

Banana breeding program at Embrapa

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ABSTRACT

The principle factors related to the banana breeding are described: botanic classification, cultivars, origin and evolution of the banana, reproductive systems, sterility and partenocarpy, polyploidy, inheritance of characteristics. To solve the problems caused by fungus, bacteria, virus, nematodes and insects, high stature and low productivity of some cultivate, it has been used the creation of new resistant varieties by means of the genetic program. The breeding program consisting of the following stages: formation, characterization and evaluation of wide germoplasma collection, introduction and selection of clones, improvement for hybridization, improvement for mutation, somatic hybridization and genetic transformation. The principle results obtained in the breeding program are: the morphologic characterization of the germoplasma, allowing the identification of promising genotypes and its recommendation to the producers; the obtainment of resistant hybrid tetraploid (Pome type) to the yellow and black sigatokas and to the Panama disease, with reduced stature and cycle and high productive; the genetic improvement of diploid AA, whose pollen has been used in the improvement of the commercial cultivars and for the own hybrid diploid; the evaluation of the cultivars and hybrid in different ecosystems, allowing to identify potential varieties to be recommended as a local and national status for several ecosystems; obtainance of hybrid of Silk cultivar (AAB) with the diploid (AA) 'Lidi', by means of the employment of somatic hybridization for eletricfusion, although, none cultivar was obtain from this technique.

KEY WORDS: *Musa* spp., hybrids, diploids, triploids, tetraploids.

INTRODUCTION

Cultivated in more than 80 tropical countries, mainly by small farmers, the banana culture has an important social and economic position throughout the world. Brazil is the third largest world producer of bananas (second most consumed fruit in the country), with an estimated output of 5,92 million tons, in a cultivated area of 528 thousand hectares (FAO, 2000). Bananas are cultivated all over the country, and almost all the fruit produced is traded in the internal market. Hence, the effective consumption of bananas is around four million tons, as the post-harvest losses reach up to 40% of the total production. The majority of the banana farmers are small producers, who have banana planting areas as additional sources of income. Besides being a permanent source of food and income, this crop's importance lies on its ability to settle men power in the field.

As in any species cultivated in large areas, the banana is affected by many phytosanitary

problems caused by fungi bacteria, virus, nematodes and insects. The fungi are very important infectious agents that cause diseases such as Fusarium wilt, yellow and black sigatokas which constitute the biggest problems affecting banana crops around the world. Among the bacteriosis, Moko disease stands out, although little is known about its sources of resistance. The "bunch top" virus, still not present in Brazil, is classified as the greatest virus etiology problem for bananas. The most important nematode for the banana crop is the *Radopholus similis*, and the weevil borer (*Cosmopolites sordidus*) is the most harmful pest. Pests and diseases are responsible for severe losses in banana production, which, depending on the factors involved, can reach up to 100%, and, in many cases, with no means of control.

The banana crop in Brazil is peculiar in relation to climate diversity, cultivars and commercialization modes. With the exception of some plantations in the States of São Paulo. Minas

Gerais. Santa Catarina. Goiás and Rio Grande do Norte, the producing areas use low levels of capitalization and technology. The majority of the plantations present low yield potential, with a national average yield of around 11,1 t/ha/year.

One of the strategies used to solve problems is to develop new varieties resistant to main diseases, nematodes and pests through breeding programs planned to generate superior genotypes such as the program being presently developed by Embrapa. The use of resistant cultivars is one of the most effective ways of disease control as it does not depend on the effort of the producer during the plants growing phase, it is not harmful to the natural environment and, generally, is compatible with other management techniques.

Besides increasing productivity and fruit quality, an improved cultivar (resistant to diseases, nematodes and pests) will result in a cost effective production, by reducing the use of pesticides and other culture management expenses.

BOTANICAL CLASSIFICATION

The banana crop *Musa spp* presents a fascicled root system, absence of vascular cambium and typically trimerous flowers, being included in the class of the Liliopsida and in the Zingiberidae subclass of the Lillanae (Cronquist, 1981) super order. The presence of the colored compound tepal and an adherent and inferior ovary includes it in the order of the Zingiberales (Scitamineae) (Takhtajan, 1953). Eight families belong to this order: Musaceae, Cannaceae, Marantaceae, Zingiberaceae, Lowiaceae, Costaceae, Heliconiaceae and Strelitziaceae (Belalcázar and Carvajal, 1991), and different numbers of genera (Simmonds, 1973): Lowiaceae, a genus (*Orchydantha*); Cannaceae, genus (*Canna*); Musaceae, two genera (*Musa* and *Ensete*); Strelitziaceae, four genera (*Strelitzia*, *Heliconia*, *Ravenala* and *Phenakospermum*); Marantaceae, 25 genera, the outstanding *Calathea* with diverse ornamental species and the Zingiberaceae, 45 genera. The Zingiber is the most important commercial genus.

Although Simmonds (1973) identified two genera (*Musa* and *Ensete*) in the Musaceae family, it is presently known that such family has more than

two genera divided into three subfamilies: Heliconioideae, Strelitzioidae and Musoideae. The *Ensete* and *Musa* genera belong to the subfamily Musoideae, and the *Musa* genus belong to the group of edible bananas. According to Valmaoyor et al. (1991) this genus was created by Karl Linné, probably to pay tribute to the Roman Antonius Musa, physicist of the first emperor of Rome, Octavius Augustus.

The *Musa* genus is subdivided in the Australimusa, Callimusa, Rhodoclamys and Eumusa sections, based on the number of chromosomes. The genome with 11 chromosomes is characteristic of Eumusa and Rhodoclamys, while the 10 chromosome genome characterizes the Callimusa and Australimusa. The Eumusa section presents the largest geographical dispersion and includes the following species: *Musa schyzocarpa* (Simmonds), *M. basjoo* (Siebold), *M. itinerans* (Cheesman), *M. nagensium* (Prain), *M. flaviflora* (Simmonds), *M. sikkimensis* (Kurz), *M. cheesmani* (Simmonds), *M. balbisiana* (Colla), *M. acuminata* (Colla) and *M. halabanensis* (Meijer) (Tezenas du Montcell, 1988).

The *Musa* genus was classified initially by Linné, into two species: *Musa sapientum* and *Musa paradisiaca*. The first species includes bananas that are consumed *in natura* or raw while the *Musa paradisiaca* species includes those normally consumed in stews or fried. This classification, without any scientific basis, is notably artificial/unsubstantiated (Simmonds, 1966).

According to Cheesman (1948), *M. sapientum* corresponds to a clone in Trinidad known as 'Silk Fig' and to the 'Cambur Manzano', in Venezuela, while *M. paradisiaca* corresponds to the Plantain 'Dominico' from Venezuela. He concluded that Linné's classification is incomplete since it mentions only the clones, leaving out the diverse species and varieties of *Musa*. A scientific classification of bananas was presented by Simmonds and Shepherd (1955), adjusting the *M. acuminata*, colla and *M. balbisiana*, colla, species which gave origin to all the other bananas. Table 1 shows a summary of the *Musa* genus classification and the corresponding species (Cronquist, 1981).

MAIN CULTIVARS

This expressive number of banana cultivars with agronomic and commercial potential is reduced drastically when consumers preference, productivity, tolerance to pests and diseases, resistance to drought, plant height and resistance to coldness are considered. The most diffused cultivars in Brazil are: Prata, Pacovan, Prata Anã, Maçã, Mysore, Terra and D'Angola which belong to the AAB group, the Nanica, Nanicão and Grande Naine, which belong to the AAA group and are produced mainly for export (Table 2). The 'Figo Cinza', 'Figo Vermelho', 'Ouro', 'Caru Verde' and 'Caru Roxa', Prata and Pacovan cultivars are produced in a small holds and are responsible for approximately 60% of

the Brazilian banana culture (Silva et al., 1999a). The 'Pacovan', 'Prata', 'Terra' and 'Mysore' cultivars have high stature. 'Maçã' is highly susceptible to the Panama disease; 'Nanica'; 'Nanicão'; 'Grande Naine', 'Terra' and D'Angola are highly susceptible to nematodes, and 'Mysore' is usually infected with BSV. All these cultivars are susceptible to moko diseases, and, with the exception of 'Mysore', to black Sigatoka as well. They are also highly susceptible to the yellow Sigatoka, with the exception of 'Maçã', 'Mysore', 'Terra' and 'D'Angola'. 'Prata' was introduced by the Portuguese since 1500 and, for this reason, the Brazilians, specially from the northeastern and northern regions, demonstrate a clear and constant preference for the 'Prata' flavor. It has small fruits of sweet to slightly acid flavor.

Table 1 - Summary of classification of *Musa* genus presenting the species in the different sections (Cronquist, 1981).

Class	Liliopsida
Subclass	Zingiberidae
Super-order	Lilianae
Order	Zingiberales
Family	Musaceae
Genera	<i>Musa</i> (n=10 ou 11) <i>Ensete</i> (n=9)
Sections :	
Callimusa (n=10)	<i>M. coccinea</i> Andrews <i>M. violascens</i> Ridley <i>M. gracilis</i> Holttum <u><i>M. borneensis</i> Beccari</u>
Australimusa (n=10)	<i>M. peekeli</i> Lant <i>M. maclayi</i> F. V. Muele <i>M. augustigemma</i> Simmonds <i>M. lolodensia</i> Cheesman <i>M. textilis</i> Nee
Eumusa (n=11)	<i>M. schizocarpa</i> Simmonds <i>M. basjoo</i> Siebold <i>M. intinerans</i> Cheesman <i>M. nagensium</i> Prain <i>M. flaviflora</i> Simmonds <i>M. sikkimensis</i> Kurz <i>M. chesmani</i> Simmonds <i>M. balbisiana</i> Colla <i>M. acuminata</i> Colla <i>M. halabanensis</i> Meijer
Rhodoclamys (n=11)	<i>M. velutina</i> Wendl et Drude <i>M. sanguinea</i> Hook <i>M. ornata</i> Roxb <i>M. laterita</i> Cheesman

'Pacovan' is more rustic and productive with fruits 40% bigger, more acid than the 'Prata' and with edges that remain even after ripe. 'Prata Anã', also known as 'Enxerto' or 'Prata de Santa Catarina' has hands closer to one another, fruits of even flavor and a bottle-neck tip.

'Maçã' is the most preferred banana type by Brazilians. Its skin is thin and it has a smooth pulp

and an apple-like taste. The Cavendish subgroup cultivars ('Nanica', 'Nanicão', 'Grande Naine') also known as the 'water bananas', produce long thin fruits, arched, of yellow to green color when ripe, and a very sweet pulp. They are mostly used for export. 'Terra' and 'D'angola' produce big fruits with bigger edges and are consumed in stews or fried. 'Mysore' has a thin pale yellow skin with a slightly acid pulp.

Table 2 – Genomic group e subgroup of the main varieties of banana in Brazil. Cruz das Almas, BA, 1997.

Genomic Group	Subgroup	Varieties
AA	-	Ouro
AAA	-	Caipira ¹
AAA	Cavendish	Nanica, Nanicão, Grande Naine, Williams
AAA	Gros Michel	Gros Michel, Highgate
AAB	-	Maçã
AAB	-	Prata Anã ou Enxerto
AAB	-	Mysore
AAB	Prata	Prata, Branca, Pacovan
AAB	Terra	Terra, Terrinha, Pacova, D'Angola
ABB	Figo	Figo Vermelho, Figo Cinza
AAAB	-	Ouro da Mata
AAAB	-	FHIA 01, FHIA 18 ² , SH3640, PV42-68 ² e Pioneira ²

^{1/} Embrapa recommended variety;

^{2/} Hybrids obtained for improvement;

Source: Silva et al. (1997).

ORIGIN AND EVOLUTION OF THE BANANA

The center of origin of most banana germoplasm is located in the Asian continent. Other secondary centers are found in Eastern Africa, in some islands of the Pacific Ocean and a considerable genetic diversity exists in Western Africa (Champion, 1967). The cultivars found in these regions evolved from wild species with three chromosome levels, diploids with 22 chromosomes (2x), triploids with 33 (3x) and tetraploids with 44 chromosomes (4x), which are all multiples of the basic number or genome (n=11). The origin of triploids from diploids and of tetraploids from the triploids is easily proven by experimental crossings (Shepherd, 1984).

Interspecific crossings between *M. acuminata* Colla and *M. balbisiana* Colla gave origin to the majority of the banana genotypes presently used as a source of food and plants generated from these crossings have characteristics of both species

(Simmonds, 1973). These hybrids can present diverse ploidy numbers, resulting in cases with 20, 22, 33, 44, 55, 77 and 88 chromosomes and several kinds of aneuploidies. *M. acuminata* is seed productive, with a diverse number of subspecies, while *M. balbisiana*, is also seed productive and more vigorous.

A study developed by Cheesman (1948) explained the participation of the *M. acuminata* and *M. balbisiana* species in the origin of edible bananas. Simmonds and Shepherd (1955) evaluated these results through taxonomic studies based in fifteen morphologic characteristics of the two species. However, they did not discard the possibility of a contribution, in a small scale, from other species such as *M. schyzocarpa* originated from New Guinea where combinations such as AS and ABBS can occur. These taxonomic studies confirmed the presence of the following (genomic) groups: diploids AA and AB; triploids AAA, AAB and ABB; Tetraploids AAAA, AAAB, AABB and ABBB. This

classification is now widely used, (Figure 1). The term subgroup is used for a complex of cultivars originated by mutations of a single original cultivar (Shepherd et al., 1984), as in the case of the AAA group that contain the subgroup Cavendish and the AAB group that contain two subgroups (Prata and Terra), in Brazil.

The genotypes with the pink coloring in the sheaths, petiols and main nerves are close to the parental species *M. acuminata*, which has dark stains distributed in the pseudostem (Stover and Simmonds, 1987). According to Simmonds and Shepherd (1955), these cultivars inherited from *M. balbisiana* similar characteristics to those of wild species, with little pigmentation and an ashen aspect in the more elevated areas of the pseudostem and petiole.

REPRODUCTIVE SYSTEMS

Wild bananas are diploids ($2n=22$ chromosomes), seed productive, generally allogamous and with fertile seeds upon which their dispersion and

regeneration depend. Commercial cultivars are triploids with $2n=33$ chromosomes and do not produce seeds. The absence of seeds can be related to the intense agronomic selection for this factor, and is, therefore, an example of this species domestication process. Thus, edible bananas are normally vegetative disseminated by means of plantlets developed from the buds attached to its underground stem or rhizome (Shepherd et al., 1986).

The knowledge of the floral structure in banana breeding is very important to analyze ploidy grade, parthenocarpy, genomic group, sterility and characteristics inheritance, considering that most of the cultivars evaluated were from *M. acuminata* and *M. balbisiana*.

INFLORESCENCE

The flower axis is a continuation of the floral stalk, developed in the meristematic region of the rhizome apex. In this structure, the leaves are replaced by bracts, and the first three or four

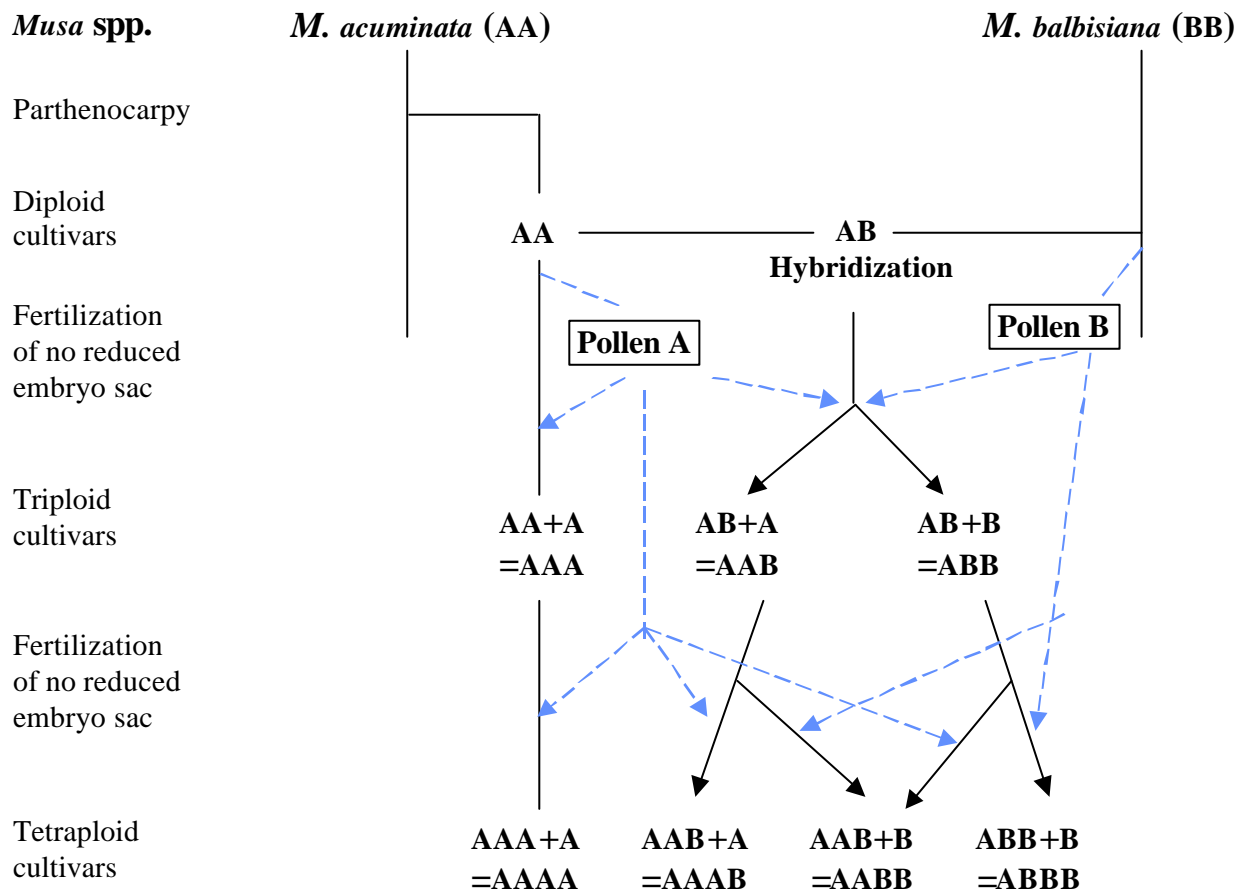


Figura 1 – Evolution process of edible bananas (Adapted to Dantas et al, 1997).

which are the biggest, do not recover the flowers. When the inflorescence emerges from the center of the pseudostem, it has a white color and 5 to 8 cm diameter, turning green afterwards (Simmonds, 1973).

After inflorescence leaves the pseudostem, it takes the same vertical course as in the plants from the *Rhodoclamys* section. On the other hand, in the *Eumusa* section, which presents edible bananas, the part of the inflorescence found outside of the pseudostem has a horizontal or pending position (Champion, 1967).

With the floral axis completely developed, the pseudostem shows three regions: the zone between the apex of the rhizome and the base of the first empty bract, without floral glomerules; the stalk, that extends itself from the first empty bract up to the first bract with feminine flower glomerules, and the third region that begins in the first hand and goes to the apex of the heart (Soto Ballesterro, 1992). The heart is formed by bracts that protect the male flowers in the nodal fascicles. These bracts fall off, exposing the male flowers that also become dry and fall off, developing, after some days, an axis with floral scars, known as cushions. This axis is denominated male rachis, and has a reduced heart in its extremity.

Each group of flowers or protected hands by a bract presents two rows of flowers or fingers, four to eight in each row, arranged alternately. There are three classes of flowers: pistilated, in the upper hands, neutral in several central fascicles, and estamined in the terminal part of the inflorescence. However, this sequence is not fixed. Thus it is possible to affirm that the sexuality tendency in *Musa* is due to the intensification of the female elements from the base of the bunch up to its apex (Leon, 1968).

FLOWERS

Flowers are irregular and classified into three groups of floral pieces: the compound tepal androecium and the gynaecium, which are inserted in the connection point of the style with the ovary, forming the epiginic flower, as the ovary is inferior, and the compound tepalis, which is formed by two tepals, and has calyces and corolla similar in shape and color. The majority of them are formed by two big and two small grown

pieces, alternated, an apex normally divided into five lobules, a conic form and an orange color. In *Musa ssp*, the term compound tepal is applied to this composed tepal which normally has a creamy color with violet spots due to the presence of anthocyanin. The smallest tepal occupies an opposite position to the compound tepal, which involves it. This piece forms the internal verticil and is denominated free tepal, due to the absence of any sign of growth with other pieces (Simmonds, 1973).

The ovary is a long, narrow and normally curved structure, with three sides in the external fingers of the hands and five in the centers (Leon, 1968). It has a straight apex where the compound tepal is inserted, and a free tepal, style and stamens which characterizes the inflorescence. In the apex, nectar is produced in abundance, attracting several insects (Fahn and Benovaiche, 1979). The ovary is trilobular with the ovules in two longitudinal rows in bananas such as Gros Michel and with four longitudinal rows in Plantains (Leon, 1968).

The spherical stigma is well developed with six lobules on its surface, while the style has a cylindrical form, with an enlargement in its bulb-like base, over which six well differentiated lobules are seen, showing the continuity with the stigma (Belalcázar Carvajal, 1991).

Female flowers are differentiated from the male by a well developed ovary and by being taller than the compound tepal. They are long, but the stamens are reduced to stamenoids with no anthers (Figure 2). The male flowers' ovary, although smaller, exceeds the compound tepal in height as well but in a smaller proportion (Simmonds, 1973).

The androecium is constituted of five to six free and introrse stamens, in series of two. One of them is transformed in stamenoid, without an anther, remaining five stamens in the male flowers, which dry and fall rapidly. The anthers are well developed, with unviable pollen in a wide range of cultivars, contrary to what occurs in the wild species. The male flowers (Figure 3) are smaller with abscission of the tepals, differentiated style and stamenoids, which distinguishes them from the clones (Leon, 1968; Simmonds, 1973; Belalcázar Carvajal, 1991).

ESTERILITY AND PARTENOCARPY

Self pollination in species or subspecies which are seed productive normally result in a smaller seed production than the cross fecundations carried out between the same species and subspecies, as certain characters can become lethal in homozygous of recessive alleles. In the banana culture, self pollination in wild forms produced a larger number of seeds than in crossings with other subspecies, although this value is lower than that obtained from crossings inside subspecies (Simmonds, 1952b)

The study of gametes anomalies to analyze the meiosis of mother cells in pollen grains and the chromosomes matching rate during metaphases has been carried out by researchers in several institutions. Some causes of sterility in *Musa* are asynapse (Wilson, 1946, Simmonds and Dodds, 1949), abortion of the embryonic sacs (Dodds, 1945), and translocation (Dodds 1943; Gowindaswani, 1962) which have been observed in the meiosis of peculiar diploid species of this genera. Irregular meiosis is more frequent when working with parthenocarpic diploids. However,



Figure 2 - Female flower of cultivar Pacovan. Cruz das Almas - BA, 2001

according to Dantas et al. (1993) this meiotic behavior of the seed productive *Musa* diploids is generally normal.

After investigating the female gametogenesis in 'Lidi', Dodds (1943) noted that this particular case of almost total female sterility did not have a straight genetic origin, but it was a consequence of particular conditions, perhaps of hormonal nature, such as the growth deficiency of the pollinic tube in the stigmas and styles, or even a defect in the fusion of the nuclei.

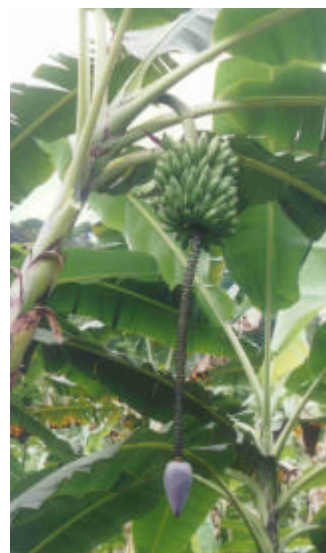


Figure 3 - Male flower of tetraploid hybrid of Pacovan. Cruz das Almas - BA, 2001.

The sterility found in the parthenocarpic diploids is frequently found in both sexes, mainly as a consequence of meiotic abnormalities due to chromosome anomalies. Partenocarpy is an independent phenomenon from the gametes sterility and therefore not associated to the polyploidy, as the partenocarpy is present in the fertile diploids (Dodds and Simmonds, 1948; Champion, 1967).

Banana fruits can be developed by two processes (Simmonds, 1952a). In the wild seed productive species, pollination is essential for the development of the fruit, while in the edible fruit species the development happens by vegetative partenocarpy, i.e., a pulp mass is developed from the external border of the lobule and placenta septum (Simmonds, 1973).

POLYPLOIDY

During the evolution process, the majority of superior plants had an increase in the basic chromosome number of the species caused by polyploidy which was frequently favorable to a better adaptation of the species to its environment. As a consequence, approximately half of the cultivated plants are polyploid, with numbers of chromosomes that are the exact multiples of the basic characteristic number for the species.

The change of the level of ploidy, from diploid to triploid, in *Musa*, resulted in the output of better fruits with greater vigor (Figure 4). The same process was observed, although less

pronounced, in the change from triploid to tetraploid (Shepherd, 1974). The triploids that involve the two *Musa* species present more variability, as *M. acuminata* contributes to the disease resistance and fruit quality and *M. balbisiana* gives them the capacity to adapt to different ecosystems. Despite the advantages presented by the highest levels of ploidy, the change from the diploid to the triploid condition will invariably result in partial or complete sterility (Soto Ballester, 1992).

The reduced triploid seed production capacity due to gametes sterility caused by matching problems generated by the triploidy of the germinative tissue, irregular or late growth of pollinic tubes in the styles of the female flower, absence of fertilization even with the development of the tube by unknown causes, and necrosis of the female flower nectary at blooming, make the commercialization of these cultivars possible. As the seeds of *Musa* are hard, banana fruits with seeds are commercially unacceptable (Dantas et al., 1993).

Genetics studies on polyploids produced a series of information. Wilson (1946) examined the meiosis of some *Musa* triploid clones and, by means of cytological analyses, verified that the suppression of the first meiotic division leads to the development of triploid gametes which

combined with haploid gametes of diploid individuals give origin to tetraploid genotypes. This cytologic behavior is in complete agreement with the evolution process in bananas.

The effects of polyploidy in bananas were observed by means of determining the cell and nucleus volumes of pollen grain mother cells from diploid, triploid and tetraploid plants. The increase of the ploidy showed a linear relation with the increase of the cell volume and with the size of the nucleolus. The relation between pollen grain size and number of chromosomes is nearly linear as well (Vakili, 1967). Simmonds and Dodds (1949) verified the effect of polyploidy in progenies produced by back crossings of three diploid hybrids, obtained by the crossing of two diploid species. Some pentaploids were produced with bigger stomata, and with stomata densities lower than those found in triploids. On the other hand, the triploids had slightly larger stomata than the diploids.

Vakili (1962) carried out a research to evaluate the induction of polyploidy in banana, using colchicine. Intact seeds and banana plantlets were treated with various colchicine concentrations which increased the mortality rate, retarded the growth of the plants and prompted the duplication of the chromosome number. The intact seeds treated with colchicine were less



Figure 4 - Diploid (AA) Monyet, triploid (AAA) 'Grande Naine' and tetraploid (AABB) FHIA 03. Cruz das Almas - BA, 2001.

affected by the polyploidy inducing agent than the plantlets, probably due to the fact that the seeds presented thick layers and an embryo in the quiescent state. Subsequently, Vakili (1967) developed new studies and discovered that colchicine can be a polyploidization inducing element in banana plants.

The process consisted of immersing diploid plantlets in a 0,5% solution of the product, resulting in the development of tetraploid plants, taller and stronger than the diploids, although with slower growth, more slanted leaves and a less developed root system. Tetraploidy affected the size and form of the fruit in some varieties without altering the size of the bunch. The duplication of the number of chromosomes also contributed to the increase of the anthocyanin concentration in the leaves of pigmented varieties. Colchicine induced female sterility was detected in treated diploid plants, transforming most of the plants in sterile tetraploids. The colchicine treatment of plantlets also gave origin to irregularities in the mitosis. Many of those tetraploid plantlets reverted themselves to diploids with the growth of the plants.

Conventional breeding has been made more difficult because of the absence of seeds in triploid banana cultivars, resulting in the absence of viable pollen and efficient natural pollinators as well. Seedless cultivars, when pollinated or produced in small quantity, can be diploid, triploid or tetraploids.

TRAIT INHERITANCE

Continuous variation is considered a particular characteristic of quantitative polygenes. Some quantitative traits such as plant height carry genotypes that can be grouped in main classes, even though continuous variation within each class may occur. Several characteristics showing continuous variation in Plantain and banana are controlled by major genes (Vuylsteke et al., 1997). Despite the low and in some cases total absence of seed production in banana crosses such as inherited resistance to black Sigatoka, nanism, albinism, apical dominancy and habit of buds formation, partenocarpy of the fingers and sterility, orientation of the bunch, wax in the pseudostem, male and female sterility, weigh of the components of the bunch and other

agronomic ones such as apical dominancy, persistency of the male bracts and hermafrodite flowers in the rachis, have already been studied. These studies concluded that such characteristics are governed by one or a few genes. (Ortiz, 1995; Vuylsteke et al., 1997) (Table 3).

Dodds and Simmonds (1948) studied sterility and partenocarpy in diploid hybrids of *Musa* and verified that partenocarpy is the result of the action of the dominant P gene, which expression is subject to the action of modifying genes. In addition, they concluded that partenocarpy is independent from the hybrid structure and the polyploidy, and that the parthenocarpic plants are not completely sterile. Subsequently, Simmonds (1953) verified that its inheritance is a more complex process, and that a minimum of three dominant genes (P1, P2 and P3) are involved in crossings among wild bananas. However, Ortiz and Vuylsteke (1992a) observed that the variation in fruit size and in partenocarpy of Plantain hybrids is due to the segregation of a single dominant gene.

The dominance of male bracts and neutral flowers in the male rachis of the bunch is controlled by complementary and independent genes, which can be affected by the environment. Results from Simmonds (1953) confirm this observation. Data from co-segregation of two peculiar crossings (Plantain x Calcutta 4) were used to estimate the recombination frequency among co-segregated loci in each hybrid diploid population. In the French Plantain, the fraction of recombination among the loci which control the dominance of male bracts and neutral flowers is of 13%. There is a straight genetic linkage in the same chromosome (and not pleiotropism) which is responsible for the unstable association between these two characters (Ortiz, 1996).

The presence of 2x chromosomes pollen in *Musa* diploid suggests that the unilateral polyploidization (2x X x) can be the origin of triploid plants. This led to the belief that a dominant gene controls the development of the 2x chromosomes pollen. New introgressions from alleles of diploid species to polyploids can occur in unilateral or bilateral polyploidization (2x x 2x) (Ortiz, 1997).

Fouré et al. (1993) verified that male sterility in Plantain diploid hybrids can be due to the interaction of the sensitive cytoplasm in the Plantain

with at least three recessive nuclear genes in the banana. A typical test cross ratio (fertile male: sterile male) is expected when the hybrid (plantain x banana) is used as a female. However, when Calcutta was used as a female genitor, segregation was not observed (all male sterile). Therefore, sterility in *Musa* is a genomic, chromosome (numerical and structural) and gene regulated characteristic.

Several accounts on the banana genetic resistance to main pests and diseases are found in the literature. The yellow Sigatoka seems to have two components of resistance. The greatest of them, genetically controlled, affects disease latency, while the smallest is a field resistance component based on a high leaves output speed which helps maintaining a larger foliar green area (Shillingford, 1974). The genetic base of the resistance is not simple. Recessive genes are probably partially responsible for the resistance of wild *Musa acuminata*. In addition, highly susceptible parentals can generate resistant hybrids (Shepherd, 1990).

The inheritance to black Sigatoka is governed by three loci with recessive/additive effects in Plantains and Calcutta 4. The model consists of a major gene (dominant allele for susceptibility to the disease) and two other independent loci with favorable additive effects. Moderately resistant phenotypes correspond to homozygous alleles recessive/favorable to the three loci. The dominant allele is always present in the diploid susceptible hybrid. When one or two smaller additive loci were homozygous, the homozygous of the smaller recessive allele provided susceptibility. The favorable effect of the allele for resistance is balanced by the negative effect of the susceptibility allele in each small additive locus. A clear effect of the dosage on tetraploid progenies with high frequency of hybrids with resistance was observed (Ortiz and Vuylsteke, 1992a; 1992b). High resistance has occurred only in AA and AAA genotypes. With the evaluation of M-53 (diploid hybrid AA) selfed progenies, it was observed that, although hidden in the parentals, the high resistance characteristic was dominant in the F1 generation and that in the interaction between resistances, partial resistance predominated (Fouré, 1993). However, Rowe (1984) related that *M. acuminata ssp. Malaccensis* resistance to black Sigatoka is controlled by several dominant genes.

Larter (1947) suggested that immunity to *Fusarium* was under the control of a dominant gene in tetraploid descendants obtained by the cross of Gros Michel with a diploid access. The study of segregation in progenies derived from crosses among three susceptible *Musa* sp. with *Pisang Lilin* (Lidi) suggested the presence of a dominant gene only for the resistance to race 1 in Lidi (Vakili, 1965a). However, for race 4, the immunity seems to be under the regulation of polygenes (Rowe, 1991).

Rowe and Richardson (1975) reported that *M. acuminata* resistance to moko disease in banana was controlled by recessive genes. However, it was verified that the resistance to the race that attacks tomato was dominant in *M. acuminata* spp. banksii, and recessive in *M. acuminata* spp. microcarpa (Vakili, 1965b).

Radophilus similis nematode resistance is controlled by one or more dominant genes. Therefore, it is possible to incorporate the resistance to nematodes from the access Pisang Jary Buaya (Rowe, 1991) in diploids and tetraploids. Table 3 shows 27 phenotypic characteristics and kinds of gene actions in *Musa*.

HERITABILITY

The ratio between genetic and phenotypic variation (broad sense heritability (h^2)) was estimated for several characteristics in populations and hybrids derived from the French Plantain X Calcutta 4 crossings. Large values were found ($h^2 = 0,8$) for plant height, bunch weigh, fruit length and diameter; intermediate values (h^2 between 0,4-0,8) for height of the tallest sucker at flowering, number of leaves at flowering, newest leaf with blotches and total leaf area affected by the black Sigatoka; low values (h^2 between 0.1-0.4) for height of the tallest sucker during harvesting, number of hands and fingers; and very low values ($h^2 = .10$) for the fruit filling period (Ortiz, 1995). Heritability, in the restricted sense, was not calculated due to the low genetic variability observed for the majority of the traits or to the strong genotype/environment interaction. In other words, high values of heritability indicate that the character is not strongly influenced by environmental factors or that its expression depends mainly on the genotype.

BREEDING METHODOS

Banana Variability

The basic prerequisites for a research program aiming at the production of new cultivars has been the development, characterization and evaluation of a wide germplasm collection. Both the increase of the desired variability and the elimination of the undesired variability are important phases in

a genetic breeding plan using germplasm. In general, when this variability is well adapted to the work of the breeder, there is no need to either induce mutations or use other modern approaches such as genetic transformation.

One important procedure to avoid the introduction of new diseases and/or pests in the collection of germplasm from other countries is the *in vitro* meristems culture. However, the

Table 3 – Phenotypic traits and types of gene action in *Musa*.

Traits	Types of gene action	Reference
Albinism	Two complementary recessive genes	Ortiz and Vulsteke (1994b)
Waxy in pseudostem	One recessive gene, plus additive genes changing the expression	Ortiz et al. (1995c)
Dry matter contents in the fingers	Additive genes	Ferris and Ortiz ¹
Apical dominance	One major recessive gene in plantains	Ortiz and Vulsteke (1994c)
Male and female fertility	Recessives genes interacting with cytoplasm sensitive	Ortiz, (1995); Fouré et al. (1995)
Margins form of the petiole	Duplicate genes with dominant effect	Ortiz (1995)
Blak pseudostem blotches	Modifier gene due to recessive suppressor	Ortiz, (1995)
Blotches in the pseudostem	Two independents genes with dominant epistasis in plantains and one complementary additional recessive gene in banana	Ortiz (1995)
Dwarfism in Cavendish	One dominant gene with modifier gene interaction	Rowe and Richardson (1975)
Dwarfism in type French plantains	One major recessive gene for short false internodes with modifiers affecting plant height	Ortiz and Vulsteke (1995b)
Bunch orientation	Three loci with threshold effect of dominant genes	Ortiz (1995)
Fruit parthenocarpy	Three independent complementary dominant genes One segregating locus in plantain hybrids and Calcutta 4	Simmonds (1952) Ortiz and Vulsteke (1992)
Fruit ripening period	Transgressive segregation due two complementary genes or partially dominant gene(s) toward long shelf life	Ferris and Ortiz ¹
Persistence of male bracts	Two loci with complementary dominant genes, which are independent of the genes for persistence of hermaphrodite flowers in plantains	Ortiz (1995)
Persistence of hermaphrodite flowers and male bracts	Two independent loci with complementary and dominant genes in bananas and plantain-banana hybrids	Simmonds (1952); Ortiz (1995)
Bunch weight	Epistatic interactions increase yield in poliploid hybrids	Ortiz and Vulsteke (1993)
Red pigmentation in leaves	Modifier gene interaction due to recessive suppressor	Ortiz (1995)
Pollen presence with 2n chromosomes	Dominant gene	Ortiz (1997)
Banana weevil resistance	Gene(s) with incomplete/partial dominance toward resistant parent in the diploid plantain-banana hybrids	Ortiz et al. (1995b)
Bacterial wilt (moko disease) resistance	Several recessive genes	Vakili (1965b); Rowe and Richardson (1975)
<i>Fusarium</i> wilt resistance	One major dominant gene for race 1. Polygenic system for race 4	Larter (1947); Vakili (1965a). Rowe (1991)
Burrowing nematode resistance	One or more dominant genes	Rowe (1991)
Yellow Sigatoka resistance	Recessive genes in <i>M. acuminata</i> ssp. <i>burmanica</i> Dominant genes in <i>M. acuminata</i> ssp. <i>malaccensis</i> Multiple genes with dosage effects in <i>M. acuminata</i> ssp. <i>microcarpa</i> e ssp. <i>errans</i>	Shepherd (1990) Rowe (1984)
Black Sigatoka resistance	One major recessive gene and two additive minor genes with dosage effect in plantain –banana hybrids	Vakihi (1968) Ortiz and Vulsteke (1994 a)
Fruit size and weight	Larger fruits in polyploids due to epistasis. Several dominant genes in <i>M. acuminata</i> ssp. <i>malaccensis</i> .	Ortiz and Vulsteke (1993) Rowe (1984)

^{1/} FERRIS, S.; ORTIZ, R. IITA-ESARC. Personal Communication. Kampala, Uganda, 1997.

biggest threat of this type of culture is the introduction of virus diseases, specially the bunchy top disease which occurs in India, Philippines and possibly in Indonesia. The presence of virus particles in the plantlets in *in vitro* culture is still possible thus indexation is recommended for the main virus diseases in banana germplasm exchange.

There are 43 banana collections in the world, in 33 different countries (a few examples are: Honduras, Jamaica, Philippines, Guadelupe, Camaroon, Cuba, Colombia and Brazil) (Silva et al., 1997). The Honduras collection, developed by the United Fruit Company in 1959 is the biggest and constitutes the basis for the "Fundación Hodureña de Investigación Agrícola -Fhia" program. This collection includes approximately 850 accesses (with more than 200 diploids), brought mainly from the Philippines, Malaya, Indonesia and New Guinea (Rowe, 1985).

The main banana germplasm collection in Brazil can be found at the Embrapa Cassava and Fruit Crop, Cruz das Almas, Bahia. This collection has been enriched and expanded in the last years by means of national introductions and international collections from countries such as India, Philippines, New Guinea and Hawaii - 1982; Venezuela and Equator-1983; Martinica, Guadelupe, Thailand, Malaya and Indonesia-1985. Presently, around 280 accesses, including species and wild subspecies, cultivars, and hybrids which are being maintained under field conditions (Silva and Shepherd, 1991) belong to this collection.

The most important banana genetic variability is found in the diverse wild forms of *M. acuminata* and in the AA cultivar group. This species includes seven subspecies, some of which are still not well defined. Each subspecies has its own distribution system in Asia and Oceania. Although their morphological differences are pronounced, they cannot be classified as distinct species since fertile hybrids can be obtained from all subspecies. The AA cultivars also show significant morphological diversity, many presenting sterility or low fertility (Shepherd et. al., 1986).

Diseases

As in any species cultivated in large areas, the banana is affected by several phytosanitary

problems caused by fungi, bacteria, virus, nematodes and insects. Fungi, infectious agents of extreme importance, cause diseases such as Fusarium wilts (Panama disease), and leaf spots (yellow and black Sigatoka) becoming the biggest problems for banana crops in the world. Among the bacteriosis, wilt or moko disease stands out, about which little is known in relation to sources of resistance. The "bunchy top" virus, still not found in Brazil, is considered the major viral etiology problem for crops. The nematode of great importance for bananas is the *Radopholus similis*, and the weevil borer caused by the *Cosmopolites sordidus* is the pest that causes most damage. Pests, diseases, and nematodes are responsible for severe losses in the production of bananas, which, depending on the factors involved, can achieve up to 100%, considering that, in many cases, there is no alternative control.

Yellow Sigatoka is caused by *Mycosphaerella musicola*, Leach, the perfect or sexual form of *Pseudocercospora musae* (Zimm) Deighton. Sexual spores (ascospores) and asexual spores (conidia) are produced. While black Sigatoka is caused by *Mycosphaerella fijiensis* Morelet (sexual phase) or *Paracercospora fijiensis* (Morelet) Deighton (asexual phase). The sexual phase is considered the most important for the increase of the disease, considering that, a large number of ascospores are produced in structures denominated pseudotecius.

The search for resistant varieties by means of selecting inside existing genetic resources and by means of generating new varieties through hybridization is nowadays the main approach to Sigatoka control. At least five breeding programs, including one in Brazil, is presently investigating resistance to yellow or black sigatokas (Silva et al., 1998a). However, the Brazilian program gives more attention to yellow Sigatoka.

Many resistant cultivars have vertical resistance to yellow Sigatoka (Shepherd, 1990; Cordeiro, 1997), as the 'Pioneira', launched by Embrapa, the 'Mysore' and the two tetraploid hybrids such as the PV03-44 (produced in Embrapa) and the FHIA-18 (produced by the Fhia). In the banana resistance to yellow Sigatoka evaluation, under field conditions, the following classes were found: highly susceptible, slightly susceptible, resistant and highly resistant.

Presently, the great majority of the studies concentrate on the black Sigatoka (*M. fijiensis*) with no variations in the methodology and the parameters used (Meredith and Lawrence, 1970; Fouré, 1982; Fouré et al, 1984; Fouré, 1985). Several characteristics such as period of the disease development number of functional leaves at flowering and harvesting and disease severity were considered by these studies. According to Fouré (1993), four distinct phenotypes can be observed among the evaluated genotypes: highly resistant, partially resistant, susceptible and highly susceptible.

Fusarium wilt, caused by *Fusarium oxysporum* f. sp cubense, E. F. Smith, is a soil fungus with a high survival rate, making the disease control even more difficult. It is a serious problem for banana crops in the world, specially in Brazil, where these cultivars are susceptible and sometimes highly susceptible to the pathogen. This fungus is a limiting factor for the Maçã variety, which is very much appreciated in the Brazilian market.

Experience has shown that Fusarium wilt must be controlled through the use of resistant varieties. Thus, the evaluation of banana genotypes resistant to this disease, aiming at new cultivars with this characteristic, has been a priority in Panama disease control. This experience was reported by Cordeiro et al. (1993a; 1993b) during the development of new methodologies for resistance evaluation in diploid, triploid and tetraploid genotypes.

Moko disease or bacterial wilt, caused by *Ralstonia solanacearum*, race 2 (*Pseudomonas solanacearum* (Smith), is another serious problem affecting banana crops. According to Wardlaw (1961), banana varieties are susceptible to bacterial wilt. However, Stover (1972) evaluated 345 banana genotypes and concluded that 34 of them had some degree of resistance and that the Pelipita (ABB) variety was highly resistant. Resistant diploid were also selected (Silva et al., 2000) in evaluations conducted in the Amazon region. These authors agree that cultivars show peculiar degrees of susceptibility to this disease. Hence, the persistent bracts varieties are less susceptible to the infection by insects (low bacterial infection), while some Plantains show some field resistance. Presently, the absence of

efficient phytosanitary control techniques has led to a selection of varieties resistant to moko disease by breeding (INIBAP, 1994).

There is no knowledge of resistance to nematodes among commercial banana cultivars. However, resistance to *R. similis* has been found in *Musa* AA diploids in Honduras (Pinochet and Rowe, 1978; Pinochet, 1988). Davide and Marasigan (1992) selected banana genotypes with different degrees of resistance to *R. similis* and *M. incognita*. AA diploid hybrids have been selected as moderately resistant to *R. similis* and *M. incognita* under greenhouse conditions at Embrapa, in Cruz das Almas, BA (Costa et al., 1997b).

Diploid variability seems to be adequate for immediate breeding purposes. There are morphological variations in height, in the vigor of suckers, in the number of hands per bunch, in the size of the fingers and sources of resistance to the main pests, diseases and nematodes. However, in some cases, as in the Cavendish cultivars, these characteristics cannot be transferred to the tetraploids due to the absence of seeds production in the crossings between diploid (AA) and Cavendish triploids (AAA).

Banana breeding programs carried out in different sites have, in general, the following objectives:

1. To develop banana types resistant to pests, diseases and nematodes such as 'Prata', 'Maçã', 'Plantains', 'Gros Michel' and 'Bluggoe' by means of conventional breeding methodologies, reducing height, crop cycle and increasing yield.
2. To develop banana varieties resistant to pests, diseases and nematodes such as Cavendish' and 'Maçã', by biotechnology, reducing height, crop cycle of the crop, and increasing yield.
3. Identify genotypes with the best agronomic characteristics regarding yield and fruit quality.

Introduction and selection of clones

Acquiring promising germplasm introduced from other regions can fulfil the same purposes of a breeding program in obtaining superior varieties. Hence, the introduction is considered a breeding method, as it supplies the genetic variability necessary to obtain new cultivars

and/or select clones (Elliott, 1958; Allard, 1971).

The low genetic variability of a crop represents an eminent risk due to either the absence of new cultivars or its disappearance caused by a disease. This is what occurred in the past with the Latin American banana crop for export, based only on the 'Gros Michel' cultivar, which is susceptible to the "Panama disease". Nowadays the banana export business runs similar risks since it is practically based on a single banana clone of the Cavendish subgroup, the 'Grande Naine' (Janick, 1998).

Somaclonal variations occur in a much higher level in banana species than in the majority of the other cultures, probably due to the meiotic instability, which is not only common in tissue culture but are also observed in the field although in small frequencies (Silva and Shepherd, 1991; Withers, 1992).

As crops are vegetative propagated and genetic breeding is relatively difficult, *in vitro* somaclonal variation to obtain new genotypes should be considered. However, this technique is limited in banana culture, being used rationally and broadly in germplasm handling as apical buds. The strong genotype dependence of the *Musa* genus makes it unfit for cell suspension, protoplast and anther culture (Vuylsteke, 2001).

The somaclonal variation used in breeding has the following advantages and disadvantages:

Advantages:

1. The development of new and stable variants;
2. High variation frequency;
3. The development of variability in agronomic characteristics.

Disadvantages:

1. Uncontrollable and imperceptible variations;
2. Variability is not new and apparently useless;
3. Nature and frequency of variability depend on the genotype and other factors;
4. Some variations are unstable and not inherited.

In vitro and field somoclonal variations produced dozens of cultivars of the (AAA) genomic group, a Cavendish subgroup, and of the AAB cultivars such as Pacovan, a 'Prata' mutant. Thus, the

selection of superior clones can contribute significantly to the increase in production and quality of the banana fruits. Lichtemberg (1997) emphasized the importance selecting natural mutants for banana crops in Israel, South Africa, Australia and Spain (Canaries). These countries cultivate little more than 40.000 hectares of banana, in subtropical conditions. In South Africa, this selection is carried out with the help of farmers who pre-select clones in their plantations. These clones are later studied by public research institutions (Lichtemberg, 1997). In Israel, private companies maintain breeding programs using clone selection as the most promising technique (Khayat the al., 1998).

To verify the true value of selections carried out in tropical regions, clones from Israel were evaluated in the Philippines, and they showed an 18% productivity increase over the best local selection, as well as superior quality (Khayat the al., 1998). Presently, Israel exports plantlets of these clones worldwide, including Central and South America, and, more recently, the Northeast of Brazil. Clones selected in Israel, Taiwan, South Africa, Canarias and Australia are being evaluated in the Madeira Island and South Africa (Ribeiro and Silva, 1998; Eckstein et al., 1998).

Mutants resistant to pests and diseases are easier to select than superior clones for quality, productivity, architecture and plant height. Hwang and Ko (1986) evaluated the field behavior of a diverse number of banana mutant clones of the Cavendish subgroup in Taiwan and obtained resistant genotypes to *Fusarium oxysporum* f. sp. Cubense (race 4), responsible for the Panama Disease by meristem culture.

Breeding by hibridization

Banana genetic breeding research started in three peculiar locations: Trinidad, in 1922, by the Imperial College of Tropical Agriculture, Jamaica, in 1924, by the Department of Agriculture and Honduras, in 1930 by the United Fruits Company. The main objective was to produce a banana with all the qualities of the 'Gros Michel', but resistant to the Panama disease (Shepherd, 1974). In the early 30's, the first tetraploid hybrid was developed by crossing between a (AAA) triploid the 'Gros Michel' as the female genitor and the wild diploid species *Musa acuminata*, a subspecies of

Malaccensis. Thus, a hybridization system for breeding some triploid banana cultivars was developed (Shepherd, 1992).

Mutation induction, genetic transformation and conventional hybridization are techniques possible to be used in banana culture. Presently, however, only hybridization has shown good results.

However, due to female sterility in edible bananas, basic genetic variability is not sufficiently reliable. There must be other hybridization options in the generation of new cultivars with improved traits. Experience has shown that female sterility is not totally absolute. The majority of them can produce, in all levels of ploidy, seeds under controlled pollination, with a great or small frequency. This production is generally more evidenced after fertilization with haploid A pollens originated from wild forms, cultivars and AA hybrids constitution. (Dantas et al., 1997).

In the development of female gametes, meiosis is predominant in hybrids, wild species or cultivars. However, two other processes can provide megaspores and embryonic sacs with the maternal chromosome number duplicated (double restitution). These two last patterns are developed due to the non reduction of chromosomes in the meioses which occur in diploids, being more frequent in triploids and less common in tetraploids. (Dantas et al., 1997)

The fertilization of embryonic sacs with the maternal chromosome numbers duplicated can lead to viable seeds but useless plants. The fertilization of non reduced diploids or triploids sacs can contribute to desirable results in hybridization programs aiming at new triploid or tetraploid genotypes. This occurs directly or by subsequent secondary crossings. According to Dantas et al. (1997), there are four hybridization classes:

1. Triploids which are the result of diploids X diploids crossings, with the original diploid male parental recombination only. The seeds obtained from females AA parentals, with the A pollen, normally contain embryos which are equally diploids, originated from egg cells with only 11 out of the 22 maternal chromosomes. Exceptionally, egg cells may contain all the 22 maternal chromosomes or have that number duplicated (44), which, after having been fertilized by haploid pollen, will generate triploid and pentaploid embryos, respectively. Theoretically, the same alternatives exist from AB plants, in natural cultivars or synthesized hybrids. Triploid cultivars supposedly evolved by this option.
2. Tetraploids which are the result of triploids X diploids crossings, with the diploid male parental recombination only. A triploid cultivar with low female fertility can produce embryos and hybrids with around 22 and 33 chromosomes due to the unbalanced meiosis (embryonic sacs with 11 to 22 chromosomes, plus 11 chromosomes of the haploid pollen), as well as embryos and hybrids with 44 chromosomes (33 plus 11) or 77 chromosomes (twice 33, plus 11). In practice, however, tetraploid hybrids carry 44 chromosomes that can be used as commercial cultivars becoming the second option for breeding by hybridization. It is important to emphasize that the pollen contributes with only a quarter of the new genotype, in each fertilization of this kind. Therefore, it is basically a process of implantation of additional characteristics without causing other representative alterations. Hence, the tetraploid hybrid always carries characteristics of the parental female triploid, including those related to the taste of the fruit.
3. Tetraploids are the result of crossings among tetraploids with segregation in the two parents. Spontaneous or synthesized tetraploid bananas contain even numbers of multiples of 11 chromosomes and, theoretically, they should be more fertile than the triploids. The diploid pollen of the tetraploids, however, has a reduced power that rarely allows for self fertilization or the fertilization of other tetraploids. The pollinic tubes in the style of diploid and tetraploid plants are in small numbers grow slower than the tubes developed by the haploid pollen. Consequently, the production of secondary tetraploids is rarely practicable, due to very low seed production.
4. Triploids which are the result tetraploids X diploids crossings and segregation in the two parentals. By using the A pollen of diploids, it is frequently possible to obtain good seed performance and, consequently, hybrid secondary triploids.

The most common procedures used in breeding, taking into consideration the mechanisms that lead to the development of entirely new triploid genotypes (first and fourth classes of hybridization mentioned), as well as those mechanisms which modify the existing triploids (second class of hybridization) are discussed next. Of fundamental importance for the three production classes of polyploid hybrids mentioned before is breeding at the diploid level, which is in agreement with the comments below.

Breeding at the diploid level

Results from a banana breeding program, independently of its objectives (triploids or tetraploids production), depend basically on the quality of the parental diploids used in the generation of the desirable hybrid. They are fundamental for the incorporation of characteristics of agronomic value.

The desirable characteristics of *M. acuminata*, however, are not joined together in a single individual, but distributed among many basic diploid accesses and, therefore, an additional hybridization, recombination and selection at the diploid level, involving subspecies of *M. acuminata* and its cultivars are necessary in any conventional breeding program.

The objective of germplasm breeding is to concentrate, in a single genotype, the largest number of desirable characteristics, such as parthenocarpy, good number of hands, long fingers, well formed bunch, and resistance to pests, diseases and nematodes.

Diploid breeding is a simple process involving the cross of selected parentals for desirable characteristics and male and female gametes which are the result of regular meiosis, thus developing diploid hybrids (ex: 2X x 2X P 2X -primary). Another diploid breeding alternative consists of obtaining hybrids derived from 3X x 2X P 4x and 2x crossings, with subsequent enrichment by means of crossings among these hybrids (ex: 2X (derived 1) x 2X (derived 2) P 2X improved (Vuylsteke et al., 1993; Tomekpe et. al., 1995).

According to Horry et al. (1993) diploids can also be improved by means of pure lines with subsequent formation of hybrids between lines.

In the three types of breeding, the process develops by means of successive output of generations of diploid hybrids accompanied by a continuous selection of the best genotypes resultant from all the crossings planted in the field. Several kinds of crossings, involving wild species, cultivars and hybrids should be carried out to obtain male improved parentals, which are used in triploid or tetraploids breeding. It is important to have basic diploid accesses with good combining ability. An example of this combining ability can be found in some accesses of *M. acuminata* ssp. *banksii*, which produces bigger fruits than those found in other wild forms, and which incorporates the resistances to other desirable characteristics in simple hybrids, without remarkable losses in the size of the fruits.

In practice, the basic diploid germplasm consists of a variety of wild forms and fertile cultivars from the AA group, sufficient to satisfy all breeding objectives. The AA germplasm contributes to the resistance to several diseases and other favorable characteristics. Although *M. balbisiana* presents resistance to diverse diseases and pests and is distributed in a wide area in Asia and in the nearby islands, it shows little variability in several traits such as form and size of the fruits. In addition, there are other known parthenocarpic diploid forms of this species. Despite being the most accessible diploid in banana genetic breeding, the AA germplasm must also be considered as other species of the sections (Eu-) *Musa* and *Rhodochlamys*, of great affinity with *M. acuminata*, which can donate genes to this species by means of a series of backcrossings from the F1 hybrids. The other species to be considered are: *M. flaviflora* Simmonds; *M. halabanensis* Meijer; *M. ochracea* Shepherd and *M. schizocarpa* Simmonds, from the Section (Eu-) *Musa*; *M. laterita* Cheesman; *M. ornata* Roxburgh and *M. velutina* Wendl. & Drude, from the Section *Rhodochlamys*.

Some diploid accesses possess hermaphrodite and male flowers, which makes anther elimination (emasculation) necessary to control the crossed pollination. Meanwhile, the majority of the bananas and diploids helpful to the breeding process have male and female flowers.

Breeding programs conducted in Honduras (Rowe and Rosales, 1993), Jamaica (Shepherd,

1974), Nigeria (Vuylsteke et al., 1993), Cameroon (Tomekpe, 1995) and Guadelupe (Horry et al., 1993) have generated diploid hybrids with a large number of fruits, low stature, and resistance to yellow and black Sigatoka, the Panama disease and nematodes (*Radophilus similis*). Some of these hybrids, such as the M-48, M-53 and M-61 produced in Jamaica and with a large number of fingers and resistant to the Panama disease and yellow and black sigatoka, the SH3263 of low stature, productive and resistant to *Radophilus similis* and the SH3362 with low stature, productive and to Race 4 of *Fusarium* and produced in Honduras, were incorporated into the breeding program at Embrapa.

Production of triploids from diploids and tetraploid x diploid crosses

Triploids are presently the most used banana cultivars. Studies about the output of new 3X cultivars, from diploid stock plants, however, have been performed in a small scale, although this is the supposed evolution pattern for the existing triploid cultivars. These studies, initially depend on the identification of diploids with substantial output of viable and not reduced embryonic sacs, with some of the qualities of the desirable cultivar. These qualities are necessary, as this diploid will contribute with two thirds of the genotype of each triploid produced.

As for the production of new triploids, AAB and AAA triploids must be considered separately, as their problems and perspectives are distinct.

Production of AAA triploids from the AA diploids

Despite the existence of many old cultivars of the AAA group, literature has shown little experimental evidence on the output of those cultivars. Among cultivars and parthenocarpic AA constitution hybrids, the production of triploid embryos is an unusual event, although, in Jamaica, some crossings have occurred inside the AA group, in which a proportion of triploid hybrids, involving crossings between *M. acuminata* ssp *banksii* and the cultivar Paka (Shepherd, 1976), with bunches larger than the ones produced by the diploids, but with no commercial value, was found. On the other hand,

many other crossings among AA genotypes no triploid appeared. Dodds (1943) reported that, in crossings of several AA cultivars, only two hybrids obtained from the parental female 'Lidi', with *M. acuminata* ssp. *Malaccensis* pollen, were triploids.

In summary, Dantas et al. (1997) reported that the production of AAA triploid genotypes via crossings between diploids is a methodology which does not offer good perspectives for the production of AAA group cultivars due to the following disadvantages:

- 1) Few AA diploids are capable of generating triploid embryos, resulting in a serious limitation for the used variability;
- 2) Generally, the small number of triploid plantlets produced occurs simultaneously with a large number of diploids, becoming necessary to distinguish plantlets with different levels of ploidy.

Production of AAB triploids from AB diploids

The existing cultivars from the AAB and ABB groups, supposedly evolved by means of fertilization of AB hybrids ovules by pollen A and B, respectively. However, all ten AB hybrids studied showed high sterility, being incapable of producing haploid spores (pollen or embryonic sacs) due to the lack of complete chromosomes pairing at meiosis (Dodds, 1945; Dodds and Pittendrigh, 1946; Dodds and Simmonds, 1946). Nevertheless, some hybrids produced non reduced spores, which produced chromosome duplication in the majority of the cases. However, the tetraploid pollen observed in young stages was useless. The non reduced embryonic sacs were rarely viable, except for the recovery of few seeds. The germinated plantlets were generally pentaploids by the addition of a haploid nucleus of the pollen to the tetraploid egg cell. An exception was found in Trinidad, where an AB hybrid produced many seeds by using the pollen of the parental species. The large number of plants obtained from these seeds, as well as the few recovered from other AB parentals, were a mixture of triploids and pentaploids. None of the parentals produced any back crossed diploid plant (Shepherd, 1976). Based on this exception, the Embrapa Cassava and Fruit Crop Breeding Program developed a project aiming at developing a new and wider prospect of AB hybrids female fertility and at identifying those

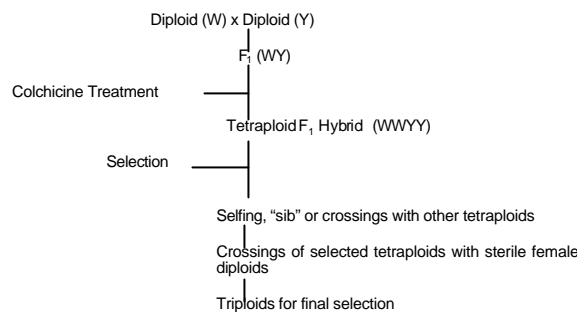
capable of producing a large number of AAB triploids. AB hybrids with a high seed output and with germinated plants constituted of mixtures of triploids and pentaploids, easily discriminated by their distinct morphological aspects (Shepherd et al., 1986) were expected. However, the AB hybrids synthesized at Embrapa did not confirm such expectations. The problem was that many AB hybrids presented a high capacity to produce diploid and aneuploids by backcrossings. Pentaploid plantlets were commonly found, but triploids were rare. Only two plantlets showed the desired behavior (mixture of triploids and pentaploids), becoming the most promising hybrid from the Bluggoe cultivar, in which viable seeds always include a small proportion of diploids, after being pollinated by the A pollen.

The advantages of producing new AAB genotypes by means of AB + A fertilization are:

- 1) Since the female AB parentals can recover 15 or more triploid plants per pollinated bunch, the unit cost of those plants will be relatively low;
- 2) By using few constant parental females and contributing with all of its chromosomes in the development of the hybrid, the variability liberated will be limited mainly by the contribution of the A pollen, which will affect the efficient use of the available space in the field;
- 3) The methodology utilizes the AA germplasm improved by two successive stages, an advantageous process, considering that the *M. balbisiana* species present little useful variability.

The main disadvantage of this approach is that any of the favorable characteristics of a synthesized AAB cultivar such as productivity and resistance to pests and diseases may be associated with fruits with different taste from that of the other cultivars of the group which has already been fully accepted by the consumer market (Shepherd et al., 1986).

Despite being laborious, an alternative route for triploid production is the duplication of chromosomes of the AA or AB promising genotypes by the colchicine treatment followed by tetraploid x diploid crosses, as proposed by Vakili (1967). The author verified that tetraploidy in *M. acuminata* and *M. balbisiana*, could be easily induced by colchicine which is a method being used by the banana breeding program in the Cirad-Flhor (Montcel et al., 1995):



Another method for obtaining triploids would be by means of tetraploids X diploids crossings, considered by some breeders as the last phase of a breeding program. In the Gros Michel subgroup, many tetraploid (AAAA) hybrids generated in Jamaica and in Honduras have been used as parentals of a triploid generation.

Considering that this process segregates the two parentals (tetraploid and diploid), Rowe and Richardson (1975) suggested that a broadly liberated variability would bring more benefit to the breeder. However, Dantas et al. (1997), considered the broadly liberated variability by the 4X x 2X crossings disadvantageous due to the fact that a large number of plants is needed for a useful selection. On the other hand, another advantage of the method consists in using a diploid selected germplasm in two successive phases. The resulting triploids would have self-sterility, an additional advantage, in contrast with the tetraploids, which present the theoretical possibility of selfing and, consequently, the production of seeds may or may not occur (Rowe, 1985).

AAB triploid crossing products resulting from ABB with AA are more interesting to Brazilian breeding programs. Few tetraploid hybrids have been produced by Bluggoe cultivar x *M. acuminata* crossings used in the production of secondary triploids. Another source of AABB tetraploids to be exploited is based on 'Pisang Awak' pollinations, a cultivar from the ABB group which produces many seeds. Unfortunately, new accesses from the Awak type are relatively sterile.

Tetraploid Production from Triploids

Since the beginning of the banana genetic breeding research programs, tetraploid production from triploids has been widely used. Recently it has been restricted to the output of

tetraploid hybrids from AAA triploids, of the Gros Michel subgroup, pollinated by A pollen. The application of this method to other cultivars or subgroups depends on their capacity to produce seeds after adequate pollination, on their seed germination potential and on the presence of the tetraploid within the recovered hybrids.

In the production of tetraploids from triploids ($3X \times 2X \rightarrow 4X$), the following objectives can be reached by using a determined pollen A (Dantas et al., 1997):

- 1) Resistance to black Sigatoka (all the important cultivars of the AAA, AAB and ABB groups);
- 2) Resistance to yellow Sigatoka (subgroup Gros Michel, subgroup Prata and 'Prata Anã');
- 3) resistance to race 2 of Panama disease (subgroup Gros Michel, 'Maçã', subgroup Bluggoe and other possible cases);
- 4) Resistance to nematodes (more relevant in relation Plantain and Bluggoe subgroup);
- 5) Resistance to the weevil borer (subgroup Plantain);
- 6) Low stature ('Maçã', 'Mysore', Subgroups Prata and Plantain);
- 7) Short production cycle (subgroup Gros Michel cultivars of the AAB group);
- 8) Greater number of hands ('Maçã', 'Prata Anã', and subgroup Prata); bigger fruits ('Maçã', 'Mysore', 'Prata Anã' and subgroup Prata).

The artificial production of tetraploid and heptaploid hybrids (four to seven multiple of $X = 11$, respectively) from triploids took place more than 50 years ago, in Trinidad, when three main alternatives in the behavior of the mother cell of the embryonic sac were observed. The alternatives are similar to the those verified in AB hybrids (Shepherd, 1974):

- 1) In the great majority of the ovules, an unbalanced meiosis occurs, common in triploids of any sort. The resultant spores are rarely viable, producing haploid, diploid or aneuploid functional embryonic sacs with intermediate number of chromosomes;
- 2) Without meiosis and alterations in the chromosomes, the mother cell develops a triploid sac;
- 3) Without meiosis and with chromosomes duplication, the mother cell develops an hexaploid sac.

After pollination by haploid pollen, these alternatives can give origin to embryos and hybrids that are diploids ($2X$), triploids ($3X$), aneuploids (between 23 and 32 chromosomes), tetraploids ($4X$), and heptaploids ($7X$) respectively. In breeding, desirable hybrids are tetraploids with a complete genotype of the parental female triploid and the majority of its characteristics, besides other helpful genes of the parental male triploid. The tetraploid production is an approach used for quick results, since adequate diploid germplasm is available. Proper selection of the parental diploid should confer resistance to the diseases and modify other characteristics of the plant.

Rowe and Rosales (1995) proposed another plan for tetraploid production (secondary) from primary and secondary triploids which involves the use of three peculiar diploids: $3X$ (primary) \times $2X$ (a) \Rightarrow $4X$ (primary) \times $2X$ (b) \Rightarrow $3X$ (secondary) \times $2X$ (c) \Rightarrow $4X$ (secondary). From this sequence of crossings, was obtained the SH 3386, a secondary tetraploid, descendant from the Gaddatu (ABB), a clone from the Philippines used as primary triploid. This system can be applied successfully in the ABB banana breeding, but not in dessert bananas and Plantains, due to the loss of desirable agronomic characteristics during the process.

The most prominent studies on the fertility of diverse female triploid parentals were carried out by Cheesman and Dodds (1942) and Shepherd (1960). These studies confirmed the differences in fertility among the evaluated cultivars, as well as in the proportions of plantlets resultant from the several triploids. Some of these varieties, such as the ones from the Cavendish (AAA) subgroup were almost totally sterile, while the most fertile belonged to the ABB group. The yield of seeds were generally lower with the *M. Balbisiana* pollen, than with the *M. acuminata*. 'Caru Roxa' (AAA) pollen and 'Mysore' stood out due to the large number of underdeveloped or empty bad seeds. In addition, the low germination frequency suggested that their viability is restricted. These and other results are summarized in Table 4.

Three cultivars from the AAB group were already evaluated in regards to seed output. Results showed that they produced a larger number of seeds than the 'Gros Michel'. The seeds from 'Prata' germinated very well and the ones from 'Maçã'

and 'Mysore' were less viable, consequently, tetraploids from 'Prata' and 'Mysore' hybrids were generated. As for the 'Bluggoe' (ABB), only a few tetraploid plants were weak.

Although Shepherd (1960) failed in obtaining seeds from Horn Plantain ('Pacova') and French Plantain ('Terra') diploid crossings, tetraploid hybrid from these cultivars were obtained in Honduras (Rowe and Rosales, 1993) Nigeria (Vuylsteke et al., 1993) and Cameroon (Tomekpe, 1995), showing the female fertility of these genotypes as well as the viability of the resultant tetraploid plants.

Breeding programs have developed triploids from crossings of improved tetraploids with diploids ($4X \times 2X \Rightarrow 3X$). However, the

recommended hybrids are tetraploids. Table 5 shows some hybrids recommended by the Fundación Hondureña de Investigación Agrícola. These hybrids, with the exception of the Bluggoe type AVP-87 and FHIA-15, which are unpopular among Brazilian farmers, were introduced in Brazil and are being evaluated in many regions.

Breeding by mutation

The production of an improved cultivar by any method is a process based on generation and use of genetic variability, helpful genotype selection and evaluation in order to demonstrate the superiority of the selected genotypes for specific agronomic characteristics. These stages are extremely time consuming when traditional

Table 4 – Female fertility of several banana triploid varieties after pollination with A pollen.

Variety and genomic group	Production of good seeds	Germination	Hybrid types ^{1/}
Gros Michel (AAA)	Few	High	4x to 7x
Cavendish subgroup (AAA)	None (\pm)	-	-
Red/ Caru Roxa (AAA)	Enough	Low	2x to 3x
Mysore (AAB)	Few	Low	4x and few 7x
Pome /Prata (AAB)	Few	High	4x in mix
Silk /'Maçã (AAB)	Few	Low	One but no 4x
Plantain (AAB)	None	-	-
Bluggoe/ Figo (ABB)	Enough	Low	Few 4x, mix 2x to 3x
Awak Legor/ +'Gia Hui (ABB)	a lot	Low	4x

Source: Cheesman & Dodds (1942); Shepherd (1960).

^{1/}2x - diploids; 3x - triploids; 4x - tetraploids; 7x - heptaploids.

methods are used. However, biotechnology has provided several alternatives to fast increase the genetic variability (Roux et al., 1994; Perea and Constabel, 1996). One of these techniques, *in vitro* mutagenesis, aims at correcting defects of one or few genes in genotypes of interest, thus, it is considered a small adjustment in the termination of a variety. Mutation breeding goes through the following phases: establishment of the *in vitro* culture, generation of the adventitious explants, mutagenic treatment (increase of the variability), regeneration of plants, acclimatization in nurseries, field selection and multiplication (Perez Ponce, 1998). Chemical agents such as Ethylsulphate methane (EMS) (Jamaluddin, 1994), and high levels of citocinines (Tujillo and Garcia, 1996) and 2,4 D (Beer and Visser, 1994) are used as mutagenics. Another highly used mutagenic agent is cobalt (60 radiation) (Pérez Ponce and Orellana, 1994; Smith et al, 1994).

In vitro selection identifies plants which tolerate several kinds of stresses caused by herbicides, low temperatures, aluminum, manganese, salinity and pathogens toxins (Smith and Drew, 1990). A micropropagation methodology is used to select *in vitro* mutant plant species. In addition, the characteristic to be selected should express itself both *in vivo* and *in vitro* (Constantin, 1984). Tolerance to heavy metals, herbicides, salinity, and resistance to diseases are among the *in vitro* characteristics found in the banana culture (Tulmann-Neto et al., 1990).

The selection of variants resistant to diseases has been emphasized in the past years (Shepherd, 1990). This technique is based on the culture of the parasite (or its toxin) with the explant, in contaminant-free conditions, which favors the fast recognition of tolerant genotypes.

Table 5 - Characteristics of cultivars recommended by Federación Hondureña de Investigación Agrícola.

Hybrid	Genealogy	Characteristics
FHIA-01 (AAAB)	'Prata Anã' (AAB) x SH 3142 (AA)	Resistance to black Sigatoka, races 1 and 4 of Fusarium wilt, nematode (<i>Radopholus similis</i>), tolerance to low temperatures and unfavorable conditions of raining and soil fertility; strong plants with good architecture; large bunches and fruits with good texture and taste.
FHIA-02 (AAAA)	'Wilhams' (AAA)x SH 3393(AA)	High resistance to black Sigatoka, present the same plant height of 'Valery', however the bunch characteristics are inferior to this cultivar
FHIA-03 (AABB)	SH 3386 (ABB) x diplóide SH3320 (AA)	Generated from the clone Gaddatu ABB, it is more rustic and productive than the Bluggoes. Present low stature and bunches that reach to 50 kg and resistance to moko disease and drought.
FHIA-15 (AAAB)	Descendent of Maqueño	Resistance to race 1 of the <i>Fusarium</i> present tolerance to black Sigatoka.
FHIA-17 (AAAA)	Highgate hybrid (AAA) (mutant of 'Gros Michel')	Good characteristics of bunch, resistance to race 1 of Fusarium wilt and more tolerant to black Sigatoka than the Grande Naine, however its plants are higher than that cultivar, mainly in second cycle.
FHIA-18 (AAAB)	Prata Anã hybrid (AAB)	High production, resistant to yellow and black sigatokas and fruits present good taste.
FHIA-21 (AAAB)	French Plantain Hybrid (AAB)	Resistance to black Sigatoka and produce larger bunches than to the others False Horn cultivars.
FHIA-23 (AAAA)	Highgate hybrid (AAA)(mutant of 'Gros Michel')	Good characteristics of bunch, resistance to race 1 of Fusarium wilt and more tolerant to black Sigatoka than the Grande Naine, however its plants are higher than that cultivar, mainly in second cycle.
AVP-67 (AAAB)	Crosses involving plantain French type (AAB) and Maqueño (ABB)	Good characteristics to use unripe or ripe, resistant to race 1 Panama disease and tolerance to black Sigatoka.
SH 3640	Prata Anã (AAB) x SH 3393	High production and resistance to black Sigatoka, partial resistance to yellow Sigatoka and ripe fruits present very good taste.

Source: Rowe and Rosales (1993; 1995)

As for the selection of plants tolerant to toxins, Pegg and Langdon (1986) suggested that tests of pathogenic comparisons should be carried out among the isolates from different localities in order to choose the most pathogenic, as the differences in the expression of pathogenicity are related to several pathogens in different cultures (Balardin et al., 1990; Henz et al., 1987; Maas, 1985).

Toxins are substances of molecular weight less than 2000, which interfere with the normal development of the plant and correspond to the main components of metabolites produced by the microorganisms. Due to these characteristics, they can be used successfully as selective agents during the selection process (Walton and Earle, 1984). Most of the times, a direct correlation between

tolerance of explants to toxins and plant resistance to disease is observed (Toyoda et al., 1989).

An undesirable factor in banana micropropagation is the appearance of somaclonal variations. However, these variations can help in the generation and selection of adequate genetic variability. The TCI 215-1 dwarf mutant of GCTCV-215-1 was selected by this technique (Tang and Hwan, 1994). As it is a rapid process which does not require adequate climatic conditions, banana breeding by mutation has been used in the Austria (IAEA), Honduras, Australia, South Africa, Colombia, Costa Rica, Cuba, Brazil, Nigeria, Sudan, Pakistan and Malaya (Roux et al., 1994).

Through the use of high levels of cytokinin in apical meristems, genotypes more resistant to yellow Sigatoka than the original cultivars were obtained (Tujillo and Garcia, 1996). Cobalt 60, with 15 to 60 Gy radiation was used to obtain clones from the Grande Naine and Parecido el Rei cultivars, the hybrid SH 3436, which is more resistant to black Sigatoka, and the Maçã and Gros Michel cultivars, which have resistance to *Fusarium* (Pérez Ponce and Orellana, 1994). *In vitro* mutation with gamma rays induction in doses which varied from 10gy to 60 Gy and/or EMS (Jamaluddin, 1994) selected earlier, shorter and higher yielding 'Grande Naine' (Fatom-1) and Pisng Rastali (AAB Maçã) clones. The stability of these potential characteristics and the true condition for resistance to *Fusarium* need to be confirmed by more intensive selection. Gamma ray mutation was also employed by Matsumoto and Yamaguchi (1991) in the development of mutants from 'Nanicão', an aluminum resistant cultivar.

These results show that mutation induction and tissue culture are techniques used to obtain desirable agronomic characteristics, mainly resistance to the Panama disease. However, only few commercial cultivars have originated by this methodology.

Somatic hybridization

Low seed production, which occurs in the majority of the triploid cultivars and diploid crossings during the generation of tetraploids, has been one of the main problems in conventional banana genetic breeding. In the 'Maçã' cultivar, for instance, the few seeds produced do not germinate whereas in the Cavendish subgroup cultivars no seeds have been obtained by this crossing (Shepherd et al., 1994). Somatic hybridization is a very powerful and easy technique to use in vegetative propagated plants and in those with high heterozygosity (Zuba and Binding, 1989; Vardi and Galun, 1989; Sihachakr and Ducreux, 1993). It allows the introduction of poligenic resistance to diseases originated from wild plants of other species or genera (Zuba and Binding, 1989).

The first experiments with protoplasts were carried out in the 70's, and their use in plant genetic breeding programs has been intensified since. The protoplast technique can be used to

obtain plants from a single cell, to incorporate exotic genes by electroporation and to create somatic hybrids by cell fusion (Panis et al., 1995).

Somatic embryogenesis of suspension cells (Novak et al., 1989; Dhed'a et al., 1991) and of protoplast culture (Megia et al., 1992; 1993; Panis et al., 1993) have also been carried out in banana plants. Plant regeneration from the AAA and ABB genotypes were obtained by suspension cells. Regeneration of plants by protoplast culture was initially obtained with success by the Bluggoe (ABB) cultivar (Sagi et al., 1994). More recently Matsumoto and Oka. (1997b) regenerated protoplasts from the triploid cultivar Maçã(AAB).

Embrapa Genetic Resources and Biotechnology has developed techniques to regenerate plants from cells or callus of the cultivars Maçã (AAB) (Matsumoto and Hirao, 1995) and Nanicão (AAA) (Conceição et al, 1998) cultivars, from protoplasts of the 'Maçã' (Matsumoto and Oka, 1997a) and from somatic hybrids of the 'Maçã' (AAB) X 'Lidi' (AA) (Matsumoto et al., 1998).

Genetic transformation

Transgenic plants became a reality in the beginning of the 1980's (Potrykus, 1991). Since then, dozens of distinct vegetal transgenic species were produced, using a heterogeneous range of genes, and a restricted number of transformation systems. Electroporation, Agrobacterium and biobalistics are the three genetic transformation systems responsible for developing almost all the transgenic plants so far. Among the results obtained by transgenics in several species, as resistance to virus, fungi, insects, tolerance to herbicides, increase of post-harvest longevity, production of vaccines, antibodies from animal hormone, and increased protein quality are the most representative.

Although transgenic plants from hundreds of species have been developed in the last two decades, only little more than a dozen have already been commercialized. Licensing to commercialize transgenic plants took place in 1994. Since then, the planted area with transgenics around the world grew from 1,7 for 44,2 million hectares (James, 2000). Dozens of transgenic varieties are being developed in several

projects, and the perspective is that many of them may reach the market in the first decade of the 21st century.

Sagi et al. (1994) reported on the first successful production of bananas by genetic transformation, after observing transient expression of the β -glucuronidase (*uidA* or *gus*) genes of the cv. Bluggoe (ABB group) being transformed by electroporation, without using regenerated plants. The first accounts on transgenic bananas were separately done by two groups in 1995 (Sagi et al., 1995; May et al., 1995).

Transgenic plants from the cv. Bluggoe and from embryonic cells transformed by biobalistics with the plasmid pWRG1515, and selected in culture media supplemented with 50 mg L⁻¹ of hygromicine antibiotics were obtained by Sagi et al. (1995). Transformed plants of the Cavendish subgroup (AAA Grande Naine) were obtained by May et al. (1995) from thin disks of the rhizome and from apical meristems using the *Agrobacterium* system (strain LBA4404) with the binary plasmid pBI141 selected in culture media supplemented with 100 mg L⁻¹ of canamicine and 500 mg L⁻¹ of carbeniciline.

After these first successful accounts on genetic transformation and regeneration of transgenic bananas, other works were carried out, such as the development of plants from 'Three Hand Planty' of Plantain with the gene that expresses an antimicrobe peptid (Dm-AMP1) (Remy et al., 1998) from the Grande Naine cultivar, with *nptII* (neomycin phosphotransferase) and *uidA* genes, from Banana Bunchy Top Virus (BBTV) genes by a transformation mediated by biobalistics (Becker et al., 2000) and from the cv. Bluggoe with the *uidA* and *gfp* (green fluorescent protein) genes by a transformation mediated by biobalistics (Dugdale et al., 2000). More recently, Ganapathi et al. (2001) obtained plants from cv. Rasthali (AAB) with *als* (acetolactate synthase) and *gusa/int* (β -glucuronidase contained an intron) genes by a transformation mediated by *Agrobacterium*.

RESULTS

Germplasm

Banana genetic breeding activities at Embrapa started in 1983, with international and national germplasm collections and introductions (Alves,

1993; Dantas et al., 1993) which gave rise to the Banana Germplasm Bank comprising 283 accesses out of which 87% are cultivars and 13% wild plants. Among the wild plants, *Musa acuminata* and *M. balbisiana* predominate. Accesses from the AAB genomic group, such as Prata, Pacovan, Prata Anã, Maçã, Mysore and Terra, Brazil's most representative cultivars, occur with greater frequency (36%) followed by the accesses from the AA and AAA groups represented by the 'Ouro' and by the Caru Verde, Caru Roxa, São Tomé, Nanica, Nanicão and Grande Naine cultivars. Accesses from AB, ABB, AAAB and AAAA groups are seldom found. However, the banana germplasm is a good representative of the *Musa* genus, with good breeding possibilities (Silva and Shepherd, 1991; Carvalho, 1996; Carvalho et al., 1996, Silva et al., 1997c).

Every germplasm is characterized morphologically (Carvalho, 1996; Silva et al., 1999b). Diploids were also submitted to a molecular characterization by RAPD and microsatellites (Souza et al., 2000; Paz and Silva, 2001). Accesses from this germplasm are being maintained both under field conditions and *in vitro*, and the exchange is been done through micropropagated apical buds. The complete list of the banana germplasm with a description of sinonimia, genomic group and origin is found in Silva et al. (1997c).

The evaluation of the banana germplasm resulted in the identification of promising (AA) diploids (the wild Calcutta, Madang and Malaccensis and cultivars Lidi, Sinwobogi, Tjau Lagada, Tuu Gia and Heva and hybrids M-48, M-53, M-61, F2P2 and F3P4) (Tables 6 and 7) used in the breeding program. Also allowed the identification of cultivars and hybrids with other ploidy, good agronomic characteristics and/or resistance/tolerance to pests and diseases such as the (AAB) cultivar's Pacovan, Prata Anã, Caipira and Thap Maeo and of the (AAAB) hybrid FHIA-18, which is already recommended to farmers, and two (AAAB) SH3640 and FHIA 21 that will be recommended.

Conventional Genetic Breeding

The banana genetic breeding program has the objective of developing productive genotypes resistant to yellow and black sigatocas and to Panama disease, with reduced stature by means of (AA) diploids improved with commercial

triploids crossings, and by evaluating and selecting new tetraploid varieties in different banana producing regions in the country. The new hybrids produced are also being evaluated for tolerance to nematodes and weevil borer (Silva et al., 1998a).

(AA) Diploid Genetic Breeding

The production and evaluation of diploids in Brazil started in 1983. In its initial phase (1983-87), there were basically wild species such as *M. acuminata* (subspecies banksii, burmanica, malaccensis, microcarpa and zebrina) and cultivars such as Heva, Lidi, Sinwobogi, Tjau Lagada and Tuu Gia (Tables 6 and 7). The first hybrids originated from crossings between these genotypes and presently all diploids employed in the program are improved hybrids which are productive and resistant to diseases. Pollen from these (diploid) genotypes are used for self fertilization and fertilization of commercial cultivars.

The synthesized hybrid is agronomic evaluated in two phases (Silva et al., 1998a). Resistance to Panama disease is evaluated according to the method proposed by Cordeiro et al. (1993a), and resistances to yellow and black Sigatoka, according to INIBAP (1994). Tables 8, 9 and 10 show the main characteristics of 31 diploid hybrids selected during the period from 1995 to 2000.

Generation of tetraploids from triploids

Banana tetraploid hybrids (AAAB) are obtained by means of crossing (AA) diploids with triploids (AAB) cultivars (Prata and Maçã types). Embryo culture has been employed to increase the rate and germination uniformity of tetraploid seeds, and to recover at the same time a greater number of hybrids.

In the beginning of tetraploid production, in 1983, male genitors, wild diploids and the available diploid cultivars were used. Among these, 'Lidi' was the most commonly used cultivar due to its pollen efficiency. Subsequently, a series of promising hybrids in terms of size and fruit quality was generated from the male genitor M-53. Nowadays, hybridization has been done with the 31 diploid hybrids selected, using average to low stature diploids in crossings with tall plants ('Pacovan' and 'Prata Comun'). Hybrids with desirable characteristics independent from the stature are selected in crossings with 'Prata Anã'.

Breeding involving cultivars of the 'Maçã' type started in 1993. In obtaining and evaluating tetraploids of this kind, it must be considered that the 'Maçã' cultivar presents problems with seed output and that the program based itself on the triploid variety Yangambi n° 2, which grows fruits with flavor similar to 'Maçã', but with reduced number of seeds when pollinated with diploids

Table 6 - Characteristics of diploid genotypes (AA) of banana plants used in the initial phase of the banana breeding program. *Embrapa Mandioca e Fruticultura*, Cruz das Almas, BA, 1993.

Genotype	Height of plant	Number of finger/bunch	Length of the fingers (cm)	Disease reaction ^{1/}		
				Panama disease	Yellow Sigatoka	Black Sigatoka
Calcuta	Low	120	8	R	R	R
Madang	High	130	12	R	MR	-
Malaccensis	Low	170	8	-	R	-
Lidi	Low	90	11	R	R	MR
Sinwobogi	Medium	100	10	-	S	-
Tjau Lagada	High	180	9	R	S	MR
Tuu Gia	Medium	70	18	R	R	R
Heva	Medium	60	17	S	MR	MR

Source: Dantas et al. (1993).

^{1/} R: Resistant; MR: Moderate Resistance; S: Susceptible.

Table 7 - Characteristics of banana (AA) diploid hybrids introduced in 1995. Embrapa Mandioca Fruticultura, Cruz das Almas, BA, 1995.

Genotype	Height of plant	Number of finger/bunch	Length of the fingers (cm)	Disease reaction ^{1/}		
				Panama disease	Yellow Sigatoka	Black Sigatoka
M-48	High	140	18	R	R	MR
M-53	High	170	16	R	R	MR
M-61	Medium	180	16	R	R	-
F ₂ P ₂	Medium	96	12	-	R	-
F ₃ P ₂	Medium	80	13	-	R	-

Source: Carvalho (1996).

^{1/} R: Resistant; MR: Moderate Resistance; S: Susceptible.

(Shepherd et al., 1994). Dozens Maçã type hybrids, some tolerant to the Panama disease and with excellent fruit flavor have been produced. Nine of these genotypes are being presently

evaluated in Petrolina, in the state of Pernambuco and Janúba, in the state of Minas Gerais, Brazil.

From 1998 to 2001, Pioneira, PV03-44, JV03-

Table 8 - Characteristics of (AA) diploid hybrids selected in Cruz das Almas, BA, in the period of 1990 to 1995.

Code ^{1/}	Height of plant (m)	Number of fingers		Length of figers		Fertility		Resistance ^{2/}		
		Med.	Max.	Med.	Max.	Female	Male	PD	YS	BS
0116-01	3,0	137	215	11,5	14	4	2	-	R	R
0304-02	2,8	105	161	10,9	13	2	2	-	R	R
0323-03	2,9	101	168	14,6	15	2	1	-	R	R
0337-02	2,5	97	126	12,8	15	2	2	-	R	R
1304-04	3,5	152	228	11,5	14	3	3	-	R	R
1304-06	3,1	155	216	12,6	14	4	2	-	R	R
1318-01	2,6	120	125	13,0	15	4	4	-	R	R
1319-01	2,8	218	230	10,5	15	2	3	R	R	-
1741-01	2,6	94	130	13,5	14	2	2	-	R	-
2803-01	1,8	84	120	13,9	18	1	2	R	R	R
4223-03	2,6	89	134	12,6	16	2	2	-	R	R
4223-06	3,2	104	134	13,3	18	2	2	-	R	R
SH3263	2,1	112	142	13,0	16	2	4	-	R	R
TH03-01	2,3	96	139	13,7	19	2	3	R	R	R

^{1/}The two first numbers correspond the female genitor, the following numbers correspond the male genitor and the last numbers are the selection codes. The numbers between parenthesis correspond to first (I) e second (II) cycles.15: Madu; 41:0304 (03: Calcutta x 04: Madang); 42: M 53 (Hybrid selected in Jamaica); 52: Kumburg; 54: 0104 (Hybrid - 01: Borneo x 04: Madang); 58: 0305 (Hybrid - 03: Calcutta x 05: Pahang); 73: Khai; 79: 2803 (28: Tuu Gia x 03: Calcutta);

^{2/}Notes based on disease infection at flowering and at harvest of bunch. Number 1 correspond high to susceptability and 8 to high resistance. First number obtained at flowering and second at harvest.

Table 9 - Characteristics of (AA) diploid hybrids selected in Cruz das Almas, BA, in the period from 1997 to 1998.

Code ^{1/}	Height of plant (m)	Number of fingers		Length of fingers		Yellow Sigatoka Resistance ^{2/}
		Medium	Maximum	Medium	Maximum	
4252-03 (I)	1,7	51	104	6	9	8/8
(II)	2,2	108	149	8	9	8/8
4252-04 (I)	1,7	77	90	9	12	8/8
(II)	2,2	87	122	10	11	8/8
4279-06 (I)	2,6	80	98	12	14	8/8
(II)	2,6	82	133	14	14	8/8
7341-03 (I)	1,7	109	150	12	14	8/7
(II)	2,2	157	203	11	13	8/7
4215-02 (I)	2	80	106	8	11	8/8
(II)	2,1	86	124	9	14	8/9
4154-01 (I)	1,7	76	140	12	14	7/5
(II)	2,1	117	156	13	14	7/5
4154-06 (I)	3,6	140	145	10	12	8/8
(II)	3,7	160	165	10	12	8/8
4154-08 (I)	2,0	94	131	14	16	7/4
(II)	2,5	109	160	12	15	7/5
5854-03 (I)	2,5	140	164	12	13	8/7
(II)	3,0	116	185	10	11	8/8

^{1/}The two first numbers correspond the female genitor, the following numbers correspond the male genitor and the last numbers are the selection codes. The numbers between parenthesis correspond to first (I) and second (II) cycles. 12:Lidi; 41: 0304 (03: Calcutta x 04: Madang); 42: M 53 (Hybrid selected in Jamaica); 49: M48 (Hybrid selected in Jamaica); 50: M61 (Hybrid selected in Jamaica); 56: 0301-02 (03: Calcuttax 01: Boneo; 73: Khai; 79: 2803 (28: Tuu Gia x 03: Calcuta); 80: 0504-01 (05: Pahang x 04: Madang); 85: 1503-01 (15: Madu x 03: Calcuta);

^{2/}The notes are based in the female and male fertility, 1 correspond to no production of pollen and/or seed and 5 correspond to a large quantity produced. ^{3/} Notes based on disease infection at flowering and at harvest of bunch. Number 1 corresponds to high susceptibility and 8 to high resistance. First number obtained at flowering and second at harvest.

Table 10 - Characteristics of (AA) diploid hybrids selected in Cruz das Almas, BA, in the period of 1999 to 2000.

Code ^{1/}	Height of plant (m)	Number of fingers		Length of fingers		Fertility ^{2/}		Yellow Sigatoka Resistance ^{3/}
		Med.	Max.	Med.	Max.	Female	Male	
4279-10 (I)	1,83	66,8	90	9,5	10	1,25	2,0	8 / 8
(II)	2,14	112,5	163	12	16	1,5	2,0	8 / 8
4279-13 (I)	2,00	77,8	100	17,6	20	1,0	2,2	8 / 7
(II)	2,82	84,8	121	16,4	18	1,0	2,0	8 / 8
5680-01 (I)	1,74	59,8	90	7,6	12	3,2	3,0	8 / 8
(II)	2,48	107,8	168	8,4	11	3,2	2,8	8 / 7
4249-05 (I)	2,90	103,4	115	17,3	19	1,4	2,0	8 / 7
(II)	3,40	113,4	165	15,6	19	2,0	2,0	8 / 8
4249-04 (I)	2,94	90,8	126	19,6	23	1,8	5,0	8 / 7
(II)	3,50	153	166	20,5	23	2,0	2,5	8 / 7
7341-01 (I)	1,46	70	90	5,8	7	2,5	2,0	8 / 8
(II)	2,06	124,6	180	9,4	12	2,4	2,0	8 / 8
5012-02 (I)	2,06	102	145	11	13	1,8	2,0	8 / 7
(II)	2,08	104,8	146	11	12	1,8	2,25	8 / 7
4285-02 (I)	2,30	120,5	130	15,3	16	1	2	7 / 7
(II)	3,50	100,0	100	14,0	14	1	2	7 / --

^{1/}The two first numbers correspond the female genitor, the following numbers correspond the male genitor and the last numbers are the selection codes. The numbers between parenthesis correspond to first (I) and second (II) cycles. 12:Lidi; 41: 0304 (03: Calcutta x 04: Madang); 42: M 53 (Hybrid selected in Jamaica); 49: M48 (Hybrid selected in Jamaica); 50: M61 (Hybrid selected in Jamaica); 56: 0301-02 (03: Calcuttax 01: Boneo; 73: Khai; 79: 2803 (28: Tuu Gia x 03: Calcuta); 80: 0504-01 (05: Pahang x 04: Madang); 85: 1503-01 (15: Madu x 03: Calcuta);

^{2/}The notes are based in the female and male fertility, 1 correspond to no production of pollen and/or seed and 5 correspond to a large quantity produced; ^{3/} Notes based on disease infection at flowering and harvest of bunch. Number 1 correspond to high susceptibility and 8 to high resistance. First number obtained at flowering and second at harvest.

15, YB42-21, ST42-08, PC42-01, ST12-31, FHIA-01, SH3640, FHIA-18, PV42-142, PV42-129, PV42-85, PV42-01, PV42-68 and PV42-53 tetraploid hybrids, from the AAAB genomic group, FHIA-03, from the AABB genomic group, Bucanner, Ambrosia and Calypso, from the genomic group AAAA, Nam, Caipira and Grande Naine cultivars, from the AAA genomic group, Prata Anã, Pacovan, Prata Comun, Thap Maeo and Figue Pomme Naine, cultivars from the AAB genomic group, and 'Ouro da Mata', from the AAAB genomic group, were evaluated, in two cycles, in Cruz das Almas, Bahia, Brazil. The reaction from the majority of the genotypes to the black Sigatoka was also evaluated in Manaus, in the state of Amazonas, in the same period (Table 11).

All the evaluated hybrids were superior or equivalent to its respective parent regarding bunch production. However, hybrids from the Pacovan variety (PV03-44, PV42-142, PV42-129, PV42-85, PV42-81, PV42-68 and PV42-53) exceeded its respective parent, with the exception of the PV03-44, which was equivalent. The Prata Anã hybrids (Pioneira, FHIA-01, FHIA-18 and SH3640) were superior to the their genitor, with the exception of the Pioneira, that was equivalent. The PC42-01 hybrid performed better than its genitor, the Prata Comun variety. The JV03-15 a 'Prata Java' hybrid and the ST42-08 and ST12-31 hybrids, from the Prata São Tomé variety, exceeded the Prata Comun, their equivalent genitor, while the YB42-21 (hybrid from Yangambi nº 2 cultivar) performed better than the Figue Pomme Naine, variety with same type of the genitor of the referred hybrid (Table 11).

These results show that, after being adequately tested and approved in other environments, all the hybrids evaluated by the program had the potential for release as varieties at the local and national level.

Evaluation of cultivars and hybrids in different ecosystems

Since the beginning of the banana breeding program, the genotypes selected in Cruz das Almas, Bahia, Brazil, have been sent to the diverse regions in Brazil to be agronomic evaluated for fruit quality. Experiments with banana

improved genotypes were already being conducted in Una - BA, Petrolina - PE, Manaus - AM, Rio Branco - AC, Alfredo Chaves - ES, Janaúba - MG, Belém - PA and Bacabal - MA. These evaluations resulted in the recommendation of the PA12-03 (Pioneira) hybrid and the Caipira and Thap Maeo varieties (Silva et al., 1996; 1998a).

More recently the Pioneira, PV03-44, Nam, Caipira, FHIA-01, FHIA-18, SH36-40, Grande Naine and Prata Anã genotypes were evaluated in the following regions: a) Southeast: Piracicaba, Registro and Brotas in the state of São Paulo; Lavras, Paracatu, Caratinga, Viçosa and Janaúba, in the state of Minas Gerais state; b) South : Maquiné, in the state of Rio Grande do Sul and Itajaí, in the state of Santa Catarina ; c) Center-West : Cuiabá and Guarantã do Norte, in the state of Mato Grosso; d) Northeast: Cruz das Almas, Guanambi, Juazeiro, Casa Nova and Ilhéus, in the state of Bahia, and São Vicente Férrer, in the state of Pernambuco . Table 12 shows the results of the evaluations in five localities with the following conclusions:

1. 'Grande Naine', genotype of the Cavendish subgroup, showed the lowest stature and, together with the hybrid SH3640, was the most productive in all environments.
2. The FHIA-01, FHIA-18 and SH3640 hybrids exceeded, in productivity, the 'Prata Anã' variety (female genitor), in all environments.
3. FHIA-01 and the FHIA-18 also exceeded 'Prata Anã' in number of fruits per bunch, while the SH3640 exceeded in fruit length, in all the environments.
4. Pioneira, which is also a 'Prata Anã' hybrid, was, in the first cycle, the earliest genotype in all environments.

These results, associated with the information on fruit quality and resistance to disease, led to the recommendation of the FHIA-18 hybrid in December 2000. With the good performance of the SH3640, in Lavras, southern Minas Gerais, Epamig (Minas Gerais State Research Company) and Embrapa Cassava and Fruit Crop decided to recommend it in 2001.

Through collaborative research developed in farming areas in the states of Bahia (Três Pancadas/Ibicaraí, Cocão/Wenceslau Guimarães and Una) and Pernambuco (Siriji/São Vicente Férrer) 11 tetraploid hybrids were evaluated with ten AAAB of the

Table 11 - Resistance to black Sigatoka, means and standard errors of the finger weight (kg), number and length of finger (cm) in two production cycles of 29 banana plants genotypes. Cruz das Almas - BA, 2001.

Genotypes	Yellow Sigatoka reaction ¹	Agronomics characteristics					
		Weight of Bunch (kg)		Number of fruits per bunch		Length of fruits (cm)	
		Cycle I	Cycle II	Cycle I	Cycle II	Cycle I	Cycle II
FHIA 18	R	25,2±2,1	25,6±10,0	147,5±11,0	136,0±23,0	17,9±1,7	17,5±2,3
Thap Maeo	R	21,6±3,7	28,7±6,0	207,3±37,2	233,0±34,2	12,0±0,9	14,1±1,6
FHIA-01	R	22,0±2,5	21,8±3,4	143,0±15,1	147,0±15,0	17,0±1,3	18,4±0,6
Grnade Naine	S	19,2±4,6	22,7±5,9	134,0±16,8	152,0±38,0	16,5±2,9	19,2±1,7
SH3640	S	18,5±3,3	25,1±5,6	110,0±12,6	128,0±18,9	16,5±1,7	19,6±1,3
ST12-31	S	17,0±3,9	29,9±13,6	107,0±11,8	129,0±30,9	15,0±3,6	18,9±2,8
Bucaneiro	-	17,2±3,5	29,8±6,6	119,0±11,1	173,0±32,0	17,5±2,1	20,3±1,7
PV42-53	R	17,9±2,3	21,7±3,3	97,9±12,4	102,0±23,6	17,2±1,1	18,6±1,2
PC42-01	R	16,0±3,6	24,3±7,2	88,0±11,0	105,0±3,7	17,0±1,4	18,0±2,7
PV42-68	R	16,9±2,5	29,6±5,5	90,1±13,6	120,0±14,9	17,7±1,7	20,4±1,4
PV42-81	R	16,3±1,8	22,8±6,8	88,6±10,5	109,0±21,4	18,4±1,2	20,3±1,6
PV42-129	S	14,8±2,9	22,1±7,8	105,3±15,2	125,0±29,5	17,1±1,5	18,9±2,2
YB42-21	-	11,3±6,3	22,7±6,0	70,0±20,5	107,0±21,9	14,8±1,1	16,6±1,2
Ambrosia	-	12,5±4,7	30,0±5,7	119,5±25,3	172,0±24,5	20,0±3,1	20,3±2,3
FHIA-18	R	13,8±2,7	21,1±4,4	123,0±14,7	147,0±10,7	15,0±3,8	15,8±1,8
PV42-142	R	15,1±1,1	22,2±3,2	92,5±13,9	98,0±14,1	17,1±1,3	20,3±1,6
PV42-85	R	14,2±1,2	21,5±5,6	87,9±11,2	115,0±27,6	16,3±1,5	18,1±2,4
Pacovan	S	13,0±2,3	16,2±5,1	94,8±12,5	93,0±15,6	15,9±1,6	17,4±1,7
Calypso	-	9,8±4,5	25,7±8,4	74,0±21,3	147,0±35,5	16,0±4,6	19,2±2,4
ST42-08	R	11,0±2,2	21,2±3,3	87,0±21,1	99,2±14,3	15,0±1,5	20,0±3,4
Prata Anã	S	11,4±1,3	15,0±4,0	109,0±14,4	142,0±24,1	12,8±0,8	13,9±1,7
Ouro da Mata	S	10,1±1,7	14,7±3,6	96,9±11,4	116,0±24,9	16,1±5,4	14,2±1,0
Figue P.Naine	-	10,1±1,6	14,9±3,1	114,0±13,6	11,0±11,2	11,5±1,3	13,5±1,7
Caipira	R	9,1±2,4	19,4±5,1	112,0±21,3	162,0±40,4	16,8±4,6	13,6±1,6
Prata Comum	S	9,1±2,1	13,6±4,2	100,5±9,1	110,0±14,0	11,5±1,1	14,1±1,4
PV03-44	S	9,6±1,5	16,1±3,5	97,0±8,2	103,0±9,3	13,4±1,1	15,7±1,9
Nam	-	8,4±1,6	19,4±4,3	93,0±12,0	148,0±32,4	11,5±0,6	14,3±0,9
Pioneira	S	8,1±1,2	14,2±2,4	96,2±7,4	106,0±12,7	12,4±0,7	15,1±1,6
JV03-15	S	4,2±2,1	19,0±4,8	70,0±16,8	121,0±15,0	9,3±3,2	15,9±2,0

¹ The black Sigatoka evaluation was carried out at Embrapa Amazônia Ocidental, Manaus, Amazonas State; R: resistant and S: susceptible.

'Prata' type (PV03-44, PV42-68, PV42-81, PV42-85, PV42-142, PV42-143, PC42-01, ST12-31 and ST42-08) obtained at Embrapa Cassava and Fruit Crop and the SH3640 introduced from Honduras, and one AABB of the Bluggoe type (FHIA-03) also introduced from Honduras. Either the local Pacovan or the Prata Comun cultivar was used as control.

All the genotypes were resistant to yellow Sigatoka, except the SH3640. The Pacovan hybrid PV42-68 was selected in three units, Cocão/Wenceslau Guimarães, Una and São Vicente Férrer. Its average bunch weight was 18,8 kg, 15,9 kg and 19,3 kg, respectively ranking, first, second and third in the preference of farmers. This genotype was not evaluated in Ibicaraí, BA; where it could probably have been selected, considering the superior characteristics listed by the farmers from the units where it was evaluated.

The ST12-31 hybrid from Prata São Tomé ranked first in Ibicaraí, BA and São Vicente Férrer, PE, where it produced bunches of 14,7 kg and 23,7 kg, respectively. Although susceptible to Panama disease, according to evaluations carried out in Cruz das Almas and in Siriji in the Farm Novo Mirim in the state of Pernambuco, the PC42-01 hybrid from the 'Prata Comum' was selected in all the units.

Fruit flavor and size together with its resistance to yellow Sigatoka were the most important characteristics, according to farmers, for the selection of a hybrid. Bunch production, although important, was not considered a decisive factor in the selection.

The PV42-68, the PV42-142, the ST12-31 and the PC42-01 were selected by farmers in Bahia and Pernambuco as the most promising genotypes (Silveira, 2000). Based on the behavior of the genotypes and their resistance to Panama disease and to yellow Sigatoka, in Cruz das Almas, and resistance to black Sigatoka evaluated in Manaus, AM, the PV42-68 and

PV42-142, were selected as the best genotypes. PV 42 68 hybrid was recommended in 2001.

Non Conventional Breeding

Although Matsumoto et al. (1999) have shown the viability of *in vitro* selection of mutants from the resistant to *Fusarium* Maçã cultivar, and despite the attempts of the Nuclear Center for Energy in Agriculture at the University of São Paulo (CENA-USP), and the Pernambuco Research Company jointly with Federal University of Pernambuco (IPA/UFP) to develop resistance to Panama disease for the same cultivar using radiation, no 'Maçã' mutant with reasonable agronomic characteristics and resistant to the Panama disease was identified.

Using the ethylmethanesulphate mutagenic, by means of electric fusion somatic hybridization, hybrids from the (AAB) Maçã cultivar with the (AA) diploid 'Lidi' were obtained (Matsumoto et al., 1998). The results although promising are still preliminary.

Presently, Embrapa Genetic Resources and Biotechnology is working towards the production of transgenic bananas. Within this context, a system of embryogenic cells in suspension has already dominated the Maçã (AAB) cultivar, and transgenic plants from this cultivar have already been obtained by transformation mediated by biobalistics, expressing the gus gene, the marker genes nptII and Ahas, plus the gene of interest Magainin2, that expresses peptid with an antimicrobe activity (Morais et al., 1999a; 1999b; 1999c; 2000). One of the main objectives of the Biotechnology research and development applied to the genetic breeding of bananas at Embrapa is to increase the number of cultivars in which the system, cultivation and transformation of embryogenic cells in suspension, with subsequent regeneration of transgenic plants dominate. Consequently, results from this research aim at offering the conventional genetic breeding program at Embrapa Cassava and Fruti Crop new possibilities for the Brazilian banana germplasm development.

Table 12 - Means (m) and standard errors (s) of the plant height (PH) in centimeter, bunch weight (BW) in kilogram, number of fruit by bunch (NF), length of finger (LF) in centimeter and production cycle period (PC) in days of 29 banana plants genotypes. Cruz das Almas - BA, 2001.

Genotypes	Traits									
	PH		BW		NF		LF		PC	
	n	s	m	s	m	s	m	s	m	s
Lavras-MG										
Pioneira	235,0	±3,3	5,1	±0,7	73,3	±1,5	13,7	±0,4	491,1	±12,4
Caipira	305,0	±2,9	6,8	±0,7	98,0	±3,7	13,3	±0,6	702,0	±14,2
SH3640	236,0	±2,8	10,8	±0,9	77,1	±1,3	16,7	±0,4	612,0	±13,3
FHIA-01	231,0	±2,9	9,4	±0,8	90,0	±1,8	15,6	±0,4	528,6	±12,9
FHIA-18	260,0	±2,9	7,3	±0,8	88,6	±1,8	14,3	±0,4	522,6	±12,6
Prata Anã	229,0	±3,1	6,0	±0,7	80,6	±1,6	13,9	±0,3	513,6	±12,9
G. Naine	195,0	±2,0	12,9	±0,9	101,9	±5,5	17,4	±0,6	600,6	±13,8
Nam	241,0	±2,2	5,5	±0,6	73,2	±1,5	13,3	±0,3	591,3	±13,3
PV 03-44	325,0	±3,9	4,1	±0,6	62,6	±1,4	13,2	±0,5	577,8	±12,9
Viçosa-MG										
Pioneira	219,0	±10,9	9,0	±1,9	90,0	±13,0	15,5	±1,7	418,3	±9,1
Caipira	240,0	±42,2	11,8	±4,6	109,0	±26,0	16,8	±2,7	652,7	±104,6
SH3640	262,0	±14,1	13,2	±2,2	111,0	±15,0	19,9	±3,1	466,9	±29,7
FHIA-01	254,0	±23,1	13,1	±2,9	120,0	±18,0	18,8	±2,4	483,8	±50,4
FHIA-18	240,0	±18,8	11,4	±2,2	136,0	±16,0	16,4	±1,7	434,0	±15,6
Prata Anã	239,0	±10,1	6,0	±1,9	112,0	±12,0	14,8	±1,9	474,0	±16,7
G. Naine	205,0	±25,0	15,8	±3,4	127,0	±22,0	22,8	±8,5	521,3	±65,9
Nam	257,0	±29,0	9,4	±1,6	109,0	±17,0	15,3	±1,3	503,8	±62,7
PV 03-44	292,0	±14,4	6,4	±1,7	87,0	±13,0	15,1	±3,1	477,0	±21,4
Cruz das Almas-BA										
Pioneira	213,5	±32,3	8,1	±1,2	96,2	±7,4	12,4	±0,7	340,1	±16,8
Caipira	186,0	±26,8	9,1	±2,4	112,0	±21,3	16,8	±4,6	386,5	±49,4
SH3640	247,5	±33,1	18,5	±3,3	110,0	±12,6	16,5	±1,7	355,6	±10,2
FHIA-01	230,0	±16,2	22,0	±2,5	143,0	±15,1	17,0	±1,3	383,6	±14,1
FHIA-18	171,5	±37,9	13,8	±3,6	123,5	±15,1	15,0	±4,0	388,8	±61,2
Prata Anã	225,0	±17,1	11,4	±1,3	109,0	±14,4	12,8	±0,9	402,5	±20,1
G. Naine	178,5	±19,5	19,2	±4,6	134,0	±16,8	16,5	±3,0	376,0	±34,2
Nam	181,5	±19,7	8,4	±1,6	93,0	±12,0	11,5	±0,6	377,6	±26,3
PV 03-44	261,0	±17,1	10,8	±1,4	106,0	±8,2	13,8	±1,1	368,5	±22,3
Guanambi-BA										
Pioneira	205,4	±14,8	9,1	±1,8	95,2	±14,7	15,6	±1,3	309,7	±12,1
Caipira	250,2	±10,2	14,6	±1,7	158,4	±15,3	14,1	±1,1	358,6	±8,1
SH3640	261,3	±12,0	17,5	±2,0	115,4	±9,5	19,7	±1,6	369,8	±8,6
FHIA-01	238,1	±12,5	15,8	±2,7	124,4	±14,7	17,8	±1,3	382,3	±11,8
FHIA-18	241,1	±16,0	14,7	±2,7	131,5	±18,7	18,4	±1,4	359,5	±14,1
Prata Anã	252,4	±7,7	12,7	±2,8	108,5	±14,9	17,2	±1,7	372,1	±15,8
G. Naine	233,0	±9,2	30,3	±3,9	167,3	±14,8	23,9	±1,5	340,0	±7,3
Nam	232,5	±10,5	9,5	±1,4	101,9	±6,2	13,8	±1,1	336,7	±10,1
Jaíba-MG										
Pioneira	198,2	±18,5	14,2	±1,5	95,8	±8,8	14,2	±12,0	324,6	±13,4
Caipira	310,4	±12,4	23,9	±3,3	181,4	±23,0	11,9	±6,2	379,9	±75,9
SH3640	290,1	±13,1	36,9	±4,1	137,7	±15,6	16,8	±15,3	375,6	±15,3
FHIA-01	269,5	±17,6	33,1	±7,7	156,0	±15,8	16,1	±22,8	354,2	±22,8
FHIA-18	247,7	±19,8	26,7	±3,4	144,6	±14,6	15,6	±12,0	350,3	±12,0

^{1/}The black Sigatoka evaluation was carried out at Embrapa Amazônia Ocidental, Manaus, Amazonas State; R: resistant and S:susceptible.

CONCLUSION

1. Microsporogenesis and macrosporogenesis occur normally in seed productive wild banana diploids, although occasional abnormalities in the meiotic pairing are observed due to the synapsis problems of the pair of chromosomes. Meiotic anomalies are more frequent in parthenocarpic diploids, resulting in high levels of sterility. However, the parthenocarpic plants are not completely sterile.
2. Parthenocarpy is an independent phenomenon of the gamete sterility resulting from the action of three dominant genes subjected to the action of modifiers. It is not associated with polyploidy, since the parthenocarpy is present in the diploids.
3. There are three main conditions regarding the origin of megaspores in banana with a subsequent development of an octa nucleated embryonic sac, with meiosis as the predominant process. The other behavior of mother cells in the embryonic sac are due to the production of megaspores and embryonic sacs with the number of maternal chromosomes or the production of megaspores and embryonic sacs with the maternal chromosome number duplicated, both as a result of the absence chromosomes reduction in meiosis.
4. The female sterility of the existing cultivars is normally not absolute and the majority, at all levels of ploidy, is able to produce seeds in controlled pollen application. Fertilization of non reduced diploids or triploid embryonic sacs, with haploid A pollen, supplies a practical base for hybridization programs aiming at new triploid and tetraploid genotypes.
5. In the production of new triploid or tetraploid banana cultivars, the methodologies based in crossings offer better perspectives for spontaneous or induced mutations.
6. Among the hybridization methodologies, the production of tetraploid hybrids from triploid cultivars is the fastest approach for obtaining new productive cultivars resistant to main diseases and pests, and acceptable in the consumer markets.
7. Complementary studies on the production of triploids from diploids or by means of crossings between tetraploids and diploids, can offer an alternative methodology.
8. Generally, the AA diploid germplasm is fundamental to genetic breeding independently from the hybridization methodology adopted in the production of polyploids.
9. The application of biotechnology associated or not with hybridization will be of great value to

- banana genetic breeding in the production of hybrids from partial or completely sterile plants.
10. (AA) diploid banana breeding for productivity and resistance to diseases is a reality, as the number of genotypes of this nature generated by Embrapa (Brazil), Fhia (Honduras) and Cirad-Flhor (France) can attest.
 11. Breeding by hybridization has generated commercial tetraploid hybrids from the Prata' (AAAB), 'Gros Michel' (AAAA), 'Plantain' (AAAB) and 'Bluggoe' (AABB) types', with good agronomic characteristics and resistance to diseases.
 12. Banana breeding by hybridization of the Maçã type for resistance to Panama disease is a viable practice.
 13. Non conventional breeding using mutation and somatic hybridization, although widely used in recent times, has not been efficient in the production of mutants and/or somatic hybrids with good agronomic characteristics.

RESUMO

Programa de melhoramento genético da banana na Embrapa

São descritos os principais fatores relacionados com o melhoramento genético da bananeira: classificação botânica, principais cultivares, origem e evolução da bananeira, sistemas reprodutivos, esterilidade e partenocarpiã, poliploidia, herança de caracteres e herdabilidade. Para solução dos problemas causados por fungos, bactérias, vírus, nematóides e insetos, pelo porte elevado e baixo potencial de produtividade de algumas cultivares, tem-se utilizada a criação de novas variedades resistentes a doenças e pragas, mediante programa de melhoramento genético, que possibilita a obtenção de genótipos superiores, constando das seguintes etapas: formação, caracterização e avaliação de ampla coleção de germoplasma, introdução e seleção de clones, melhoramento por hibridação, melhoramento por mutação, hibridação somática e transformação genética. Como resultados relevantes obtidos no programa pode-se citar: a caracterização morfológica do germoplasma, permitindo a identificação de genótipos promissores e a sua recomendação aos produtores; a obtenção de híbridos tetraplóides resistentes às sigatokas amarela e negra e ao mal-do-Panamá, com porte e ciclo reduzidos e produtivos; o melhoramento

genético de diplóides AA, cujo pólen tem sido usado no melhoramento de cultivares comerciais e dos próprios híbridos diplóides; a avaliação de cultivares e híbridos em diferentes ecossistemas, permitindo averiguar que muitos apresentaram potencial para serem recomendados e lançados como variedades nos âmbitos local e nacional, desde que devidamente testados e aprovados nos diversos ambientes; obtenção de híbridos da cultivar Maçã (AAB) com o diplóide (AA) 'Lidi', mediante o emprego de hibridação somática por eletrofusão, que apesar de promissores são ainda preliminares em relação ao uso como cultivares.

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Received: June 18, 2001;

Accepted: October 11, 2001.