

The Improvement and Testing of *Musa*: a Global Partnership

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DR Jones

INIBAP's Mandate

The International Network for the Improvement of Banana and Plantain (INIBAP) was established in 1984 and has its headquarters in Montpellier, France. INIBAP is a nonprofit organization whose aim is to increase the production of banana and plantain on smallholdings by:

- initiating, encouraging, supporting, conducting, and coordinating research aimed at improving the production of banana and plantain;
- strengthening regional and national programs concerned with improved and disease-free banana and plantain genetic material;
- facilitating the interchange of healthy germplasm and assisting in the establishment and analysis of regional and global trials of new and improved cultivars;
- promoting the gathering and exchange of documentation and information; and
- supporting the training of research workers and technicians.

Planning for the creation of INIBAP began in 1981 in Ibadan with a resolution passed at a conference of the International Association for Research on Plantain and Bananas. In May 1994, INIBAP was brought under the governance and administration of the International Plant Genetic Resources Institute (IPGRI) to enhance opportunities for serving the interest of small-scale banana and plantain producers.

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Cover illustration: Symptoms of black leaf streak/black Sigatoka disease on a leaf of a highly susceptible 'Cavendish' cultivar growing on Aitutaki Island, Cook Islands (photo: DR Jones, INIBAP).

INTERNATIONAL NETWORK FOR THE
IMPROVEMENT OF BANANA AND PLANTAIN

inibap

Plantain Breeding at IITA

R Ortiz¹, D Vuylsteke²

Plantain and Banana in Africa

Plantain and banana are important food crops in the humid forest and mid-altitude zones of sub-Saharan Africa, providing more than 25% of the carbohydrates for approximately 70 million people in the region (Vuylsteke et al. 1992). In addition to being a staple food for rural and urban consumers, plantain and banana provide an important source of rural income, particularly for the small holders who produce them in compound or home gardens (Dorosh 1988; Nweke et al. 1988). The gross value of annual production exceeds that of several other main food crops, such as maize, rice, cassava, and sweet potato (Vuylsteke et al. 1993b).

The bulk of cultivated *Musa* are triploids ($2n = 3x = 39$). Almost completely sterile, they develop fruit by parthenocarpy. The most important cultivars vary in their genomic constitution, which is generally as follows: dessert banana (AAA), East African highland banana (AAA), plantain (AAB), and cooking banana (ABB). The genome of cultivated clones is derived from the diploid wild species *M. acuminata* Colla. and *M. balbisiana* Colla., which contributed the A and B genomes, respectively (Simmonds 1962).

Although Southeast Asia is considered to be the center of origin of *Musa*, a remarkable diversity of plantain and banana exists in sub-Saharan Africa (De Langhe 1961; Ortiz et al. 1993c; Swennen, Vuylsteke 1987). Each of the different types is grown in a distinct subregion. Thus, the AAB plantain cultivars are predominant in the humid lowlands of West and Central Africa, while AAA cooking and beer banana types are prevalent in the East African highlands (Swennen, Vuylsteke 1991a). The former region harbors the world's greatest variability of plantain and is thus considered a secondary center of plantain diversification. Similarly, East Africa is considered a secondary center of diversity for banana of the *Musa* AAA group. This secondary diversification is the result of somatic mutations and human selection during the long history of cultivation of the crop in the region (De Laigue 1904).

Sub-Saharan Africa produces about 35% of the world bananas and plantains, about 70 million t. a⁻¹. Pest and disease pressure on the crop has been increasing over the past

15 years due to intensification of production (Wilson 1988). Also, rising population pressure on the land has led to altered farming practices, among which shortening fallow periods are most conspicuous. These rapid agricultural changes provide the context for IITA's work in this area, particularly that aimed at developing appropriate and ecologically sustainable technology for resource, crop, and pest management in *Musa* farming systems (Vuylsteke et al. 1993c).

Production Constraints

Black leaf streak/black Sigatoka leaf spot disease, caused by *Mycosphaerella fijiensis* Morelet, is considered to be the most serious constraint to plantain and banana production in sub-Saharan Africa (Swennen et al. 1989). The disease was accidentally introduced to this continent some two decades ago and spread rapidly, first in Central and West Africa, and later in East Africa (Wilson, Buddenhagen 1986). The pathogen can cause severe leaf necrosis, reducing yields by 30-50% (Mobambo et al. 1993). All plantain cultivars are susceptible to black leaf streak/black Sigatoka (Hahn et al. 1989), as are, at least, some of the most widely grown banana cultivars in East Africa (Vuylsteke, Swennen 1988). Other major diseases, mainly in East Africa, are Fusarium wilt and banana bunchy top virus (Swennen, De Langhe 1989).

An important insect pest of plantain and banana in Africa is the banana weevil (*Cosmopolites sordidus* Germar), whose larvae bore tunnels in the corm, thus weakening the plant. Plantain and the East African highland banana are susceptible to the insect, though its effects have not yet been quantified (Vuylsteke et al. 1993c).

Another formidable obstacle to plantain cultivation in West and Central Africa is the rapid yield decline that occurs after a year or two when the crop is grown in open field plantations (Wilson 1987). In contrast, plantations of dessert banana remain productive for many years, as does plantain within home or village compound gardens, where it benefits from regular application of organic matter in the form of household refuse (Nweke et al. 1988). The causes of this problem are complex. In East Africa, declining yields are undoubtedly related to reduced soil fertility and mineral deficiencies in the poorer soils. Yield decline under open field conditions can be arrested somewhat through mulching (Wilson et al. 1987). In addition to soil fertility problems, damage caused by the banana weevil and nematodes generally worsens with time (Swennen et al. 1988). Also, plants lodge easily under field conditions as a result of factors linked to the plant's basic physiology. These include poor root development, slow ratooning, and "high mat" (a tendency of the plant base to grow out of the soil). These problems can probably be ameliorated through plant breeding (Ortiz, Vuylsteke 1994a).

Postharvest losses of plantain are a serious deterrent to expanded production in some African countries (Wilson 1987; 1988). Oversupply during the main production season is the primary cause, but losses can also be attributed to poor methods of harvesting, transporting, and storing the fruit (Ferris et al. 1993a,b; Thompson et al. 1992).

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Musa Breeding and Genetics at IITA

Research on plantain at IITA began in 1973. In 1979, the center of plantain research was transferred to the Onne station, in the more suitable ecological niche of the humid forest zone. IITA included plantain and banana among its mandate crops in 1987 and the Plantain and Banana Improvement Program (PBIP) was created in 1991 as a full part of the Crop Improvement Division of IITA (Vuylsteke et al. 1990b).

IITA was encouraged by several African governments to launch an urgent campaign to control black leaf streak/black Sigatoka. IITA scientists were impelled by the need to save the plantain for the millions of smallholders who depend on it for subsistence and livelihood. In 1987, a *Musa* genetic improvement program was initiated at IITA which aimed to incorporate durable host-plant resistance to black leaf streak/black Sigatoka leaf spot disease into plantain (Swennen, Vuylsteke 1991b). The Institute was aware, however, that the development of resistance to streak virus in maize (Efron et al. 1989), which is a much simpler plant to cross, took 10 years. IITA had expected at least another 10 years to develop improved plantain germplasm resistant to black leaf streak/black Sigatoka. Therefore, plantain scientists at Onne Station assessed the potential of Sigatoka-resistant cooking banana cultivars (ABB group) as a possible substitute crop (short-term approach) (Hahn et al. 1990), and at the same time they started a long-term endeavor: breeding plantain through a combination of conventional and innovative cross-breeding techniques (Vuylsteke et al. 1994a).

Adoption of the resistant cooking banana cultivars, which were tested in West Africa in collaboration with NARS and INIBAP, has so far been limited, because of West Africans' lack of familiarity with them and their clear preference for plantain (Vuylsteke et al. 1992).

High-Yielding Plantain Resistant to Black Leaf Streak/Black Sigatoka

The problems in developing improved *Musa* germplasm have been attributed to difficulties associated with banana breeding at the genetic and practical levels. Genetic improvement has been severely hampered by lack of genetic variability and low levels of female fertility within the germplasm collections. Genetically speaking, plantain is a triploid, having three sets of chromosomes instead of the two carried by many crop plants (which are diploids). Moreover, space (6 m² per plant) and time (1.5 years from seed to seed) requirements also impede breeding progress. These problems were alleviated by screening the germplasm for rearity (Swennen, Vuylsteke 1988; 1990), ploidy manipulations (Vuylsteke et al. 1993f) and intensive application of tissue culture techniques (Swennen et al. 1992; Vuylsteke, Swennen 1993). By the end of 1989, the future of plantain was already beginning to look bright—there had been a breakthrough in the crossing of susceptible plantain cultivars with resistant diploid banana clones (Swennen, Vuylsteke 1993). Patience, vision, and core commitment by IITA management have also fostered success.

The development of tropical *Musa* plantain hybrids (TMPx)

Thirty sources of black leaf streak/black Sigatoka resistance were identified among the more than 400 *Musa* accessions screened in the field gene bank (Swennen, Vuylsteke 1991a). Most of the resistant accessions are diploid *M. acuminata* (AA), both wild and cultivated types, and ABB cooking banana cultivars.

The 119 different plantain cultivars in the Onne collection were screened for female fertility by hand-pollinating these with a wild banana (Swennen, Vuylsteke 1993). Twenty-nine French and eight False Horn plantain cultivars were identified as female-fertile because they produced viable true seed. This is the highest number of seed-fertile plantain clones reported so far. Also, seed production rates in plantain at the Onne station seem to be higher than previously expected (Swennen et al. 1991). Average seed-set ranged from less than 1 to more than 20 seeds per bunch, depending on the cultivar (Jenny et al. 1993; Vuylsteke et al. 1993f). The relatively high female fertility of plantain at Onne was one of the key elements in breeding advancement.

The initial breeding approach involved producing primary tetraploid progenies via $3x \times 2x$ crosses, in which the triploid female plantain (AAB) produced $2n (-3x)$ eggs (Vuylsteke et al. 1993d). The bulk of the pollination effort was on two plantain cultivars (Obino l'Esrai from Nigeria and Bobby Tannap from Cameroon) of which 3750 plants (2.25 ha) were rapidly propagated by in-vitro culture techniques. The main pollen parent was a wild relative, *M. acuminata* ssp. *burmannicoides* clone Calcutta 4, which is highly resistant to black leaf streak/black Sigatoka (Swennen, Vuylsteke 1993). Its resistance was readily inherited in its offspring, but not its poor bunch characters (Vuylsteke et al. 1993f).

More than 250 hybrids were field-established in early evaluation trials in 1989-90. From these, 20 tetraploids were selected for in-vitro multiplication and further field evaluation in preliminary yield trials and in comparison with their plantain parents. Selection criteria were partial resistance to black leaf streak/black Sigatoka and good agronomic and fruit quality characters (Vuylsteke et al. 1993g). Genotype response to black leaf streak/black Sigatoka infection was measured by recording the number of the youngest leaf spotted (YLS), counting down from the first (top) unfurled leaf, on plants at flowering, being the time at which leaf production stops. Increasing YLS values indicate the presence of more healthy leaves on the plant and, hence, greater resistance to the fungus (Craenen 1994). After intensive testing from 1989 to 1992 at Onne station in the humid forest zone near Port Harcourt, Nigeria, a total of 14 TMPx genotypes were further selected for distribution to agricultural research programs. On average, it took 1000 seeds, produced from hand-pollination of 200 plants (0.12 ha), to obtain one selected tetraploid hybrid per year.

The TMPx clones in Table 1 are identified by their original cross/progeny number. All the TMPx have higher levels of black leaf streak/black Sigatoka resistance than their susceptible plantain parents, as determined by an evaluation of the youngest leaf spotted. Selected TMPx may show a gain of up to six leaves without black leaf streak/black Sigatoka spotting when compared with the fungicide-treated parents (Table 1). Host response to black leaf streak/black Sigatoka in the partially resistant TMPx germplasm is based on

Table 1. Agronomic evaluation of 14 selected TMPx clones at Onne, Nigeria, from 1990 to 1992¹ (after Vuylsteke et al. 1993g).

Genotype	Parents ²	YLS	PH	HTSh	DFV	BW	H	F
		#	cm	cm	days	kg	#	#
TMPx 548-4	OL x C4	10.0 ^{ns}	330 ^{ns}	280 ^{ns}	126 ^{ns}	16.7 ^{ns}	6.5 ^{ns}	103 ^{ns}
TMPx 548-9	OL x C4	11.0 ^{ns}	290 ^{ns}	310 ^{ns}	131 ^{ns}	10.5 ^{ns}	0.0 ^{ns}	00 ^{ns}
TMPx 582-4	BT x C4	11.0 ^{ns}	300 ^{ns}	270 ^{ns}	135 ^{ns}	14.3 ^{ns}	7.5 ^{ns}	124 ^{ns}
TMPx 1621-1	OL x C4	7.5 ^{ns}	340 ^{ns}	280 ^{ns}	120 ^{ns}	13.8 ^{ns}	6.5 ^{ns}	90 ^{ns}
TMPx 1658-4	OL x PL	9.3 ^{ns}	320 ^{ns}	185 ^{ns}	134 ^{ns}	21.5 ^{ns}	7.5 ^{ns}	123 ^{ns}
TMPx 2637-49	OL x C4	11.6 ^{ns}	360 ^{ns}	225 ^{ns}	127 ^{ns}	16.6 ^{ns}	6.4 ^{ns}	95 ^{ns}
TMPx 2796-5	BT x PL	10.0 ^{ns}	335 ^{ns}	205 ^{ns}	123 ^{ns}	21.3 ^{ns}	6.5 ^{ns}	101 ^{ns}
TMPx 4479-1	BT x C4	9.0 ^{ns}	295 ^{ns}	290 ^{ns}	114 ^{ns}	13.2 ^{ns}	6.0 ^{ns}	88 ^{ns}
TMPx 4698-1	OL x C4	10.5 ^{ns}	345 ^{ns}	275 ^{ns}	130 ^{ns}	20.0 ^{ns}	8.5 ^{ns}	124 ^{ns}
TMPx 4744-1	OL x C4	10.0 ^{ns}	360 ^{ns}	235 ^{ns}	125 ^{ns}	11.5 ^{ns}	6.0 ^{ns}	89 ^{ns}
TMPx 5511-2	OL x C4	10.0 ^{ns}	345 ^{ns}	165 ^{ns}	118 ^{ns}	17.8 ^{ns}	6.0 ^{ns}	87 ^{ns}
TMPx 5700-1	OL x C4	7.5 ^{ns}	340 ^{ns}	250 ^{ns}	109 ^{ns}	10.6 ^{ns}	6.0 ^{ns}	06 ^{ns}
TMPx 6930-1	OL x C4	10.8 ^{ns}	330 ^{ns}	240 ^{ns}	138 ^{ns}	17.5 ^{ns}	7.5 ^{ns}	121 ^{ns}
TMPx 7002-1	OL x C4	13.0 ^{ns}	340 ^{ns}	270 ^{ns}	120 ^{ns}	17.5 ^{ns}	6.0 ^{ns}	115 ^{ns}
Bobby Tannap ³		7.0	340	171	92	14.0	7.8	100
		± 0.2 ⁴	± 2	± 3	± 0	± 0.2	± 0.1	± 1
Obino l'Ewai ³		7.0	370	188	92	12.4	7.2	91
		± 0.2	± 2	± 5	± 0	± 0.2	± 0.1	± 1

¹YLS = youngest leaf spotted, PH = plant height, HTSh = height of tallest sucker at harvest, DFV = days to fruit-filling, BW = bunch weight, H = number of hands, F = number of fingers.

²OL = Obino l'Ewai; BT = Bobby Tannap; C4 = Calcutta 4; PL = Pisang Lilin.

³Non-fragile treated plots.

⁴Mean ± standard error (n=200).

ns, **, * = nonsignificant or significant at $P = 0.05$ or 0.01 , respectively, for multiple comparison within columns of TMPx genotypes with the respective plantain parent. Critical values of Student's F distribution were adjusted by Sidak's multiplicative inequality based on $\alpha = 1 - (1 - \alpha)^{1/c}$, in which α is the level of significance required for statistical testing and c is the number of comparisons between the hybrids and the plantain parents (11 and 3 for OL and BT, respectively).

slower or delayed disease development. The ensuing reduction of leaf spot damage results in a larger healthy leaf area during fruit-filling time and, thus, larger and heavier fruit. Black leaf streak/black Sigatoka resistance results from the interaction between a major recessive gene and two modifiers with additive effects (Ortiz, Vuylsteke 1994c).

The tetraploid hybrids show close phenotypic resemblance to their respective female plantain parents. All hybrids display parthenocarpic fruit development and all but one, TMPx 4698-1 (Fig. 1), have erect fruit orientation like their plantain parents. Most hybrids have a pendulous bunch orientation, except TMPx 582-4 and TMPx 4744-1, which have a subhorizontal bunch. Neutral flowers are deciduous in all hybrids, except TMPx 4744-1, which has semipersistent neutral flowers. TMPx 548-9 (Fig. 2), 582-4, 4479-1,

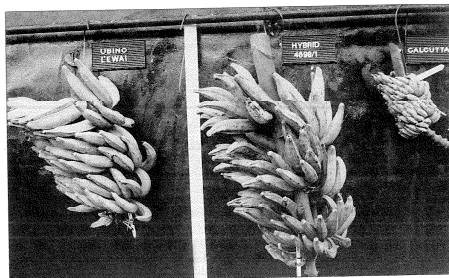


Figure 1. Black leaf streak/black Sigatoka-resistant tetraploid hybrid TMPx 4698-1 (middle) derived from a cross of the susceptible triploid plantain cultivar Obino l'Ewai (left) with the highly resistant wild diploid banana Calcutta 4 (right).

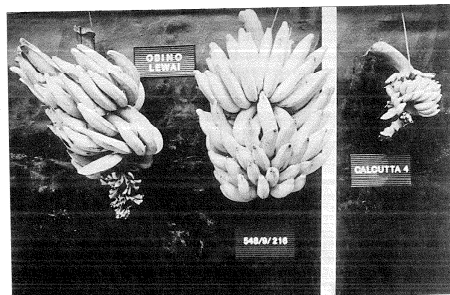


Figure 2. Black leaf streak/black Sigatoka-resistant tetraploid hybrid TMPx 548-9 (middle) derived from a cross of the susceptible triploid plantain cultivar Obino l'Ewai (left) with the highly resistant wild diploid banana Calcutta 4 (right).

4698-1, 4744-1, 5511-2, 6930-1, and 7002-1 exhibit male bud imbrication. The TMPx germplasm develops one or two suckers freely while further suckering is inhibited. Regulated suckering is a highly desirable trait for perennial plantain production. In contrast to the plantain parents, which are female-fertile but male-sterile, the hybrids are female- and male-fertile and can be used as parents to produce secondary triploid hybrids by $4x \times 2x$, and vice versa, crosses.

The agronomic performance of TMPx clones, along with that of the fungicide-treated plantain parents, is shown in Table 1. TMPx germplasm is shorter in plant stature and has a taller sucker (follower) at harvest, both of which are desirable traits. Most of the TMPx clones yield more than their fungicide-treated plantain parents. Most hybrids have fewer hands, but they generally have a higher number of fruit per bunch, which is an important component of yield.

TMPx 548-9 (Fig.2), also known as PITA-2, is an exceptional hybrid. This clone has a cylindrical pendulous bunch and large parthenocarpic fruit with yellow-orange pulp. The fruit is heavier and thicker than the fruit of its plantain parent Obino l'Ewai. The performance of TMPx 548-9 was compared with that of its plantain parent in an independent replicated clonal evaluation trial at Onne (Mobambo et al. 1993). The susceptible plantain parent was maintained under fungicide and nonfungicide conditions. TMPx 548-9 flowered 56 to 97 days earlier than Obino l'Ewai, with or without fungicide. Yield of the TMPx 548-9 plant crop (33.5 t.ha^{-1}) was 43% higher than that of the fungicide-treated plot of Obino l'Ewai (23.5 t.ha^{-1}) and 100% higher than in the nontreated plot of Obino l'Ewai (15.7 t.ha^{-1}). It is inferred that black leaf streak/black Sigatoka resistance is not the sole component of higher yield in TMPx 548-9. High yield could also be partly due to the manifestation of heterosis in this tetraploid hybrid (Vuysteke et al. 1993d).

Plantain-derived diploids with black leaf streak/black Sigatoka resistance (TMP2x)

Diploid, triploid, and tetraploid progenies were obtained from the $3x \times 2x$ crosses (Vuysteke et al. 1993d). In plantain-derived F_1 populations, diploids represented about 80%, but there was a significant difference between plantain cultivars in the proportions of different ploidies. This suggests a difference in the rate of $2n$ egg production between cultivars.

While tetraploid hybrids are of immediate interest as potential new cultivars, the plantain-derived diploids (TMP2x) play an increasingly important role in germplasm enhancement at the $2x$ level and as a source of plantain alleles (Vuysteke, Ortiz 1993). Since 1991, crosses between tetraploid hybrids and diploids were performed to produce secondary triploids. The latter approach was pursued mainly to reintroduce male sterility, as such avoiding seed-set in the hybrids (Vuysteke et al. 1993e).

All the TMP2x have higher levels of black leaf streak/black Sigatoka resistance than their susceptible plantain parents, as determined by an evaluation of YLS. Host response to black leaf streak/black Sigatoka in the partially resistant TMP2x germplasm is based

on slower or delayed disease development. Less leaf spot damage ensues. Such host response seems to provide more durable resistance.

Two exceptional Bungenoisan-derived hybrids, TMP2x 2348-6 and TMP2x 2348-7, were initially selected due to their high bunch weight and large parthenocarpic fruits (Table 2). In addition, these two fertile TMP2x have short to medium plant size and low apical dominance, i.e. improved suckering behavior.

Other promising diploid hybrids derived from Bobby Tannop and Obino l'Ewai were evaluated in preliminary yield trials in both plant and ratoon crops. Growth and yield characteristics of the average diploid population, the selected TMP2x and of the male parent Calcutta 4 are listed in Table 3.

All selected TMP2x do not show the high apical dominance of plantain. Moreover, TMP2 x 597-2, 1199-6, 2625-20, 2829-62, and 9722-1 develop only one or two suckers freely while further suckering is inhibited. Such regulated suckering is a highly desirable, dominant trait for perennial plantain production. In contrast to their plantain parents, which are male-sterile, all the diploid hybrids, except TMP2x 9722-1, are male-fertile. Figure 3 shows the bunch of TMP2x 1518-4 along with those of its plantain (Bobby Tannop) and wild banana (Calcutta 4) parents. Another two clones, TMP2x 1297-3 and TMP2x 1605-1, were also selected in the period 1989-1991. TMP2x 1297-3 and 1605-1, resistant to black leaf streak/black Sigatoka, have low apical dominance and high-yielding subhorizontal (to almost pendulous) bunches with parthenocarpic fruit. TMP2x 1605-1, a diploid derived from the giant plantain Ntanga-2, is male-sterile but female-fertile. TMP2x 1297-3 is a diploid derived from the French Reversion somaclonal variant of Agbagba. TMP2x 1297-3 offers the opportunity to breed in the previously inaccessible False Horn plantain gene pool. For example, TMP2x 1297-3 was crossed with its full-sib triploid TMPx 1112-1 to produce tetraploid and diploid segregating offspring in which directional selection for large fruit size was applied. This breeding scheme aims

Table 2. Agronomic evaluation of four selected TMP2x clones and average plantain-derived diploid population performance in early evaluation trials at Onne, Nigeria, in 1989/1990¹ (after Vuysteke and Ortiz, pers. comm.).

Clone	YLS #	PH cm	HTSh cm	BW kg	H #	F #	FW g	FL cm	FC cm	DFF days
TMP2x 2348-6	10	2.7	2.9	13.9	8	126	98	15	10	139
TMP2x 2348-7	9	2.2	2.9	15.2	7	117	130	15	10	165
TMP2x 597-2	9	3.6	1.9	7.8	7	108	72	18	10	118
TMP2x 1448-1	9	3.5	3.2	6.0	8	161	35	11	9	181
2x population ²	8	2.5	2.5	4.3	6	92	46	11	10	115
		± 0.2	± 0.1	± 0.1	± 0.7	± 0.2	± 6	± 6	± 1	± 6

¹YLS = youngest leaf spotted, PH = plant height, HTSh = height of tallest sucker at harvest, BW = bunch weight, H = number of hands, F = number of fingers, FW = fruit weight, FL = fruit length, FC = fruit circumference, DFF = days to fruit-filling.

²Mean \pm standard error.

Table 3. Agronomic evaluation of 10 selected TMP2x clones in comparison with the performance of their diploid male parent Calcutta 4 and the average plantain-derived diploid population in preliminary yield trials at Onne, Nigeria, 1991-93 (plant and raton crops)¹ (after Vuysteke and Ortiz, pers. comm.).

Clone	YLS #	PH m	HTSh:BW kg	H #	F #	FW g	FL cm	FC cm	DFF days	
TMP2x 597-2	10	3.6	3.5	7.4	6	94	79	15	10	125
TMP2x 1199-6	10	3.1	2.5	8.3	7	125	65	16	9	115
TMP2x 1448-1	9	3.1	3.3	5.8	7	122	50	13	8	125
TMP2x 1518-4	10	3.4	2.6	5.6	9	146	54	13	6	103
TMP2x 1549-5	10	3.6	2.8	5.8	10	187	35	13	7	117
TMP2x 1657-4	9	3.1	2.7	7.9	8	155	48	13	8	125
TMP2x 2625-20	12	2.6	2.6	7.9	8	112	69	14	10	149
TMP2x 2829-62	11	3.4	2.8	5.5	8	137	35	14	7	123
TMP2x 4600-12	12	2.9	2.5	6.1	7	107	55	11	8	143
TMP2x 9723-1	13 ²	0.0	0.0	5.7	0	149	90	14	7	87
2x population ²	8	2.6	2.2	3.0	6	95	32	11	7	117
Calcutta 4 ³	+0.2	+0.2	+0.1	+0.2	+0.1	+4	+2	+0.3	+0.2	+3
	-3	2.2	2.8	0.8	7	113	6	5	3	104
	+0.2	+0.2	+0.3	+0.2	+15	+2	+1	+1	+4	

¹YLS = youngest leaf spotted, PH = plant height, HTSh = height of tallest sucker at harvest, BW = bunch weight, H = number of hands, F = number of fingers, FW = fruit weight, FL = fruit length, FC = fruit circumference, DFF = days to fruit-filling.

²Mean \pm standard error.

³Highly resistant to black leaf streak/black Sigatoka; leaf spots not readily observed.

to develop improved *Musa* germplasm with fruit size and quality like the preferred Palao Horn plantains.

Most of the improved plantain-derived diploids were crossed with primary TMPx tetraploids to assess their breeding value. Several secondary triploids, combining short to medium plant stature, high levels of resistance to black leaf streak/black Sigatoka, low apical dominance or regulated suckering, pendulous bunches with parthenocarpic fruit, and acceptable yields, were identified in the segregating populations. This was expected because promising tetraploid hybrids (TMPx) were earlier selected even when using a wild nonselected male parent such as Calcutta 4. Segregation for plant size was expected because all hybrids are carriers of the recessive dwarfism gene. Moreover, TMP2x should produce offspring with more plantain-like characteristics than progenies derived from Calcutta 4, because TMP2x have 50% plantain genes in their genome. This may be important for improving fruit quality in the plantain breeding population.

Registration of improved germplasm

Registration of improved germplasm, combining black leaf streak/ black Sigatoka resistance, high yield, and other useful attributes, is actively pursued in order to place

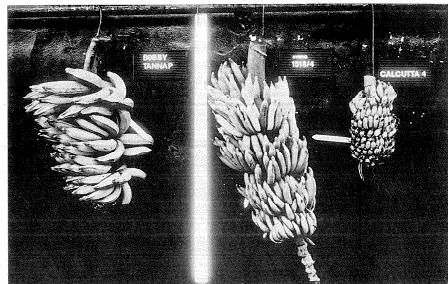


Figure 3. Black leaf streak/black Sigatoka-resistant plantain-derived diploid hybrid TMP2x 1518-4 (center), obtained from crossing the 3x-susceptible Bobby Tannap plantain (left) with the highly resistant wild 2x Calcutta 4 banana (right).

this new technology in the public domain and to provide landmarks of technological advancement. Fourteen improved tropical *Musa* plantain hybrids (4x), now referred to as TMPx, were registered in the journal HortScience (Vuysteke et al. 1993g). Registration of some plantain-derived diploid hybrids is in preparation.

Gaining Insight into the *Musa* Genome

Genetic information is required to develop scientific breeding strategies. However, few genetic studies have been undertaken in *Musa*, despite the importance of the crop. Moreover, others have claimed that "formal genetic studies of nearly or quite sterile triploids are impossible", illustrating the absence of inheritance studies in plantain and banana during the past 40 years. As a consequence, very few genetic markers were available in *Musa* before 1992.

Several characteristics of the crop make genetic analysis of *Musa* difficult. The low rate of hybrid progenies recovered after interploidy/interspecific crosses, resulting in small sample sizes, is the major obstacle to genetic analysis. Nevertheless, the production of testcross segregating populations, obtained from triploid (heterozygous parents) x diploid (homozygous recessive parent) crosses, and of diploid plantain-banana hybrids have made genetic analysis in *Musa* possible (Ortiz 1992). Genetic analyses in the TMP2x were simplified due to disomic inheritance. As such, the plantain and banana genomes, which were inaccessible until recently, were investigated.

Widening plantain variability through segregation in the plantain genome

Variation in growth and yield parameters, qualitative morphological traits, and black leaf streak/black Sigatoka reaction was observed within the same tetraploid family obtained from crosses of plantain with Calcutta 4, a true-breeding line (Vuylsteke et al. 1993f). This observation was surprising, as it was expected to have an entirely uniform progeny from the combination of unreduced female gametes and the male gametes of a homozygous species. Therefore, the occurrence of genetic segregation in the triploid plantain genome during the modified megasporogenesis leading to 2n egg formation was inferred (Ortiz, Vuylsteke 1994c). Hence, the production of megasporocytes and embryo sacs with the maternal chromosome number does not necessarily imply that these carry the intact maternal genotype (Vuylsteke et al. 1993f).

This inference challenged the commonly accepted premise about the $3x \times 2x$ breeding approach, in which the $3x$ female genome is apparently fixed with recombination only possible from the $2x$ male parent. Due to the occurrence of segregation and recombination in the $2x$ female plantain genome, much more variability can be recovered from crosses on plantains.

Genetics of traits

The genetics of resistance to black leaf streak/black Sigatoka (Ortiz, Vuylsteke 1994c) and to banana weevil, dwarfism (Ortiz, Vuylsteke 1993b), albinism (Ortiz, Vuylsteke 1994b), apical dominance and suckering behavior (Ortiz, Vuylsteke 1994a), fruit parthenocarpy (Ortiz, Vuylsteke 1992), bunch orientation (PBIP 1993), pseudostem waxiness (PBIP 1993), male and female sterility (PBIP 1993), bunch weight, its yield components and other agronomic quantitative traits (Ortiz, Vuylsteke 1993b) were elucidated.

Continuous variation has been considered as a feature of quantitative polygenic traits. However, there are several traits, e.g. plant height, for which 'major genes' can be grouped into classes, but within each class there is continuous variation. Several traits showing continuous distribution in plantain and banana are mainly affected by major genes (Ortiz et al. 1994a). For example, major genes controlling dwarfism, black leaf streak/black Sigatoka resistance and apical dominance are inherited as recessive genes in the plantain germplasm (Ortiz, Vuylsteke 1993b; 1994a,c).

Trisomic inheritance and genome differentiation

It is generally accepted that plantain is a triploid derived from interspecific crosses between the diploid species *M. acuminata* and *M. balbisiana*, which contributed the A and B genomes, respectively. Consequently, the AAB genome designation was given to plantain due to its interspecific origin and based on a putative differentiation between the A and B genomes. Genome differentiation was investigated with the aid of genetic-marker segregation in diploid populations derived from $3x$ (plantain) $\times 2x$ (wild banana) crosses. Most of the markers analyzed fitted a trisomic rather than a disomic ratio (Ortiz, Vuylsteke 1994d). Hence, plantain has a trisomic pattern of inheritance because each

linkage group occurs three times instead of twice. Furthermore, there was no preferential pairing between the homologous chromosomes of the A genome, but random distribution of the paired chromosomes to the cell poles during anaphase I of the first meiotic division. This suggests that there is no genome differentiation between *M. acuminata* and *M. balbisiana* and, therefore, the AAB genomic designation for plantain should be discontinued or replaced with a more specific genetic characterization when necessary. This was further supported by analysis of male and female fertility in diploid hybrids (PBIP 1993). Pollen production and stainability, as well as seed-set after interdiploid crosses, were the criteria to determine whether the hybrids were sterile or fertile. Under the hypothesis of genome differentiation, AB hybrids should be sterile. Thus, if this hypothesis were correct, 1/3 of the diploid hybrids should be both male- and female-sterile. The high percentage of fertile hybrids, however, suggests that this hypothesis was not true.

Genetic analysis and choice of selection methods for population improvement

There were significant differences between different plantain populations for the fruit-filling period, bunch weight, and fruit parameters (PBIP 1993). This suggests that selection for fruit size in plantain could be possible. Significant differences within populations were also observed for bunch weight and its components. Therefore, improvement within populations will be effective to select genotypes with increased bunch weight. However, the difference between clones of the same population may be the result of ploidy levels. Significant contrasts between ploidy levels in each family were found for bunch weight and fruit size (Vandenhout 1993). On average, tetraploids yielded more than diploids. Tetraploid hybrids have high-yielding bunches with large fruit, but have the same number of hands and fingers as their full-sib diploids. This suggests that tetraploidy is the optimum ploidy level for maximum expression of bunch and fruit weight in plantain-banana hybrids. Tetraploidy enables the expression of higher order interactions (tri- and tetraallelic), which is important for maximizing yield in vegetatively propagated polyploid crops (Peloquin, Ortiz 1992). Results suggested that selection between families would be the most effective method to improve plant height, bunch weight, and fruit weight and its components. Individual selection within ploidy level for each family would also be effective for almost all traits.

Phenotypic recurrent selection schemes for traits with additive gene action, recurrent selection with progeny testing for traits with nonadditive gene action, and reciprocal recurrent selection for traits with both additive and nonadditive gene action are employed to improve the plantain gene pool at the diploid level.

Looking ahead: breeding in other *Musa* gene pools

The next breeding target of IITA is the amelioration of the East African highland beer and cooking banana (Vuylsteke et al. 1993b). This seems to be possible because several seed-fertile cultivars have been identified, multiplied in vitro and field-established in

large pollination blocks at Onne. It is important to note that TMB x 612-74, a hybrid resistant to black leaf streak/black Sigatoka derived from Bluggoe, a cultivar very popular in Malawi, has already been selected (Fig. 4). It has high bunch weight (>17 kg/bunch) and big fruit (>200 g) at Onne (Vuysteke et al. 1993c).

Equal Partnership with NARS in Research:

Clonal phenotype, which corresponds to a specific genotype, can vary from year to year in the same location and/or from location to location in the same year in the same agroecological zone. This phenomenon, which influences genotype ranking in different environments, is known as genotype-by-environment (GxE) interaction. Breeding programs aim to identify genotypes that have high and stable yield in a range of environments across a targeted region.

Multilocal trials are required to assess yield stability (Ortiz 1993a; Ortiz et al. 1994b) and durability of black leaf streak/black Sigatoka resistance (Vuysteke et al. 1993a) of the plantain-banana hybrids across environments. Multilocal evaluation trials (METs) and advanced *Musa* yield trials (AMYTs) were set up with local partners in Africa and tropical America (Table 4). IITA and NARS contributed equally by dedicating their own resources towards the success of this joint testing activity. IITA provides planting materials, field designs, data analysis, as well as individual and group training, and research guides (Gauhl et al. 1993; Swennen, Ortiz 1994). Trials are monitored with

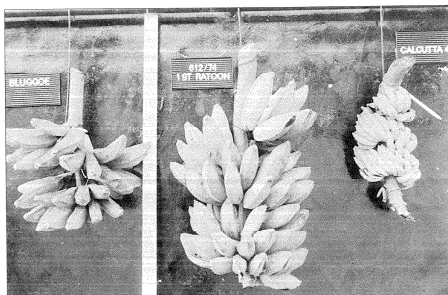


Figure 4. Black leaf streak/black Sigatoka-resistant tetraploid hybrid TMBx 612-74 (middle) derived from a cross between the triploid susceptible cooking banana cultivar Bluggoe (left) with the highly resistant wild diploid banana Calcutta 4 (right).

frequent visits. NARS run the trial with their own budgets. This demonstrates commitment of both partners in the implementation of the flow of materials and technology transfer. This option for equal partnership in research redefines the function of CGIAR's breeders in relation to scientists in national programs, who must mobilize the local expertise available within NARS (Ortiz 1993b).

The results from the Humid Forest Stations of IITA at Onne (Nigeria) and M'Balmayo (Cameroon) showed that TMBx 1658 1 (Fig. 5) had high and stable yielding clones in this agroecology (Ortiz et al. 1994a). Moreover, this clone and its half-sib TMPx 2796-5 (Fig. 6) were the highest yielding hybrids at the IITA station in the derived savanna or transition zone, which had a dry season 3 months longer than at the Onne station. Plantain hybrids often outyielded not only their susceptible plantain parent, but also other ABB cooking banana cultivars with less susceptibility to black leaf streak/black Sigatoka (Ortiz, Vuysteke 1993a; Ortiz et al. 1994a).

The black leaf streak/black Sigatoka reaction of 20 genotypes across West and Central Africa was recorded (Ortiz et al. 1993b) and results are shown in Table 5. Plantain and Valery (AAA 'Cavendish') are equally susceptible to black leaf streak/black Sigatoka. They had about 50% of their standing leaves showing final stages of disease development

Table 4. Institutions involved in multilocal evaluation trials (METs) and advanced *Musa* yield trials (AMYTs).

METs	
<i>Nigeria:</i>	NIHORT-Ibadan, NRCRI-Umudike, BSDP-Oqou Ibono, ADP-Akure, A&RD-Calabar, TTS/FDA-Ugwuogba-Oji River, Pamol-Ibadan, Shell-Warri, Shell-Bori, NAOC-Green Rivers Project, and IITA Stations at Ibadan and Onne
<i>Benin:</i>	UNIBEN-Benin City
<i>Cameroon:</i>	CRDP-Nyumbé and IITA Station at M'Balmayo
<i>Uganda:</i>	ESARC(IITA)-Namulonge
<i>Ghana:</i>	CRI-Assin Fosu
<i>Dominican Republic:</i>	Fundacion de Desarrollo Agropecuario, Inc.-Sto Domingo
<i>Cuba:</i>	INIVIT-Villa Clara, Sto Domingo
<i>Australia:</i>	QDPT-South Johnstone
AMYTs	
<i>Nigeria:</i>	NPQS-Ibadan and ADPs at Umuahia and Owerri
<i>Côte d'Ivoire:</i>	IDEFOR-Abidjan
<i>Ghana:</i>	University of Ghana-Kade
<i>Burundi:</i>	IRAZ-Rugombo
<i>Uganda:</i>	UNBRP-Kawanda
<i>Tanzania (Zanzibar):</i>	Ministry of Agriculture-Kizimbani
<i>Malawi:</i>	Ministry of Agriculture/Agricultural Experiment Stations at Songwe and Mkondezi
<i>Kenya:</i>	ICIPE-Ungoye

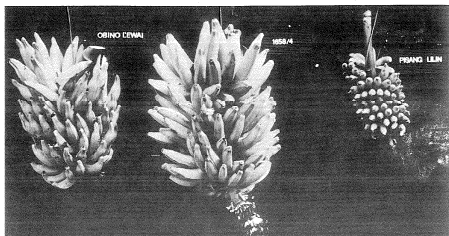


Figure 5. Black leaf streak/black Sigatoka-resistant tetraploid hybrid TMPx 1658-4 (middle) obtained by crossing the susceptible triploid plantain cultivar Obino E'wai (left) with the highly resistant cultivated diploid banana Pisang Lilin (right).

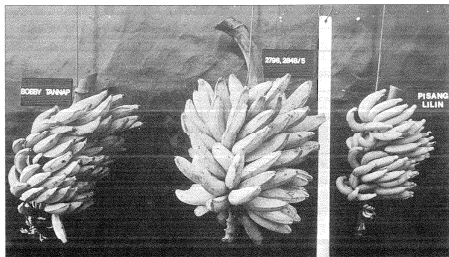


Figure 6. Black leaf streak/black Sigatoka-resistant tetraploid hybrid TMPx 2796-5 (middle) obtained by crossing the susceptible triploid plantain cultivar Bobby Tannap (left) with the highly resistant cultivated diploid banana Pisang Lilin (right).

whereas ABB cooking banana cultivars, such as Bluggoe and Cardaba, initially considered as potential substitutes for plantain, are less susceptible to black leaf streak/black Sigatoka. Although there were significant differences in host response to black leaf streak/black Sigatoka among the hybrids, they show partial resistance to the disease, i.e. slow disease development in their leaves. None of the TMPx hybrids have the high level of

resistance (considered as hypersensitivity) of Calcutta 4. However, this type of resistance is more prone to breakdown. A more durable resistance in the TMPx germplasm might be expected because its host-plant response to black leaf streak/black Sigatoka slows the progress of an epidemic without inhibiting its initiation.

Regression analyses, following the linear model $BSR_i = \mu + \beta_j I_j + \delta_{ij}$, were used to describe the genotypic response to black leaf streak/black Sigatoka (BSR_{ij}) of the i^{th} clone in the respective j^{th} environment (Ortiz et al. 1993a). In this equation, μ is the clonal mean over all the environments, β_j is the regression coefficient which measures the response of the i^{th} clone to varying environments, δ_{ij} is the deviation from the regression of the i^{th} clone at the j^{th} environment, and I_j is the environmental index. Calcutta 4, Pisan Lilin, and TMBx 612-74 were not included in the analysis because they did not show any leaf with black leaf streak/black Sigatoka-induced necrotic spots in specific environments (Table 5). A practical way to select stable genotypes resistant to black leaf streak/black Sigatoka is illustrated in Figure 7.

The vertical lines are one $LSD_{0.05}$ above and below YLS grand mean and at the grand mean, whereas the horizontal line is drawn parallel to the X axis from the slope $\beta=1$. Thus, eight spaces were defined. Cultivars and experimental hybrids in the right lower part of the graph should be considered as having stable black leaf streak/black Sigatoka resistance. Indeed, TMPx 5511-2 had a $\beta = 0.40$, which was not significantly different from zero. This means that the environment and the genotype-by-environment interaction did not affect the expression of black leaf streak/black Sigatoka resistance in this clone. The TMPx clones had different β values which indicate that selection for stable black leaf streak/black Sigatoka resistance may be possible in this population. On average, the Obino l'Ewai derived hybrids had higher black leaf streak/black Sigatoka resistance stability than those derived from Bobby Tannap. This was not surprising because the susceptible black leaf streak/black Sigatoka response of Obino l'Ewai is more stable than that of Bobby Tannap.

Independent analyses of variance combined over environments in each group of materials were carried out to compare the improved TMPx germplasm versus their susceptible parents (Obino l'Ewai and Bobby Tannap) and other reference cultivars. Thus, estimates of variance components were obtained, and coefficient of variability (CV) and broad-sense heritability (ratio of genetic variance to total phenotypic variance) were calculated for the hybrids and the susceptible cultivars (Table 6).

Broad-sense heritability estimates show clearly that the improved germplasm has on average a more stable as well as more heritable variation for black leaf streak/black Sigatoka response than the natural susceptible germplasm. Thus, a faster progress through selection for black leaf streak/black Sigatoka resistance may be expected in the TMPx population than in the natural germplasm. The CVs indicated that on average the recorded YLS was smaller, but more variable, in the natural germplasm than in the improved TMPx germplasm.

In general, the improved germplasm has a more stable response to black leaf streak/black Sigatoka than the natural *Musa* germplasm.

Table 5. Percentage of leaves spotted (ILS¹) with black leaf streak/black Sigatoka in multiblocational trials in West and Central African locations (rainy season of 1993) (after Ortiz et al. 1993b).

Clone	Ghana		Cameroon		Nigeria		Congo			
	Assin-Fosu	M'Balmayo	Omé	Umuokike	Calabar	Bori	Obrikom	Akure	Bandan	X-ISE ²
546-4	27	12	20	14	38	0	10	25	16	18.4
948-9	20	24	34	11	11	11	6	23	28	19.0
958-4	25	28	38	23	21	29	5	28	22	24.0
1021-1	30	20	42	19	40	17	19	28	33	28.0
1085-4	38	37	34	25	38	28	9	36	19	29.0
2481	24	0	14	14	11	2	0	19	17	12.0
2607-40	30	0	18	9	21	12	0	14	10	15.0
2796-5	21	16	18	7	18	9	10	14	9	14.2
4088-1	94	9	20	13	18	12	10	17	12	15.0
8911-2	91	4	17	35	22	0	6	25	6	16.0
0199-1	55	0	15	20	22	0	22	12	3	16.0
012-14	20	50	62	33	42	60	46	44	0	13.4
Agbagba	54	82	46	33	46	64	41	44	52	48.0
B. Tannop	61	80	58	45	53	50	45	44	51	48.0
B. Tannop Ewai	64	70	65	49	67	65	34	49	60	59.0
Valery	44	49	32	43	38	53	38	32	44	40.0
Bogoso	44	52	38	41	36	38	40	60	44	42.0
Chavabou	n/a	17	13	3	11	11	10	17	10	12.0
Phenig Lilla	0	0	0	0	0	0	n/a	n/a	0	0.0
Calabata 4	38	26	28	22	30	22	18	28	24	24
SE	+4a	+4b	+4b	+4b	+4b	+4b	+4c	+4b	+4b	+4bc

Means followed by the same letter within the column of clone mean and the row of environmental means are not significantly different according to Student-Neuman-Keuls' multiple range test at $\alpha = 0.05$.
¹ILS = (YLS) net number of standing leaves (NSL) ILS = 100 X (NSL/YSL) (NSL = number of leaves standing at harvest, YLS = number of leaves spotted at harvest).
²Standard error = $0.01^{1/2}$, where α is the standard deviation of the sample and n is the number of observations.
³Leaf spots not readily observed.

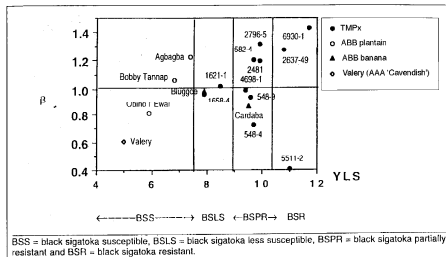


Figure 7. The relation between black Sigatoka/black leaf streak response, as measured by youngest leaf spotted (YLS), and stability (B) (after Ortiz et al. 1993a).

Table 6. Genetic variance (σ^2_G), genetic-by-environmental variance (σ^2_{GE}), phenotypic variance (σ^2_P), broad-sense heritability (H^2), youngest leaf spotted (YLS, mean \pm standard error) and coefficient of variability (CV, %) in improved hybrid (TMPx) and natural susceptible germplasm.

Germplasm	σ^2_G	σ^2_{GE}	σ^2_P	H^2	YLS	CV
TMPx	0.983	1.189	2.172	0.45	9.8 ± 0.3	11.1
3x susceptible Musa	0.773	1.902	2.675	0.29	6.3 ± 0.5	21.9

Chemical and Host-Plant Resistance Strategies to Control Black Leaf Streak/Black Sigatoka

Though it is possible to control the disease with fungicides, these are too expensive for the smallholders who grow most of the crop in Africa. In addition, fungicide applications are hazardous to health in the village homesteads. Moreover, as was pointed out recently "fungicides are the most abundant pollutants in water downstream from banana plantations, where they can kill fish or prevent them from reproducing. Anything to reduce pesticide use in the tropics is strongly to be welcomed" (New Scientist, 17 April 1993, p.9). Therefore, resistance breeding was considered by IITA as the most

appropriate component intervention to control black leaf streak/Sigatoka leaf spot (Vuylsteke et al. 1993c).

Potential farmer gains were quantified by using two different black leaf streak/black Sigatoka control strategies: fungicide treatments and improved hybrids resistant to black leaf streak/black Sigatoka. Results of preliminary evaluation of hybrids along with their susceptible plantain parent under fungicide and nonfungicide protection were used to compare both control strategies (PBIP 1993). For a proper assessment of gains, the potential yields ($t \cdot ha^{-1} \cdot a^{-1}$) of TMPx black leaf streak/black Sigatoka-resistant plantain hybrids and those obtained by their plantain parent Obino l'Ewai with and without fungicide treatment were determined. The potential yield of untreated Obino l'Ewai was used as the reference point (Fig.8). The best hybrid, TMPx 548-9, had a 225% increase in yield over untreated Obino l'Ewai, whereas fungicide control increased yields of Obino l'Ewai by only about 70% in comparison with untreated Obino l'Ewai. Moreover, farmers require an investment of at least US\$750 $ha^{-1} \cdot a^{-1}$ to provide chemical protection against black leaf streak/black Sigatoka disease, while few suckers of the hybrids may be provided free-of-charge or at a minimum cost by NARS for further multiplication.

A rough estimate was made of the advantages of host-plant resistance over chemical protection to black leaf streak/black Sigatoka leaf spot disease taking into consideration the cost/benefit ratio at rural markets in the area near Onne Station. In normal periods, 6 kg of plantain costs about N 30 (30 N = 1 US\$ in December 1993), while in periods of scarcity, a 6 kg bunch costs up to N 50. During periods of scarcity, a farmer's gains by increasing production due to chemical protection might be as high as US\$1100. This may

decrease to US\$370 in normal periods when plantains are abundant in the rural markets. With the TMPx black leaf streak/black Sigatoka-resistant germplasm, the gains may be about US\$6050 in periods of scarcity and US\$3630 in normal periods. Therefore, host-plant resistance through the utilization of favorable genes could have a comparative advantage over fungicides of 10:1 in normal periods of supply and 5.5:1 in periods of scarcity.

A Holistic Approach for Sustainable and Perennial *Musa* Production

The main areas of PBIP research (breeding and genetics, biotechnology, postharvest studies, and agronomy) are independent yet interrelated because of a holistic approach that has been adopted to develop sustainable plantain/banana production and utilization systems (Vuylsteke et al. 1993b). Single-component interventions do not provide complete solutions to the different constraints affecting *Musa* production in sub-Saharan Africa. Indeed, PBIP's breeding philosophy summarizes our long-term goal: "to develop not only disease- and pest-resistant materials, but also better cultivars for the smallholders who produce plantain and banana in compound or home gardens and large field plantations". Better cultivars must have high and stable yields, adaptation to target areas, resistance to/tolerance of biotic and abiotic stresses, and acceptable quality.

Genotype-by-cropping system interaction

The main components of the biological production system, plant and environment, are normally discussed independently. However, plant-environment interactions must be considered in the development of sustainable systems. Therefore, improved technological packages, i.e. better cultivars in combination with improved crop husbandry practices (Ruhigwa et al. 1993; Wilson, Swennen 1987), must be refined by adaptive testing in target areas with NARS partnership. Integration of cropping systems and crop improvement technologies will achieve maximum interaction between these components.

Early research on plantain agronomy at IITA (1973-87) focused on crop husbandry techniques for long-term productivity (Vuylsteke et al. 1993b,c). The essential role of soil organic matter for sustainable production of plantain was established. Innovative production systems, such as alley cropping with multispecies hedgerows were developed (PBIP 1992). The aim was to provide the adequate environment in which an improved genotype, adapted to the target agroecology, will sustain its yield without degrading the resource base. Currently physiological investigations aim to determine leaf stomata conductance and leaf transpiration and their correlation with potential drought tolerance (Ekanayake et al. 1994a,b).

Based on considerable knowledge accumulated during 10 years of agronomy and physiology research, IITA published a manual on plantain cultivation in West Africa (Swennen 1990), which has been distributed among national programs, participants in IITA training, and farmers.

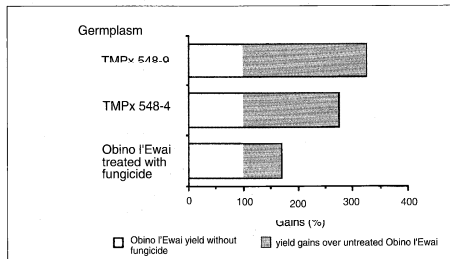


Figure 8. Yield gains from fungicide application vs host-plant resistance to control black leaf streak/black Sigatoka. Obino l'Ewai without fungicide treatment was the reference point ($0.7 t \cdot ha^{-1} \cdot a^{-1} = 100\%$) to determine gains in potential yield with respect to fungicide treatment and to Obino l'Ewai derived tetraploid hybrids TMPx 548-4 and TMPx 548-9 (after PBIP 1993).

Postharvest research

There are many postharvest quality attributes related to fruit palatability and durability that have to be satisfied before any improved varieties are adopted by farmers (Eggleston et al. 1992a,b; Ferris 1993a). The project has undertaken the appraisal of consumer preferences and postharvest properties of new varieties to ensure their acceptability and successful introduction (Ferris 1993b). Research has focused on the consumer acceptability of new, black leaf streak/black Sigatoka-resistant hybrids by investigating fruit palatability (flavor, texture, response to cooking procedures) and durability (shelf life, ripening, handling, and storage aspects) (Ferris et al. 1994). For example, fully matured fruits of Obino l'Ewai and TMPx 548-9 were evaluated by a taste panel of trained Nigerian assessors for a preliminary consumer acceptability test at Onne (Vuytsteke et al. 1993g). Green and yellow peel-colored fruits were fried to prepare chips and a local Nigerian dish 'dodo', respectively. Taste and texture were similar regardless of peel color, but the fruit color of TMPx 548-9 was less preferred. Nevertheless, TMPx 548-9 was rated as "good".

In-vitro culture techniques and biotechnology for *Musa* germplasm enhancement

Efforts to propagate, conserve, and breed cultivated *Musa* are fraught with many obstacles (slow propagation, low reproductive fertility, lack of genetic variability) specific to the biology of this vegetatively-propagated crop (Asiedu et al. 1992). Hence, plant tissue culture and molecular genetic methods are increasingly being used as techniques for the handling and improvement of *Musa* germplasm (Vuytsteke, Swennen 1992).

At IITA, shoot-tip culture is routinely used for the propagation, exchange, and conservation of *Musa* germplasm (Vuytsteke 1980). For example, micropropagation has been pivotal in the rapid deployment of the breeding program by supplying large numbers of plants of female and male parents for the crossing blocks and of promising new hybrids for the evaluation trials. Also, more than 300 new *Musa* accessions were introduced as shoot-tip cultures during the past 8 years (Vuytsteke et al. 1990a). Embryo culture and rescue techniques (Vuytsteke et al. 1990b) are still applied to increase the germination rate of the true seed produced during breeding. More than 10,000 seeds obtained from plantain crosses are handled in the tissue-culture laboratory at Onne each year (PBIP 1993).

Technical assistance in the development of tissue-culture laboratories has been provided to national research institutions in Nigeria (Vuytsteke, Swennen 1989). This is another area in which PBIP envisage a sustainable institutional development. Tissue culture (TC) laboratories will play an important role for distribution and multiplication of clean and improved planting materials of selected genotypes. Main limitations in NARS laboratories are chemicals and power supplies. To overcome this constraint, support of the international and regional donor community is sought jointly. It is expected that these TC facilities will be used not only for *Musa*, but also for other crops

that benefit from in-vitro culture techniques. This approach may enhance the sustainability of the system. It is also envisaged that, in the near future, requests for planting materials sent to IITA could be handled by these TC labs. By selling improved in-vitro propagules of different crops, NARS could recover their operational costs. This may increase the chance of success and survival of the TC labs that will be set up with the support of international and regional donors.

More advanced studies in cell and molecular biology are currently under way. Somaclonal variation in plantains derived from shoot-tip culture has been described in detail by IITA scientists (Vuytsteke, Swennen 1990; Vuytsteke et al. 1988; 1991; 1994b). Work on the genetic stability of plants regenerated from cell suspensions (Dhedea et al. 1991; 1992) and from cryopreserved cultures is currently ongoing in collaboration with the Catholic University of Leuven (KUL), Belgium (De Smet 1993). In the area of new molecular techniques, the use of molecular markers (RFLPs, VNTRs, RAPDs) for phylogenetic studies (Jarret et al. 1993), construction of a linkage map, marker-assisted selection in breeding, and mapping Sigatoka resistance loci is being investigated in collaboration with the USDA/ARS at Griffin, Georgia. DNA-fingerprinting techniques are also being used to detect somaclonal variants and elucidate the origin of somaclonal variation in collaboration with the University of Birmingham, England and KUL.

Exchange of Information with NARS

Limited access to basic information is still perceived as being a major problem of plantain and banana researchers in Africa (Wilson 1989). Hence, in early 1993, IITA began publishing *MusaAfrica*, a plantain and banana newsletter for Africa. The newsletter is sent to collaborators and *Musa* scientists as a means of maintaining links with NARS and to provide rapid transfer of information and technology. Also, partners in the multilocational testing have a forum in the newsletter for exchange of information and ideas as well as for description of improved germplasm. This offers the opportunity to develop networking. A key element for success of this network is the development of horizontal relationships between the network partners with PBIP scientists as catalytic agents.

Potential Impact of TMPx Germplasm in African Agriculture

Based on information provided by the Technical Advisory Committee of the Consultative Group on International Agricultural Research (CGIAR), the potential impact of the TMPx germplasm in the African rural economy was calculated. The annual gross value of *Musa* production in Africa is about US\$2.8 billion. Only 1% of the total production goes to the export trade. Therefore, the 22% yield increase that may be possible by using TMPx germplasm might have a potential impact of US\$6.2 billion.

The total investment of IITA in plantain breeding in the development of TMPx germplasm has consisted of 2 core scientists (breeder and tissue culturist) for a 5-year period (1987-1992). Five years is about the time from an initial cross to the registration of a hybrid. IITA costs for international staff, local staff support, travel, supplies and other expenses, was about US\$2 million for the 5-year period. The ratio of potential impact to investment, therefore, was established as US\$6.2 billion/US\$2 million. In other words, for US\$1 invested by IITA in the development of 14 improved TMPx hybrids with black leaf streak/black Sigatoka resistance, the African economy might have a yearly gain of about US\$3100.

In conclusion, genetic improvement of plantain and banana, by a modified but conventional cross-breeding approach, is an important component in the scientific path towards sustainable production of *Musa* cultivars. The impact of this research depends on the incorporation of TMPx germplasm as an integral component of *Musa* production systems. This requires that NARS scientists, working closely with international agricultural research centers (IARC's), transfer this improved germplasm along with other sustainable technologies to farmers of developing countries. In this regard, the International *Musa* Testing Program (IMTP), under the auspices of INTDAG, must provide the opportunity for the evaluation of improved germplasm developed by different programs (CIRAD, EMBRAPA, FHIA, IITA, TBRI, etc.), which should be freely available for extensive testing, local selection, and cultivar release by NARS scientists.

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