-01 across sites, this genotype can be classified as tolerant to both races 1 and 4 of fusarium wilt.

PV 03.44 had a score of 2.1. This score was under the overall average and lower than that of the susceptible references. However, its yield was usually low with a general average of 8.4 kg and a maximum average of 11.3 kg in South Africa.

PA 03.22 also had a low score in sites infested with race 4; its score was 2.3. Similar to PV 03.44, this score was also under the general average across sites and lower than that of the susceptible references. However, its yield was also usually very low with an overall average of 6.4 kg and a maximum average of 11.6 kg in South Africa.

FHIA-17 average score was 2.5 across sites, the same for FOC race 1 and race 4 sites. This score is above the general average and in Taiwan it had a score of 4.5. Yet FHIA-17 was the best yielding genotype of all in these trials. It had an average weight of 25.2 kg across sites and maximum yields of 43.8 kg in Taiwan. Our results indicate that it is very tolerant to race 4 during the plant crop. It remains to be tested whether following generations retain this level of tolerance.

FHIA-03 had an average score of 3.09 which means that it had around one third of the vascular tissue showing discolouration on both types of sites. These scores are above the total average score when all the participating genotypes are considered and they certainly indicate susceptibility. Moreover, in some sites, FHIA-03 had very high discolouration scores (4.3 to 5.7) as in the Canary Islands and Australia. Despite its high discolouration scores, surviving plants of this hybrid gave some very good yields with an average of 20.4 kg across sites and maximum average of 29.8 kg. The only exception to the high yield was in the Canary Islands. In this particular case, the data were collected from only two plants.

FHIA-23 had an average score of 3.1, well above the general average indicating susceptibility. Moreover, in Australia and the Canary Islands it had an average above 4, which indicates high susceptibility to sub-tropical race 4. Despite its high discolouration scores, this hybrid was the second best yielding of all FHIA hybrids with an average bunch weight of 22.3 kg and a maximum average yield of 46.8 kg in Taiwan.

Landraces

Bluggoe was the genotype with the highest discolouration scores on both types of sites. It had a score of 3.5 on race 1 sites and 4.1 on sites with race 4, indicating high susceptibility to both races of FOC. Burro CEMSA, an ABB collected by INIVIT and classified by them as a somaclonal variant of Bluggoe had less discolouration scores than Bluggoe. It had a general average of 2.8 in sites with race 1 and an average score of 2.9 in sites with race 4. The other genotype that showed susceptibility to both races was Gros Michel with a score of 3.2 for race 1 sites and 3.5 for race 4 sites. Another landrace that showed higher than the general average discolouration scores for both types of sites was Saba. On sites with FOC race 1, Saba had a general average of 2.8 and on sites infested with FOC race 4, a score of 3.

Some landraces had a very specific reaction to the race of FOC. For example, Yangambi Km 5 and Pisang Mas. Yangambi had a score of 2.1 which was lower than the general average for sites with race 1 but it had a score of 4.1 well above the average for sites with race 4. The difference, although less marked, was also evident for Pisang Mas. On sites with race 1, Pisang Mas had a score of 2.06 while for sites with race 4 it had a score of 3.5.

Landraces that consistently had scores lower than the average, were Cultivar Rose and Pisang Nangka. It should be noted that the Cavendish clone Williams that was included as the race 4 susceptible reference was a dwarf somaclonal variant and therefore, data from this clone should not be considered reliable.

Stability of clones

Linear regressions of agronomic traits on the environmental index were performed. For a genotype to be considered stable, the slope (β) needs to be at least equal to one, meaning that the change in the genotype average changes in the same measure as the environment. The four FHIA hybrids proved to be the most stable genotypes across environments. Moreover, they generally had b values higher than one which indicated a better than expected performance of the hybrids in the various environments. Somaclonal variant GCTCV 119 was also very stable for bunch weight ($\beta = 1.18$ $p < 0.01$), and number of hands ($\beta = 1.17$ $p < 0.05$). Somaclonal variant GCTCV 215 was very stable for the average weight of fruits ($\beta = 1.41$ $p < 0.01$) and had good stability for the number of hands ($\beta = 0.701$ $p < 0.05$) and for bunch weight ($\beta = 0.710$ $p < 0.05$).

The case of number of days to harvest was slightly different, as the number of days to harvest is greatly dependent on the type of climate and local practices. The regression index b , is therefore, generally close to one and always highly significant ($\beta < 0.01$). The strong correlation indices (R^2) between the genotype averages for days to harvest and the environmental indices for this trait corroborate this fact.

Figure 4 plots the bunch weight versus the slope (β) that classifies for stability. FHIA-17 and FHIA-23 were the most stable and best yielding cultivars across sites followed by FHIA-03. FHIA-01, on the other hand, although it yields well, had too much variability from site to site. GCTCV 119 was also classified as stable although its bunch weights were slightly lower than the general average. Figure 5 plots the average internal symptom score versus the slope (β). In this case, the lower slope means that the genotype shows relatively fewer symptoms in a site where the other genotypes showed more severe symptoms. In other words, the average score of the genotypes does not increase at the same rate as the site average, which is expected from a resistant or tolerant genotype.

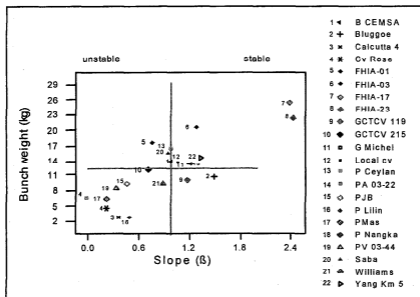


Figure 4. Comparative behaviour of genotypes according to bunch weight and stability.

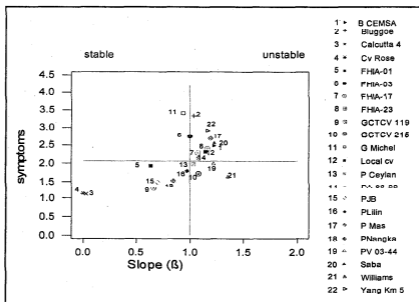


Figure 5. Comparative behaviour of genotypes according to internal symptoms and stability.

An improved cultivar that deserves special reference is GCTCV 119, which had the lowest discolouration score for both FOC races and good yields under good management. FHIA-01 also showed a good stability and had lower than average symptoms for both FOC races.

Acknowledgments

IMTP Phase II represents the effort of many dedicated researchers and breeding programmes, national agricultural research programmes and funding agencies around the world. Special thanks to the improvement programmes of FHIA, CNPMF/EMBRAPA, TBRI and INIVIT that donated germplasm; to the site evaluators that did the fieldwork, and to Suzy Bentley from CRC for Tropical Plant Pathology Laboratories for her contribution to the DNA fingerprinting.

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