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Problems and prospects of banana breeding in India

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ABSTRACT

Banana breeding programme in India involves maintenance of various genetic resources of banana, of which triploids constitute the maximum share over diploids or tetraploids. RAPD studies conducted in these clones exhibit many distinct genotypes. During a hybridization programme, although many crosses were made, seed set and seed germination were relatively poor in many crosses. Male fertility in banana hybrids could be assessed by pollen output per anther; pollen viability and pollen size, which vary from cross to cross, and also from ploidy to ploidy. Ploidy levels in hybrids are estimated by phenotypic appearance (scoring technique) and confirmed either by stomatal density, size and number of chloroplast per guard cell pair or root tip mitosis. However, flow cytometry appears to be the most reliable method in many disputed cases. Generation of parthenocarpic hybrids depends largely upon selection and utilization of parents with parthenocarpic pedigree in a breeding programme. Evaluation of hybrids and parents indicated the nature of inheritance with respect to plant height and suckering habit but no definite trend could be ascribed to the traits of bunch orientation. Diploid x Diploid breeding approach has led to identification of a superior triploid hybrid, NPH 02-01, while Triploid (with AB) x Diploid approach has led to the development of a promising diploid hybrid H.212 and a triploid hybrid H.96/7 (ABB). Similarly, the Triploid x Diploid breeding programme resulted in development of many potential tetraploids that need further improvement. Innovative breeding approaches through *in vitro* mutation breeding and *in vitro* polyploidazation resulted in the development of many potentially useful variants. Breeding for resistance against biotic stresses such as *Fusarium* wilt and nematodes holds promise in banana, and, biochemical mechanisms for resistance in resistant genotypes / hybrids have been elucidated.

Key words: Banana, breeding, India, problems, prospects

INTRODUCTION

Banana is one of the most important fruit crops of India grown over an area of about 0.49 million hectares, annually producing about 11 million tonnes which account for 44.35 % of the total national fruit production. Poovan (Mysore -AAB), Cavendish cultivars (Robusta and Dwarf Cavendish – AAA), Silk (Rasthali-AAB), Karpooravalli (Pisang awak-ABB) and Virupakshi (AAB), 'Nendran' (Plantain - AAB) and Ney Poovan (AB) are the important cultivars grown commercially in India. As banana production is limited by threat from pests and diseases, banana breeding was also started in India in line with that in other countries such as Honduras, Cameroon, Brazil, etc.

The breeding programme initiated at the then Central Banana Research Station, Aduthurai, during 1949 in Tamil Nadu state, was the earliest among systematic banana improvement efforts in India. The Tamil Nadu Agricultural University (TNAU) at Coimbatore, since its formation in 1971, is vigorously and actively pursuing *Musa* improvement programmes. Similarly, Banana Research Station (BRS), Kannara, Kerala Agricultural University (KAU) is also engaged in banana improvement and is concentrating on the improvement of 'Nendran'. Besides, National Research Centre on Banana (NRCB) (ICAR), Trichy, established in 1994, is also involved in crop improvement in banana. In this paper, problems and prospects of banana breeding in India are highlighted.

Genetic resources in *Musa*

The breeding programme started as early as 1949 at Aduthurai did not yield desirable results. However, a lot of cytogenetical information useful for formulating better strategies for *Musa* improvement programmes was

Table 1. Musa genetic resources available at various centres in India (%)

Genome	NRCB, Trichy	TNAU, Coimbatore	IIHR, Bangalore	BRS, Kovvur (AP)	RAU, Pusa, Bihar	GAU, Gandevi, Gujarat	KAU, Kannara
AA	9	7	15	9	1	5	8
AB	7	10	25	8	2	5	8
BB	6	2	20	1	4	-	2
AAA	14	10	-	16	21	12	12
AAB	25	28	13	30	36	24	40
ABB	25	30	20	28	31	22	21
AAAA	1	1	5	2	2	-	3
Others	13	12	2	6	3	32	6
Number of accessions	942	125	69	51	75	74	121

accumulated (Anon, 1968). Detailed investigations on morphological characters and taxonomic status of South Indian banana were earlier published by Venkataramani (1949) and Jacob (1952). Several South Indian banana were found to be more closely related to *M. balbisiana* than *M. acuminata* as revealed by the metroglyph (Raman *et al*, 1968). Bhakthavatsaulu and Sathiamoorthy (1979) and Sathiamoorthy *et al* (1979) also elaborated upon the genomic status and breeding potential of many South Indian banana. Many centres in India maintain the various genetic resources (Table 1), which reveal that many clones available are triploid, and that too, under 'AAB' category.

As many synonyms exist for each clone and are differently named at various regions, much confusion prevails on the nomenclature of banana clones. In this situation, molecular characterization helps to identify clones unambiguously. Molecular characterization of different clones has been attempted by various authors in India. Jagannath *et al* (2003) characterized many AA and AB diploids using RAPD markers. An overall similarity of 63% was found among the diploid cultivars. Greater diversity was seen within the AA group than within the AB group. Many natural mutants, such as 'Ambala Kadali', 'Erachi Vazhai', 'Thattila Kunnan', 'Vennetu Kunnan' and 'Rasa Kadali' showed significant variation from their parental genotypes (Fig 1).

Rekha *et al* (2004) made an attempt to study the variability between the 17 AB cultivars available at Indian Institute of Horticulture Research, Bangalore by using RAPD markers. Of the 80 primers that were screened, 16

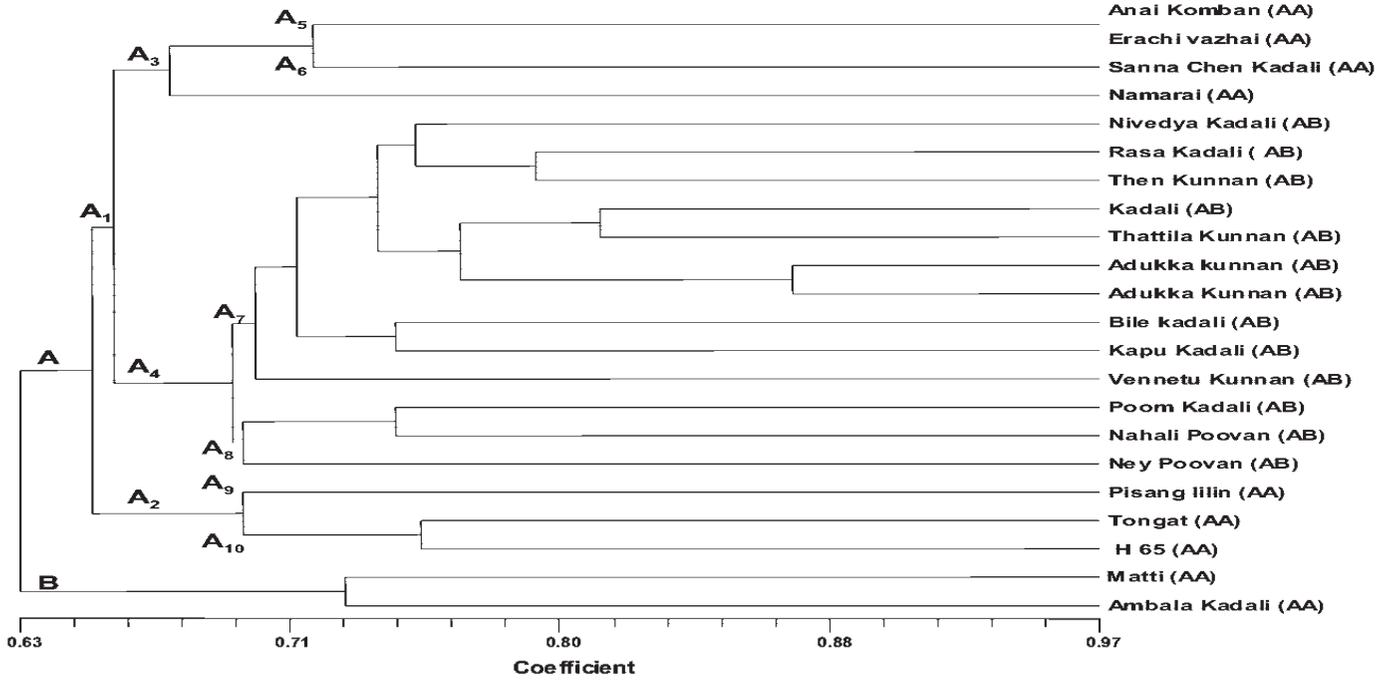


Fig 1. Dendrogram of diploid cultivars of banana

showed polymorphism. Scorable polymorphic bands were analysed using cluster analysis and principal component analysis. Results showed that there were two distinct clusters separating the Ney Poovan types and Kunnan types. Variability, attributed to somaclonal variation and natural mutations, was also observed in each group.

Menon *et al* (2003) assessed genetic diversity in 35 morphologically distinct indigenous banana cultivars representing five genomic groups, viz., AA, AB, AAA, AAB and ABB available at the genebank of Kannara (KAU), employing RAPD analysis and found that the cultivars were grouped in clusters that generally represented the taxonomic classification based on morphological characters.

Cluster analysis among 'B' genome rich accessions of banana at NRCB, Trichy (Annual Report, 2004-05) was done with Bluggoe (ABB), Monthan (ABB) and Peyan (ABB) and two sub-clusters were observed under each type.

Crossing technique

Pollinations are generally carried out between 7.00 and 10.00 A.M. Undehisced anthers from male flowers are collected and twisted gently to force them to dehisce. Using a soft sable hair brush, pollen grains are prised out and smeared gently over the stigmatic surface of those female flowers that open on the day of pollination. Pollinated flowers are covered with a soft-cloth bag.

Seed set and germination

Unlike in other crops, seed set is very low in many varieties of banana. On hybridization the seed yield is reported to be influenced by time of pollination, fertility variations between basal and distal hands, the apical bias within a fruit (Shepherd, 1960) and genomic make up (Simmonds, 1962). Most of the seeds (74.9%) are found in the distal one-third of the fruit bunch, 20.9 % in mid one-third and the rest (4.2%) in the proximal or basal one-third.

Using 'Matti' as the female parent and seven diploids as male parents, 368 crosses were made but only 66 hybrids were obtained (Sathiamoorthy, 1987). Krishnamoorthy (2001) on the other hand crossed 7830 flowers and obtained a total seed yield of 1096 of which 91.5 % were good seeds while the rest were empty floats or bad (as designated by Shepherd, 1960). Among the various combinations such as AA x AA, AB x AA, and 3*x* x 2*x* crosses, he found that, in general, AB x AA crosses yielded more seed compared to the rest of the cross combinations, and, very low seed set in AA x AA crosses. This might be due to the fact that increase in *balbisiana* genome increased seed yield and factors for seed sterility increased with *acuminata* genome.

Climatic factors are also known to decide success of pollination and seed set in banana. Krishnamoorthy (2001) observed that one of the reasons for obtaining relatively higher number of hybrids in his study might be the time of germination i.e., February to March under Coimbatore conditions since, during this season, bright sunny weather and cooler nights prevail. Seeds can be mechanically extracted from ripe fruits. The seeds are soaked in water for a week before sowing in seed pans kept in a mist chamber. Germination is low and erratic, both during winter and warmer months (May-June).

Pollen fertility

Pollen fertility is an important factor for a clone to be used as the male parent in a hybridization programme. Male fertility in banana clones / hybrids is assessed by pollen output per anther, pollen viability and pollen grain size. Studies on pollen production and male fertility status of several clones by Sathiamoorthy (1973, 1987) revealed the diploid AA cultivars Ambalakadali, Erachivazhai, Pisang lilin and Tongat to be more polleniferous than the triploids AAA, AAB and ABB. Krishnamoorthy (2002) assessed male fertility in many newly developed hybrids and found that diploid progenies generally produced good pollen grains with stainability and germinability and were good enough to be considered as potential parents. Further, he also found that lack of pollen production identified in some AB and AA hybrids was due to meiotic aberrations, confirming that male sterility in *Musa* is not exclusive to triploids. Damodaran (2004) assessed male fertility in 'Peykunnan' derived hybrids and found that 'Peykunnan' had poor male fertility but high female fertility, whereas, the reverse was true in the case of pollen parents Pisang Lilin and Erachi Vazhai. Hence, the presence of high female and male fertility in 'Peykunnan' derived crosses might be due to inheritance of these traits from their female and male parents respectively.

Sathiyamoorthy *et al* (1979) and Krishnamoorthy (2002) found no consistence relationship between stainability and germinability values. It is obvious that acetocarmine staining of pollen is not a reliable index of fertility, since acetocarmine stains only the cytoplasm of the pollen grains as living or dead (Vakil, 1958).

Evaluation of male fertility status of bananas based

on pollen size would be of considerable importance, as, the potential of a male parent depends on the quantum of fertile haploid spores it can produce. Dodds (1943) reported, based on Darlinton's (1937) ratio of haploid, diploid and tetraploid spores, that in a banana the pollen diameter of 129.00 µm or less is haploid, 147.60 µm is diploid and 230.00 to 240.00 µm is tetraploid, whereas, Sathiamoorthy (1987) arrived at the mean diameter of haploid, diploid and tetraploid pollen grains as 122.10, 139.20 and 222.22 µm, respectively, under Coimbatore conditions. This brings out the fact, that in banana, pollen diameter is influenced by environment too. Krishnamoorthy (2002) found that diploid hybrids produced 56.65 % pollen grains with a mean diameter of 101.12 µm, and 96.88 % of pollen grains had a diameter of 65.20 to 121.70 µm and these correspond to the haploid size mentioned by Sathiamoorthy (1987). Diploid hybrids / parents produced the maximum percent of haploid pollen grains (Table 2) indicating their potential for use as male parents.

Table 2. Haploid, diploid and tetraploid spores produced (%) by banana clones (based on Darlington's Ratio) (Sathiamoorthy, 1987)

Clone Type	Haploid	Diploid	Tetraploid
Wild species	61.7 – 70.3	–	–
Diploid cultivars	22.1 – 61.0	4.0 – 26.2	–
Triploids (AAA)	7.3 – 32.0	5.8 – 16.5	1.2 – 4.0
(AAB)	10.0 – 20.0	12.0 – 20.0	2.7 – 13.3
(ABB)	4.0 – 28.0	3.5 – 12.0	0 – 4.5
Tetraploids	1.7 – 9.5	11.0 – 28.3	–

Krishnamoorthy (2002) also found that some synthetic triploid hybrids produced more than 50 % haploid pollen grains as; these can be used as male parents in future programmes. He also found that some tetraploid hybrids derived from 'Karpooravalli' as female parent produced more than 90 % diploid pollen grains offering greater scope for use as the male parent in crossing programmes.

Ploidy assessment of hybrids

Studying ploidy levels of hybrids obtained from different cross combinations is a must in banana breeding because of potential production of diploid, triploid, tetraploid, hyperploid and aneuploid hybrids. Ploidy levels are estimated by phenotypic appearance and confirmed either by root tip mitosis or stomatal density, size and number of chloroplast per guard cell pair. Sathiyamoorthy (1973) and Vandenhout *et al* (1995) classified banana clones into diploids, triploids and tetraploids based on stomatal density and stomatal size, respectively, as indicated below:

Ploidy level	Stomatal density Sathiamoorthy (1973)	Stomatal size (mm^2) Vandenhout *et al* (1995)
Diploid	40.0 – 50.0	1250
Triploid	30.0 – 40.0	1250 – 1840
Tetraploid	9.0 – 15.2	1840

Krishnamoorthy (2002), while assessing the ploidy status in his hybrids, found that stomatal density in different ploidy levels did not concur with the reports of Sathiamoorthy (1973). Hence, the range of stomatal density of respective ploidy parents was taken as the criterion for ploidy assessment. Based on this criterion, all the diploid hybrids were found to fall in the range of parental diploids, whereas a few diploids, triploids and tetraploids exceeded the range. Hence, stomatal size as well as number of chloroplast per guard cell pair was also taken as criteria. However, stomatal size in the exceeded diploid, triploid and tetraploids was found to be in the ranges observed in parental diploid, triploid as well as tetraploids. But, in respect of chloroplast number per guard cell pair, most of the diploids fell within the range of diploid parents or triploid parents. Vandenhout *et al* (1995) observed that a high density of small stomata correlated with low ploidy level. Besides stomatal size and density were also influenced by genotype effect within the same ploidy level. Similarly, the chloroplast numbers in guard cell pair was influenced by ploidy level i.e, it increased with increase in ploidy level.

Root tip mitosis study is considered as one of the reliable methods or confirming ploidy status in banana. Krishnamoorthy (2002) carried out root tip mitosis in all the 36 parthenocarpic hybrids to confirm the ploidy of each hybrid assessed by stomatal characters. The ploidy level of 34 hybrids was in accordance with ploidy level assessment based on stomatal characters. One of the hybrids, H-02-01, which had stomatal characters similar to that of tetraploids, had only 22 chromosomes, i.e., a diploid. Another hybrid, H-02-21, did not fall under any of the ploidy ranges of parental cultivars-either triploid or tetraploid-but root mitosis confirmed it as a tetraploid. This serves as a caution to banana breeders to confirm the ploidy of any new hybrid by root mitosis as well.

Flow cytometry analysis is considered as the most superior and reliable method to confirm ploidy status, especially, in the case of most disputed cases, because of its precision, rapidity and it does not require dividing cells. Precision is higher here because of analysis of the nuclear DNA itself, which is least disturbed by environmental factors (Dolezel *et al*, 1997). Young cigar leaves of selected

Table 3. Confirmation of ploidy through flow cytometry

Hybrid / parent	Parentage	Ploidy status determined by		
		morphological scoring	stomatal density	flow cytometry
NPH.02.01	H.201 x Anaikomban	42 (AAB) 3x	55.53 (2x)	AAB (3x)
H.02.08	H.201 x Erachi vazhai	46 (AB)	72.30 (AB)	AAB (3x)
H.03.11	H.02.32 x Pisary Lilin	54 (AABB) 4x	46.05 (AABB) 4x	4x
H.03.13	Peykannan x Erachi vazhai	56 (AABB) 4x	13.16 (AABB) 4x	4x
H.03.15	H.02.32 x Pisang Lilin	42 AAB (3x)	(12.76 (AAB) (3x)	3x

hybrids are analysed for their ploidy level by measuring the size of nuclear genome by this high-throughput method. The cigar leaves were cut using a sharp sterile blade for a length of 15-20 cm, cleaned gently with sterile distilled water and wrapped with partially wetted sterilized Whatman No. 3 filter paper. The samples were then packed in zipped polyethylene covers and sent to the Laboratory of Molecular Cytogenetics and Cytometry, Czech Republic, for ploidy analysis. Flow cytometry ploidy assay involved preparation of suspensions of intact nuclei from small amounts of leaf tissue and the analysis of fluorescence intensity after staining with DAPI. Chicken Red Blood Cell (CRBC) nuclei were included in every sample as an internal reference standard. Ploidy of individual plant was estimated based on the ratio of peaks corresponding to G1 nuclei of *Musa* and CRBC.

Damodaran (2004) employed this method for the first time in banana hybrids in India to confirm ploidy status of some hybrids. Results indicated that two hybrids, viz., NPH-02-01 and H-02-08, of doubtful ploidy status, were confirmed as triploids (AAB), instead of diploids, as evident through stomatal density analysis and morphological scoring (Fig 2, Table 3).

At NRCB, Trichy , too, this method was employed to confirm ploidy status of 36 ambiguous accessions of banana.

Evaluation of hybrids for parthenocarpy

Although success in banana breeding depends on production of seed upon crossing, hybrids should be parthenocarpic to find acceptance as edible banana. Sathiamoorthy (1987) evaluated 66 hybrids and found that only 23 were parthenocarpic and observed that a high degree of parhenocarpy, to an extent, suppresses seed set, although parthenocarpy and fertility are reported to be genetically independent characters. Krishnamoorthy (2002) and Krishnamoorthy and Kumar (2005) subjected all the hybrids to assessment for parthenocarpy by bagging the female flowers. Out of 312 seedlings evaluated, only 36 hybrids were found to be parthenocarpic (Table 4).

Among these 36 hybrids, higher rates (by number) of parthenocarpic hybrids were obtained from diploid x diploid and triploid x triploid crosses.

In crosses between H 201 and AA diploids (i.e., diploid x diploid), many hybrids were found to be non-parthenocarpic by Krishnamoorthy (2002) indicating the

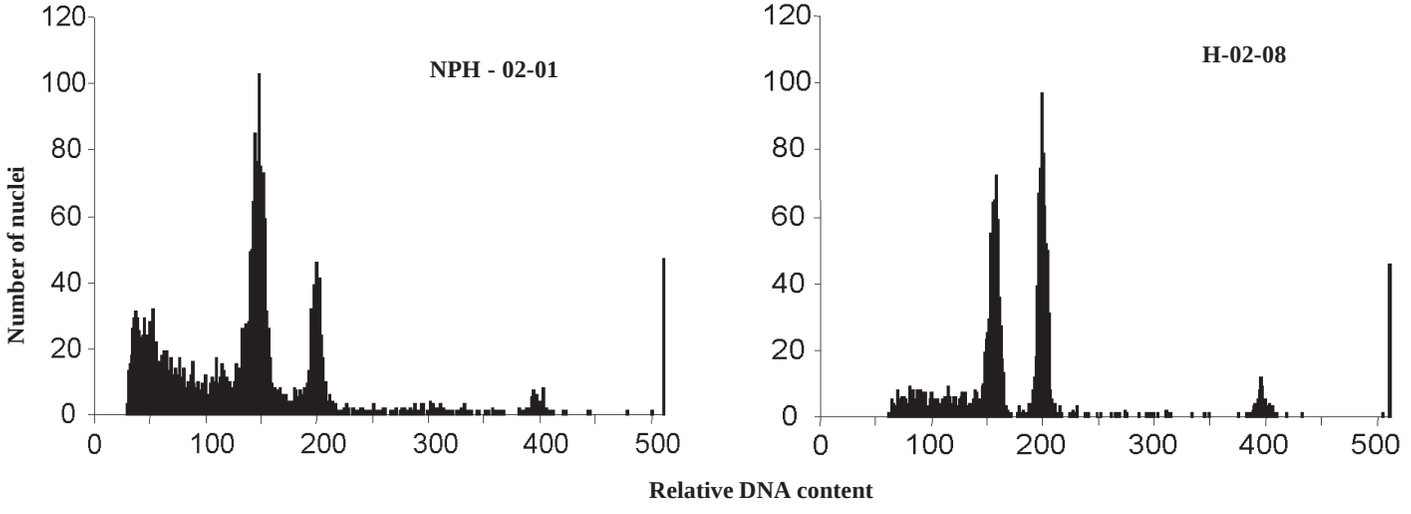


Fig 2. Flow cytometry analysis of selective banana hybrids for ploidy confirmation

Table 4. Ploidy distribution in hybrids obtained from various cross combinations

Name of the cross	No. of hybrids obtained	No. of hybrids in each ploidy level				No. of parthenocarpic hybrids	No. of non-parthenocarpic hybrids
		2x	3x	4x	5x		
Diplod x Diploid	257	196 (76.27)	-	55 (21.40)	6 (2.39)	15 (5.84)	242 (94.16)
Triploid x Diploid	12	-	1 (15.39)	12 (76.92)	-	4 (30.77)	8 (61.54)
Triploid x Triploid	42	1	-	35 (83.33)	6 (16.67)	17 (40.48)	25 (59.52)

Figures in parantheses indicate % hybrid recovery

influence of female parent, which is highly female fertile because of the influence of B genome contained in it. From the pedigree of hybrid H 201, when traced back, it was evident that one of its parents, cv. Barelichina (ABB) used as female parent, had contributed to the non-parthenocarpic nature. In this background, it may be inferred that the parthenocarpic hybrids obtained from crosses involving H 201 and diploid parents, or, in other crosses involving diploid parents, might be due to a series of complementary dominant genes which were derived from edible natural diploids, which are variously heterozygous in their genetic composition for these genes. From the breeding point of view, it is an important consideration, as, parthenocarpic progenies could be readily sorted out in a population from crosses involving these diploid parents.

In the case of triploid x diploid crosses, out of four parthenocarpic hybrids (Table 4), one (H-02-16) was found to be triploid with AAB while all other hybrids were found to be tetraploid with AABB genome. The triploid hybrid could have arisen from an egg containing AB genome from 'Karpooravalli' and one 'A' genome from Pisang Lilin, while, rest of the tetraploids could have arisen from unreduced gametes of ABB combining with one 'A' genome from the male parent.

In the case of triploid ´ triploid crosses (excepting H-02-20 and H-02-24), all were found to be tetraploids with AABB genome, thus again revealing the ability of 'Karpooravalli' to produce unreduced gametes/egg cells to combine with 'A' genome from the triploid (Red Banana / Robusta). Puskaran (1991) also obtained 32 tetraploid hybrids using 'Karpooravalli' as the female parent with Pisang Lilin, indicating the ability of 'Karpooravalli' to produce unreduced egg cell during megasporogenesis.

Damodaran (2004), on the other hand, found that all the hybrids were parthenocarpic. Selection and utilization of parents with parthenocarpic pedigree in the breeding programme might have contributed to enhanced parthenocarpy occurrence as he used all the parthenocarpic hybrids of Krishnamoorthy (2002) for further improvement. It also confirms the role of dominant genes in controlling parthenocarpy (Simmonds, 1953).

Two of the 25 hybrids, viz., NPH-02-01 and NPH-02-02 which were found to be non-parthenocarpic in phase I generation by Krishnamoorthy (2002) were, however, found to be parthenocarpic in phase II evaluation by Damodaran (2004). This suggests the possibility of reversion of non-parthenocarpy to parthenocarpy in some cases. Reversion of gene for parthenocarpy to non-parthenocarpy in the first vegetative generation was earlier reported by Simmonds (1953). This peculiar phenomenon should serve as a caution to banana breeders to carefully consider potential non-parthenocarpic hybrids lest they revert to parthenocarpy in subsequent vegetative generations.

Evaluation of hybrid seedlings

Apart from parhenocarpiness, hybrids need to be evaluated for desirable horticultural traits. The evaluation consists of phase I (seed to harvest stage) and phase II (sucker to harvest stage). The full inherent potential of hybrids cannot be derived from the Phase I as it is seed propagated and, therefore, it takes relatively more time for corms to develop from seeds. Hence, evaluation of the phase II sucker propagated hybrids is essential to assess the plant's fullest potential (Sathiamoorthy, 1987; Krishnamoorthy, 2002).

Krishnamoorthy (2002) observed that hybrids involving dwarf parents, such as H 201 and or Pisang Lilin, were also dwarf. This indicates that the gene for dwarfness was derived either from H 201 or Pisang Lilin. It is also interesting to note that H 201 had one of the parents as Pisang Lilin and, hence, Pisang Lilin is the source of dwarf gene. Ortiz and Vuylsteke (1995) suggested that dwarf hybrids could easily be produced if only dwarf diploid male parents with better horticultural qualities were involved in the breeding programme. The importance of dwarfness as indicated by Simmonds (1966) was the advantage conferred by it in terms of higher yields at high densities, enhanced weed control, reduced sucker pruning, less damage from

Tetraploid H-02-30
Karpooravalli x Red Banana

Height : 490 cm
Girth : 105 cm
No. of roots : 288/mat
Bunch not supported

Fig 3. A tall statured tetraploid hybrid (AABB)

wind and easy harvest. Krishnamoorthy (2002) observed tetraploids to be always very tall (Fig 3). Damodaran (2004) observed the segregation pattern of height of the progenies of H-02-32 (AABB) x Pisang Lilin (AA) which ranged from as dwarf as 149 cm to as high as 411 cm.

This indicated that hybrids segregated for dwarf and tall stature of plants. The possibility of obtaining dwarf hybrids stems from the fact that dwarfism in banana is controlled by a recessive gene (Ortiz and Vuyletske, 1996) which should have been derived from dwarf parent Pisang Lilin. In rest of the crosses, the height of the progenies was over 400 cm, indicating tallness to be dominant and the recessive gene from the 'A' genome of Pisang Lilin could not express itself in tetraploid hybrids.

In *Musa*, suckering behaviour is influenced by apical dominance. Inhibition of lateral bud growth due to growth substances released by the terminal bud has been considered as a limiting factor for perennial productivity (Ortiz and Vuylsteke, 1996). Krishnamoorthy (2002) found that the mean number of suckers per mat in diploid hybrids was more than in triploid or tetraploid hybrids. Even in diploid AA x diploid AA crosses, the number of suckers per mat was more (9.00) than in diploid AB x diploid AA crosses (7.00). Damodaran (2004), on the other hand, found that the number ranged from two to four in diploids, seven to eight in triploids and five to eleven in tetraploids. This indicates that tetraploids of the present cross combination had higher suckering ability than diploids, a trait otherwise not common among diploids. Better suckering ability in tetraploids may be attributed to heterotic vigour manifested in the 3n x 2n cross- combination. The hybrids H-03-07 and H-03-06 produced only one and two suckers, respectively, while the other hybrids from the same parents produced more number of suckers. This might be due to the poor penetration of the "Ad" allele or due to the variable expressivity of the allele. The "Ad" gene has incomplete penetrance, genetic specificity and variable expressivity (Ortiz and Vuylsteke, 1994).

Bunch orientation is an important trait in plantain and banana. Pendulous bunches are more symmetrical than sub-horizontal or oblique and horizontal or erect bunches and are, therefore, better suited for transportation. Studies by Krishnamoorthy (2002) and Damodaran (2004) revealed that inheritance of bunch orientation character is a complex trait and needs further systematic investigation (Table 5).

The duration of hybrids as assessed by days to flowering, filling and harvest varied widely within and between cross combinations in banana. Total duration varied from 268 to 716 days in hybrids (Krishnamoorthy, 2002). Diploids had shorter duration, while tetraploids had a longer duration. Wherever a long duration parent viz., 'Karpooravalli' was involved, hybrids invariably had longer duration. This suggests that to breed varieties for shorter duration, the progenies need to be further backcrossed to shorter duration parents only.

Diploid breeding

Banana breeding is essentially a diploid breeding. The long-held conclusion of Dodds (1943) that progress is dependent on breeding superior diploids and, then, selecting new commercial hybrids from tetraploids derived from crossing these diploids onto 'Gros Michel', was based on

Table 5. Expression of Bunch hang traits in banana hybrids

Name of the hybrid / parent	Genome	Bunch hang	Parent	Bunch hang	Parent	Bunch hang
H.02.027	(AABB)	P	Karpooravalli (ABB)	O	Red banana (AAA)	P
H.02.25	(AABB)	H	Karpooravalli (ABB)	O	Red banana (AAA)	P
H.02.26	(AABB)	O	Karpooravalli (ABB)	O	Red banana (AAA)	P
H.02.06	(AB)	P	H.201 (AB)	H	Anaikomban	O
H.02.09	(AB)	H	H.201 (AB)	H	Pisang 4 lin	H
H.02.11	(AB)	O	H.201 (AB)	H	H.110	H

P : Pseudoutous Orientation, H : Horizontal Orientation, O : Oblique Orientation

Table 6. Potential synthetic diploid hybrids developed at Tamil Nadu Agricultural University using Matti (AA) as female parent

Hybrids	Ploidy level	Height (cm)	Number of Suckers / mat	Bunch weight (kg)	TSS (%)	Duration (days)	Reaction to Sigatoka	Reaction to Burrowing nematode
H 21	2x	302	15.0	105	20.0	339	R	HR
H 59	2x	280	6.1	19.3	26.3	320	R	R
H 65	2x	355	9.3	17.8	19.5	327	HR	R
H 82	2x	301	6.7	8.3	20.5	350	S	-
H 89	2x	301	18.4	12.3	24.0	372	R	-
H 103	2x	303	10.0	16.7	27.1	333	HR	-
H 109	2x	311	17.6	20.7	23.7	369	R	R

HR: Highly Resistant, R: Resistant, S: Susceptible

historical limitations of diploid accessions. These accessions (in all the germplasm collections) are agronomically so inferior that there was little expectation that diploids with adequate bunch sizes could be developed from breeding them. The continuous, primary emphasis in breeding has been to develop diploids with desirable agronomic qualities and, then, to cross theses agronomically improved hybrids with disease-resistant clones to synthesize diploids with combinations of agronomic excellence and disease resistance.

Diploid breeding at TNAU, Coimbatore, primarily involved the use of vars. Anaikomban, Namarai, Erachi vazhai, Pisang lilin and Tongat (all AA) as male parents with Matti (AA) as the female parent (Sathiamoorthy *et al*, 1979, 1988) led to development of potential synthetic diploid hybrids (Table 6). Many of the synthetic diploids have been found to have good resistance to burrowing nematode, Sigatoka leaf spot and *Fusarium* wilt.

Cultivar Matti showed higher female fertility than other diploids which were generally female sterile. A plausible explanation for high seed fertility of Matti could be attributed to its localised cultivation in Kanyakumari district of Tamil Nadu near the foot hills of Southern Travancore wherein, even now, its wild ancestor *M. acuminata* sub sp. *burmanica* could be traced. This wild species is highly female fertile and is self propagated through seeds. Matti would have arisen as a clonal selection for parthenocarpic type from this wild species while probably retaining the seed fertility trait.

Development of potential synthetic diploid hybrid H 201(AB)

Robusta as male parent has been used almost simultaneously and independently in Honduras and India. It readily crossed with both *M. acuminata* and *M. balbisiana*. Hybrid progenies were also obtained when it was crossed with a synthetic tetraploid of AABB genome (Bareli Chinia x Pisang Lilin). Except for one diploid, all the progenies consisted of non-parthenocarpic diploids. The parthenocarpic diploid hybrid, designated as H 201, was of the genome AB and was dwarf, non-polleniferous but highly seed fertile and exhibited high resistance to panama wilt, nematodes and sigatoka diseases (Sathiamoorthy, 1987). Its use in evolving new AAB or ABB forms appears to be worthwhile.

Diploid x Diploid breeding approaches

Although extensive inter-diploid crosses were made at TNAU, Coimbature since 1971, with the primary objective of synthesizing new diploid forms as stated earlier, hybridisation work did not always result in pure diploid progenies. Frequently, due to single and double restitution, occurrence of triploid and penta polyploids was also encountered among hybrids the generated (Table 4). However, production of triploid hybrids through 2x x 2x breeding approaches will be very useful.

Krishnamoorthy (2002) crossed H 201 (AB) x Anaikomban (AA) to develop new triploid combination viz., NPH-02-01 (AAB) (Fig 4) which is a pome type, parthenocarpic, resistant to *Fusarium* wilt (race 1) and nematodes (Fig 5) and possesses desirable horticultural

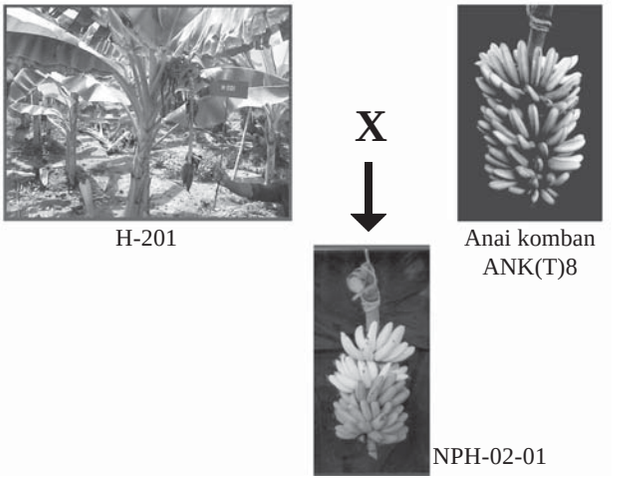


Fig 4. Pedigree of hybrid NPH - 02-01

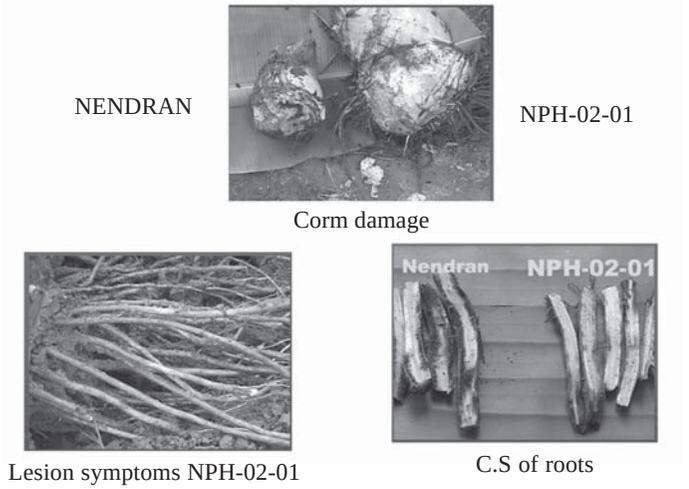


Fig 5. NPH-02-01 hybrid corm and roots exhibiting no damage by nematodes

traits such as better bunch weight (19.00 kg) and 11.00 hands. This hybrid is now under multi-locational testing in different parts of Tamil Nadu.

Triploid x Diploid breeding approaches

The 'Pome' cultivars are usually grown in cool, mid hill ranges of Tamil Nadu where they develop their characteristic flavour and taste. An attempt was made to improve a pome cultivar called 'Kallar Ladan'. One of the AB hybrids from the cross Kallar Ladan (AAB) x *M. balbisiana* clone Sawai (BB) was used to cross with cultivar Kadali (AA) to develop an AAB hybrid. This was later released for commercial cultivation as CO 1 banana (Azhakiamanavalan et *al*, 1985). This belongs to 'Pome group' and closely resembles Virupakshi (AAB), another popular 'Pome' banana in the hills of Tamil Nadu. Plants of CO1 are medium tall (2.7 M). The bunch weighs 10.5 kg on an average with 7 hands and 80-85 fruits. Fruits have TSS of 22-24°brix. Crop duration is 14 - 15 months. Just before full ripening, fruits exhibit acidic taste but become sweet at full ripening.

Triploid x diploid breeding attempted at KAU also led to the release of two hybrids, viz., BRS-1 (Agniswar x Pisang lilin) and BRS – 2 (Vannan x Pisang Lilin). BRS-1 (AAB) is 100 days earlier than Rasthali with significant difference in bunch weight. It has been released for homestead cultivation in Kerala as it is resistant to sigatoka leaf spot. BRS-2 (AAB) is a medium statured hybrid, tolerant to leaf spot and Panama diseases, rhizome weevil and nematodes. The average bunch weight is 14 kg, with 8 hands and 118 fruits in a crop duration of 314 days.

This suggests that Triploid x diploid approach to breeding is a viable method to produce new hybrid combinations. In this direction, Krishnamoorthy (2002) took up hybridisation between triploid parents 'Karpooravalli', 'Red banana', 'Nendran' and Rasthali with already identified, potential male diploids / synthetic diploids. Among the triploid female parents, higher seed-set as obtained with 'Karpooravalli' while the remaining commercial triploids 'Rasthali' (AAB), 'Red banana' (AAA) and 'Nendran' (AAB) did not set seed. Presence of female fertility in AAB genome group of plantains has been reported in some African countries where high rainfall with high relative humidity may have favoured pollen germination and fertilization (Swennen and Vuylsteke, 1993 and Vuylsteke et al. (1993). Therefore, the climate of Coimbatore where relative humidity is generally not very high may be a possible cause for poor or nil fertility in 'Rasthali' (AAB) and 'Nendran' (AAB). The stigmas in female flowers of these cultivars were dry and completely blackened on crossing, indicating that female flowers are not compatible for hybridisation. Similarly, though the stigma of Red banana showed stickiness indicating receptivity, it could not set any seed under Coimbatore conditions. It may be due to post-pollination incompatibility. In contrast, Karmacharya *et al* (1992) reported maximum seed production in Agniswar, a synonym for Red banana under Kerala conditions. This indicates the influence of climate on fertilization and development of embryos. This poses a challenge to breeders to attempt induce female fertility by some means in clones that are otherwise female sterile.

Triploid x Diploid breeding sometimes produces promising diploids too (Table 7).

The hybrid H-212 (Fig.6), a diplod (AB), resembles Ney Poovan (AB) and possesses resistance to nematodes and sigatoka leaf spot. Hence, this hybrid has been tested with Ney Poovan (Table 8) and found to be promising. Hence, this hybrid is also under multilocation testing now.

Table 7. Performance of triploid and tetraploid hybrids obtained from 'Karpooravalli' (AAB) crosses

Hybrid	Male parent	Bunch wt (kg)	No. of fingers	TSS (°Brix)	Total crop duration	Reaction to nematodes
H 212	Pisang Lilin (AA)	12.52	160	31.00	363	Tolerant
H 02-21	Red Banana (AAA)	18.00	192	19.20	458	Resistant
H 02-34	Red Banana (AAA)	15.00	116	18.50	550	Resistant

Table 8. Comparative performance of H.212 *vs* cv. Ney Poovan

Trait	H-212	NEY POOVAN
Genome	AB	AB
Male parent	Pisang Lilin	-
Height (cm)	311	360
Girth (cm)	66	85
Bunch weight (kg)	13	10
Number of fingers	160	130
TSS (%)	31	26
Total duration (days)	363	335
Reaction to nematodes	Tolerant	Susceptible

Further, 3n x 2n breeding programme at TNAU resulted in identification of a promising hybrid designated as 96/7 (Fig 7). This was evolved by crossing 'Karpooravalli' x H. 201 (3n x 2n) and its attributes are tabulated (Table 9). The fruit colour is bright yellow without any ashy coating. The female parent (Karpooravalli) has fruits with ashy coating that masks the brightness of yellow colour. The ploidy and genome were assessed to be ABB, similar to female parent. This cultivar is now under small scale field-testing.

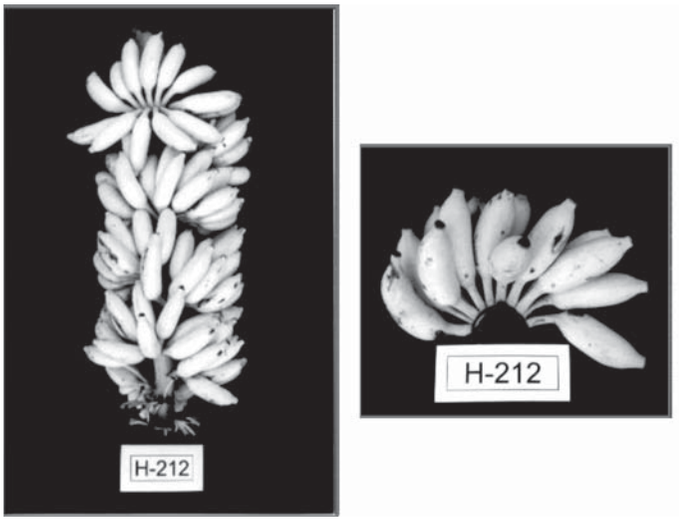


Fig 6. Bunch traits of Hybrid H 212 (AB)

Karpooravalli X H-201

H-96/7

Under now multi location testing for yield potential and resistance to nematodes

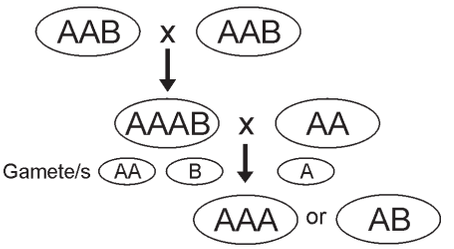
Fig 7. A promising triploid hybrid (ABB)

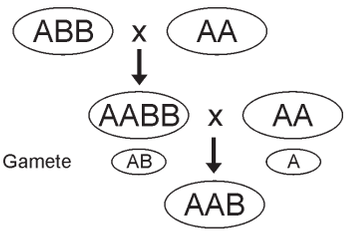
Table 9. Comparison of Hybrid 96/7 with the female parent 'Karpooravalli' (ABB)

Sl. No.	Characters	Hybrid 96/7 (ABB)	Karpooravalli (ABB)
1.	Plant height (cm)	3.10	3.25
2.	Plant girth (cm)	98.00	93.50
3.	Bunch weight (kg)	28.50	24.00
4.	Hands/bunch	12.00	10.00
5.	Fruits/bunch	202.00	186.00
6	Fruit weight (g)	93.50	84.10
7	Total soluble solids (°Brix)	21.5	19.0

Development of primary tetraploids and their further improvement

Hybridisation between triploid cultivars and dipoloid cultivars often results in many primary tetraploids. As tripoids are preferred over tetraploids because of superior vigour and yield potential, these primary tetrapoids are subjected to further crossing with potential diploids to develop secondary triploids, as indicated below :





Damodaran (2004) attempted crossing triploid Peykunnan (Syn. Pisang Awak) with many diploids (AA) to develop primary tetraploids (Table 10).

Table 10. Promising potential tetraploids in banana

Hybrid	Parent	Genome	Bunch weight (kg)	TSS(%)	Male fertility	Female fertility	*Fusarium* wilt	Nematodes
H-02-34	Karpooravalli x Red banana	AABB	13.5	20.05	P	P	R	R
H-03-05	Karpooravalli OP	AABB	11.0	24.10	P	P	R	R
H-03-13	Karpooravalli x Erachi Vazhai	AABB	16.0	23.50	P	P	R	R
H-03-17	Karpooravalli x Pisang Lilin	AABB	13.0	20.15	P	P	R	R
H-03-19	Peykunnan x EV	AABB	17.5	22.50	P	P	R	R

P = Present; R = Resistant

These tetraploids need to be further improved by crossing again with diploids to improve bunch weight. One attempt by Damodaran (2004) to cross H-02-02 (AABB), hybrid between Karpooravalli (ABB) and Red Banana (AAA) with Pisan Lilin (AA) has resulted in development of H.03-15 (AAB), a primary triploid with good bunch weight (14.50 kg) and resistance to wilt and nematodes suggest that extensive hybridization between potential tetraploids and diploids has to be pursued in relatively large numbers so that there are more chances of producing of new triploids with novel genomic background.

Breeding may not be always successful in this direction. Kavitha (2005) crossed a primary tetraploid (H.02.32 – AABB) with Pisang Lilin (AA) and obtained, again, tetraploid hybrid (AABB) only. This warrants the use of potential tetraploids as male parents (if male fertile) with potential diploids and triploids to develop new triploid combinations.

In vitro mutation and selection

Improvement of commercial triploids like cvs. Robusta and Rasthali through sexual hybridisation is very difficult as these are female sterile. Therefore, mutation breeding was initiated in 1995 with cultivars like Robusta (AAA) and Rasthali (Silk –AAB). These were subjected to 3 Kr, 4 Kr and 5 Kr dosage of g - irradiation (Baskar Rajan, 1998). Among the three dosages used, 3 Kr was found to be optimum (more than 50% of survival was observed). After three subcultures *in vitro* plants were established, rooted and hardened. Plants obtained *in vitro* were planted in field for further evaluation (Baskaran *et al*, 2000).

Recently, Kumar *et al* (2004) reported the potential of *in vitro* mutation breeding with cvs. Robusta, Rasthali, Nendran and Poovan through gamma rays and EMS (ethyl methane sulphonate) and isolated many economic mutants (Table 11).

Table 11. Economic mutants isolated in banana through *in vitro* mutation breeding

Mutant	Desirable attribute observed
Nendran	
Ne-IMVo-5	Heavier bunch (13.0kg) Larger fruits (269.3g)
Ne-IMVo-7	Heavier bunch (14.0kg)
Ne-IMVo-8	Good bunch and finger traits Heavier bunch (14.5kg) Larger fingers (Weight : 302g, Length : 27.1cm)
Poovan	
Po-IMVo-1	Heavier bunch (16.0kg) Larger fruits (114.2g)
Po-IMVo-2	Heavier bunch (15.5kg) More no. of fingers in the bunch (176) Attractive fingers
Po-IMVo-3	Heavier bunch (15.5kg)
Po-IMVo-4	Lesser plant height (2.2m)
Po-IMVo-7	Heavier bunch (16.0kg)
Po-IMVo-10	Lesser plant height (2.2m)
PROMISING MUTANTS AND THEIR DESIRABLE ATTRIBUTES	
Robusta	
Ro-IMV$_4$6-1-1	Heavier bunch (30.5kg)
Ro-IMV$_4$6-1-2	Heavier bunch (30.0kg)
Ro-IMV$_4$6-1-3	Heavier bunch (28.5kg)
Ro-IMV$_4$6-2-1	Heavier bunch (30.5kg)
Ro-IMV$_4$6-2-2	Heavier bunch (29.5kg) Lesser plant height (1.73m)
Rasthali	
Si-IMV$_4$ 6-2-4	Lesser plant height (1.98m)
Si-IMV$_4$ 6-2-5	Heavier bunch (13.0kg)
Si-IMV$_4$ 10-5-1	Lesser plant height (1.96m)
Si-IMV$_4$ 10-5-3	Lesser plant height (1.90m)
Si-IMV$_4$ 11-6-5	Heavier bunch (13.5kg)

In vitro polyploidy breeding

In vitro ploidy breeding programme was taken up at TNAU to improve some potential diploid cultivars which are not amenable to conventional breeding methods due to sterility but are otherwise resistant to many biotic stresses with a good yield potential (Ganga, 2001 and Ganga *et al*,

2002). Diploid cultivars cultivars viz., Sannachenkadali (AA), Anaikomban (AA), Kunnan (AB) and Thattillakunnan (AB) were treated with anti-mitotic agents like colchicine ($C_{22}H_{25}N0_6$) (Fig 8) and Oryzalin (3,5-dinitro N_4-N_4 – dipropylsulfanilamide) (Fig 9). Relatively, oryzalin is found to be effective in producing higher frequency of tetraploids (Fig 10). A totall of 41 tetraploids were obtained, 16 each from Sannachenkadali and Anaikomban, 12 from Kunnan and 13 from Thattillakunnan. Uma *et al* (2003) evaluated the response of diploid cultivars to induction of tetraploidy and the efficiency of polyploidizing agents at NRCB, Trichy. In the first trial, 'Kunnan' and 'Matti' shoot tips were soaked in colchicine for 24 h and in oryzalin for 72 h, respectively. In the second trial, these were, respectively, cultured in an initiation medium in which colchicines and oryzalin had been incorporated. MS medium fortified with 3.0 mg/l of BAP was used for culture initiation. The same medium with reduced level of BAP (2.0 mg/l) was used for monthly subculture. After the third subculture, explants were rooted on MS basal medium without any growth regulator. Induction of polyploidy was verified.

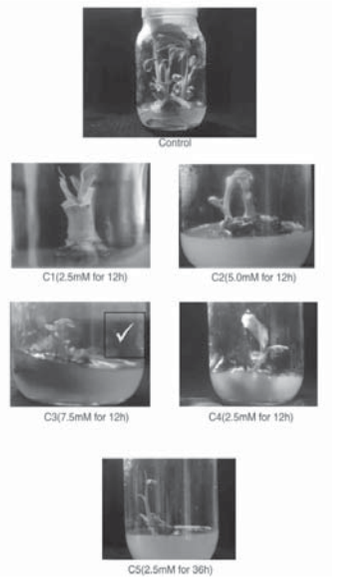


Fig 8. Shoot proliferation in colchicine treated cultures

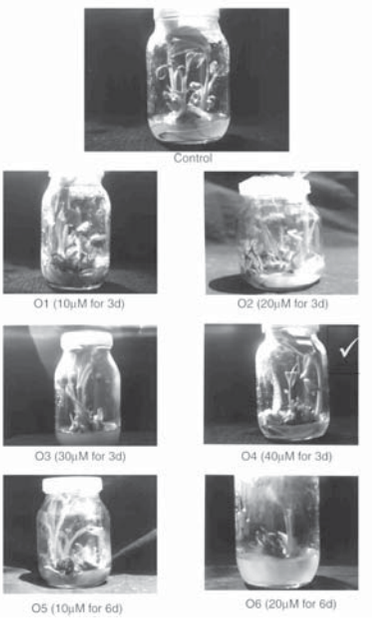


Fig 9. Shoot proliferation in oryzalin treated cultures

Breeding for resistance to Fusarium wilt

Fusarium wilt caused by *Fusarium oxysporum* f. sp. *cubense* causes yield loss in South India from 2-90% (Thangavelu *et al*, 1999). This warranted initiation of breeding for resistance to fusarium wilt at TNAU.

Potential diploids like Anaikomban (AA), Matti (AA), Namarai (AA) and Pisang Lilin (AA) and new synthetic diploid hybrids such as H-201 (AB) and H-65 (AA), developed at TNAU, were screened for resistance to *Fusarium* wilt by Gunavathi *et al* (2003). Of the 20 genotypes screened, synthetic diploid hybrids H-65, H-109, H.103 and H-201, the parents 'Anaikomban', 'Pisang lilin' and 'Tongat' and the commercial cultivar 'Robusta' showed resistance to fusarium wilt, while the others exhibited susceptible reaction.

In breeding for resistance to any disease, the basic requirement is availability of an efficient screening technique, which should clearly distinguish resistant genotypes from susceptible ones. At TNAU, screening was taken up against the pathogen *Fusarium oxysporum* f.sp. *cubense* race 1 employing root inoculation technique (Fig 11). (Gunavathi, 2000 and Damodaran, 2004). In this method, roots are directly exposed to the conidial suspension, which helps in better and easy colonization of

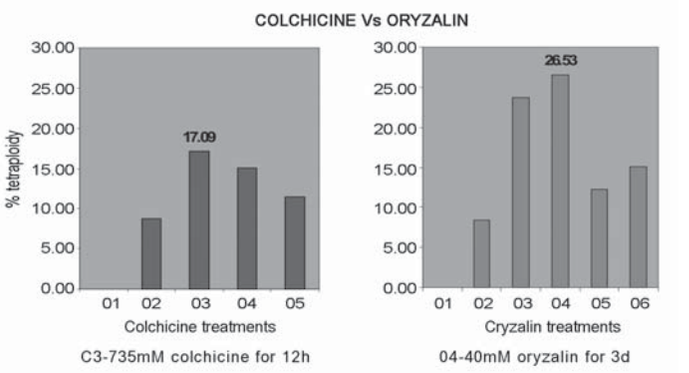


Fig 10. Effect of Colchicine vs. Oryzalin on induction of tetraploidy in banana diploid clones

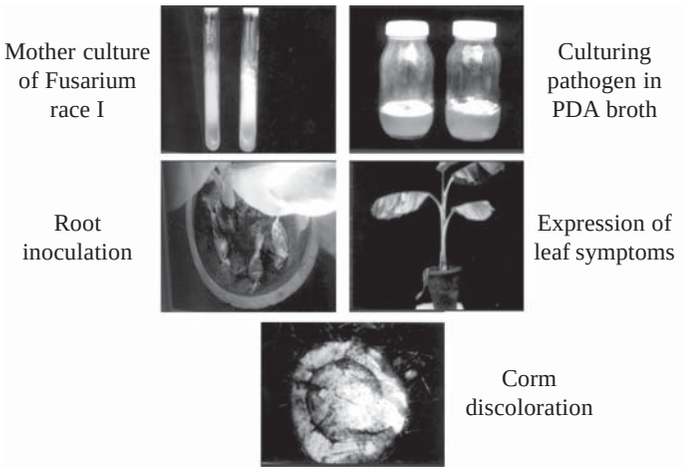
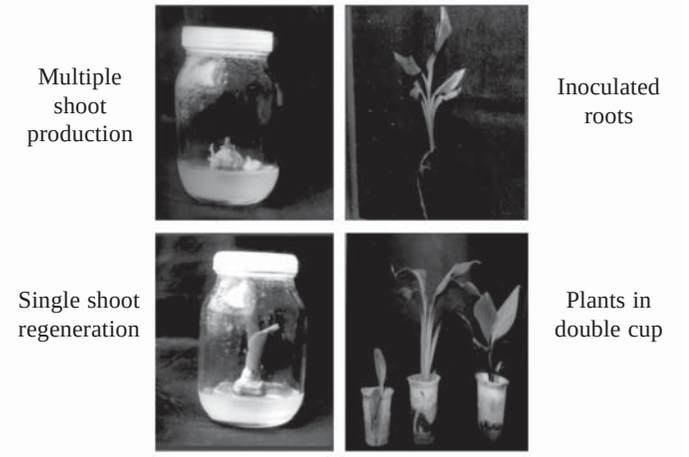


Fig.11 Root inoculation technique against *Fusarium* in banana



UI : Un inoculated, R : Resistant, S : Susceptible

Fig .12 *In vitro* screening of selected Phase I & phase II hybrids by double cup method for *Fusarium* wilt (Race 1) resistance

the fungus in the susceptible host (Sun and Su, 1984). Apart from pot screening, *in vitro* screening of hybrids was also attempted to test the utility of double cup method, as described by Brake *et al* (1995) (Fig 12).

Damodaran (2004) screened many hybrids and from these, 15 hybrids exhibited resistance, while others were susceptible. A critical analysis of inheritance pattern of hybrids revealed that resistant diploid, triploid and tetraploids hybrids had either Pisang Lilin or Anaikomban as one of the parents (Table 12). This indicates that dominant genes govern resistance. Further, when a particular parental combination was taken into account, for example H-201 with Anaikomban or Pisang Lilin, none of the progenies showed resistance although the male parents used were resistant to *Fusarium* wilt. This may be due to the heterozygous nature of the parent or due to the polygenic inheritance of resistance genes with modifying effect. Vakili (1965) stated that a single dominant gene governs resistance to *Fusarium* wilt while using one homozygous parent as the resistant source. Further, recovery of resistance genes from susceptible x susceptible combinations may be due to transgressive segregation and heterozygous nature of the parents.

Table 12. Screening of banana hybrids for resistance to *Fusarium* wilt (race 1) under pot culture by root inoculation technique

Hybrid	Parentage	Genome	Disease Score	Status
H-02-06	H 201 × AN	AB	5	HS
H-02-08	H 201 × EV	AAB	1	R
H-02-09	H 201 × PL	AB	1	R
H-02-10	H 201 × H 110	AB	1	R
H-02-11	H 201 × H 110	AB	3	S
H-02-17	KV × PL	AABB	5	HS
H-02-18	KV × PL	AABB	2	S
H-02-19	KV × EV	AABB	1	R
H-02-20	KV × RB	AABB	6	HS
H-02-36	KV × RoO	AABB	5	HS
NPH-02-01	H201 x AN	AAB	1	R
NPH-02-02	H201 x AM	AB	2	S
	Parents			
H201		AB	1	R
Anaikomban (AN)		AA	1	R
Ambalakadali (AM)		AA	1	R
Erachi Vazhai (EV)		AA	2	S
Pisang Lilin (PL)		AA	1	R
H110		AB	3	S
Karpooravalli (KV)		ABB	5	HS
Red Banana (RB)		AAA	5	HS
Robusta (RO)		AAA	1	R

R- Resistant: S- Susceptible: HS- Highly susceptible

Screening of hybrids under *in vitro* screening by Damodaran (2004) confirms beyond doubt the claims of varying degree of resistance/susceptibility of a cultivar under different situations in banana. The major problem encountered in resistance screening programme in pot culture was limited availability of suckers from the field for testing. However, in *in-vitro* culture, production of multiple shoots from a single sucker enabled easy regeneration and increased sample number for evaluation.

Breeding *Musa* hybrids resistant to nematodes

Banana production is severely threatened by many different types of soil nematodes. Among the various nematodes, lesion producing nematodes such as *Radopholus similis* and *Pratylenchus coffeae* are considered to be economically important nematode pests of banana (Rajendran *et al*, 1980). Sundararaju (1996) reported that the burrowing nematode exhibited severe root rotting,

Table 13. Variation in root damage caused by nematodes in banana hybrids / parents

Sl. No.	Hybrid / parent	Genome	Resistant reaction	Total no. of roots	No. of dead roots	No. of functional roots	Dead roots (%)	Feeder roots	Root lesion (%)
1.	H-02-34	AABB	R	8.50	0	8.5	0.00	1.5	10.00
2.	H-201	AB	R	11.00	0	11.0	0.00	1.5	2.50
3.	NPH-02-01	AAB	T	16.50	0	16.5	0.00	1.0	0.01
4.	Karpooravalli	AAB	T	10.50	3	7.5	28.63	2.0	17.00
5.	Anaikomban	AA	T	8.50	1	7.5	12.50	2.0	5.00
6.	H-02-32	AABB	S	9.00	4	4.5	44.44	3.0	37.50
7.	Red banana	AAA	HS	11.00	5.5	5.5	50.00	3.5	38.00
	C.D *(P=0.01)*			1.081	0.78	1.021	19.298	0.661	5.669

resulting in 25-35% reduction in yield. Thus, the foremost aim in banana and plantain improvement is to enhance quantitative and qualitative traits besides developing hybrids with increased resistance/tolerance to major nematodes (Kumar and Soorianathasundaram, 2002). Initial screening carried out at TNAU resulted in the identification of potential diploids like Anaikomban (AA), Matti (AA), Namarai (AA) and Pisang Lilin (AA) (Sathiamoorthy and Balamohan, 1993). Further screening among diploids, showed that cultivars Amabalakadali (AA), Erachivazhai (AA), Venneettu Kunnan (AB), Adakka Kunnan (AB), Poomkadali (AB), Kadali (AB), Ney Poovan (AB) had moderate resistance (Gowen *et al*, 1998).

Screening of banana hybrids/cultivars for resistance to nematodes requires an effective screening technique that should distinguish the genotype as susceptible, tolerant or resistant to various nematodes. The INIBAP method (Fig 13) largely encompasses the ability of the genotype to resist nematode infection based on root and corm damage assessment besides its ability to tolerate a high population

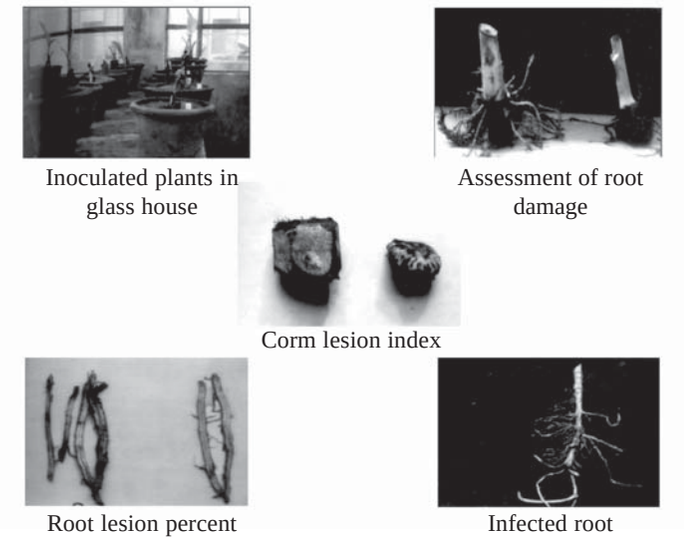


Fig 13 . Screening of banana against nematodes based on root and corm damage

of nematodes. Though the nematode can live in soil, it cannot enter the roots of resistant hybrids and multiply fast (Gowen, 1994).

As the nematode population inflicts damage directly to the root system by causing lesions, assessment of root and corm damage is important (Speijer and Gold, 1996). Besides, root number and percent dead roots are considered critical in the assessment of nematode damage as Gowen (1993) reported that the rate of root destruction is not directly related to nematode population density in the root system as a whole, but, to the number of individual colonies on the roots.

Evaluation of banana hybrids by Krishnamoorthy (2002) and Damodaran (2004) revealed significant difference in the ability of hybrids and parents to produce more number of functional roots (Table 13). Resistant/ tolerant hybrids produced thick and healthy roots besides more functional roots and the least number of dead roots to overcome nematode infection. However, assessment of soil in their mat showed a considerable amount of nematode population. Thus, it is evident from the above facts that these hybrids possessed an inherent ability to overcome the invasion by nematodes. Increase in number of functional roots in these hybrids compared to either of the parents was attributed to heterosis.

Damodaran (2004) observed that resistant x resistant crosses produced both resistant as well as susceptible hybrids. This indicated that resistance to nematodes is under polygenic control and segregation for resistance and susceptibility was expected because of the heterozygous nature of the parents studied.

Higher resistance in hybrids NPH-02-01 and H-02-08 may be attributed to triploidy (AAB) with the hybrid having inherited the entire genome of AB from H-201 and the resistant 'A' genome from Anaikomban or Erachi Vazhai, respectively.

Table 14. Biochemical activities in the roots of banana hybrids and parents inoculated with *Fusarium* under pot culture conditions (Damodaran, 2004)

Sl. No.	Hybrid / parent	Genome	Category	Total phenols (µg g^{-1})	Proline (µg g^{-1})	Lignin (%)	Peroxidase (ΔA $min^{-1}g^{-1}$)	Polyphenol oxidase (ΔA $min^{-1}g^{-1}$)	PAL (nmol $min^{-1}mg^{-1}$)	β glucannase (µg min^{-1} g^{-1})
1.	NPH 02.01 (H2= 1x Anaikomban)	AAB	R	620.0	586.0	1.39	4.55	0.75	14.80	312.82
2.	H.201	AB	R	610.5	583.0	1.22	5.55	0.83	15.10	321.68
3.	Anaikomban	AA	R	566.5	498.0	1.13	4.40	0.73	12.05	301.89
4.	H.02.34 (AABB) (Karpooravalli x Red banana)	AABB	R	641.0	608.5	1.28	0.51	0.66	13.93	337.21
5.	H.02.32 (AABB)	AABB	S	414.5	324.9	0.74	1.92	0.23	2.98	231.50
6.	Karpooravalli	ABB	S	478.7	453.5	0.75	1.62	0.20	4.71	153.39
7.	Red Banana	AAA	S	184.5	151.5	0.50	1.23	0.22	4.53	147.77
8.	H.02.06 (H.2=1 x Anaikomban)	AB	S	241.2	167.0	0.24	0.44	0.12	1.35	164.50
	CD *(P=0.05)*			75.4	6.7	0.04	0.17	0.04	0.61	22.76

In other tetraploid hybrids such as H-03-05, H-03-10, H-03-11, H-03-13, H-03-17 and H-03-19, Damodaran (2004) observed higher amount of resistance as these had inherited the extra resistant 'A' genome from diploid parents Pisang Lilin or Erachi Vazhai. In this situation, the tolerant genome ABB might have interacted with resistant genome 'A' resulting in resistant AABB genome, implying the non-allelic genic interaction. Rowe and Rosales (1996) indicate that one or more dominant alleles control genetic resistance to burrowing nematode.

It is interesting to analyse results in pot culture experiments which clearly showed that many of the hybrids claimed to be resistant under field conditions were found to be tolerant or susceptible under pot culture. This is normally expected, as a high population of nematodes is bombarded in to the root zone for forceful infection (Dosselaere *et al*, 2003). Besides, in the pot, nematodes are inoculated in 45 days old seedlings when the roots are young and tender. However, under field conditions a high population of nematodes is encountered normally only at the time of shooting and harvest (Jean *et al*, 2002), when the plants possess a larger and longer root system, with increased biochemical and enzyme activity, resulting in more resistant reaction to nematodes. This might be the probable reason why some hybrids that exhibit resistance under field conditions become susceptible when inoculated artificially.

Role of biochemical markers in resistance to *Fusarium* wilt and nematodes

When a pathogen infects the host tissue, a small number of specific gene, producing mRNA's that permit synthesis of similar number of specific proteins, are induced (Vera Conejero, 1989). Many of these proteins are enzymes such as phenylalanine ammonia lyase, polyphenol oxidase and peroxidase, b-1-3 glucanase (Vidhyasekaran, 1993) that are involved in synthesis of low molecular weight substances such as phytoalexins, phenols and lignin, which

Table 15. Biochemical contents in the roots of banana hybrids and parents inoculated with nematodes under pot culture conditions

Sl. No.	Hybrid / parent	Genome	Category	Total phenols (µg g^{-1})	Proline (µg g^{-1})	Lignin (%)	Peroxidase (ΔA $min^{-1}g^{-1}$)	Polyphenol oxidase (ΔA $min^{-1}g^{-1}$)	PAL (nmol $min^{-1}mg^{-1}$)
1.	H-02-34	AABB	R	722.4	421.5	0.35	13.05	0.86	11.60
2.	H-201	AB	R	605.0	536.5	1.32	6.23	0.87	11.35
3.	NPH-02-01	AAB	T	656.7	404.0	1.27	16.45	1.10	12.35
4.	Anaikomban	AA	T	636.5	438.5	1.30	6.23	0.72	10.20
5.	Karpooravalli	AAB	T	444.0	342.5	1.02	4.41	0.30	3.45
6.	H-02-32	AABB	S	414.4	211.3	0.53	4.22	0.46	5.25
7.	Red banana	AAA	HS	139.5	219.0	0.53	3.55	0.22	2.06
	C.D *(P=0.01)*			8.9	2.9	0.06	0.448	0.036	0.536

are inhibitory to fungal pathogens (Bell, 1981; Vidhyasekaran, 1993). Hence, analysis of these biochemical markers, which provide a mechanism for resistance to fungal pathogens, is very essential. Damodaran (2004) established that *Fusarium* wilt resistant hybrids / parents had higher levels of these enzymes than the susceptible ones (Table 14) confirming the role of biochemical markers in conferring resistance. Similarly, assessment of these biochemical markers (Damodaran, 2004) in resistant, tolerant and susceptible hybrids/ parents revealed their significant role in conferring resistance against nematodes in banana (Table 15).

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