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2007 - '08



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तिरुचिरापल्ली - ६२० १०२, तमिल नाडु, भारत



NATIONAL RESEARCH CENTRE FOR BANANA
(Indian Council of Agricultural Research)
Thogamalai Road, Thayanur Post
Tiruchirapalli - 620 102, Tamil Nadu, India

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National Research Centre for Banana
Thogamalai Road, Thayanur Post
Tiruchirapalli - 620 102
Tamil Nadu, India

Published by : **Dr.M.M. MUSTAFFA**
Director

Edited by : **Dr.M. Mayilvaganan**
Dr.M.M. Mustafa

Compiled &
Photographs (Gen.) by : **Mr.P. Ravichamy**

Cover design by : **Dr.M. Mayilvaganan**
Dr.M.M. Mustafa

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Tel : 0422-2450133
E-mail : sakthi_press@yahoo.co.in

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PREFACE

I am proud to present the Annual Report for the year 2007-'08. The Centre continued to progress by developing research achievements to solve the production constraints for the benefit of banana growers and stakeholders. The major initiatives and research accomplishments are presented in this Annual Report as below:

Banana improvement group has developed a barcode for the NRCB released variety 'Udhayam' and also characterized 57 banana accessions using IRAP primers. *Musa acuminata* ssp *burmannica*, a wild species, was collected from the Western Ghat regions of Nagarcoil. Two putative bands linked to Sigatoka leaf spot resistance have been identified and development of SCAR markers is in progress. Protocols have been developed to isolate the seed DNA for conservation purpose. A hybrid progeny of Anaikomban x Pisang Jajee found to be promising for leaf industry because of its superior leaf quality.

The production group has found that soil application of Fe and B with foliar application of Zn along with recommended dose of NPK produced 32% higher yield in Ney Poovan. Soil test based nutrient tailoring fertilizer equation for Karpuravalli and Poovan have been developed. Plants under 4500 plants ha⁻¹ recorded significant yield with higher leaf N and K in top 15 cm soil. Soil at deeper depth recorded higher soil P. Screening of banana varieties for salt stress indicated Karpuravalli and Poovan are tolerant with higher chlorophyll content and chlorophyll stability index.

Crop protection group identified many bio-control agents for the control of banana weevils and nematodes. The Centre has released eco-friendly bio-agents *Beauveria bassiana* and *Verticillium lecanii* for the control of weevils and aphids respectively. Identified five species of *Cynobacterium* for the control of *P. coffeae*. Genetic diversity analysis of *Foc* isolates indicated two main clusters. BBTV-REP genes, coat protein gene of CMV and complete gene of BSMysV infecting Poovan were cloned and sequenced. ECS for Virpupakshi and Poovan varieties have been developed.

Developed protocol for dry stem juice, which has potential as health supplement. Standardized protocol for retting of banana fibre using sodium hydroxide. Developed protocol for cold stabilization of banana wine at lower temperature, which has yielded good quality wine.

The Centre has commercialized two bio-control agents and many value added technologies. Three short-term training courses and one winter course were offered for the benefit of entrepreneurs and researchers. Three television and four radio talks were broadcasted to disseminate the technologies developed by the Centre.

I specially acknowledge Dr.C.K. Narayana, Principal Scientist of the Centre for translating the Executive Summary into Hindi.

I acknowledge Dr. M. Mayilvaganan and his team involved in bringing out this Annual Report in time.

Tiruchirapalli
10th October 2008



(M.M. Mustaffa)
Director

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राष्ट्रीय केला अनुसंधान केंद्र के स्थापना के पंद्रह साल के दौरान यह केन्द्र ने केले का उत्पादन एवं उत्पादकता बढ़ाने और इस व्यवसाय पर आधारित किसानों एवं अन्य भाग्यधारियों का जीवन स्तर उठाने में अनुसंधान द्वारा निरन्तर प्रयास कर रही हैं। इस दिशा में चार मुख्य विषयों पर अनुसंधान की जा रही हैं, जैसे की फसल सुधार, उत्पादन प्रौद्योगिकी, तुड़ाई उपरान्त प्रबंध एवं मूल्य वर्धन और फसल संरक्षण। बदलते परिवेश में बहुविभागीय कार्यक्रम के द्वारा परस्पेक्टिक प्लान-विजन: २०२५ में निर्धारित की गयी लक्ष्य को प्राप्त करने के लिए केन्द्र अपनी पूरी ताखत जुटा रही है। भविष्य कार्यक्रम फसल उत्पादन के खर्च कम करने एवं लागत में कटौती लाने कि दिशा में होगी। पिछले साल (सन् २००७-२००८) के दौरान पाई गयी उपलब्धियाँ इस प्रकार है:

फसल सुधार

तमिल नाडु के तिरुनेलवेली एवं नागरकोईल जिलों में की गई जनन द्रव्य की खोज में पश्चिम घाट के इलाके में मूसा अक्युमिनाटा Spp बरमानिका पाया गया जो जंगिली जाति में शामिल है। राष्ट्रीय केला अनुसंधान केन्द्र के जीन बैंक में १४ नया संकलनों को शामिल किया गया। शीत भंडारित सांबराणि मोंधन किस्म के कंद या TC पौदों का प्रक्षेत्र मुल्यांकन में पौदों की वृद्धि या पैदावार में कोई फरख नहीं पाया गया। इसके अलावा मैक्रो साटिलाइट पालिमरफिज़म असेसमांडट द्वारा यह साबित हुआ कि जनन द्रव्य की जनन स्थिती मे कोई बदलाव नहीं आया।

मैक्रो साटिलाइट पालिमरफिज़म पर आधारित बार कोडिंग द्वारा रा.के.आ.के में विकसित 'उदयम' किस्म में 150 bp और 290 bp मात्रा के दो अलील पाया गया जिस से यह पता लगा की उदयम एवं कंताली किस्मे एक वर्ग के और करपूरवल्ली एवं अंकुर किस्मे दूसरे वर्ग के है। 57 जनन द्रव्य संकलनों का IRAP मारकर द्वारा स्वभाव वर्गीकरण में यह पाया गया कि 22 संकलन एक वर्ग और ३५ संकलन दूसरे वर्ग के है। कावेंडिस उपवर्ग के केलों का विभिन्नता जांच ने के लिए RADP प्राईमर से ज्यादा IRAP प्राईमर उपयोगी है। 60% माइचूर बीज में से DNA को अलग करने की विधी को मनकीकृव किया गया। केले में मोम स्वभाव का अनुमान लगाने में SSR प्राईमर का उपयोग असफल पाया गया।

1.3 kb (RPAD प्राइमर OPC-4 से एवं 1.5 kb (OPK-1 और OPK से) साइज के दो पुटेटीव बाइंड को पहचाना गया जो सिगाटेका पत्ता धब्बा रोग के प्रतिरोधक शाक्ती से जुडे हुए बै। इस

बाइंड को सीक्वेंस करके मारकर निर्मित किया गया। लवणता के प्रति सहिष्णुता के बारमें किये गये अनुसंधान में यह पाया गया की रस्थाली एवं रोबस्टा किस्म के explant, 1% NaCl से ज्यादा लवणता सहन नहीं कर सकता है।

केले के झड़ों से टोटल RNA को निकालने में Modified cetyl trimethyl ammonium bromide एवं Lithium chloride तरीके ज्यादा उपयोगी पाये गये। इस टोटल RNA से mRNA और cDNA सिंथासाइज किया गया।

फसल उत्पादन

ग्रांड नाइन किस्म हर घट्टे में दो कंद, 2 मी X 3 मी कि दूरी में रोपण करके 150% निर्धारित खाद की मात्रा फर्टिगेशन के द्वारा देने से पौदे को कद, पत्रियों को संख्य एवं पेडंकल की लंबाई में वृद्धि पायी गयी। विभिन्न घनता एवं दूरी से पौदों की लंबाई, पत्तियों की संख्या, पुत्तियों की संख्या, पत्तियों का विस्तीर्ण, घेरा लीफ एरिया इंडेक्स एवं फिल्लाक्रान में भिन्नता पायी गई। पौदों की घनता में वृद्धि करने से फोटोसिंथेसिस की प्रक्रिया कम हुये लेकिन क्लोरोफिल की मात्रा में कोई फरख नहीं पडा। हर घट्टे में एक पौदा रोपण करने से झड़ों की संख्य में बढ़ौत्री पाई गई। पत्तियों के बीच दूरी बढ़ाने से पत्तियों में अधिक N एवं K और कम P, Ca एवं Mg पाया गया। मिट्टी के ऊपरी १५ से.मी हिस्से में अधिक N एवं K पाया गया जब की 16-30 से. मी. नीचे अधिक P पाया गया।

निर्धारित NPK मात्रा के साथ साथ यदी मिट्टी में Fe एवं B का उपयोग एवं पौदे पर Zn छिडकाव करे तो पौदे का घेरा, लंबाई, पत्तियों की संख्या, हस्थो की संख्या एवं हस्थों में फलों की संख्या में वृद्धि पाई गई। इससे उत्पाद में 32% की एवं मुनाफे में प्रति हेक्टेर रु 44.700 की बढ़ौत्री नेपूवन किस्म के केले में पाया गया। सूक्ष्म धातुओं के साथ साथ गंधक का उपयोग करने से पत्तियों में सभी धातुओं की मात्रा में बढ़ौत्री हुई। मिट्टी के जाँच पर आधारित Nutrient Tailoring एवं fertilizer adjustment equations का निर्माण करपूरवल्ली एवं पूवन प्रजातियों के लिए किया गया।

पौद दैहिकीय अनुसंधान में यह पाया गया की नेन्द्रन कंद के अंदरी हिस्से में हर अवस्था में N, P, K, Ca एवं Mg अधिक मात्र में होती है। रस्थाली एवं नेपूवन केले में टोटल खाद्य पदार्थ निर्माण में करंट फोटोसिंथेसिस का योगदान 52.58% एवं 52.73% पाया गया। सबा एवं करपूरवल्ली किस्मों में प्री-फलवरिंग (फूल - निकलनेसे पहले) भंडारण किया हुआ खाद्य पदार्थ का योगदान ज्यादा है। इन में करंट फोटोसिंथेसिस का योगदान सिर्फ 7% है। रोबस्टा एवं करपूरवल्ली केले में फूल निकलने से पहले पानी का तनाव उत्पन्न करने से घेर क



वजन पर असर पड़ता है। इम्बोगान किस्म में सब से ज्यादा मोम तत्व पाया गया। लवण तनाव के सहन शक्ती पर की गई अद्ययन में यह पाया गया की 'सबा' ज्यादा सहिष्णु है। पौद जीव रसायन अद्ययन से यह अनुमान लगाया गया की सूत्र क्रिमी ग्रस्थ (*Praty lenchus cofeac*) केले के झडों में फिनाल एवं एन्जाईमज की मात्रा बढ़ जाती है जो सूत्रकिमी सहिष्णुता संबंधित जीनस को उत्तेजित करता है।

तुडाई उपरां प्रौद्योगिकी

सामान्य तापमान पर रस्थाली एवं करपूरवल्ली केला शीघ्र पकता है जब की अच्छा रंग आने के लिए 22°C तापमान जरूरी है। कम तापमान पर केले को पकने में अधिक समय लगता है। नोनी (*Morinda citrifolia*) रस 2% के मात्रा में केले के रस के साथ मिश्रित किया जा सकता है। ज्यादा मात्रा में नोनी मिलानेसे पेय कढवा और अस्वादिष्ट हो जाता है। इस की भंडारण क्षमता ६ महिने तक है। केले का डंठल एवं डंठल का रस सुखाकर पाऊडर बनाकर उसके रसायनिक गुणों का जाँच किया गया। इस पाऊडर से काइपसूल एवं स्वास्थ्य पेय बनाया गया। जो गुरदों की पथर गलाने एवं मलबद्धता को हटाने में उपयोगी पाया गया। केले के तने का रेशा निकालने का नया रसायनिक विधी पहचाना गया। इस रसायनिक विधी से अती शीघ्र रेशा निकाला जा सकता है और इस रेशेका गुणवत्ता भी अन्य तरीकों से निकाली गई रेशा से अधिक है। यह यार्न बनाने में ज्यादा काम आयेगा। केले से वाईन बनाने क विधी में भी सुधार लागा गया। वाईन फरमेंटेशन करते समय तापमान को 10°C से 13.5°C पर रखने से वाईन की गुणवत्ता एवं अलकोहॉल की मात्रा बढ़ जाती है।

पादप रक्षण

टिशू कलचर पौदों में झड-गांठ सूत्रकिमी का प्रकोप ज्यादा गया था। सूत्रकिमी क रोखधाम में EPN एवं अन्य सूत्रकिमी नाशकों के मुखाबले *Heterorhabditis indica* ज्यादा उपयोगी पाया गया। केले के झडभाग से आइसोलेट किये ४५ सूडोमोनास स्ट्रेनस में से १६ सूडोमोनास फ्लूरसेन्स जाती के पाये गये और इनमें ४ आइसोलेटस सूत्रकिमी को कंट्रोल करने और पौदे की वृद्धी में सहायता करने वाले थे। ट्राईकोडरमा विरिडी एवं ट्राईकोडरमा हारशियानाम नीम की खली के साथ उपयोग करने से झड-गांठ सूत्रकिमी का अच्छा नियंत्रण हुआ। साइनोबॉक्टीरियम के ५ स्पीशिस एवं VAM (*Glomus faciculatum*) सूत्रकिमी के नियंत्रण में उपयोगी पाये गये। सूत्रकिमी के साथ साथ अगर विषाणु का भी प्रकोप हो तो पैदावार काफि कम हो जाता है। पिसांग जरिबूया, पिसांग लिनिन, मट्टी, तिरुवनंतापुरम, मनोरंजितम एवं FHIA-23 रूट-नाट निमाटोड के प्रती सहिष्णु पाये गये।

GC/MS द्वारा केले के शीत में से ६-८ वोलाटाइल रसायनों का पहचान किया गया। वीरुपाक्षी केले के शीत से निकला वोलाटाइल अन्य किस्मों के मुखाबले कम आकर्षक रहा। तना छेदक कीट के आकर्षण में semio-chemicals जैसे सिनिओल, पैनिन, क्रिसोल एवं हेकसानोल-१ ज्यादा प्रभावी रहा।

टी. विरिडे के कोयंबतूर आइसोलेट से कईटिनेज जीन को निकाल कर pGEM-T वेक्टर में क्लोन किया गया। तेनी जिले में बाक्टीरिया (*Erwinia Spp*) से होने वाली कंद गलन का निश्चित पहचन किया गया। २२ नान-पाथोजेनिक फूजेरियम आइसोलेटस का मालिकुलर अनालाइसिस किया गया। महाराष्ट्र, तमिलनाडु एवं बिहार में किये सर्वे से यह पता लगा की जलगाव, तेनी, वैशाली एवं मुझफरपूर में बंचीटाप फैल रहा है। BBrMV ग्रस्थ नपूवन और रोबस्टा केले में ज्यादा खाद देने से पैदावार में आनेवाले नष्ट को कम कर सकते है। Multiple Virus resistance के जिन कनट्रक्ट को बनाने के लिए प्राईमर डिजाइन किया गया। विरुपाक्षी एवं पूवन के लिए ECS का निर्माण किया गया।

प्रौद्योगिकी हस्थान्तरण

EPF, *Beaveria bassiana* और *Heterorhabditis Indica* जैसे बयोकंट्रोल एजेंट किसानों को वितरण किया गया। केले का मूल्यवर्धित पदार्थ एवं फफूदी और विषाणु रोगों के पहचान के बारे में ३ प्रशिक्षण आयोजित किये गये। इथेरल से केले पकाने की विधी और जूस एवं फिग बनाने की विधी पर प्रौद्योगिकी हस्थान्तरण की गयी। इस साल के दौरान २ टेलिविजन और ३ रेडियो टाक प्रसारित किये गये। केन्द्र ने विभिन्न स्थानों पर १२ प्रदर्शिनियाँ आयोजित की।

संबंध, सहयोग एवं मानव संसाधन

केन्द्र ने विभिन्न कार्यक्रमों के लिए CICY (Mexico), FAO (Rome) DAE, NBPGR. जैसे संस्थाओं के साथ संबन्ध एवं सहयोग किया। केन्द्र के वैज्ञानिकों एवं कर्मचार्यों को प्रशिक्षण एवं शिक्षा के लिए भेजा। शिक्षा एवं प्रशिक्षण के अंतरगत ४७ स्नातकोत्तर विद्यार्थियों को अपने प्रोजेक्ट कार्य के लिए मार्गदर्शन किया गया।

रेवेन्यु जनरेशन

तीन टेखा अनुसंधान प्रयोजनाओं के माध्यम से रु. ५.७३ लाख, विषाणु परिक्षण से रु.६ लाख एवं प्रशिक्षण द्वारा रु.४०,००० केन्द्र ने कमाया। प्रक्षेत्र के उत्पाद के बिक्री द्वारा रु.४.५ लाख का आय मिला।



3 Executive Summary

The National Research Centre for Banana (NRCB) has undertaken research and developmental activities during the last one and half decade after its establishment in 1993 to increase the production and productivity of banana and to raise the socio-economic status of the banana farmers and other stakeholders involved in banana industry. The four major thrust areas of research are: Crop Improvement, Production Technology, Post-Harvest Management and Value Addition and Crop Protection. The Centre is working on multidisciplinary approaches to alleviate the problems faced by the banana growers and will be working on the future thrust areas in research as envisaged in the Perspective Plan-Vision 2025. The main focus of research on banana is to minimize the cost of inputs for maximum production and productivity of banana. The salient achievements of the NRCB for the research year 2007-'08 are presented in this annual report.

Crop Improvement

An exploration survey for banana germplasm in Tirunelveli and Nagarcovil districts of Tamil Nadu has confirmed the occurrence of a wild species, *Musa acuminata* ssp. *burmannica* in the eastern slopes of Western Ghats. Fourteen new accessions were added to NRCB gene bank during this year. Field evaluation of cryopreserved Sommarani Monthan with tissue cultured and conventional sucker controls showed no significant differences for major growth and yield parameters. In addition, microsatellite polymorphism assessment also indicated that the cryopreserved *Musa* germplasm was genetically stable. Using microsatellite polymorphism for bar-coding of NRCB variety 'Udhayam' indicated two alleles with approximate sizes of 150 and 290 bp unique to Udhayam and comparison with local varieties viz., Karpuravalli, Kanthali and Ankur-II using 12 primer pairs, which produced 39 alleles with a mean of 3.25 alleles per primer pair. The cluster analysis showed Udhayam and Kanthali in one cluster and Karpuravalli and Ankur in other. A set of 57 accessions were characterized with six IRAP primer sets, which grouped into two main clusters with 22 accessions in cluster-I and remaining 35 in cluster-II with 58% similarities. The IRAP primers were found better suitable than RAPD primers to study the variability within the Cavendish subgroup. A protocol has been standardized to isolate DNA from 60% mature seeds with liquid state endosperm and devoid of seed coat. SSR primers failed to detect the genetic expression of waxiness trait in banana.

Field evaluation of plants cv. Rajeli derived from ECS and meristem culture for growth, yield and molecular traits revealed no difference between the plants. In evaluation of hybrids, among eleven progenies, four were parthenocarpic, three were seeded and others yet to complete the crop cycle. A progeny of Anaikomban x Pisang Jajee (Hybrid 07/05) had larger leaf area (width 90 cm) and found promising for leaf industry. Callus could be induced in Robusta, Grand Nain and Ney Poovan, but ideal callus was obtained only in Robusta with growth regulators like picloram and zeatin. Eight percent survival was observed in field planted Rasthali and Nendran plantlets developed from ECS and assessment of genetic diversity is in progress. Two putative bands of 1.3 kb (by RAPD primer OPC-4) and 1.5 kb (OPK-1 and OPK-11) linked to Sigatoka leaf spot disease resistance have been identified. A band of 1.3 kb has been sequenced and SCAR marker designed for the sequenced fragment was synthesized, which is being validated in both parents and progenies. *In-vitro* screening of Rasthali and Robusta to salt stress revealed ex-plants of both cultivars failed to survive above 1% NaCl concentration while negative correlation was observed with number of leaves and roots produced at 0.25% and 0.5% salt concentration as compared to control. Among the cvs. Robusta explants were more adaptable than Rasthali with higher peroxidase and polyphenol oxidase activity.

Modified cetyl trimethyl ammonium bromide and lithium chloride methods were found suitable for isolating total RNA from banana root samples and from total RNA, mRNA was isolated and cDNA was synthesized. LD₅₀ for Ethyl Methane Sulphonate was determined for shoot meristem, proliferating bud and ECS explants of Rasthali banana. Pollen shape, size, fertility and germinability of 37 diploid, triploid and tetraploid accessions were completed. Pollen fertility varied with the ploidy level. Pisang Jari Buaya, Pisang Lilin, Matti, Thiruvananthapuram and Manoranjitham and FHIA-23 were found resistant to *M. incognita*.

Crop Production

In Grand Nain variety, planting of two suckers per hill at 2 x 3 m spacing with 150% RDF fertigation recorded the highest plant height, more leaves and longer petiole length. Plant height, girth, healthy leaves, number of suckers, leaf area, leaf area index, phyllochron, days taken from planting to bunch emergence showed significant differences. Though the photosynthetic activity decreased with increasing plant population, there was no significant difference in chlorophyll content. Rooting pattern was significantly higher in plants under single plant. Plants under wider spacing (1.8



x 3.6 m) recorded significantly higher leaf N and K than P, Ca and Mg contents. Top 15 cm soil recorded higher N and K, whereas 16-30 cm depth recorded higher soil P.

Soil application of Fe and B and foliar application of Zn with recommended dose of NPK produced the highest pseudostem height, girth, total number of leaves, hands and fingers per bunch and 32% more yield with an additional profit of Rs. 44,700/- per ha in Ney Poovan. Application of sulphur along with micronutrients recorded more leaf nutrients than without sulphur application and soil application of sulphur reduced the soil pH from 8.7 to 7.6 in the rhizosphere. Soil test based nutrient tailoring and fertilizer adjustment equations for Karpuravalli and Poovan have been worked out. The fertilizer adjustment equations were tested in different multilocation trials, wherein highest cost of Rs. 4,17,230/- per ha was worked out with the highest B:C ratio (24.02), when the yield target was fixed at 38 t/ ha.

Major nutrients (N, P, K, Ca and Mg) content were higher in the inner core of the corm in all phenological stages of Nendran. In Rasthali and Ney Poovan, current photosynthetic contribution was 52.58% and 52.73% respectively for the bunch development whereas in Saba and Karpuravalli, pre-flowering storage contributed more for the bunch development and bunch photosynthesis was less than 7%. Water stress imposed at flowering stage and 30 and 60 days after flowering decreased the bunch weight more in Robusta and Karpuravalli. Anaikomban recorded the highest chlorophyll content among 24 diploids studied. Imbogan, a diploid, recorded the highest chlorophyll and epicuticular wax. Screening of banana varieties to salt stress indicated Karpuravalli and Poovan recorded higher chlorophyll content while Saba recorded better Chlorophyll Stability Index and more proline content and higher lipid peroxidation in Nendran. Total phenol content and activities of enzymes increased from third day after inoculation of *Pratylenchus coffeae* in banana indicating triggering of nematode resistance genes.

Post-Harvest Technology

Rasthali and Karpuravalli ripened faster at ambient temperature and lower temperature (18 and 20 °C) storage increased the green life and yellow life while the optimum temperature for ripening is 22 °C. Banana juice blended with Noni juice at 2% could be stored for 6 months. Dried stem and stem juice powders were prepared and biochemically characterized to develop as health supplement. Study on retting of pseudostem fibre indicated chemical retting with NaOH gave better results with

lesser time for retting, better color, highest tenacity and suitability for yarn making. For wine preparation, fermentation at 24 °C was quicker but increased alcohol concentration was observed at lower temperature (10 and 13.5 °C).

Crop Protection

Heavy infestation of root-knot nematode was observed in tissue-cultured banana seedlings. The *Heterorhabditis indica* gave better control of nematodes over other EPN and nematicides. Out of 45 *Pseudomonas* strains isolated from the rhizosphere of banana; only 16 belonged to *P. fluorescens* and four were effective in increasing plant growth and controlling the nematode population. Antagonistic fungi *Trichoderma viride* and *T. harzianum* with neem cake effectively controlled the root knot nematode, *M. incognita*. Five species of Cyanobacterium were most effective in controlling the *P. coffeae*. Similarly, VAM (*Glomus fasciculatum*) was also found effective in controlling the nematode and increasing the plant growth. BSV and nematodes infection had more impact in reducing the growth parameters of banana.

Six to eight volatile components including naphthalene were identified from leaf sheaths by GC/MS. Leaf sheath volatiles of Virupakshi attracted lesser weevils than other banana cvs. Semio-chemicals such as cineole, pinene, cresol and hexenol-1 showed good response to banana stem weevil. The yield loss in hill banana due to stem weevil was 65-95%. Many bio-control agents like white and green muscardine fungi, *Beauveria bassiana* and *Metarhizium anisopliae* were isolated from soils collected from various districts of Tamil Nadu. Ten to 70% mortality of banana stem weevil was observed against non-pathogenic *Foc*. Field evaluation of *Beauveria brongniartii* at 1×10^{10} cfu/20 g/trap, trapped 0.2-2.6 corm weevil/trap and 0.1-1.2 stem weevil/trap.

The chitinase gene from Coimbatore isolate of *T. viride* was isolated and cloned into pGEM-T vector. In Theni district of TN, severe occurrence of *Erwinia* spp. causing bacterial rot was recorded. Molecular analysis of 22 non-pathogenic *Fusarium* isolates was done and the size of the amplified product of ITS region of all the isolates ranged from 325 to 370 bp and the IGS region of 11 isolates ranged from 1144 to 1875 bp. Amplification of ITS region of rDNA in 50 pathogenic *Fusarium* isolates yielded amplicon size of 600 bp. In ISSR analysis, out of 19 primers tested, only 7 primers gave consistent scorable bands. Using these 7 ISSR primers, genetic diversity analysis of 37 *Foc* race-1 isolates revealed 3 main clusters and many sub-clusters. Similarly, genetic diversity analysis of 28 *Foc* race-2 isolates



with 10 race-1 isolates using the same primers indicated two main clusters.

Survey revealed incidences of bunchy top viral disease in Jalgoan (Maharashtra), Theni (Tamil Nadu) and Vaishali and Muzafarpur (Bihar). Application of higher doses of fertilizers to BBrMV infected Ney Poovan and Robusta plants proportionately increased the growth parameters and bunch weight. BBTv rep gene have been cloned and sequenced and *in silico* analysis revealed 91.5-97.9% homology with amino acid sequences of South-Pacific group. Coat protein gene of CMV (Hosur and Jalgoan isolates) and complete genome of BSMysV infecting Poovan have been cloned and sequenced. CMV coat protein gene was cloned into pMAL-expression vector and transformed into *E. coli*. BSV integration pattern in Poovan plants were attempted using 40 RAPD, 2 SSR and 2 ISSR markers and one of the ISSR markers was found to be useful as it produced polymorphic bands. Primers designed for developing gene construct for multiple virus resistance yielded product of 200 bp for BBrMV, CMV and BBTv and were cloned in sense direction. The intergenic regions of six components of BBTv were amplified with designed primers and a promoter construct has been made with IR region of BBTv-5 by replacing the CaMV 35S promoter from pBI121 vector. ECS for Virupakshi and Poovan have been developed with high number of non-embryogenic cells. BBTv antisense replicase gene construct has been developed using pCAMBIA2301 binary vector.

Transfer of Technology

Technologies like chaffy grain formulation of EPF, *Beauveria bassiana* and EPN, *Heterorhabditis*

indica were released to the banana farmers. Three short training courses were offered on value added products and molecular detection of fungal and viral diseases. Technologies on artificial ripening of banana using Ethrel and banana juice and fig preparation have been transferred to two clients. Three television programmes and four radio talks were broadcasted during the period. The Centre participated in 12 exhibitions to disseminate the technologies to banana growers and stakeholders.

Linkages, Collaboration and HRD

The Centre has collaboration with CICY (Mexico), FAO (Rome), Department of Atomic Energy (GOI), DBT and NBPGR (New Delhi) for various research activities. The scientists of the Centre were deputed to trainings and pursue higher studies to update their knowledge in their respective fields of research. The technical personnel and other staffs were also sent for various training to enhance their capability. Under education and training, 47 graduate and post-graduate students of various universities have been guided for research project work on various aspects of banana.

Revenue Generation

Through three contract research projects, an amount of Rs. 5.73 lakhs have been generated to the Centre. A sum of Rs. 9 lakhs were generated for virus testing of banana samples by various tissues culture companies and other agencies and Rs. 40,000 was generated through trainings. A total of Rs. 4.5 lakhs was generated through sales of farm produce.



4 Introduction

The National Research Centre for Banana was established on 21st August 1993 at Tiruchirapalli, Tamil Nadu by the ICAR, New Delhi with an aim to increase the production and productivity of bananas and plantains through mission mode basic and strategic research approaches. This Centre is located at 11.50° N latitude and 74.50° E longitude, 90 m above MSL and receives 800 mm rain annually. The climate is warm and humid and the average min and max temperature are 25 and 35 °C respectively. Infrastructure facilities like library, ARIS Cell, meeting and exhibition halls, green houses, quarantine lab and net houses are also established in the Centre. The Centre's laboratories and administrative building are situated in an eight acre land near the farm and staff quarters have been constructed both in the office premises and also in the city. The Centre has a research farm with 36 ha land and has good ground water resources as well as canal water from Kavery river.

The Centre works on four major thrust areas of research viz., Crop Improvement, Crop Production, Post-Harvest Management and Crop Protection. With the funding during 8th, 9th and 10th five year plans and also through externally funded projects, the Centre has well-equipped laboratories for tissue culture, biotechnology, soil science, nutrient management, physiology and biochemistry, entomology, nematology, fungal, bacterial, viral pathology and post-harvest technology research.

In nineties, the Centre had more focus on collection of banana germplasm from primary and secondary sources and conservation in field gene bank. The genetic resources team has made 10 explorations to collect the wild banana germplasm from the North - Eastern states, Western Ghats and Andaman and Nicobar islands. At regular intervals, exotic banana accessions were also introduced from International Transit Center (ITC), Belgium through NBPGR, New Delhi. At present, the Center has reoriented its research priorities based on the QRT and different RAC recommendations. The Centre has completed seven in-house research projects and 11 are in progress in the 11th five year plan. In addition to Centre's in-house projects, 26 external projects funded by AP-Cess fund of ICAR, NATP, DBT, NHB and INIBAP were carried out. The Perspective Plan and Vision 2025 based on the research priorities and also inputs from QRT and RAC was published. The Centre conducts two meetings of Institute Research Council to review the on-going research projects and also to incorporate the RAC recommendations. The vision of the Centre

is to increase the production and productivity of bananas and plantains to meet the growing need in India.

The mandate of the Centre is:

- ❖ To undertake the basic and strategic research for developing the technology to enhance the productivity and utilization of banana
- ❖ To develop improved cultivars through traditional and biotechnological methods and conserve the diversity
- ❖ To serve as national repository of germplasm and information related to banana and plantain and also to disseminate the knowledge to improve the production and productivity
- ❖ To provide leadership and coordinate the network research for generating location specific variety technology and for solving specific constraints on banana and plantain production
- ❖ To collaborate with relevant national and international agencies in achieving the above objectives.

Salient Achievements

Crop Improvement

Up to 2007-'08, around 1100 accessions have been collected from both indigenous and exotic sources, which are maintained in the Centre's gene bank at Tiruchirapalli and the satellite gene bank at Agali. Banana field gene bank of the Centre is the 'National Repository' for banana in India. Collections were made through 10 explorations from all the regions of India. In the germplasm collection, two wild banana species from Andaman and Nicobar islands and a wild species viz., *Musa acuminata ssp burmannica* from TBGRI, Kerala have been added to the Centre's collection. The genomic status of collected accessions has been assigned based on their morphotaxonomic characterization using score card developed by Stover and Simmonds. The collections include AA, AB, BB diploids, AAA, ABB, AAB triploids, AAAA, AAAB, AABB, AB BB, AAAh tetraploid collections and Fe'i bananas. Among the collections, 92 accessions are highly resistant and 25 are resistant to Sigatoka leaf spot disease. The collected germplasm has been narrowed down to 310 by eliminating the synonyms using both morphotaxonomic and molecular markers viz., RAPD, IRAP and SSR. Three promising selections have been identified and being evaluated. NRCB-selection 1, Udhayam, which belongs to Pisang Awak sub group, is a high yielder, tolerant to Sigatoka leaf spot and nematodes. A protocol with



modified MS media without growth regulators for embryo culture has been standardized for Pisang Awak (ABB), Bluggoe (ABB), Pome (AAB), wild *Musa balbisiana*, *M. nagensium* and *M. ornata*. More than 12 hybrids have been developed through breeding, which are being evaluated under field condition. Embryogenic cell suspensions (ECS) for five different commercial varieties viz., Rasthali, Nendran, Ney Poovan, Robusta and Grand Nain have been developed and regeneration protocol from ECS for Nendran and Rasthali has been standardized. A Pisang Awak hybrid resistant to Sigatoka leaf spot has been identified. Different cultivars of Cavendish sub-group were distinguished using duplex RAPD markers and Thella Chakerakeli (AAA) could be differentiated from other AAA clones using RAPD primers. The Inter Retro transposon Amplified Polymorphism (IRAP) markers differentiated 57 accessions at both genomic and sub-group level. Putative diagnostic RAPD markers linked to Sigatoka and nematode resistance have been identified for early screening of hybrid progenies. FHIA-23 was found highly susceptible to pseudostem weevil in various multilocation trials. The carotenoid content in different bananas ranged between 86 and 1626 mg per 100 g pulp and the highest was recorded in Thiruvananthapuram (AAB).

Kanai Bansi, *Musa balbisiana*, Athiakol, Bhimkol and Aittakola were identified as resistant to burrowing and root lesion nematodes. Bhimkol (BB), Athiakol (BB), Elavazhai (BB) Sapkal (ABB), Dudhsagar (AAA), Pisang Lilin (AA) and Pisang Jari Buaya (AA) were resistant while Nendran was highly susceptible to pseudostem weevil (BSW).

Crop Production

Application of 25% N as FYM + 50% N as neem cake + 25% N as inorganic fertilizer increased the yield by 20 per cent in Rasthali, Poovan, Robusta, Monthan and Karpuravalli cultivars. Application of organic manures reduced the time taken for flowering, maturity and total crop duration in all cultivars. Weed free condition up to 9 months gave an additional income of Rs. 26,600/- in Karpuravalli banana. Poovan plants supplied with 20 liter water/day/plant and 75% N (150 g N/plant) as fertigation increased the yield by 20% with maximum net profit and a benefit cost ratio of 1.96. A combination of distillery sludge 2.5 kg + 1 kg vermicompost + 1 kg neem cake + 2.5 kg poultry manure per plant recorded the maximum growth and yield parameters in Rasthali and Karpuravalli bananas. The organic manure applied plants had less incidence of Sigatoka leaf spot disease while the inorganic treatment had severe incidence of leaf spot disease. Application of gypsum 2 kg/plant + FYM 15 kg/plant + 120% recommended K in saline

sodic soil increased the yield by 51% over control in Nendran and Rasthali bananas. Application of 15 kg rice husk ash or 5 kg poultry manure per plant resulted in an additional profit of Rs. 23,750/ha and Rs. 34,250/ha respectively in Poovan banana. Paired row planting system, which accommodated 4,500 plants/ha, increased the productivity and fruit quality with 75 per cent recommended fertilizers dose as fertigation in Robusta, Grand Nain and Red Banana. But, in first ratoon crop of Robusta and Grand Nain, the growth and yield parameters were significantly higher in conventional system than paired row planting. Application of VAM and phosphobacteria was found superior to *Azospirillum* for increasing the banana growth and yield parameters. Rice husk ash was superior as compared to vermicompost and poultry manure in increasing the bunch weight of Rasthali and also in the ratoon crop. Application of 15 kg rice husk ash + 25 g VAM + 80% recommended NPK gave an additional profit of Rs. 32,500/ha. Soil application of Fe and B along with foliar spray of Zn under high pH soil condition, increased the bunch weight of Ney Poovan banana by 43.5% over control, resulting in an additional net profit of Rs. 38,000/ha. Research on soil test based nutrient tailoring for banana has resulted in developing fertilizer adjustment equations for Rasthali and Ney Poovan. Photosynthetic activity was more in Rasthali at flowering stage than other commercial cultivars of banana. Eight drought tolerant accessions were identified based on leaf water retention capacity. The leaf longevity was more in Robusta than Nendran, Rasthali and Ney Poovan banana varieties.

Post-Harvest Technology

Pre-packaging in 400 gauge LDPE bags, low temperature storage, use of ethylene absorbents and pre-storage treatments have resulted in extension of shelf life up to 3 months in Robusta, Grand Naine, Rasthali and Ney Poovan bananas. Several value added products like flower *thokku*, peel *thokku*, fruit pickle, fig, biscuits, jam, ready to serve beverages and functional foods like *chapathi*, bread and health drink have been developed. Many of these technologies have been commercialized. Banana and Jamun juice blend was the best among the blends made with other fruit juices rich in antioxidants. Poovan juice blended in 1:1 ratio with beet root juice was found more acceptable. Pickle from the waste peels of Nendran having equivalent quality as that of flower pickle has been prepared, which could be stored for 8 months.

Crop Protection

The burrowing nematode (*Radopholus similis*) was present in few pockets of Tamil Nadu,



Maharashtra, Gujarat, Karnataka and Kerala, whereas the root-lesion nematode (*Pratylenchus coffeae*) and root-knot nematode (*Meloidogyne incognita*) were present in all banana growing states. Application of 500 g neem cake per plant reduced the root lesion nematode. Application of *Trichoderma viride* effectively controlled the root knot and root-lesion nematodes. Mass production technique for *Paecilomyces lilacinus*, (an egg parasite of root-knot nematode) using banana petiole and pseudostem has been developed. Flower extracts of *Tagetes erecta* was highly effective against nematodes. Application of *T. viride* and *P. fluorescens* were found superior in controlling the nematodes and increased the plant growth parameters of Robusta. Integration of *P. lilacinus* with either neem cake or *Tagetes* or *S. torvum* is useful for effective management of root-knot nematode.

Artificial diet has been developed for banana stem weevil. Swabbing 0.06% Chlorpyrifos 20 EC on the pseudostem to a height of 1.2 m during 5th and 8th months completely controlled BSW. Treating suckers with Monocrotophos 36 EC (14 ml/liter) followed by soil application of Carbofuran 3G at the rate of 30 g per plant each at 4th and 7th months after planting found effective against corm weevil. Pseudostem split trap swabbed with chaffy grain formulation of *Beauveria bassiana* trapped the stem and corm weevils better than traps swabbed with maize flour formulation. Banana pseudostem split traps swabbed with 25 ml solution containing EPNs such as *Heterorhabditis indica*, *Steinernema glaseri* and *S. abbasi* trapped 63.12% and 36.8% stem and corm weevils respectively and the mortality was high (44%) due to *H. indica*. *B. bassiana* (isolate 5) collected during survey gave 83.3% mortality of weevils. Banana leaf sheath volatiles eluted with hexane and methanol gave EAG response.

Diseases such as wilt, *Erwinia* rot, Sigatoka leaf spot and peduncle rot (5 to 25 %) were prevalent in all banana growing states. Septoria leaf spot (*Septoria eumusae* = *Mycosphaerella eumusae*), eye spot (*Drechslera* sp.) and pitting disease were recorded for the first time in India. A new wilt like disease caused by Triclotmataceae fungus of Basidiomycetes has been identified. 141 nit mutants of *Foc* were developed from 100 *Foc* isolates and 9 different VCG have been identified. Cross reaction between race 1 and race 2 of *Foc* has been observed in VCG analysis which was confirmed in pot culture experiment. Diversity of *Foc* isolates has been studied using RAPD, PCR-RFLP analysis of IGS region and the sequence analysis of rDNA-ITS region. Different isolates of *Colletotrichum musae* have been characterized morphologically and based on amplicon size of rDNA-ITS region. Screening of germplasm and entries from International Musa

Testing Programme revealed 17 accessions as highly resistant to Sigatoka leaf spot disease. A fusaric acid detoxifying strain of *Pseudomonas fluorescens* was isolated. Propiconazole (0.1%) or Hexaconazole (0.1%) alternated with Chlorothalonil (0.25%) controlled Sigatoka leaf spot disease and increased the yield significantly. Anthracnose disease of banana was controlled by spraying of 25% percent leaf extract of *Solanum torvum*. Application of *Trichoderma viride* (10^9 /ml) (or) *Pseudomonas* spp (10^6 /ml) (or) *Bacillus* spp. (10^6 /ml) (or) Propiconazole (0.1%) spray was also effective in controlling the disease anthracnose. Three applications of *T. harzianum*, *P. fluorescens* and *B. subtilis* each 10 g per plant at the time of planting, 3rd and 5th months after planting significantly reduced the wilt incidence. *P. aerogenosa* and *P. viridiflavus* were effective in controlling crown rot disease. Population of *T. viride* mass multiplied in rice chaffy grain was stable even after 6 months of storage.

Viral diseases viz., Banana Bunchy Top (BBTV), Streak (BSV) and Infectious Chlorosis (CMV) were present in the entire banana growing areas, while Bract Mosaic (BBrMV) was restricted to South India. Recent surveys indicated widespread occurrence of BBTV and BBrMV in Coorg and Wyanad districts. A yield loss of 49 per cent due to BSV was recorded in Poovan. Three aphid vectors including *Pentalonia nigronervosa* transmitted BBrMV and mealy bug vector *Ferrisia virgata* transmitted BSV. All the banana viruses could be detected from their vectors by either PCR or RT - PCR. Polyclonal antiserum to BBTV was produced and ELISA technique has been standardized for detection. NA probe based and PCR based diagnostic techniques have been developed for all the banana viruses. A multiplex PCR technique has been developed for detecting three banana viruses simultaneously. A quick PCR based technique has been developed to detect the EPRV's present in the host genome. Complete genome of BSV infecting Poovan has been cloned and sequenced which revealed that the virus is closely related to BSOLV, but there was a deletion of 450 bp in the genome. Promoter sequences from BBTV were cloned and sequenced. Both sense and antisense BBTV cp gene constructs and a BBTV replicase construct have been prepared in pBINAR /pCAMBIA 2301 vectors respectively for transformation. Embryogenic calli has been obtained for Hill banana and Poovan for transgenic research. Dig-labeled RNA probes have been prepared and assessed for detection of BBrMV by NASH.

Transfer of Technology

Suckers and tissue culture plants of NRCB variety, 'Udhayam' were supplied to the progressive



growers and tissue culture companies. Bunchy top virus indexed Hill banana plants have been supplied to the hill banana growers of lower Pulney hills. A virus testing lab was developed based on NRCB technology at BTC, Dept of Horticulture, Government of AP, Hyderabad. Virus testing of mother plants and tissue cultured plants from different tissue culture industries is done on contract service mode. Technologies on value added products were transferred to several clients. Virus indexing trainings were imparted to technical personnel of many tissue culture companies, scientists, assistant professors and students involved in banana research. Banana value added products training were offered to 'Mahabanana' personnel, Maharashtra and other beneficiaries. The Centre has participated in many exhibitions to disseminate the technologies to the stakeholders. Radio talks on different production technologies of banana were broadcasted and 22 video programmes were recorded by Department of Agribusiness Management, Ministry of Agriculture, New Delhi for dissemination of the technology at national level. DBT has recognized and accredited the molecular virology and tissue culture laboratories for testing banana viruses and genetic fidelity under Accredited Test Laboratory (ATL) under the National Certification System for Tissue Culture raised Plants (NCS-TCP).

HRD and Education

The scientists have been deputed regularly to undergo training and to pursue higher studies to update their skill and knowledge. The technical personnel and other staff members were also deputed for various training to enhance their

capability. Under education and training, over 300 M. Sc., B. Tech. and M. Tech. students belonging to different universities have been guided for their project work in various aspects of banana. The science day is regularly celebrated and many school children have been invited on that occasion to show cause the research on banana.

Linkages and Collaboration

The Centre has developed good linkages with international institutes viz., INIBAP, France; CIRAD, France; KUL, Belgium; IAEA, Austria and QDPI, Australia. It collaborates with different national research institutions for different activities viz., NBPGR, New Delhi; BARC and CIRCOT, Mumbai; IICT, Hyderabad; IIHR, Bangalore; NHB and DBT New Delhi and all SAUs. Tissue culture industries involved in banana mass propagation, farmers, exporters, banana federations, State Horticulture and Agriculture departments and self-help groups are linked with the Centre for various research and developmental activities. NRCB also co-ordinates with AICRP (Tropical Fruits) Centers working on banana. The new technical programmes for banana under AICRP-TF were made in consultation with NRCB scientists.

Revenue Generation

A gross revenue of Rs. 6.13 lakhs was generated from consultancy, contract research projects, contract services and trainings. A total of Rs. 9 lakhs have been received for virus indexing of tissue culture plants from various tissue culture companies and other agencies. This year through the sales of farm produce, the Centre has generated Rs. 4.5 lakhs.

BUDGET DETAILS (REVISED ESTIMATE) FOR THE YEAR 2007-'08

PLAN		
Sl. No.	Head of Account	Amount (Rs. in lakhs)
1	Pay & Allowances	0.00
2	Travelling Allowances	4.00
3	Human Resource Development	3.00
4	Contingencies	70.00
5	Equipments	45.00
6	Works	30.00
7	Library books	2.00
TOTAL		154.00

NON PLAN		
Sl. No.	Head of Account	Amount (Rs. in lakhs)
1	Pay & Allowances	115.00
2	Travelling Allowances	3.00
3	Overtime Allowances	0.10
4	Contingencies	36.15
5	Works (Maintenance)	5.75
TOTAL		160.00

5 Research Achievements

5.1 CROP IMPROVEMENT

5.1.1 Genetic Resources Management

5.1.1.1 Exploration and collection

During the reporting period, an exploration was conducted in the eastern slopes of Western Ghats including Tirunelveli and Nagarcoil districts to study the genetic wealth of *Musa* and its allied genus *Ensete*.

5.1.1.2 Occurrence of wild *Musa* species

The present exploration has confirmed the occurrence of *Musa acuminata* ssp. *burmannica*. This exploration has resulted in the addition of 14 new accessions to the NRCB field genebank including wild and landraces.

5.1.1.3 Evaluation of cryopreserved germplasm

Cryopreserved Sommarani Monthan supplied by NBPGR was regenerated under *in vitro* conditions and evaluated under field conditions along with tissue culture cryo control and conventional sucker. Both growth and yield parameters were recorded for the plant crop (Table 1). Results indicated that no significant differences existed for the major growth and yield parameters.

Simultaneously, microsatellite polymorphism was assessed in the cryopreserved Sommarani

Monthan using the same controls under field conditions along with a green mutant as references. Among the nine primer pairs tested, seven primers pairs namely AGMI-33,34, AGMI-35,36, AGMI-67,68, AGMI-93,94, AGMI-95,96, AGMI-129,130 and MbSSR 1-149 amplified products resulting in discrete repeatable amplicons and two primer pairs viz., AGMI-103,104 and MbSSR 1-146 failed to produce any amplification. From the seven primers pairs considered for genetic analysis, a total of 13 alleles were identified with a mean of 1.86 alleles per primer. All the alleles were found to be monomorphic. This indicated that the cryopreserved material was genetically stable and neither cryopreservation nor regeneration after a time lag was able to cause genetic changes.

5.1.2 Characterisation

5.1.2.1 Udhayam DNA fingerprinting

A total of 12 primer pairs were used to assess the microsatellite polymorphism in the NRCB released variety 'Udhayam' using local Karpuravalli, Kanthali and Ankur II as reference cultivars. All the 12 primer pairs tested (100%) amplified products resulting in discrete, reproducible amplicons and were considered for the genetic analysis. From the 12 primer pairs that produced discrete and repeatable amplicons, a total of 39 alleles were identified with a mean of 3.25 alleles per primer pair (Fig. 1). Out of 39 alleles scored, 10 were monomorphic (25.64%) and the rest were polymorphic (74.36 %). Two alleles were unique to Udhayam and their approximate sizes were 150 bp (AGMI 67/68) and 290 bp (MbSSR 1-146) respectively. These alleles could be used as markers to confirm the identity.

Table 1. Growth and yield characters of Sommarani Monthan

Sl. No.	Parameter	TC control	Cryo control	Cryo material	Conventional sucker	MSE	C.D @1%	CV %
1	Pseudostem height (cm)	275.00	281.75	282.00	282.50	8.45	6.68	1.04
2	Pseudostem girth (cm)	62.00	61.50	60.75	62.00	5.45	NS	3.79
3	No. of leaves at shooting	13.75	15.00	15.50	14.75	0.33	1.32	3.89
4	Petiole length (cm)	62.75	63.00	63.75	61.25	1.12	2.43	1.69
5	Leaf area (sq. m)	14.65	15.97	16.93	15.98	345.32	1.35	3.70
6	Shooting (days)	362.25	366.50	363.50	366.00	67.90	NS	2.26
7	Bunch maturation (days)	115.00	115.00	113.75	113.00	22.78	NS	4.18
8	Total duration	477.25	481.50	477.25	479.00	42.00	NS	1.35
9	Bunch weight (kg)	15.62	16.25	15.62	15.75	0.26	NS	3.23
10	No. of hands/ bunch	8.75	8.50	8.50	8.75	0.36	NS	6.96
11	No. of fruits/ hand	13.25	13.75	13.75	13.25	0.28	NS	3.92
12	Total no. of fruits	116.00	116.75	116.75	116.00	81.25	NS	7.75

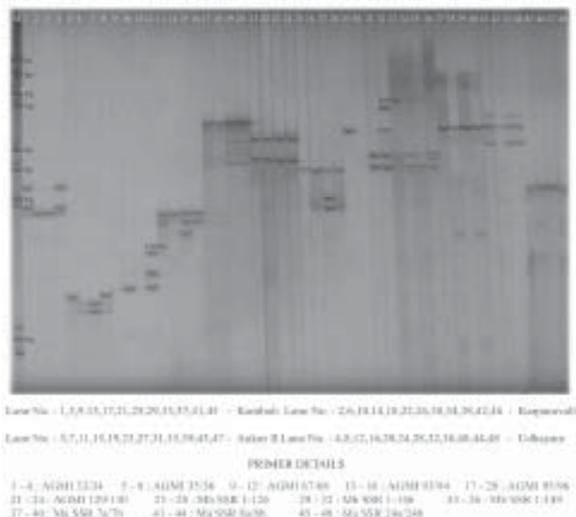


Fig.1. Micro satellite polymorphism obtained for the newly released variety Udhayam

5.1.2.2 Cluster analysis

Analysis of the dendrogram indicated two different clusters. Cluster I includes Udhayam and Kanthali sharing 70% similarities while cluster II includes local Karpuravalli and Ankur II sharing 60% similarities. However, cluster I and II shared less than 55 per cent similarities (Fig. 2).

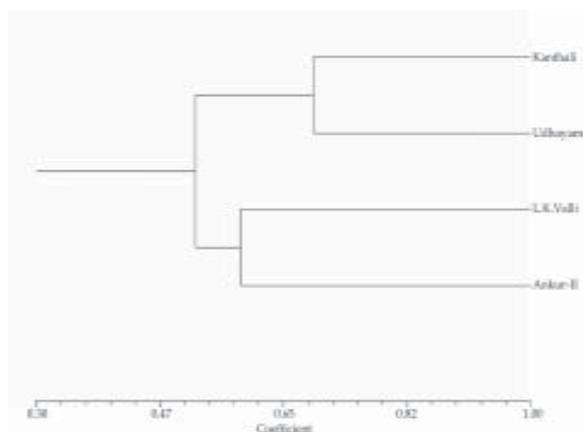


Fig. 2. Dendrogram showing the microsatellite polymorphism of the released variety Udhayam based on Simple Matching Coefficient

5.1.2.3 Core collection assessment using IRAP markers

A second set of 57 accessions were characterized using six IRAP primer combinations. The robustness of the core collection at NRCB, Trichy. The genetic similarities of the tested accessions ranged from 58-99% and are grouped into two clusters at 58% similarities (Fig. 3; Table 2).

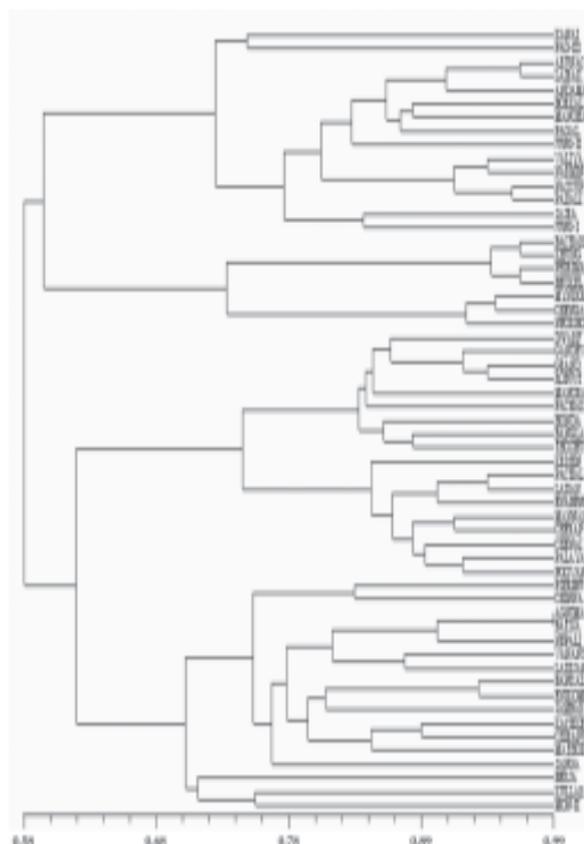


Fig. 3. Dendrogram showing the retrotransposon polymorphism of the core collection accessions based on Simple Matching Coefficient

Cluster - I

Twenty two accessions had fallen in the cluster I, which was divided into two sub-clusters namely cluster Ia and cluster Ib for easy interpretation.

Cluster - Ia

It consisted of all the diploids (AA, AB, and BB) with similarities ranging from 60-97%. Kanai Bansi (AA), a parthenocarpic diploid grouped with a wild diploid, Pagalaphad wild and shared 74% similarities. Arunachal Pradesh wild (AA) and Lairawak (AA) collected from north eastern India grouped with *M. acuminata* wild from Andaman and Nicobar islands and shared 91-97% similarities, indicating geographical isolation did not have significant effect on the genetic composition. Among the eight BB types tested, four accessions viz., Borkal Baista, Manohar and Pagalaphad wild collected from north eastern India and Jungle Kela II collected from Andaman and Nicobar islands grouped together with 86% similarities and joined with aforesaid *M. acuminata* types. This indicated more genetic affinities with each other than with other *balbisiana* types. These genetic affinities could be utilized in conventional breeding program for the development of "AB" hybrids. Sasra Bale and Jungle Kela I clustered with 84% similarities. Though

**Table 2. Cluster composition**

Clusters	Name of the accessions
Ia	Kanai Bansi (0064), Pagalapahad wild-III (1183), Acuminata wild Arunachal Pradesh (1731), Lairawak (1019), Acuminata Andaman wild, Borkal Baista (0018), Manohar (0047), Pagalapahad wild (1182), Jungle Kela II (1913), Valiya Kunnan (234), Narmine (0369), Nattupoovan (0186), Padalimoongil (0482), Sasra Bale (0449), Jungle kela I (1912).
Ib	Bacharia malbhog (0446), Khungsang wild (1168), Phirima wild (1186), Beeji Kela (1914), Myndoli (0138), Chengalikodan (0701), Neokhom (0731).
IIa	Dwarf Cavendish (0165), Gandevi collection (0370), Grandnaine (0580), Robusta (0200), Manoranjitham (1419), Pachakappa (1065), Honda (0057), Bangladesh Malbhog, Thozhuvan (0700), Krishnavazhai (0023), Pachaladan (0190), Ladan (0242), Ennabenian (0489), Mannan (0499), Cherapadathi (0160), Cheni Champa (0015), Palayankodan (0192), Poovan (0197).
IIb	Pisang Seribu (0579), Chinali (0483), Agnimalbhog (0059), Battisapiro (0089), Nepalivannan (0117), Vananthpurani (0354), Ladisan (0421), Bankela (0453), Enikomban (0732), Sambrani Monthan (0411), Kachkel (0016), Chirapunji (0163), Mathok grang (0429), Singalaji (0137), Bersain (0090), Kullarkanai (0435), Monthan II (0479).

phenotypic variations present in all the AB types, they clustered with 90% similarities and were placed in between the BB types, proving their close genetic relationship.

Cluster - Ib

In cluster *Ib*, four BB types were grouped in one sub-cluster and all the three plantain types in another. The results suggest that though plantain sub-group expressed phenotypical traits of *M. acuminata*, they were genetically closer to BB types.

Cluster - II

All the triploids (AAB and ABB) have grouped in cluster II with 60-92% similarities with two sub-clusters.

Cluster -IIa

All the AAA and AAB accessions have grouped in this cluster *IIa*. All the AAA types including Dwarf Cavendish, Giant Cavendish, Manoranjitham, a scented banana and Pachottan have grouped with soft and sweet table varieties of “Silk” sub-groups. This shows that the Silk sub-group had more genetic relatedness with “AAA” types. In general “AAA” and “AAB” Silk sub-groups expressed more than 85% similarities.

Mysore subgroup (AAB) and Pome subgroup accessions have grouped in one cluster and shared 82% similarities, which confirmed their genetic relatedness. Both Pome and Mysore sub-group members are comparatively female fertile. This might be one of the reasons for clustering in one group. Krishnavazhai, a Pome type, is little away not only from Mysore sub-group but also from its own sub-group proves its unique morphological appearance *i.e.*, black coloured pseudostem.

Cluster - IIb

Consisted of all the ABB types including, Monthan, Karpuravalli and unique ABB types. In addition, two AAB unique types have joined this group with 75% similarities. Five Pisang Awak sub-group members have grouped with an average similarity of 81-91%. All the Monthan types have grouped with 84-90% similarities, in which two accessions from Pisang Awak sub-group have also grouped. Small fruited ABB Bluggoe types *i.e.* Beula, Kullarkanai and Monthan II grouped in the cluster *IIb*, which proved their uniqueness among ABB types. Grouping of AAB unique in this cluster *IIb* needs clarification.

In general, all the test accessions had clustered according to their genomic groups and proved their individuality by showing a minimum of 4-24% dissimilarities. These results confirmed that IRAP markers could be exploited for detecting variability and eliminating synonyms in banana gene bank.

5.1.2.4 Variability within Cavendish sub-group

The insertional polymorphisms of the retro-elements and RAPD were studied in nineteen Cavendish (AAA) group accessions from the core collection of NRCB, Trichy and subjected to diversity analysis. Ten published IRAP primer combinations and 30 decamer RAPD primers were used in the present study to assess the robustness of markers IRAP and RAPD in studying the intragroup diversity. The average polymorphism exhibited by RAPD was 67.77% and IRAP was 70% indicating that there was substantial variation at the DNA level among the nineteen Cavendish group accessions. Result of RAPD and IRAP reiterates the grouping of accessions from the same geographical origin is quite common. Unusual grouping of members of two



different Cavendish subgroups namely Gros Michel and Madhukar has been commonly observed in both IRAP and RAPD. Likewise 2390-2 from ITC, Belgium was unique due to by its independent clustering in both IRAP and RAPD. Singapuri and Dwarf Cavendish were morphologically similar due to dwarf stature, which was proved in IRAP by their grouping in the same cluster. But RAPD has placed morphologically similar accessions in two different clusters. Similarly tissue culture variants of common somaclonal origin from Taiwan namely GCTCV 119 and GCTCV 215 have been grouped together in IRAP, but they were placed in two different clusters in RAPD. Hence IRAP depicted a better picture of the diversity indicating its suitability to study the genetic relatedness than RAPD (Fig. 4 and 5).

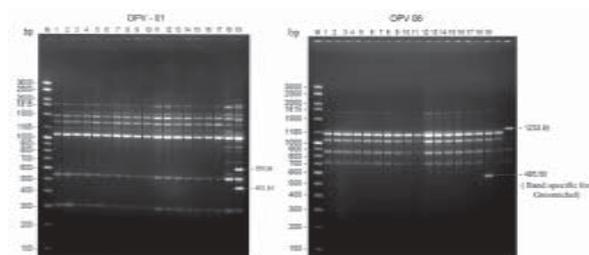


Fig. 4. DNA polymorphism for Cavendish clones (AAA) using RAPD - primers

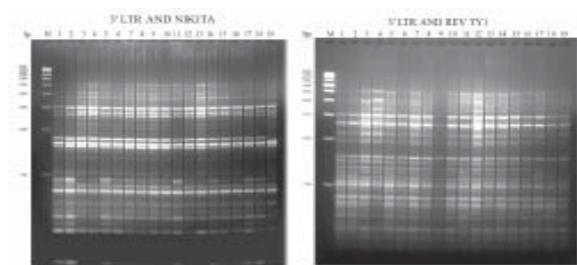


Fig. 5. DNA polymorphism for Cavendish clones (AAA) using IRAP - primers

5.1.2.5 Seed DNA for medium term conservation

For routine molecular works, availability of fresh leaf from wild species has always been a problem.



Fig. 6. 80% mature banana seeds

To overcome this, a novel protocol was developed and validated to use seed DNA for medium conservation strategy. Sixty percent mature seeds with liquid state endosperm and devoid of seed coat was found to be the best source of DNA (Fig. 6).

5.1.2.6 Genotypic expression of the phenotypic trait waxiness

Morphotaxonomic and molecular characterization using SSR markers, failed to detect the genetic expression of the trait waxiness in banana. Waxiness trait dominates over other morphological traits thus resulting in delineating green-fruited varieties from ashy types among culinary types. Twenty five out of 40 primers produced more than 5 polymorphic bands of which 4 primers were considered as indicative primers (OPA-20, OPB-04, OPB-05 and OPC-06) of trait waxiness (Fig. 7 and 8). Results suggested that RAPD method is best for differentiating waxiness, which is a general indicator for cold tolerance trait in crop plants.

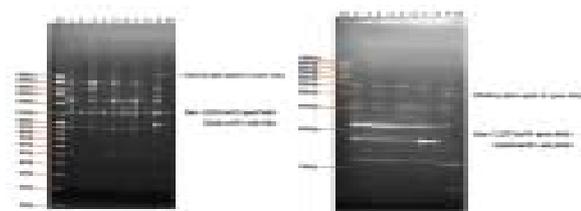


Fig. 7. RAPD polymorphism for green and its ashy mutant using primers OPB-5 & OPC-6

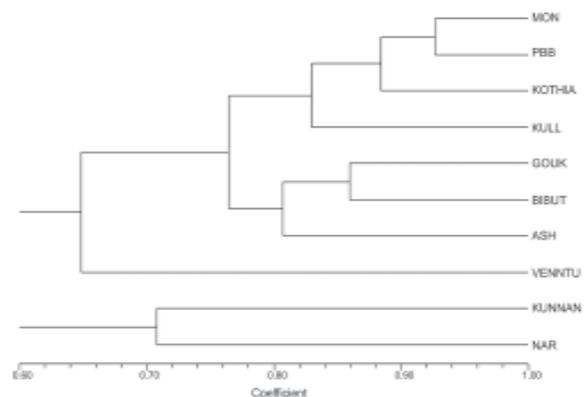


Fig. 8. Dendrogram showing the phylogenetic relatedness between green and their ashy mutants

5.1.3. Classical Breeding in Banana

5.1.3.1 Evaluation of hybrids

Among eleven progenies evaluated under field conditions, four were found to be parthenocarpic, three were seeded and rest of them yet to complete the crop cycle. All the seeded progenies had Pisang Jajee as male parents with an extended crop duration of more than 450 days. Plants were taller with > 2 m height and robust with > 60 cm girth with comparatively larger leaf area.



Hybrid for leaf industry

The hybrid progeny of the cross Anaikomban x Pisang Jajee (Hybrid 07/05) produced ill-filled fruits, but it was found promising with respect to leaf traits like area, unusual width (90 cm) and absence of tapering ends (Fig. 9). These traits along with its freeness from leaf spot diseases could be potentially exploited for the commercial leaf industry.



Fig. 9. Hybrid (Anaikomban x Pisang Jajee) suitable for leaf industry

5.1.3.2 Improvement of Rasthali through induced mutagenesis

Attempts were made to determine the LD_{50} of Ethyl Methane Sulphonate (EMS) for three different explants (shoot meristems, proliferating buds and embryogenic cell suspension) of cv. Rasthali.

Fifty floral buds of cv. Rasthali have been initiated in the callus induction medium towards the development of ECS, the basic *in vitro* system required for induction and selection of mutants without the problem of chimeras. ECS were treated with EMS at concentrations ranging from 0.02 – 0.1% uniformly for two hours. Among the various treatments tried, EMS at 0.02% and 0.04% for 2 h showed good signs of regeneration.

Proliferating buds obtained after second subculture were treated with EMS at doses ranging from 0.2 – 1.0% for different incubation periods viz., ½, 1, 1 ½ and 2 h. Proliferating buds could not sustain the EMS treatment beyond 0.8%.

When the FWG% of the shoot meristems was used in the derivation of simple linear regression

equation, there was no correlation and a negative correlation could be obtained only beyond 0.6%. This suggests that the concentration of EMS needs to be increased beyond 1.0% to determine the LD_{50} of EMS in cv. Rasthali.

From the preliminary studies, it is concluded that the concentration of mutagen and incubation time is cultivar specific and hence needs to be established in Rasthali. The method of treatment also has a significant effect on the mutagen efficiency and hence needs investigation.

5.1.3.3 Pollen studies

Comparative studies were undertaken for pollen shape, size, fertility (stainability) and germinability for selected accessions. A total 37 accessions of diploid, triploid and tetraploid clones have been studied (Table 3). Among the various methods of pollen preparation, the method followed by Wodehouse (1935) was found suitable for banana pollen. Pollen size varied with the ploidy and genome and within a variety also, there was wide variation in pollen size. Besides, pollen size was found to increase significantly under wet condition. Pollen content of banana varieties was estimated using 'Haemocytometer' technique and the optimum sample size was standardised. Pollen out put per anther for varieties comprising six different genomes (AA, AB, AAA, AAB, ABB and ABBB) was estimated. Pollen content as influenced by the age of the male bud was studied and the flowers in the 10th node of the male rachis produced large quantities of pollen per anther.

During the course of study, a male sterile form of 'Octoman' (ABB) was spotted and the abnormality leading to pollen degradation. Besides cultivars Ney Poovan (AB) Poovan (ABB) and Peykunnan (ABB) were non-polleniferous, while cultivars Balukpong (AA), Kothia (ABB), Birbutia (ABB) and Chakkia (ABB) were identified as high polleniferous. Pollen fertility as measured by their stainability was found to vary with the ploidy. Most of the edible triploids and tetraploids were over 50 percents indicating their breeding potentiality. Besides many triploids were found to have varied fertility in relation to age of the bud but showed no significant difference up to 35th node of the male rachis. Sucrose solution (10 percent) and boric acid (10 ppm) was found suitable for pollen germination, which gave best results. The cultivars AA diploids, AAA triploids and ABB triploids showed germination percentage ranging from 30.2 to 36.4, 8.6 to 14.9 and 12.4 to 24.6 respectively. Pollen germinability did not vary significantly with the age of the bud.



Table 3. List of the varieties for pollen output, stainability and germinability

Sl. No.	Variety	Nodal position	Pollen output		Stainability % (Acetocarmine)	Germinability %
			Per anther	Per flower		
1	Anai komban(AA)	9	10516	52580	69.7	34.8
2	Balukpong Wild (AA)	11	19473	97365	62.2	36.4
3	Calcutta-4 (AA)	16	14653	73265	68.7	32.6
4	Namarai(AA)	12	4578	22890	56.5	30.2
5	Ney Poovan (AB)	17	Nil	-	-	-
6	Bharat Moni (AAA)	14	7314	36570	52.2	14.9
7	Manjahaji (AAA)	16	4950	24750	55.6	12.4
8	Highgate (AAA)	14	11000	55000	52.4	9.7
9	Karpura Chakkarkeli(AAB)	12	7500	37500	72.1	18.6
10	Kottavazhai (AAB)	19	7750	38750	42.7	22.4
11	Pisang Seribu (AAB)	21	5217	26085	54.6	20.2
12	Soniyal (AAB)	35	4568	22840	30.9	12.6
13	Thozhuvan (AAB)	22	5517	27585	44.9	21.3
14	Madhuranga (AAB)	29	4217	21085	26.6	16.2
15	KallaLadan (AAB)	35	15416	77080	33.2	15.4
16	Khozhikodu (AAB)	28	3714	18570	29.7	19.1
17	Borchampa (AAB)	24	8154	40770	41.2	14.8
18	Poovan (AAB)	20	Nil	-	-	-
19	Peykunnan (ABB)	17	Nil	-	-	-
20	Ashy Batheesa (ABB)	12	5438	27190	33.6	22.4
21	Bersain (ABB)	18	4217	21085	27.8	16.4
22	Birbutia (ABB)	10	18414	92070	24.6	20.4
23	Monthan (ABB)	14	4568	22840	22.6	19.1
24	Octoman (ABB)	17	2144	10720	34.2	8.4
25	Kallumonthan (ABB)	23	4576	22880	30.4	12.8
26	Bathesa Ash (ABB)	16	4167	20835	23.5	22.4
27	Kothia (ABB)	23	19617	98085	31.3	22.4
28	Nutepong (ABB)	12	7676	38380	33.1	16.6
29	Chakkia (ABB)	24	18744	93720	32.2	24.6
30	Kaitkhullung (ABB)	26	17143	85715	26.8	12.4
31	Goukar (ABB)	29	16314	81570	34.6	16.2
32	Ennabenian (ABB)	27	3157	15785	27.8	18.4
33	Gauria (ABB)	30	14614	73070	26.2	19.6
34	Nepali Chinia (ABB)	18	13716	68580	33.1	17.3
35	Ginde (ABB)	17	14316	71580	30.4	12.8
36	Klue Tearod (ABBB)	32	10614	53070	54.1	30.4
37	Neyvannan Sawai (ABBB)	18	32450	162250	24.5	32.5

5.1.4 Molecular Breeding

5.1.4.1 Development of somatic embryogenesis

All the three cultivars namely Robusta, Grand Nain and Ney Poovan produced callus in induction medium. Nature of callus produced varied with cultivar. To improve the embryogenic response in the selected varieties, various treatments were tried.

Growth regulators like picloram, zeatin, malt extract and glutamine were added along with other components in the callus induction medium (Table 4, 5 and 6).

Irrespective of the medium tested, all three cultivars produced callus but ideal callus was obtained only in Robusta (Table 7). Whereas Grand Nain and Ney Poovan failed to produce ideal callus. Embryogenic callus with many translucent embryos were transferred to liquid medium.

Table 4. Effect of picloram on callus induction

S. No.	Cultivar	No. of explants	Picloram mg/l	Total	Response in callogenesis		
					+++	++	+
1	Robusta	15	0.25	2	-	1	1
		15	0.5	3	1	1	1
2	Grand Nain	15	0.25	1	-	-	1
		15	0.5	2	-	2	-
3	Ney Poovan	15	0.25	-	-	-	-
		15	0.5	1	-	1	-

Table 5. Effect of zeatin on callus induction

S. No.	Cultivar	No. of explants	Picloram mg/l	Total	Response in callogenesis		
					+++	++	+
1	Robusta	15	0.25	1	-	-	1
		15	0.50	1	-	-	1
2	Grand Nain	15	0.25	1	-	-	1
		15	0.50	1	-	1	-
3	Ney Poovan	15	0.25	-	-	-	-
		15	0.50	-	-	-	-

Table 6. Effect of glutamine and malt extract in callus induction

S. No.	Cultivar	No. of explants	Response in callogenesis			
			M4	M5	M6	M7
1	Robusta	5	+	++	+	-
2	Grand Nain	5	+	+	+	-
3	Ney Poovan	7	+	++	+	-

M4- MA1+150g/l Glutamine+100mg/l Malt extract

+++ - Embryogenic callus.

M5- MA1+150g/l Glutamine+150mg/l Malt extract

++ - Friable callus.

M6- MA1+200g/l Glutamine+100mg/l Malt extract

+ - Compact non embryogenic callus.

M7- MA1+200g/l Glutamine+200mg/l Malt extract

Table 7. Status of somatic embryogenesis in commercial cultivars

S.No.	Cultivars	No. of buds initiated	Callus obtained	Ideal callus	Suspension initiated	Regeneration of plantlets
1.	Robusta	65	✓	✓	✓	-
2.	Grand Nain	65	✓	-	-	-
3.	Ney Poovan	67	✓	-	-	-

5.1.4.2 Field evaluation of cvs. Rasthali and Nendran developed from embryogenic cell suspension

Cvs. Rasthali and Nendran plantlets developed from embryogenic cell suspension were planted in the farmers' field for assessing their genetic stability and 80% survival was observed. The field trial is in progress and growth parameters like height, number of leaves produced, their length and breadth are being recorded at regular intervals of one month.

5.1.4.3 *Agrobacterium* mediated transformation

Immature male flower buds of cv. Rasthali were inoculated in MS medium supplemented with 2, 4-D, IAA and NAA for induction of calli. The friable calli obtained after eight months of culture were suspended in liquid MS medium containing 2, 4-D and maintained at 80 rpm for three months for the establishment of ECS. The cell aggregates in log phase of growth was the target tissue used for *Agrobacterium* infection. The binary plasmid vector pCAMBAR chi 11 harboring chitinase gene under the influence of maize ubiquitin promoter was mobilized in the *Agrobacterium* strain LBA 4404 and then used. The vector possesses npt 11 as bacterial selectable marker, hpn as plant selectable marker and gus as reporter gene (Fig. 10). The cells were co-cultured for 4 hours and then transferred to the regeneration medium containing Cefotaxime and Basta for selection. Though 25-30 embryos have successfully germinated per petriplate, only 5-8 are likely to regenerate into plantlets and they are in various stages of development.

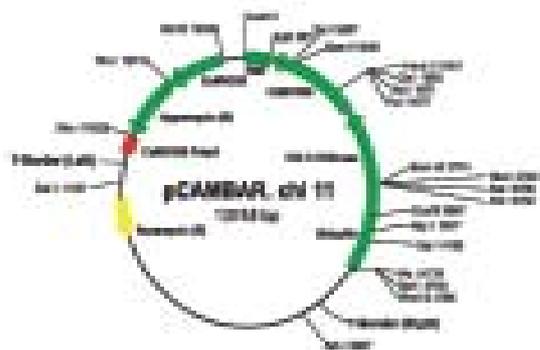


Fig. 10. Schematic diagram of the T-DNA region of the binary plasmid pCAMBAR. chi11 containing the chi 11, PAT genes. RB / LB -right / left border sequences; PAT, under the control of the maize ubiquitin promoter; PAT-coading region conferring resistance to the PPT, under the control of CaMV35S promoter

5.1.4.4 Development of markers for Sigatoka resistance

Two Sigatoka resistant varieties viz., Calcutta 4 and Manoranjitham and susceptible varieties

Anaikomban and Grand Naine were screened using 102 RAPD markers. But only 22 primers (21.6 %) produced polymorphic bands. Among 22 primers considered for the genetic analysis, only seven primers viz., OPC-4, OPC-10, OPD-7, OPD-12, OPK-1, OPK-11 and OPZ-13 produced bands specific to resistant varieties while 12 primers produced bands specific to susceptible varieties but three primers viz., OPD-16, OPK-16 and OPK-18 produced bands specific to both resistant and susceptible varieties but of different molecular weights. Two putative bands of size 1.3 kb (OPC-4) (Fig. 11) and 1.5 kb (OPK-1 and OPK-11) were identified.

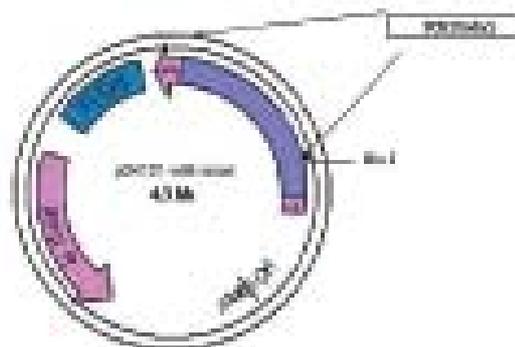


Fig. 11. Putative marker for Sigatoka resistant with primer OPC - 4 (1.3 kb) cloned and sequenced for conversion into SCAR marker

Among these two, 1.3 kb fragment was eluted, column purified and cloned in pDK 101 vector between *Xcm* I sites and *Nco* I sites. The clones were confirmed by release of insert using *Nco* I restriction endonuclease. The cloned fragment was sequenced bi-directionally using T 7 and SP 6 (vector specific) primers. The sequence was blasted in NCBI and it was found to have 76% homology with *Vitis vinifera* and *Citrus*. Hence, the SCAR marker was designed for the sequenced fragment was synthesized using Primer 3 software and is being validated both in the parents and their progenies.

5.1.4.5 *In-vitro* screening of banana cv. Rasthali for salt stress

A preliminary study was undertaken to screen the commercial varieties like Robusta and Rasthali for their salt tolerance using shoot meristems as explants. Shoot meristems were multiplied and rooted in the MS medium containing salt at various concentrations namely 0.25, 0.50, 1.0 and 2.0%. Explants of both cultivars failed to survive in salt concentrations at 1.0% and above. While negative correlation was observed with the number of leaves and roots produced at 0.25% and 0.5% NaCl compared to control. Higher the salt stress, there was a delay in root and leaf emergence and their number in both the test cultivars. Data collected using physiological parameters showed that Robusta explants are more adaptable to salt stress

Table 8. Effect of salt stress on various growth parameters after a period of 25 days

S.No	Cultivars	Concentration of NaCl	No. of leaves produced	No. of roots produced	% Change in fr. wt.	Enzyme activity (units/ml)	
						Peroxidase	Polyphenol oxidase
1.	Rasthali	Control	1.71	1.42	100%	0.07	0.007
	Robusta	Control	3.14	2.28	100%	0.048	0.005
2.	Rasthali	0.25%	1.71	1.3	32.8% Gain	0.058	0.009
	Robusta	0.25%	2.14	1.8	33.6% Gain	0.06	0.006
3.	Rasthali	0.5%	0.85	1.0	10.0% Loss	0.042	0.006
	Robusta	0.5%	1.0	1.42	19.0% Loss	0.050	0.007

when compared to Rasthali. Present results were also supported by enzymatic assay, which showed that peroxidase and polyphenol oxidase activity were more in Robusta as compared to Rasthali (Fig. 12; Table 8).

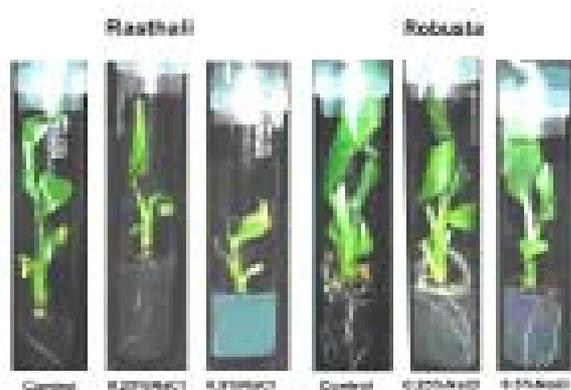


Fig. 12. *In vitro* screening of banana cv. Rasthali for salt stress

5.1.4.6 Standardization of RNA isolation

Surface sterilization with 1% SDS and DEPC treated water is essential to remove all the proteins and RNase, which will appear in the outer surface of the root sample, there by it facilitate to get good quality RNA without any degradation.

Four methods have been tried for standardizing the RNA isolation from banana root samples. By using Hi-Media RNA isolation kit, no trace of RNA was found. Little amount of RNA with lot of protein contamination was obtained in TRIZOL method. In case of modified CTAB method, pure RNA was obtained *i.e.*, without any protein contamination but the RNA quantity was less. Modified lithium chloride method gave a good quantity and quality of RNA (Fig. 13). In this method, a treatment of washing step with washing buffer (containing 100 mM Tris HCl (pH 8.0), 5 mM EDTA (pH 8.0), 0.35 M glucose, 2% PVP and 4% 2-mercaptoethanol) was added before extraction to remove the contaminants, organic molecules and excessive water. Before LiCl precipitation step, one chloroform treatment was

given to remove excess amount of phenol content. Apart from normal steps, one more step is added. *i.e.*, potassium acetate precipitation step would facilitate removal of residual polysaccharides. This method gave high quality and quantity of RNA.



Fig. 13. RNA isolated from banana root sample by modified Lithium chloride method

5.1.4.7 cDNA synthesis

mRNA and cDNA have been synthesised from the total RNA as per the protocol given above. PCR analysis was carried out in genomic DNA as well as mRNA by using the RAPD primers. In this, amplification was observed only in genomic DNA not in mRNA and this confirms that mRNA does not have any DNA contamination.

PCR was performed in the genomic DNA as well as cDNA by using RAPD primers namely OPB 4, OPB 6, OPC 8, OPC 11 and OPD 3. In genomic DNA banding pattern was observed while using aforesaid primers, whereas no banding pattern was observed in cDNA while using OPB 4, OPB 6 primers. This shows that these two primers are amplifying only the intron region of the genomic DNA. Polymorphic banding pattern was observed in genomic DNA as well as cDNA while using OPC 8, OPC 11 and OPD 3. More number of bands was appeared in the genomic DNA when compared to cDNA. This confirms that these primers are amplifying both in intron and exon region of genomic DNA (Fig. 14).

High molecular bands were observed only in the genomic DNA and *vice versa* in case of cDNA.

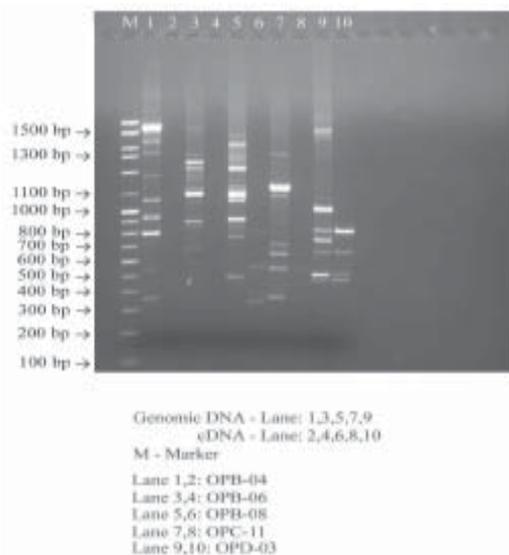


Fig. 14. Confirmation of cDNA using RAPD markers

5.1.4.8 Gene regulation studies

PCR analysis was done in genomic DNA of root and leaf samples as well as cDNA of root sample. A band was appeared with a 280 bp molecular weight in all the DNA samples and this primer can be used as control for gene regulation studies.

5.2 CROP PRODUCTION

5.2.1 Standardization of agro techniques for banana production and productivity

An experiment was conducted to standardize the nutritional requirement of banana cv. Grand Nain grown under two different plant spacing (2 x 3 m and 1.8 x 3.6 m) with two plant densities (two suckers per hill and three suckers per hill) and weekly fertigation with five levels of fertilizers *i.e.*, 50%, 75%, 100%, 125% and 150% RDF fertigation and was compared with single plant per pit at 2 x 2 m spacing and 100% RDF fertigation. Observations were recorded at vegetative and flowering stages on various growth parameters and the observations recorded at vegetative stage revealed that among all the treatments, the plants under *T5* (S1P2F5) *i.e.*, two suckers per hill at 2 x 3 m spacing and 150% RDF fertigation recorded the highest plant height (196.7 cm), girth (55.5 cm), more number of healthy leaves (14.5), petiole length (30.3) and while *T12* (S2P2F2) *i.e.*, two suckers per hill at 1.8 x 3.6 m spacing and 75% RDF fertigation recorded highest number of suckers (6.0), total leaf area (38.47) and LAI (5.94).

The data recorded at the flowering stage revealed that plant morphological characters namely plant height, plant girth, number of healthy

leaves, side suckers, leaf area and leaf area index exhibited significant differences among the different treatments.

The plant height was highest (251.2 cm) in treatment *T5* (S1P2F5) while the lowest plant height of 178.6 cm was recorded in *T20* (S2P3F5). The plants in *T2* (S1P2F2) recorded the highest pseudostem girth of 62.8 cm followed by *T7* (S1P3F2) while the least plant girth (46.2 cm) was recorded under *T20* (Fig. 15). The highest number of healthy leaves at flowering (14.7) was recorded in *T14* *i.e.*, two suckers/hill at 1.8 x 3.6 m spacing and 125% fertigation followed by *T13-S2P2F3* which recorded 14.5 leaves (Fig. 16). The least number of leaves at flowering (11.2) was recorded in *T20-S2P3F5*. The number of side suckers was highest (5.8) under two plants per hill at 1.8 x 3.8 m spacing and 75% RDF fertigation (*T12*) followed by *T11* (5.7 suckers) and the least number of 2.7 suckers were recorded under *T10-S1P3F5*.

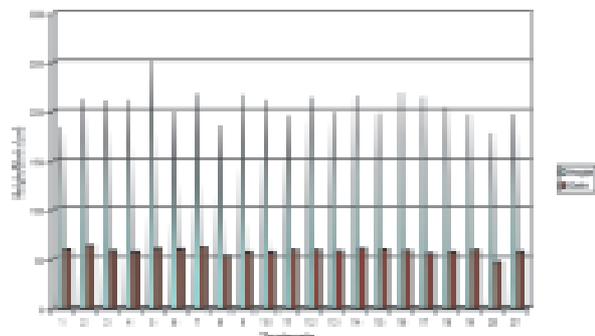


Fig. 15. Plant height and girth as influenced by high density planting and fertigation in cv. Grand Nain

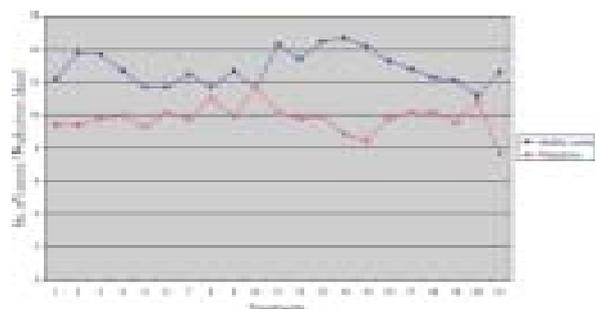


Fig. 16. Number of healthy leaves and phyllochron under different plant densities and levels of fertigation

The leaf breadth varied significantly among the treatments and *T10-S1P3F5* recorded the largest leaf width of 77.3 cm followed by *T9-S1P3F4* while the narrowest leaves (61.5) were recorded under *T20-S2P3F5*. The largest mean leaf area of 1.23 m² was recorded under *T5*- followed by *T10* (1.21 m²) and the least mean leaf area of 0.81 m² was recorded in three plants per hill at 1.8 x 3.6 m spacing and 150% RDF fertigation (*T20*) (Fig. 17). As against the mean leaf area, the total leaf area was found highest (43.42 m²) in *T16-S2P3F1* followed by *T7* (42.62 m²), while

the least (11.76 m²) was recorded in the control (T21) treatment of single plant per hill at 2 x 2 m spacing and 100% RDF fertigation. The leaf area index (LAI) was found highest (6.70) in T16 followed by T7 (6.66) and it was the least in the control (2.94) (Fig. 18).

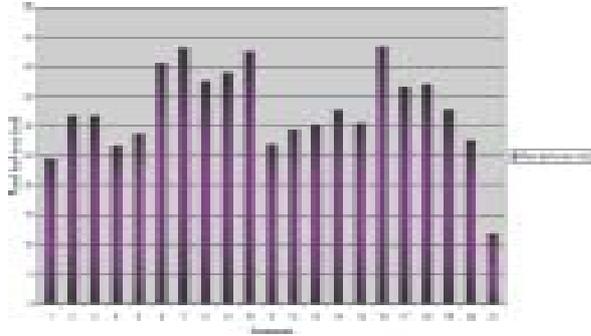


Fig. 17. Effect planting density and fertigation on total leaf area (m²) in Grand Nain

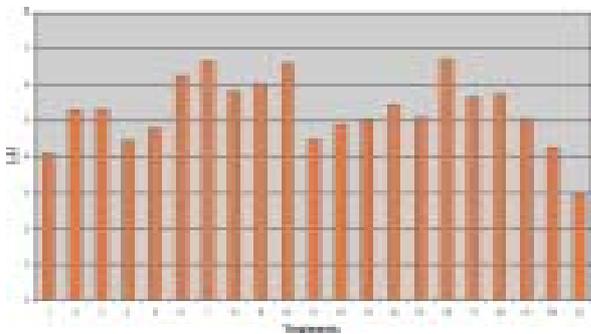


Fig. 18. Leaf area index (LAI) under different planting densities and fertigation levels

The time interval between planting to bunch emergence (shooting) varied significantly among the treatments and the plants in the control treatment (T21) took significantly lesser time for shooting (268.4) followed by T15 (285.0 days). Whereas, the plants under T10 (342.3 days) and T20 (341.3 days) took longer time for shooting. The plants in the control treatment of T21 (Control) took significantly lesser time for leaf emergence with a phyllochron of 7.69 days followed by T15 (8.42 days) while the highest phyllochron of 11.76 days was recorded in T10 (S1P3F5). The analysis of the leaf samples at flowering stage revealed that the chlorophyll pigments did not show significant difference among the treatments.

Studies on the photosynthetic activity (Pn) in different treatments revealed that the photosynthetic rate decreased with the increasing plant population. Among the three plant populations, the photosynthetic activity was higher (11.85 u mol CO₂ m⁻² s⁻¹) at single plant per hill and the least activity of 9.27 u mol CO₂ m⁻² s⁻¹ was recorded at three plants per hill. The photosynthetic activity increased with the increasing dose of fertigation up to 125% RDF fertigation but decreased later. Fertigation with 50%

recommended dose of fertilizers (F1-50% RDF) recorded the lowest activity of 7.76 u mol CO₂ m⁻² s⁻¹ and increased with the higher fertigation levels reaching the highest rate of 11.31 u mol CO₂ m⁻² s⁻¹ at 125% RDF fertigation but decreased to 9.41 u mol CO₂ m⁻² s⁻¹ at 150% RDF fertigation.

Studies on rooting pattern revealed that the root number was significantly more in plants in single plant. Soil (250 cc) and root (10 g) samples were collected from all the treatments at 9th month and the nematode population was estimated. The root-knot nematode, *Meloidogyne incognita* was the only endo-parasitic nematode recorded from all the root samples but the intensity of the populations varied between the treatments. Maximum nematode population (296/g root) was recorded from T21 (control) followed by 50% and 75% of RDF (179 & 178/g root respectively), whereas the minimum population of 85/g root was recorded from 125% RDF (Fig. 19). It was further noticed that nematode population increased when 150% RDF was given. An optimum 104.5/g root recorded in 100%RDF has been identified as below threshold level. Not much difference in nematode population was noticed between different spacing and population densities. Soil populations revealed the occurrence of root-knot nematode, *Meloidogyne incognita*, *Rotylenchus reniformis* and *Rotylenchus* sp.

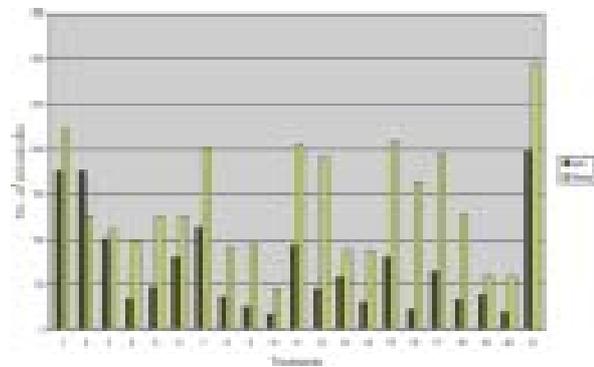


Fig. 19. Nematode population under different planting densities and fertigation levels

The leaf nutrient analysis revealed that the plants in wider spacing of 1.8 x 3.6 m had significantly higher leaf N & K, whereas the plants at 2 x 3 m recorded higher leaf P, Ca and Mg contents. Among the plant densities tried planting of two suckers per hill recorded more leaf NPK contents while higher levels of Ca and Mg were recorded in the plant density of three suckers per hill.

The soil microbial studies at flowering stage revealed that the population of bacteria, fungi and actinomycetes were higher in the spacing of

2 x 3 m. The highest microbial population was recorded in 3 plants per pit than in two or one plant per pit. The population decreased with increasing fertigation from 50% to 150% RDF.

The results on the effect of levels of fertigation on the soil NPK content revealed a positive correlation between the levels of fertigation and the soil nutrient status. The results on the influence on the soil depth and distance on the population revealed that among the two soil depths the highest microbial population was recorded in the soil depth of 0-15 cm while the population was reduced in deeper layers of 16-30 cm. Similarly among the three distances, 0-30 cm recorded the highest population of all the three microbes and the population decreased with increasing distance and the lowest population was recorded 61-90 cm at away from the plant.

The results on soil NPK contents in relation to the soil depth revealed that top 0-15 cm soil layer registered higher nitrogen and potassium contents than at 16-30 cm depth whereas, the highest soil P content was recorded in deeper layers (16-30 cm). Among the three distances, the N and P contents were found more at the closest distance of 0-30 cm than other distances, while the soil K content exhibited a curvilinear response with the highest K content was recorded in the closest distance of 0-30 cm, which decreased under 31-60 cm but was again increased at 61-90 cm.

Studies on sigatoka leaf spot disease severity and youngest leaf spotted (0%-31%) revealed that the disease severity ranged from 0.64 to 14.81. The minimum severity of 0.64 with a total reduction of 82.88% in disease severity was recorded in plants in two suckers per hill at 2 x 3 m spacing and 100% RDF fertigation (T3) while the treatment T6- S1P3F1 recorded the highest incidence of 14.81%. With regard to YLS-0, it ranged from 8 to 14.66% and the maximum value was recorded in S2P3F2 (T17). But in case of YLS-31, the maximum value of 15 was observed in four treatments namely S1P2F5 (T5), S2P2F4 (T14), S2P3F2 (T17) and T21 (control).

5.2.2 Studies on micronutrients in banana

The highest pseudostem height (289 cm), pseudostem girth (82 cm), total number of leaves (34 nos.), total leaf area (13.6 m²), number of fingers per bunch (204 nos.) and hands per bunch (12 nos.) were observed in $Fe_{soil}:Zn_{foliar}:B_{soil}$. Application of sulphur (20 g bentonite sulphur per plant) along with micronutrient treatments recorded more leaf nutrients than those without sulphur application. The leaf N ranged from 1.76 to 2.65% without S application and from 1.91 to 2.61% with S application. These values varied significantly with

varying levels of micronutrients. The leaf P without S application showed non-significant variation with micronutrients but with S application. The leaf P ranged between 0.13 and 0.33 with S application. The leaf K varied significantly in both with S application and without S application. The highest leaf K (2.05%) without S application was observed at $Fe_{soil}:Zn_{foliar}:B_{soil}$ and the highest leaf K (2.26%) with S application at $Fe_{soil}:Zn_{soil}:B_{soil}$. The leaf S content significantly varied with varying levels of micronutrients in both with S and without S. The highest leaf S (0.45%) without S application was observed at $Fe_{soil}:Zn_{foliar}:B_{soil}$ and the highest leaf S (0.53%) with S application was also observed at the same treatment. The leaf Fe ranged from 71 to 185 ppm without S application and from 78 to 169 ppm with S application with statistically significant variations. The leaf Zn significantly varied from 5 to 13 ppm without S application and from 8 to 15 ppm with S application. The leaf B significantly varied from 8 to 25 ppm without S application and from 10 to 28 ppm with S application. The leaf Mn significantly varied from 93 to 137 ppm without S application and from 108 to 150 ppm with S application. The leaf Cu significantly varied from 6 to 17 ppm without S application and from 10 to 20 ppm with S application.

The bunch weight significantly varied with varying levels of micronutrients without S application from 9.9 to 13.5 kg and with S application from 10.9 to 15.2 kg. The treatment, $Fe_{soil}:Zn_{foliar}:B_{soil}$ regarded the highest bunch weight of 13.5 kg without S application and 15.2 kg with S application. Thus, soil application of 5 g ferrous sulphate, 5 g borax and foliar spray of 0.5 percent zinc sulphate along with 20 g of sulphur per plant with recommended dose of NPK (200:30:400 g/plant) in high pH soil produced 32% more yield with an additional profit of Rs. 44,700/- per ha in Ney Poovan ratoon-I.

The interaction among the leaf nutrient elements was observed in terms of correlation coefficients (Table 9). Highly significant correlations (at 1% level) leaf N and leaf K (0.642**) and with leaf Zn (0.519**) were observed. Significant correlation (at 5% level) N with Zn and Cu were between observed. The leaf P content significantly and positively correlated with leaf K content but negatively with leaf Fe and Zn. This may be due to formation insoluble compounds of these elements with P in the soil under high pH condition. The leaf K was positively correlated (at 1% level) with leaf Zn (0.546**) and significantly correlated (at 5% level) with leaf B, Mn and Cu. The leaf Fe had positive and significant correlation with leaf Zn, B and Cu. The leaf Zn had positive and significant correlation with leaf Mn and Cu. The leaf B had significant positive correlation with leaf Cu. This correlation



Table 9. Correlation coefficient among leaf nutrients and between leaf nutrients and bunch weight

	N	P	K	Fe	Zn	B	Mn	Cu	Bunch wt.
N	1.000								0.4828**
P	0.042	1.000							0.2281
K	0.642**	0.329*	1.000						0.5802**
Fe	0.261	-0.010	0.108	1.000					0.0672
Zn	0.519**	-0.111	0.546**	0.388*	1.000				0.3473*
B	0.389*	0.142	0.413*	0.379*	0.220	1.000			0.1340
Mn	0.219	0.118	0.425*	0.171	0.332*	0.187	1.000		0.3363*
Cu	0.377*	0.121	0.389*	0.442*	0.416*	0.379*	0.268	1.000	0.3149*

study revealed that leaf K and N played major role in increasing the bunch weight in the presence of other micronutrients. Very high significant positive correlations were observed between bunch weight with leaf K (0.5802**) and with leaf N (0.4842**). The leaf Zn, Mn and Cu showed significant positive correlation with bunch weight. Other nutrients also showed positive correlation with bunch weight but they were not statistically significant.

The soil application of sulphur reduced the soil pH from 8.7 to 7.6 in the rhizosphere. Thus, the application of sulphur increased the availability of soil applied micronutrients to the crop at the desired level under high pH condition.

5.2.3 Fertilizer tailoring for targeted banana yield and sustainable soil health

The soil fertility gradients were created with suitable soil exhaust crop like maize. The significance of soil fertility gradients were analysed by proper soil testing for NPK. The banana varieties like Karpuravalli and Poovan were planted in the nutrient gradient plots according to the statistical design. In both the varieties, the whole treatments were implemented with and without organic manure applications in two sets. The data on plant growth parameters like plant height, pseudostem girth, phyllochron, etc. were collected in both the varieties under different treatment combinations. The data at 9 MAP revealed that there were statistically significant variations under different levels of NPK.

5.2.3.1 Karpuravalli

The plant height of Karpuravalli banana gradually increased with increasing levels of N. The N150 level recorded the highest plant height of 224 cm. It was 9% more than that of the control, N0 level. The P50 level recorded the highest plant height of 226.4 cm. In case of potassium gradient, the K50 recorded the highest plant height of 219.5 cm. In all the gradients of NPK, the application of organic manure recorded more height than without application of organic manure.

The highest pseudostem girth were recorded in N150 (68.3 cm), P50 (66.2 cm) and K0 (68.6 cm) in Karpuravalli banana. The application of organic manure significantly increased the pseudostem girth in the gradients of N and P but not in K. The NPK gradients significantly varied the phyllochron. The lowest phyllochron were recorded at N150 (7.6 days/leaf), P100 (7.7 days/leaf) and K50 (7.8 days/leaf). The application of organic manure along with K increased the phyllochron significantly in Karpuravalli.

5.2.3.2 Poovan

In case of Poovan banana, the highest plant height was observed in N50 gradient (169.1 cm), P50 gradient (158 cm) and K200 gradient (164.9 cm). The P gradient did not show any significant variation in plant height. The K gradient showed significant variation in plant height with and without application of organic manure but not the N and P gradients. The N150 gradient recorded the highest pseudostem girth (53.1 cm) among the N gradients. The P50 recorded the highest pseudostem girth (50.5 cm) among the P gradients and K200 recorded the highest pseudostem girth (52.8 cm) among the K gradients. The application of organic manure significantly increased the pseudostem girth in P and K gradients but not in N gradient. Among the N gradients, the N150 recorded the lowest phyllochron (6.7 days/leaf). Among the P gradients, the P100 recorded the lowest phyllochron (7.1 days/leaf) and among the K gradients, the K50 and K200 recorded the lowest phyllochron (7.9 days/leaf). The application of organic manure increased the phyllochron significantly with N gradients but not in P and K gradients.

5.2.4 Soil test based integrated nutrient tailoring for optimum banana production and sustainable soil health

Fertilizer adjustment equations were developed for Ney Poovan banana by following the 'Targeted Yield Concept'. The important factors used in development of fertilizer adjustment equations for



Table 10. Important factors arrived and used in development of fertilizer adjustment equations for Ney Poovan

Nutrient	Nutrient requirement (NR) kg / ton	Contribution from soil in control plot (CS) (%)	Contribution from fertilizer in treatment plot (CF) (%)	Contribution from organic fertilizer in treatment plot (CO) (%)
Nitrogen (N)	13.36(521/39)	58.9	70.3	19.7
Phosphorus (P ₂ O ₅)	1.59(62/39)	50.3	65.9	12.9
Potassium (K ₂ O)	26.28(1025/39)	39.5	79.4	35.4

NeyPoovan are given in Table 10. The Fig. 20 depicts the variation in Rasthali bunch size due to variation in NPK levels.



Fig. 20. Effect of NPK gradients on size of Ney Poovan banana bunches

5.2.4.1 Fertiliser adjustment equations for Ney Poovan

$$FN = (19.0 \times T) - (0.84 \times SN) - (0.28 \times ON)$$

$$FP = (2.41 \times T) - (0.76 \times SP) - (0.20 \times OP)$$

$$FK = (33.10 \times T) - (0.50 \times SK) - (0.45 \times OK)$$

Here, FN, FP and FK are nitrogen (N), phosphorus (P₂O₅) and potassium (K₂O) requirement (kg/ha) of banana cultivated in one hectare, respectively, through fertilizers. T is the target (tons/ha) of banana yield. SN, SP and SK are quantity (kg/ha) of nitrogen (N), phosphorus (P₂O₅) and potassium (K₂O) already existing in the soil, before application of fertilizer. ON, OP and OK are quantity (kg/ha) of nitrogen (N), phosphorus (P₂O₅) and potassium (K₂O) contributed from the recommended dose of organic manures applied to banana crop.

5.2.4.2 Evaluation of fertilizer adjustment equations

The above fertilizer adjustment equations were tested in multi-location trials in different farmers' fields. When the blanket recommendation of NPK was adopted, the Ney Poovan yield was 27.50 t/ha with B:C ratio of 22.31. The highest B:C ratio of 24.02 was observed with actual yield of 37.93 t/ha

when the adjustment equations were applied for yield target of 38 t/ha. The actual yield and B:C ratio increased with the implementation of adjustment equations with increasing targets, upto 38 t/ha. Beyond this yield target there was a declining trend in actual yield and B:C ratio with increasing targets. The highest cost of yield of Rs. 4,17,230/ha was worked out with the highest B:C ratio (24.02), when the yield target was fixed at 38 t/ha. The superiority of fertilizer dose at 38 t/ha target over blanket recommendation of fertilizer dose was due to the yield increase by 10.43 t/ha, the cost of yield increase by Rs. 1,14,730/ha with increase in return for every one rupee spent on fertilizer by Rs. 1.71. Thus, in Ney Poovan banana cultivation, the dose of N:P:K (255:35:459 gram/plant) worked out for a yield target of 38 t/ha was found to be optimum for sustainable soil health and wealth.

5.3 PHYSIOLOGY AND BIOCHEMISTRY

5.3.1 Physiology of flowering

Nendran corm was analysed for N, P, K, Ca and Mg at 3rd, 5th and 7th months and at flowering and harvest. In all the phenological stages, the nutrients content were higher in the inner core of the corm than outer core. The NPK and Mg content (%) were found to be higher at flowering, whereas calcium content was higher at 7th month after planting.

Contribution to bunch development by current photosynthesis, pre-flowering storage and bunch photosynthesis was studied. In Rasthali and Ney Poovan, the current photosynthesis was contributing 52.58% and 52.73% respectively to the bunch development, whereas in Saba and Karpuravalli, 66.87% and 66.13% of pre-flowering storage has contributed. In all the above tested varieties, the bunch photosynthesis contribution was only 3.14 % to 7.07%.

The starch content in Saba was 75.26% whereas in Monthan, it was 81.26%. The amylose and amylopectin content were 34.26% and 65.74% respectively in Saba, whereas in Monthan, it was observed as 36.27% and 63.73% respectively.



5.3.2 Drought stress tolerance

In a field experiment conducted at NRCB farm, water stress was imposed by withholding water for one month at flowering through drip irrigation, which decreased bunch weight in Robusta (42.07%), Karpuravalli (25%) and in Rasthali (18.83%). When water stress was imposed at 30 days after flowering for one month, the bunch weight has reduced to 18.83% (Robusta), 11.25% (Rasthali) and 27.66% (Karpuravalli), similarly when the water stress was imposed at 60 days after flowering for one month, the bunch weight decreased to 16.47% (Rasthali), 16.84% (Karpuravalli) and 25% (Robusta). Among all the three varieties tested, Robusta was more sensitive. The fruit length (11-14%) and circumference (5.75-16%) also decreased.

Twenty four diploid (AA) was phenotyped for drought tolerant traits. It was found that there was wide variation in chlorophyll content among the 24 diploid (AA) banana genotypes. In this group Anaikomban recorded the highest chlorophyll a, chlorophyll b and total chlorophyll content among all the tested genotypes and Hatidat, Kanai Bansi, Siguzani and Namarai recorded the lowest chlorophyll.

Drought resistance of plants is related to the thermostability of chlorophyll. The present result showed that there were different ranges of chlorophyll stability index among the tested diploid genotypes. Matti and Imbogon showed the highest chlorophyll stability index (80.64 and 79.06) and Tongat recorded the lowest (34.13). Imbogon, Pisang Jari Buaya and Siguzani showed higher membrane stability index (42.89, 41.48 and 41.90). The lowest membrane stability index shown by *M. acuminata* (Assam) (17.56).

Based on the epicuticular wax (0.050 mg/cm²) content, higher membrane stability index (42.89), chlorophyll stability index and membrane stability index, Imbogon apparently showed the tolerant traits for drought tolerance.

5.3.3 Salt stress tolerance

The leaf pigment analyses were done in the banana plants grown in salt affected field. The Karpuravalli and Poovan varieties showed higher total chlorophyll content (11.25 µg/g fr. wt.) and Robusta showed lesser total chlorophyll content (5.67 µg/g fr. wt.). In Saba, higher carotenoid pigments in leaf (4.27 µg/g fr. wt.) was observed, whereas in Nendran lesser carotenoid content (1.57 µg fr.wt.) was observed.

Among the eight commercial cultivars were grown in the salt affected field (EC_{1:2.5} = 8.19 dSm⁻²)

Karpuravalli, Saba, Poovan, Ney Poovan and Nendran recorded the highest leaf epicuticular wax in the highest range of 0.427-0.577 mg/cm² whereas Red Banana, Robusta, and Rasthali recorded 0.164 to 0.241 mg/cm² of leaf epicuticular wax.

When the NaCl salinity stress was imposed on four different commercial varieties, the leaves of all cultivars produced more epicuticular wax (ECW) at higher concentration of NaCl (150 mM). However, Ney Poovan and Saba produced 81% and 78% more epicuticular wax content respectively in 150 mM NaCl treatment than control. The Nendran and Robusta produced ECW in the range of 33-35% and considered to be non-responsive to NaCl stress.

The Chlorophyll Stability Index (CSI) is one of the indicators for salinity tolerance. Among the four cultivars studied, the Saba recorded better CSI through low percentage (13.07%) reduction in CSI at 150 mM NaCl indicating its tissue tolerance to salinity tolerance.

The proline content (0.37-0.68 mg/g dry wt.) was higher in NaCl (150 mM) than control treatment (0.16-0.27 mg/g dry wt.) and Nendran recorded higher proline concentration and Ney Poovan contained lower concentration of proline at higher NaCl (150 mM) treatment.

The lipid peroxidation is considered as an indicator of the extent of oxidative damage under salt stress. The lipid peroxidation was measured as content of malondialdehyde (MDA) in leaves. Among four cultivars studied, the Nendran recorded lower MDA (150.41 nmol/g fr. wt.) content at 150 mM NaCl indicating its tissue tolerance to salinity as compared to other varieties. Robusta showed higher MDA content (182.56 nmol/g fr. wt.) indicating its susceptibility to salt tolerance.

The starch content in leaf tissue was significantly decreased when treated with different concentration of NaCl (50 mM, 100 mM and 150 mM) in all the varieties (Robusta, Saba, Nendran and Ney Poovan). In Saba, a five fold decrease in starch content was observed. Similarly the total sugars and reducing sugars increased four to five fold in all the varieties in 150 mM NaCl concentration as compared to control.

The free amino acids content was observed higher in 150 mM NaCl (44.98 mg/g dry wt.) treated leaves than control (14.05 mg/g dry wt.). Robusta showed higher free amino acid content and Saba recorded lesser free amino acid content at 150 mM NaCl treatment.



5.3.4 Biochemical studies to assess triggering time of nematode resistance genes

Phenol content and activity of peroxidase, polyphenol oxidase and beta-1,3-glucanase were estimated in leaves and roots of Rasthali (susceptible) and FHIA-1 (resistant) inoculated with *Pratylenchus coffeae*. In general, the total phenol content in both roots and leaves increased from the third day after inoculation of the nematode and the highest quantity was found on 4th or 5th day. The peroxidase, polyphenol oxidase and beta-1,3-glucanase activities in roots and leaf tissues of Rasthali and FHIA-1 increased from third day after inoculation of the nematode and activity peaked in 4th day with 2-6 fold increase following which the activity levels of the enzymes decreased except the activity of beta-1,3-glucanase which remained at higher levels during 5-7 days. The level of induction of enzymes activity and phenols were higher in root tissues than in leaves. Also, the phenols content and enzyme activities in the hybrid, FHIA-1 was higher than in cv. Rasthali. The results of the study indicated that the triggering time of nematode-resistant gene(s) is on the 3rd day after inoculation.

5.4 POST-HARVEST TECHNOLOGY

5.4.1 Studies of controlled ripening in banana

Studies on the controlled ripening of banana by regulating the ripening temperature at 18, 20, 22 and 24 °C in Rasthali and Karpuravalli banana was taken up. 90% mature fruits were harvested and bunches were deheaded. The hands were dipped in Bavistin 500 ppm solution for 10 min and surface moisture was dried under fan. The fruits were sprayed with 500 ppm Ethrel solution and after drying of surface moisture, five hands were kept in CFB box and stored at each temperature of 18, 20, 22 °C and ambient temperature. Observations were recorded for green life, yellow life, total soluble solids, acidity, sugar acid ratio and organoleptic quality. The results indicated that ripening of fruits at ambient temperature was faster as compared to 18-22 °C in both the varieties. Rasthali had 2 days green life at ambient and 8 days at 18-22 °C while Karpuravalli had 3 days green life at ambient, 6 days at 18 °C and 5 days at 20 and 22 °C. Subsequent yellow life was also longer at lower temperatures than ambient thereby increasing the overall shelf life of the fruits. The data on quality changes like TSS, acidity, total sugars and sugar acid ratio also supported these findings. The overall organoleptic acceptability indicated that after ripening, the fruits stored at 22 °C had the best acceptability compared to those at 18 °C in Rasthali while in Karpuravalli

there was no significant difference in any of the temperatures.

5.4.2 Studies on blending of banana juice with beetroot, carrot and Noni (*Morinda citrifolia*) juices

Beet roots and carrots were made into shreds and blended in mixer and juice was extracted. The extracted juice was pasteurized at 100 °C and bottled for storage and blending with banana juice. Due to the unsuitability of these juices because of fermentation, it was discontinued. The Noni (*Morinda citrifolia*) juice was procured from the local market and blended with banana juice in different proportions. The beverage made was pasteurized and storage studies were carried out upto 6 months. Pure banana RTS was used as absolute control while 3% Noni beverage was used as second control and Noni at 2, 3, 6 and 9% concentrations were used for blending with banana for making beverage. Various observations like TSS, acidity, vitamins A and C, protein, total carbohydrates, phenols and overall organoleptic acceptability were recorded during storage.

The results indicated that Noni juice could be blended with banana juice up to an extent of 2% beyond which it was not acceptable organoleptically. Pure Noni juice was acceptable as a beverage only when used at a rate of 3%. It had a storage life of 6 months, with good acceptable quality. After 6 months the best treatment had a TSS of 16.6° Brix, acidity of 0.565%, vitamin A of 38.57 µg/100 g, vitamin C 1.512 mg/100g, protein 0.386%, phenols 0.036% and organoleptic score of 5.6 out of 9.0.

5.4.3 Developing dried stem and stem juice powders

The stem juice from Poovan and Karpuravalli varieties were tray dried at 70 °C and the dried material was scrapped out. It was re-suspended in water and components separated on a Florisil column. The pomace was dried and powdered in a powder mill and encapsulated. A formulation containing banana stem powder, eno fruit salt, CMC and stevia and develop as health supplement.

The results indicated that stem juice yield ranged between 86.67% in Karpuravalli and 83.3% in Poovan. After drying the juice by tray drying process, the scrap yield was 0.89% in Karpuravalli and 1.24% in Poovan. The pomace had a moisture content of 89.25% in Karpuravalli and 87.4% in Poovan. The bulk density of dried pomace ranged from 0.40 g to 0.47 g/cc. The biochemical analysis of stem juice indicated that a TSS of 2.8° Brix in Karpuravalli and 1.90° Brix in Poovan, acidity of

0.0499 and 0.030%, total sugars of 0.85 and 0.39%, phenols of 0.05% and 0.03% respectively in Karpuravalli and Poovan. The scarp analysis revealed presence of 0.23% and 0.256% acidity, 10.80 and 6.0% total sugars, 3.45% and 1.16% starch and 0.437% and 0.175% phenols in Karpuravalli and Poovan respectively. The re-hydration ratio of pomade powder was 1: 10.58 in Karpuravalli and 1: 9.09 in Poovan. Each encapsulated capsule could hold 0.14 g of Karpuravalli pomace powder and 0.11 g of Poovan pomace powder.

5.4.4 Study on retting processes of banana pseudostem fibre

For chemical retting, the pseudostem of Poovan was cut into stripes of 15 cm and boiled in 0.1%, 0.5% and 1% NaOH solution for 30 min and 1 h. For enzymatic retting, the stripes of pseudostem were dipped in 0.2% pectinase enzyme for 22 days while for microbial retting the *Aspergillus niger* was used in distilled water for 20 days for complete retting. The tap water was used as a control was no chemical or microbe was used for retting. It took 18 days for complete retting. After completion of retting the fibres were cleaned by washing and combing and were bleached using different bleaching agents like bleaching powder (0.4%), 1% citric acid, 1% oxalic acid, 1% hydrogen peroxide.

The chemical retting processes using 0.1%, 0.5%, and 1% NaOH for 30 min or 60 min showed good results with complete retting of the sheaths of the Poovan pseudostem while 1 or 2% oxalic acid or combination of EDTA or sodium phosphate did not give satisfactory results. In enzymatic retting 0.2% pectinase took 22 days for completion of retting while microbial retting using *Aspergillus niger* in distilled water took 20 days for completion of retting. The tap water without addition of any chemical or inoculum took 18 days for complete retting. Further bleaching of fibres was necessary as the colour of the fibre was dull. Among the different bleaching agents, 1% citric acid treatment for 2 h was best where the colour of the fibre was bright and the texture was soft. A combination of 0.5% NaOH and 1% citric acid gave the best fibre with good colour and fine texture. The quality analysis done at CIRCOT revealed that the 0.5% NaOH treated fibre had the highest tenacity and is highly suitable for yarn. Among the chemical component, it had the least lignin and pectin content and highest cellulose content.

5.4.5 Standardizing the optimum temperature for banana wine production

Karpuravalli fruits were ripened and juice extracted. The wine was prepared adopting the procedure standardized earlier. However, the

temperatures were maintained at three levels viz., 10, 13.5 and 24 °C. After the completion of secondary fermentation, the wines were filtered and cold stabilized at 4-8 °C in refrigerator for 15 days, then filtered and bottled. The results indicated that at 24 °C, the fermentation was completed within 8 days while at 13.5 and 10 °C, it took 18 days and 20 days respectively. The alcohol concentration also increased at lower temperature. By cold stabilization, the astringent taste of the wine mellowed down and the aroma and taste improved. The acidity of the wine was also reduced by the cold stabilization processes.

5.5 CROP PROTECTION

5.5.1 Nematode incidence

5.5.1.1 Nematode in tissue culture banana nurseries

The root-knot nematode, *Meloidogyne incognita*, is predominantly observed in tissue culture plants at the secondary hardening stage. Heavy nematode infested seedlings on transplantation to main field fail to establish since the newly formed roots were infected (Fig. 21). Preliminary survey carried out from few tissue culture companies as well as from field planting has indicated heavy root-knot nematode infestation. An integrated nematode management strategy has been suggested to TC companies for effective control of nematodes.

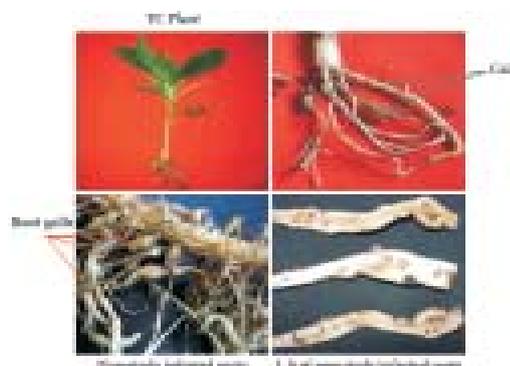


Fig. 21. Nematode infested roots of TC plant in the polybag stage itself

5.5.1.2 Interaction between entomopathogenic nematodes and root-lesion nematode

Biocontrol agents like *Pseudomonas fluorescens*, EPN, *Steinernema carpocapsae* and *Heterorhabditis indica* along with nematicides viz., Carbofuran and Rugby tested in nematode infested Robusta plants resulted that all the treatments were found effective in reducing (80%) the nematode populations with 65% increased plant growth over untreated control plants. Among the treatments, *H. indica* performed better than all the other treatments.

5.5.1.3 Isolation, identification and evaluation of *Pseudomonas fluorescens* strain

Forty five *Pseudomonas* strains were isolated from the rhizosphere of banana grown in various parts of Tamil Nadu. Among them, only 16 were identified as *P. fluorescens* by various morphological and biochemical tests. These 16 isolates were subjected to *in vitro* screening against *P. coffeae* at different concentrations. Eight isolates viz., PFNRCB4, PFNRCB6, PFNRCB9, PFNRCB12, PFM2, PFM5, PFM8 and PFM9 were found inducing higher per cent mortality in both 25% and 50% concentration. These 8 isolates of *P. fluorescens* were tested against root-lesion nematode, *P. coffeae*, in Nendran banana in pot condition. All the isolates were used alone and in combination with nematodes. Uninoculated control (nematode alone) was used to compare the treatments. At the time of termination, the plant growth parameters and nematode population from soil and roots were recorded. The results of the study revealed that the strains viz., PFNRCB6, PFM5, PFM8, PFM9 were found effective in increasing the plant growth (45 to 65%) with significant reduction in nematode populations (80 to 100%) over control plants.

5.5.1.4 Biological suppression of root-knot/ root-lesion nematodes in banana

The effect of promising bio-control agents such as *T. viride* (local isolate), *T. viride* (Pest Control Lab., Bangalore), *T. harzianum* (local isolate), *T. harzianum* (Pest Control Lab., Bangalore) in combination with neem cake was studied against root-knot nematode, *M. incognita* infesting banana in cultivar Robusta in pot condition. Both bio-agents were used alone and in combination with neem cake. Uninoculated control (nematode alone) was used to compare the treatments. At the time of termination, the plant growth parameters and nematode population from soil and roots were recorded. The results of the study have clearly indicated that all the treatments whether used alone or in combination were significantly superior to check with nematode alone in terms of plant growth. Significant reduction in nematode population and root-knot indices was recorded from all the treatments except in nematode alone inoculated plants. Among the combined treatments, plants receiving either local isolate of *T. viride* or *T. harzianum* with neem cake at 250 g/plant or the species of bio-agent isolates obtained from Pest Control Lab., Bangalore with neem cake at 250 g/plant gave superior performances in terms of plant growth with significant reduction in nematode population over nematode alone inoculated plants or individual bio-agent applied in nematode

infested plants. Thus, the integration of *T. viride* or *T. harzianum* isolates obtained either from local or from Pest Control Lab., Bangalore along with neem cake can be effectively used in the management of root-knot nematode in banana since the use of single bio-agent or organic amendment can not be very effective in the management of nematode induced disease complex.

5.5.1.5 Evaluation of the Cyanobacterium against banana nematodes

Studies on the toxic effect of cyanobacteria against nematodes, pure cultures of seven species of cyanobacterium namely *Chroococcus minutus*, *Oscillatoria carpiana*, *O. formosa*, *O. tenuis*, *O. claricentrosa*, *Phormidium corium* and *P. tenue*, obtained as marine phytoplankton from Microbiology Department of Bharathidasan University, Tiruchirapalli were mass multiplied in BG11 medium and the same culture was used.

In vitro studies carried out by using these seven species of Cyanobacterium at two levels of concentration viz., 25% and 50% against the mortality of root-lesion nematode exposed at 24, 48 and 72 h indicated that five species namely *O. formosa*, *O. carpiana*, *O. claricentrosa*, *P. corium* and *P. tenue* have recorded 80 to 90% mortality of *P. coffeae* when exposed to 72 h at 50% concentration. Mortality rate of *P. coffeae* was directly proportional to the concentration of cyanobacterium and time of exposure. *In vivo* experiment was also conducted by using all the species of cyanobacterium in pots under shade net condition in order to control the major nematode, *P. coffeae* infesting banana in cv. Nendran. The results obtained from the pot culture experiment revealed that significant reduction in root-lesion indices and final nematode population with increased vegetative growth of banana plants was recorded at the higher dose of 20 ml of cyanobacterium compared with the lower dose of 10 ml.

The present investigation have showed that cyanobacterium has the ability to kill the nematodes. Among the species tested, *O. formosa*, *O. carpiana*, *P. corium*, *O. claricentrosa* and *P. tenue* were found to be the most effective cyanobacterium which can control the nematodes significantly with improved plant growth parameters compared to nematode alone inoculated plants.

5.5.1.6 Efficacy of biofertilizers against root-lesion nematode (*Pratylenchus coffeae*)

The effect of biofertilizers such as *Azospirillum*, *Azotobacter* and Vesicular Arbuscular Mycorrhizae-*Glomus fasciculatum* alone and in combination with all biofertilizers were tested against root-lesion

nematode, *Pratylenchus coffeae* infesting banana in cv. Rasthali in pots under shade net condition. Besides, the nutritional changes like nitrogen, phosphorous and potassium in soil and root samples of all experimental treatments were also estimated.

Among the treatments, individual treatment of VAM-*Glomus fasciculatum* was found to be the most effective treatments with significant increase in plant growth (55 %) with reduction in nematode population (80%) over uninoculated control followed by the combination of VAM-*G. fasciculatum* + *Azospirillum* which showed increased trend in plant height and pseudostem girth, whereas VAM-*G. fasciculatum* + *Azotobacter* did not perform well. The nutritional changes like N, P and K in both soil and root samples of all treatments had showed only slight increase in their levels as compared to nematode inoculated control plants.

5.5.1.7 Nematode and virus interaction studies on banana

Interaction between nematodes (root-lesion and root-knot nematodes) and banana streak virus studied in cv. Poovan revealed that the plants infested with BSV alone or with nematodes had more impact in reducing the growth parameters compared to healthy plants without nematodes. The present study also ruled out that both root-lesion and root-knot nematodes had not influenced in transmitting the BSV to the healthy plant.

5.5.1.8 Soil hybridization using black soil for the control of nematodes

Minimum nematode population (*Pratylenchus coffeae*) with fewer incidences of *Fusarium* wilt symptom was recorded in Rasthali banana grown in pots containing 100% or 75% black soil as compared to normal soil or in 50/25% black soil with equal quantity of normal soil.

5.5.1.9 Varietal reaction against major nematodes

Out of 10 diploids screened against major nematodes under field condition, diploids Pisang Linin, Tonget, Pisang Berlin, Pisang Mas, *Musa acuminata*, Matti, Calcutta-4, and *Musa sikkimensis* were found highly susceptible to root-lesion nematode, *Pratylenchus coffeae*. However, diploids viz., Pisang Jari Buaya, Pisang Linin, and Matti were found resistant/ tolerant to root-knot nematode, *M. incognita*.

Promising hybrids viz., FHIA-01, 03, 17 and 23 and cultivars viz., Udhayam, Rasthali, Thiruvananthapuram, Robusta, Manoranjitham and Grand Nain screened against root-lesion and

root-knot nematodes under *in vitro* condition resulted that the hybrid FHIA-23 and cvs. Thiruvananthapuram and Manoranjitham were found resistant to root-knot nematode, *Meloidogyne incognita*, whereas all the cultivars tested were found susceptible to root-lesion nematode, *Pratylenchus coffeae*.

5.5.1.10 Field demonstration of the integrated nematode management techniques

A field demonstration was conducted out using various treatments viz., Marigold (*Tagetes*) plants as an intercrop (Fig. 22) along with promising bio-control agents, *P. lilacinus*, *P. fluorescens*, *T. harzianum*, neem cake and nematicides, Carbofuran as soil application. The plant growth parameters and nematode populations from both soil and root assessed at different periodical interval revealed that all the treatments have performed better than the non-treated plants. Among the treatments, the Marigold plants grown as an intercrop have produced higher yield with significant reduction (90%) in nematode populations over other treatments.



Fig. 22. Marigold plants (*Tagetes* spp) grown as an intercrop in Nendran banana

5.5.2 Management of Weevils

5.5.2.1 Identification of banana leaf sheath volatiles

Isolated banana leaf sheath volatiles from commercial cultivars (Poovan, Monthan, Hill banana, Karpuravalli, Rasthali, Robusta and Nendran) and identified a number of volatile components. Seven components were identified in Poovan and Robusta. Eight components were identified in cvs. Rasthali, Karpuravalli, Hill banana (Virupakshi) and Monthan. In cv. Nendran, 6 components were identified.

Green leaf volatile (hexanal) was identified by GC/HS and naphthalene was identified by GC from the volatiles collected from cv. Nendran through air entrainment technique.

Field evaluation of microbial bio-control agents

Beauveria brongniartii (1×10^{10} cfu/ 20 g/ trap) evaluated at Perumparai for 8 weeks by stem trap method. It trapped 0.2-2.6 weevils/ trap of corm weevil and 0.1-1.2 weevils/ trap of stem weevil. Weevil mortality was 71.1 % (stem weevil) and 65.7% (corm weevil) (Fig. 26). *Beauveria bassiana* (1×10^{10} cfu/ 20 g/trap) evaluated at Maharajapuram for 6 weeks by stem trap method and 73.3% stem weevil mortality was recorded.

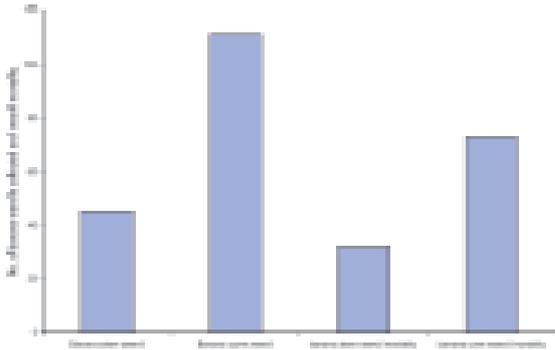


Fig. 26. Field evaluation of *Beauveria brongniartii* against banana weevils

Field evaluation of entomopathogenic nematodes against banana weevils

Steinernema massodi (1×10^8 IJs/-20g/trap) evaluated at Perumparai for 8 weeks by stem trap method. 52 corm weevil and 120 stem weevil were trapped. 1.5 -0.2 corm weevil/trap and 3.4 -0.4 stem weevil /trap. Weevil mortality of 49.2 and 32.7 per cent was recorded by stem and corm weevil respectively.

Heterorhabditis indica (1×10^8 IJs/-20 g/ trap) evaluated at Perumparai for 8 weeks by stem trap method. Trapped stem weevil 162 and corm weevil 61. Weevil mortality was 59.3 % (stem weevil), 39.4% (corm weevil) (Fig. 27).

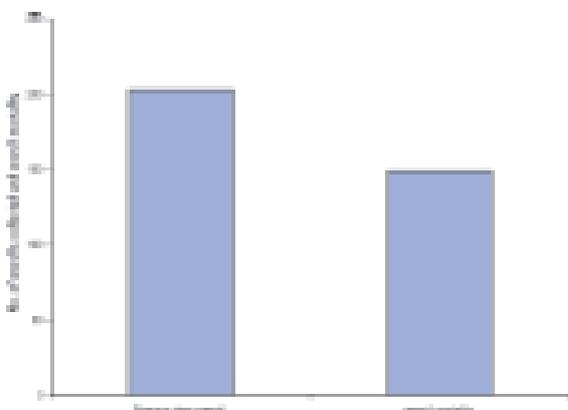


Fig. 27. Field evaluation of *Heterorhabditis indica* against banana weevil

5.5.3 Studies on Fungal and Bacterial Diseases and their Management

5.5.3.1 Cloning of IGS region of *Foc* isolates

IGS region was amplified from *Foc* collected from infected Rasthali from AP and Ney Poovan from Karnataka and the size of the amplified band was approximately 1800 bp. The PCR products were cloned in pGEM-T Easy vector, sequenced and analyzed with Clustal-W program.

5.5.3.2 Development of chitinase gene constructs

The chitinase gene from a *Trichoderma viride* of Coimbatore isolate has been isolated and cloned into pGEM-T vector (Fig. 28A). A putative construct has been prepared in pBinAR vector for the use in transgenic development for resistance to fungal pathogens.

5.5.3.3 Molecular characterization of a leaf spot pathogen and a Basidiomycete fungi, causing Trichy wilt

The ITS region of a leaf spot pathogen has been confirmed to *Mycosphaerella eumuse* based on sequence analysis. This fungi cause septoria leaf spot disease more or less similar to Sigatoka leaf spot symptoms. The basidiomycete fungi reported to cause wilt in banana has been confirmed as

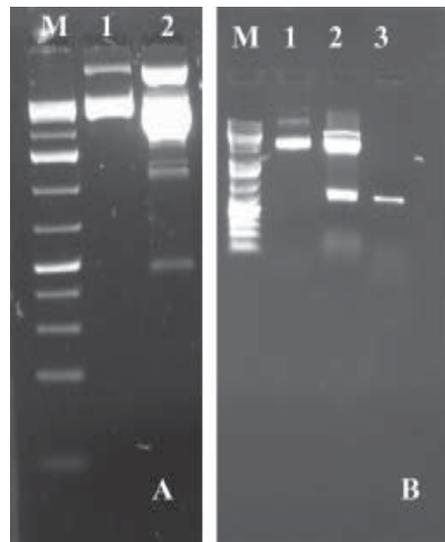


Fig. 28. A) Agarose gel electrophoresis of restriction digested chitinase gene of *Trichoderma viride*-CBE isolate, the gene had an internal EcoR1 site lane M: marker, 1: undigested, 2: digested with EcoR1

B) Restriction digestion analysis of a pGEM-T clone containing amplified ITS region of Trichy wilt fungi: lane M: marker, 1: undigested, 2: Digested with EcoR1 and 3: PCR amplified product of ITS region.

Athelia rolfsii (= *Sclerotium rolfsii*) based on cloning, sequence analysis of rDNA-ITS region of the fungi. Restriction digestion analysis of the clone is shown in Fig. 28B.

5.5.3.4 Survey, isolation and molecular identification of rhizome rot

In Jalgoan district, *Erwinina* corm rot incidence (21%) was recorded in April month planted fields. In Theni district, around 52 fields were surveyed for disease and the incidence ranged from 1.2 % to 12.25%. The fields in the areas of Rayapappanpatti, Chinnamanur, Erasi, Dharmathupatti, T. Sinthalaicherry, Uthamapalayam, Gudaloor were surveyed. The bacterium was isolated and pure cultured. The pathogenicity test has confirmed that the bacteria is soft rot causing *Erwinia* spp. PCR was performed using 16SrDNA specific primers. The 1.5 kb amplified products was cloned and sequenced.

5.5.3.5 Molecular characterization of non pathogenic *Foc* (NPF) isolates

5.5.3.5.1 rDNA-ITS-RFLP of NPF isolates

The genomic DNA of 22 non-pathogenic *Fusarium* isolates were extracted and purified.

The isolated DNA samples were quantified and were subjected to PCR.

PCR was performed to amplify ITS region of rDNA in 22 NPF isolates isolated from wilt suppressive soils. The size of the amplified product ranged from 325 to 370 bp. The PCR products of the ITS amplified region were digested with six restriction enzymes and each produced unique banding patterns (Fig. 29). Patterns that displayed fragments with similar sizes were grouped together and a letter was assigned to each specific pattern (A – G). The enzymes *Hae* III, *Eco*RI, *Hha*I, *Hinf*I, *Taq*I, and *Msp*I produced 4, 2, 4, 3, 4 and 7 bands respectively. Totally 10 ITS genotypes were observed, which could be distinguished by a six letter code designated to each isolate ITS genotype and DFCDAC (group 6) was the most common and 5 isolates of NPF was belonged to that group (Table 11).



Fig. 29. Restriction digestion with the enzyme *Taq*I

Table 11. NPF – ITS – RFLP grouping

Gp	Isolate	Source	Collection site	ITS Genotype					
				<i>Taq</i> I	<i>Msp</i> I	<i>Hinf</i> I	<i>Hea</i> III	<i>Eco</i> RI	<i>Hea</i> I
1.	208b	Karpooravalli	NRCB	A	A	A	A	A	A
2.	208a	Karpooravalli	NRCB	B	B	A	A	A	A
	178a	Monthan	NRCB	B	B	A	A	A	A
3.	177	Rasthali	NRCB	A	C	A	A	A	A
		Poovan	NRCB	A	C	A	A	A	A
4.	FHIA 23	Giant Cavendish type	NRCB	C	D	B	B	B	B
	B ₁	Bluggoe	NRCB	C	D	B	B	B	B
	Pn ₁	Pisang nanga	NRCB	C	D	B	B	B	B
5.	FHIA 23b	Giant Cavendish type	NRCB	C	E	B	C	B	B
	133C	Mortaman	NRCB	C	E	B	C	B	B
6.	140	Karpooravalli	NRCB	D	F	C	D	A	C
	138 a	Mortaman	NRCB	D	F	C	D	A	C
	138 b	Mortaman	NRCB	D	F	C	D	A	C
	K2	Kanaibansi	NRCB	D	F	C	D	A	C
	M2	Matti	NRCB	D	F	C	D	A	C
7.	T1	Tongat	NRCB	E	G	B	E	B	B
	K1	Kanaibansi	NRCB	E	G	B	E	B	B
	M1	Matti	NRCB	E	G	B	E	B	B
8.	184 C	Monthan	NRCB	B	C	A	D	A	D
		Poovan	NRCB	B	C	A	D	A	D
9.	Matti C	Matti	NRCB	F	G	B	B	B	B
10.	139 a	Monthan	NRCB	D	G	B	D	A	B

5.5.3.5.2 rDNA-IGS-RFLP analysis

The universal primer set specific for the amplification of the IGS region had not amplified all the 22 NPF isolates. Nearly 11 isolates had amplified the IGS region and the size of the amplified band was ranged from 1144 bp to 1875 bp. The IGS amplified isolates were digested with six restriction enzymes namely, *HhaI*, *HinfI*, *TaqI*, *MspI*, *HaeIII* and *RsaI*. Among the enzymes *HinfI* had not produced any bands and the bands obtained by other enzymes were not clear. So, they could not be grouped to classify the genotypic characteristics and thus finally, the resolution obtained for rDNA-IGS-RFLP was poor as compared to the rDNA- ITS – RFLP.

5.5.3.5.3 Molecular characterization of pathogenic *Foc* isolates

PCR was performed to amplify ITS region of rDNA in 50 *Fusarium* isolates isolated from different banana growing areas of Bangalore. The size of the amplified band is approximately 600 bp. The PCR products of the ITS amplified region were digested with six restriction enzymes. The enzymes *HaeIII* produced six bands, *EcoRI*-three, *HhaI*-two, *HinfI*-three, *TaqI*-three, and *MspI*-single bands respectively. Banding pattern of *EcoRI* is given in Fig. 30. Totally 4 ITS genotypes were observed, which could be distinguished by a six letter code designated to each isolate. ITS genotype AAAAAA (group 1) was the most common and consisted of 44 isolates. Four isolates were observed under group 2 (BBBBBB) and one isolate each for group 3 and 4 with ABAAAA and AAABAA respectively.

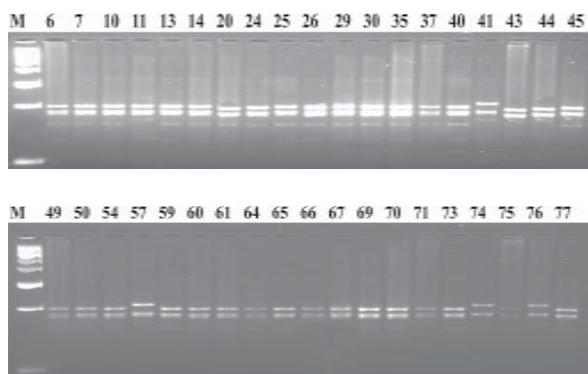


Fig. 30. Restriction digestion with *EcoRI*

5.5.3.6 ISSR analysis

Initially 4 ISSR markers were employed to optimize the PCR protocol for ISSR using 4 different isolates of *Foc*. The amplicon size ranged from 100 bp to 5 kb. All 19 ISSR primers were tested with the same 4 isolates. Out of this 19 only 7 primers gave consistent scorable bands. Then only 7 primers have been chosen for analysis.

Amplified products were scored as either '1' for the presence or '0' for the absence and a combined cluster analysis was done using NTSYS software application for fingerprints obtained with 7 ISSR primers for the 10 *Foc* isolates (Fig. 31). This resulted in two major clusters which grouped most of the Rasthali, Hill banana and Karpuravalli isolates in one cluster and Ney Poovan and Rasthali collected from Tamil Nadu in other cluster with 50% similarity between them. The similarity between two main clusters is only 15%. Result showed that there is existence of variability among Race-1 isolates of *Foc* infecting different cultivars collected from different locations.

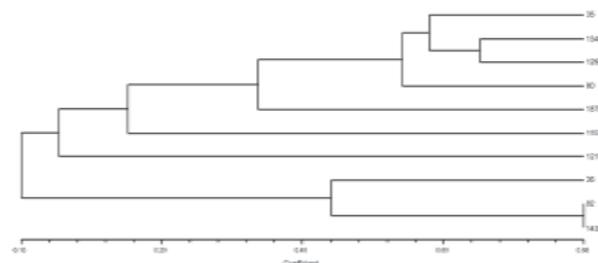


Fig. 31. A combined dendrogram for the 10 *Fusarium oxysporum f. sp. cubense* isolates using the similarity coefficients of markers generated by 7 ISSR primers. 35-Rasthali *Foc* from Tamil Nadu, 36-Rasthali *Foc* from Tamil Nadu, 134-Rasthali *Foc* from Andhra Pradesh, 80-Rasthali *Foc* from Kerala, 129-Rasthali *Foc* from Karnataka, 187-Rasthali *Foc* from Assam, 92-Neypoovan *Foc* from Tamil Nadu, 143- Ney Poovan *Foc* from Tamil Nadu, 115-Hill banana *Foc* from Tamil Nadu & 121-Karpuravalli *Foc* from Karnataka.

5.5.3.6.1 Genetic diversity among race 1 isolates based on ISSR analysis

Thirty seven representative *Foc* isolates that were collected from different parts of India were selected and subjected to ISSR analysis using the same 7 ISSR primers that were used for the above said ten isolates. This is done to analyse the diversity among race 1 isolates. Among the 37 race 1 isolates, 7 race 2 isolates were included for comparison. The banding pattern of 76 PC primers is given in Fig. 32. A combined dendrogram (Fig. 33) using 7 markers for 37 isolates was prepared using NTSYSpc software. Dendrogram revealed the presence of 3 main clusters and many sub-clusters. Two *Foc* isolate collected from hill banana in Tamil Nadu showed 90% similarity. Four isolates infecting Ney Poovan collected from Tamil Nadu and Karnataka showed 100% similarity. *Foc* isolates infecting Rasthali, Ney Poovan and Karpuravalli belonging to race 1 and *Foc* infecting Monthan belonging to race 2 grouped together in cluster 1. In cluster 2, Rasthali infecting *Foc* isolates collected from Tamil Nadu and North Eastern regions were grouped together. In cluster 3, both race 1 and race 2 isolates were grouped together. This shows that there are

lot of variability present among the isolates collected from different varieties and different states.

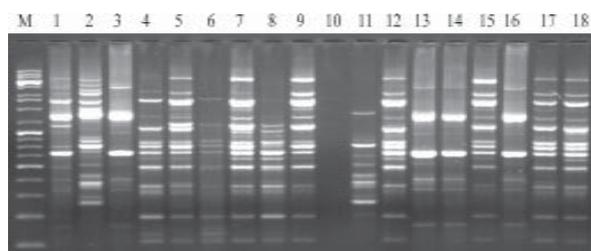


Fig. 32. ISSR profile of *Foc* using primer 76 PC

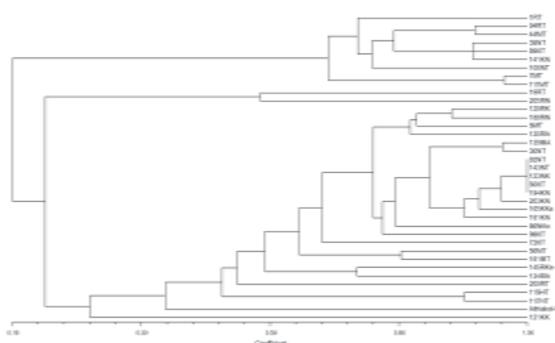


Fig. 33. Combined dendrogram for 37 *Foc* isolates using 7 ISSR primers. R-Rasthali; T-Tamil Nadu; M-Monthan; K-Karnataka; N-Ney Poovan; Ke-Kerala; K-Karpuravalli; A-Andhra Pradesh; H-Hill banana; N-North Eastern regions. The numbers given before the letters are the *Foc* isolate numbers

5.5.3.6.2 Genetic diversity among race 2 isolates based on ISSR analysis

The same set of primers used for the above experiment has been used for 38 isolates which consists of 28 race 2 and 10 race 1 isolates. Three isolates (one isolate of race 1 and two isolates of race 2) were not considered for scoring because of the faint bands produced for these isolates. Race 1 isolates have been added to compare with race 2. Banding Pattern of 75 PC primer is given in Fig. 34. A combined dendrogram (Fig. 35) for all 7 ISSR primers resulted in the formation of 2 main clusters I and II. All the Monthan isolates collected from various part of Tamil Nadu have grouped under cluster I. All the four Rathali *Foc* isolates collected South India are grouped in the same cluster I. The

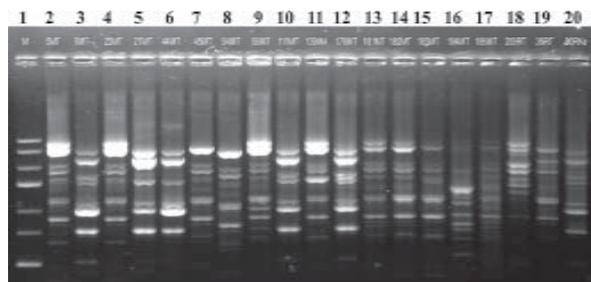


Fig. 34. ISSR profile of race 2 *Foc* along with Race 1 isolates using 75 PC ISSR primer

Ney Poovan isolates collected from Karnataka and Pudukkottai (TN) have also grouped here and the Ney Poovan isolate from Tirunelveli (TN) grouped under cluster II.

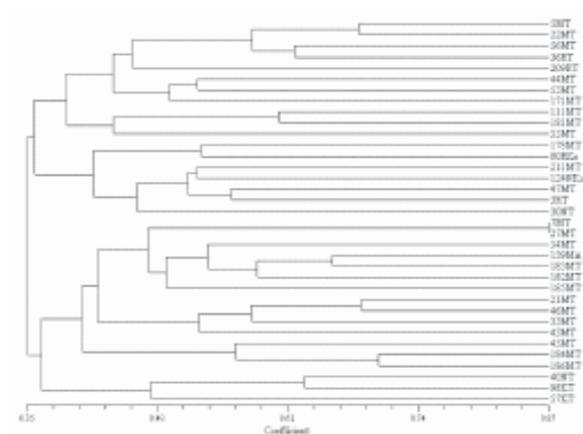


Fig. 35. Combined dendrogram for 38 *Foc* isolates using seven ISSR primers

Seven of the nine Monthan isolates collected from Madurai have grouped under cluster II along with monthan isolate collected from Andhra Pradesh (139). Two Karpooravalli isolates (98, 57) are grouped under cluster II. Only 35% similarity is observed between cluster I and II. The isolates 7 and 27 which are collected from Theni and Pudukkottai districts respectively showed 100% similarity. The result revealed that there is a wide variation among the *Foc* isolates collected from different part of India.

5.5.4 Studies on Viral Diseases and their Management

5.5.4.1 Survey for viral diseases

Survey undertaken in Jalgaon district, Maharashtra revealed high incidences of bunchy top. The incidence was more in Nimbhore, Nhavi, Chinawal. An average incidence of 28.72% was recorded in three taluks. The incidence was more in ratoon than the plant crop. In Theni district, Tamil Nadu incidence of bunchy top in two gardens was recorded to be 23.65%. A recent survey was conducted in Vaishali and Muzafarpur districts of Bihar for banana viruses revealed the presence of BBTv (upto 5%) and BSV (upto 37%).

5.5.4.2 Effect of increased dose of fertilizers on BBrMV infected plants

A field experiment on the effect of graded doses of fertilizer on growth and yield parameters of healthy and BBrMV infected plants in ratoon of cvs. Robusta and Ney Poovan was studied. In infected plants of Ney Poovan, when the fertilizers dose is increased, the bunch weight also increased proportionately whereas in healthy plants, the rate of increase was less. Irrespective of the different

fertilizer treatments, there was significant increase in height, girth, number of functional leaves, number of fingers and bunch weight in healthy plants as compared to infected plants. Similar result was observed in cv. Robusta. Significant difference was observed among the treatments due to application of graded dose of fertilizer also in ratoon crop of Robusta. Bunch weight of infected and healthy plants significantly increased with increased doses of fertilizer.

5.5.4.3 Molecular characterization of banana viruses

BBTV rep gene from Andaman, Solapur and Cumbum have been cloned, sequenced and deposited in NCBI genbank. *In silico* analysis was done, which revealed their homology between 91.5% and 97.9% with published amino acid sequences of South Pacific group. The phylogenetic analysis of rep gene of all the three isolates revealed that they belonged into South Pacific group, but not Asian group (Fig. 36).

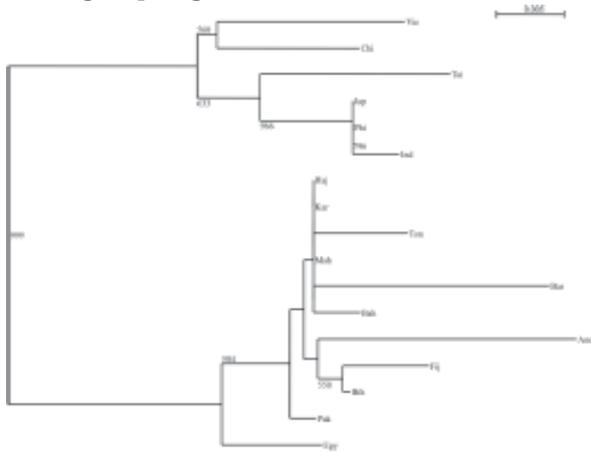


Fig. 36. Neighbour-joining phylogenetic dendrogram based upon amino acid sequences alignment of replicase encoding components of various banana bunchy top virus isolates. Numbers at nodes represent bootstrap values

BBTV truncated rep gene has been cloned for use in construct development. Coat protein gene and stem loop region of BBTV has been amplified from Foorkey diseased large cardamom and the amplicons were cloned. Partial coat protein gene of BBrMV-Coimbatore isolate has been cloned and sequenced. This was 100 % similarity with BBrMV-TRY isolate.

Coat protein gene of CMV- banana isolate from Hosur and Jalgaon isolates have been cloned and sequenced. Sequence analysis of CP gene showed high level of sequence conservation within subgroup I isolate at nucleotide (90-97%) homology at nucleotide level respectively. That clearly showed that our three CMV-banana isolates belong to CMV subgroup I.

A 586 bp length BSMysV partial fragment from infected cv. Poovan collected from Coimbatore and *M. acuminata* spp *zebrina* (Bangalore) has been amplified, cloned and sequenced for genetic diversity analysis.

5.5.4.4 BSMysV genome cloning and characterization

Complete genome of BSMysV infecting Poovan has been cloned and sequenced and the sequence was submitted in NCBI genebank. The complete 7650 bp genome had 99% sequence homology with published BSMysV sequence (Fig. 37).

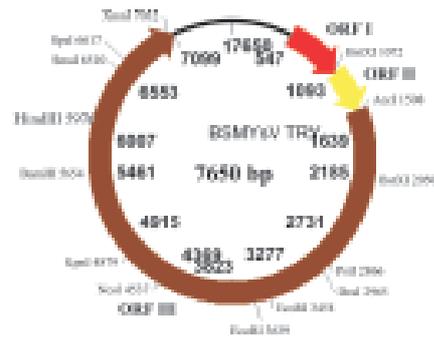


Fig. 37. Cloning and sequence analysis of the complete genome of BSMysV -Poovan isolate. Genome map indicating the organization of the virus

5.5.4.5 Expression of recombinant fused CMV viral coat protein in *E. coli*

CMV coat protein gene was cloned into pMAL-expression vector and transformed into *E. coli*. The fusion protein was expressed by induction using IPTG. Various conditions were standardized for expression. The expressed protein was in insoluble

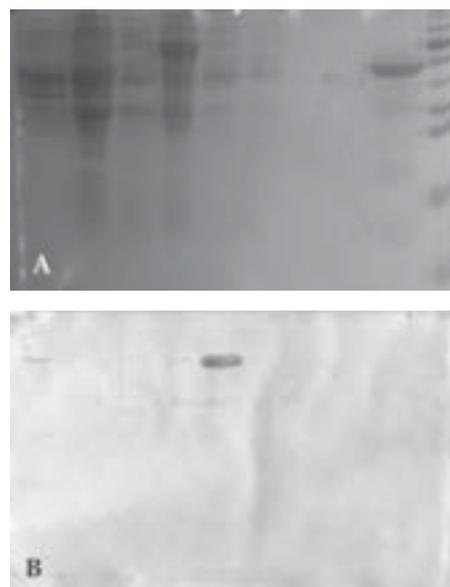


Fig. 38. Over expressed recombinant CMV coat protein resolved by SDS-PAGE (A) and confirmed using specific antiserum by western blot (B)

fraction which was confirmed through SDS-PAGE analysis (Fig. 38A). Expressed protein has also been confirmed as 'CMV-CP', through western blotting using CMV specific polyclonal antiserum (Fig. 38B).

5.5.4.6 Supply of virus indexed Hill banana mother plants

As per the QRT recommendation, healthy suckers of hill banana indexed for BBTV were supplied to Hill banana growers of lower Pulney hills and also to Dept. of Biotechnology and HC and RI, TNAU for research purpose.

5.5.4.7 Screening Poovan banana for BSV genome integration using molecular markers

An attempt was made to differentiate the BSV integration pattern in 14 Poovan plants using 40 RAPD, 2 SSR, 2 ISSR markers. Polymorphic bands were observed with RAPD primers but it needs further confirmation. One of the ISSR markers used gave polymorphic bands which might be useful to analyse the integration (Fig. 39).

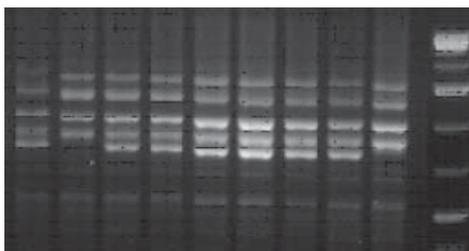


Fig. 39. Agarose gel electrophoresis of ISSR marker derived bands from BSV positive Poovan plants: Lanes 1,3,4,5,6,7& 8 - Healthy Poovan sample; L 2& 9 - Infected Poovan M - 1 kb ladder

5.5.4.8 Developing severity index for BSV

Poovan plants planted in the year 2005, was observed for expression of typical BSV symptom naturally in their subsequent ratoon crops. Initially, in the first year, the percent expression was 1.20%, later it increased to 2.8% and 4.4% respectively in the first and second ratoon. With regard to developing the severity scale for assessing the BSV damage, a 0-3 and 0-4 rating scale has been compared to assess the BSV in Poovan.

5.5.4.9 Developing multiple virus gene construct for transgenic development resistance to viruses

In order to develop gene construct for multiple virus resistance, the primers were designed for three viruses, which gave expected PCR product of 200 bp each of BBrMV, CMV and BBTV. These were cloned in sense direction individually as well as in combinations. Primers for intron sequences were also designed to develop the RNAi construct.

5.6 EXTERNALLY FUNDED PROJECTS

5.6.1 Evaluation of *in-vitro* plants derived from meristem culture and embryogenic cell suspension of cv. Rajeli (AAB)

In NRCB- BARC collaborative project to evaluate plants of cv. Rajeli derived from ECS in comparison with plants derived through meristem culture was undertaken. Twenty five plants each were planted in the field and evaluated for growth, yield and molecular traits. Morphotaxonomically there was no difference between the plantlets derived from meristem and ECS. They were found to be statistically on par with respect to yield and other related traits. Molecular characterization using SSR markers also suggested their genetic similarity.

5.6.2 Evaluation of cv. Giant Cavendish for better traits-through induced mutations using gamma rays at 3 different doses (5 Gy, 10 Gy and 30 Gy)

In NRCB- BARC collaborative project, cv. Giant Cavendish was irradiated at 5 Gy, 10 Gy and 30 Gy were evaluated at NRCB field gene bank. Plants treated with 30 Gy failed to establish in the field. 5 Gy and 10 Gy plants were evaluated with normal plants of cv. Giant Cavendish. Two years results suggested that there were no positive mutants obtained at these lower irradiation doses. Higher doses are required to obtain mutants, which will be tested for positive mutations.

5.6.3 Developing transgenic banana resistant to BSV and BBTV (cp mediated)

5.6.3.1 Genome characterization of BBTV

All six genomic components of BBTV - Hill banana isolate have been sequenced (Fig. 40). They were analyzed and compared with other isolates of the world. *In silico* analysis was done which revealed their homology between 93 and 99% nucleotide with published sequences of South Pacific group. Based

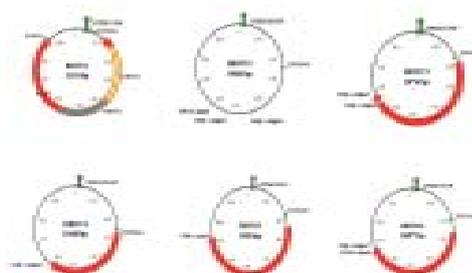


Fig. 40. Cloning and sequence analysis of BBTV: Genome organization of banana bunchy top virus - Hill banana isolate

on the phylogenetic analysis for the component 3 BBTV-Hill banana isolate, it is confirmed that the isolate belong to South Pacific group but not Asian group (Fig. 41).

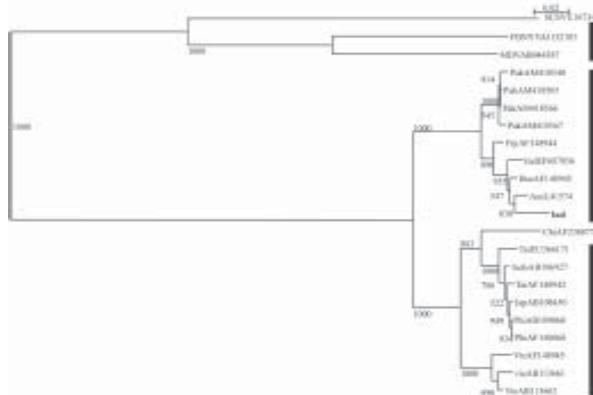


Fig. 41. Neighbour-joining phylogenetic dendrogram based upon a nucleotide sequence alignment of all available capsid protein encoding components (DNA component 3) of BBTV, showing their relationship to the newly characterized components from India (highlighted with bold letters). Bootstrap values for internal support of the branches are given along the branches. Three major clades are marked: the Asian, the South Pacific and the Nanovirus groups. Accession numbers are preceded by abbreviations for the places of origin

5.6.3.2 Developing gene constructs with BBTV cp gene

A BBTV cp gene sense construct has been prepared in pCAMBIA 1301 vector having gene for hygromycin selection. The entire cassette was released by restriction digestion and cloned into pCAMBIA 1301. This vector has GUS gene to identify putative transformants.

5.6.3.3 Promoter construct from BBTV intergenic regions

Primers were designed to amplify the intergenic regions of six BBTV components. The intergenic region was amplified and expected sized products were obtained. A promoter construct has been made with IR region of BBTV-5 by replacing the CaMV 35S promoter from pBI121 vector. A 586 bp length BSMYV intergenic region has been cloned to develop a promoter construct.

5.6.4 Developing ECS for hill banana, Poovan and transformation for transgenic development

Developed ECS for hill banana cv. Virupakshi and also for cv. Poovan. The ECS developed had high number of non-embryogenic cells. Further standardization of obtaining high quality ECS is in progress. Direct embryogenic calli were also

observed in Hill banana. The embryogenic cell suspensions are being kept in the semisolid maturation medium for embryo development. Agrobacterium strain LB4404 harboring pBIN-AR-BBTV-cp has been used for co-cultivation with the calli. The transformed calli was checked for the presence of BBTV cp gene by PCR. Out of 5 tested, one was positive.

5.6.5 Developing diagnostic kit for BSV and BBrMV

BBrMV coat protein gene was cloned into pMAL-expression vector and transformed into *E. coli*. The fusion protein was expressed by induction using IPTG. The expressed protein was found in the pellet, which was confirmed through SDS-PAGE analysis. Expressed protein has also been confirmed as "BBrMV-CP" through western blotting using potyvirus specific polyclonal antiserum obtained from DSMZ, Germany.

Non-radioactive DIG-oxigenin labelled probes were developed using DIG-high prime labelling and DIG northern starter kit and used for detecting the virus in suspected banana samples and RNA probes prepared using northern starter kit able to detect BBrMV more effectively than DIG-high prime labelled probe. Non-radioactive DIG-oxigenin labelled probes developed for two BSV fragments, which were found effective in detection. A partial fragment of 1115 bp size covering and 2.5 kb size covering HC-Pro and CI region of BBrMV was cloned into pGEM-T Easy vector sent for sequencing as part of molecular characterization.

5.6.6 Developing replicase gene construct of BBTV

The BBTV antisense replicase gene construct has been developed using pCAMBIA2301 binary vector (Fig. 42). This binary construct has been

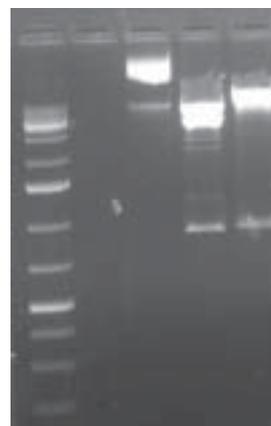


Fig. 42. Restriction digestion of pCAMBIA2301-antisense replicase gene construct M- Marker (1kb ladder) Lane 2-undigested recombinant plasmid; Lane 3, 4-digestion with *Bam*HI+ *Sac*I

mobilized from *E. coli* to *Agrobacterium tumefaciens* LBA4404 strain. The BBTV replicase gene (sense) construct has been developed in the binary vector of pCAMBIA 1301, which has hpt-II gene as selectable marker.

5.6.7 Developing ECS for Hill banana

ECS for hill banana cv. Virupakshi was developed. Phenolics were a major problem in obtaining the callus, for which three antioxidants were used to eliminate diffused product of oxidative browning in and around the explants. Charcoal

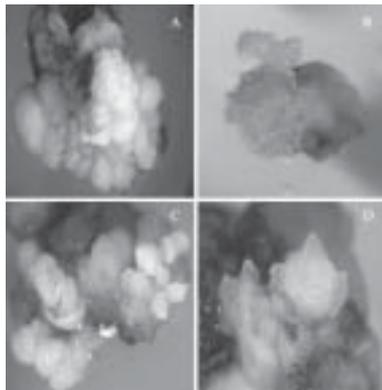


Fig. 43. Different types of calli of Hill banana obtained using immature male flower buds. A- Compact calli; B-Friable embryogenic calli; C&D- Direct somatic embryo

found better antioxidant for overcoming the blackening. Cell suspensions were obtained but with mixed embryogenic and non-embryogenic cells. Direct embryogenesis was also noticed from calli. The embryogenic cell suspensions are being kept in the semisolid maturation medium for embryo development. (Fig. 43).

5.6.8 Transformation of ECS with antisense rep gene construct and confirmation through PCR

The antisense rep gene construct was used for transformation through *Agrobacterium* co-cultivation method. The putative transformants were selected using Kanamycin incorporated selection media and two of the transformants were found PCR positive for rep gene. Gus assay performed gave positive blue color in the transformed calli.

5.6.9 Developing promoter construct from intergenic region of BSV

A promoter construct using intergenic region of BSMysV –Try isolate has been prepared by replacing the CaMV-35S promoter. Similarly another promoter construct using IR of BBTV-component 3 has been prepared. Both the promoters drives GUS gene.

6 Technology Assessed and Transferred

Rice chaffy grains formulation of entomopathogenic fungus, *Beauveria bassiana* and talcum powder formulation of entomopathogenic nematode, *Heterorhabditis indica* and CD of NRCB technology on Management of Banana Weevil were released during the Field Day on 'Eco-Friendly Management of Banana Weevil' held at Horticultural Research Station, TNAU, Thadiyankudisai on 2nd June, 2007.

The technology of artificial ripening of banana using Ethrel was transferred to M/s Sivagami Cold Storage Pvt. Ltd., Viralimalai, Trichy and provided consultancy services in establishing commercial ripening chamber.

The technology of banana juice and fig preparation was transferred to M/s Yendal Health Foods Pvt.Ltd, Pattiveeranpatti, Dindigul dt.

Television Talk

S. No.	Topic	Name of the Scientist	Date(s) of Broadcast
1.	Nutrient management in banana	K. J. Jeyabaskaran	16 & 17. 01. 2008
2.	Management of banana weevils	B. Padmanaban	07. 02. 2008
3.	Value addition in banana	C. K. Narayana	14. 02. 2008

Radio Talk

S. No.	Topic	Name of the Scientist	Date(s) of Broadcast
1.	Micronutrients in banana	K. J. Jeyabaskaran	04. 05. 2007
2.	Corm rotting in banana	R. Selvarajan	22. 05. 2007
3.	Management of banana weevils through eco-friendly approaches	B. Padmanaban	25. 08. 2007
4.	Banana by-product	C. K. Narayana	13. 11. 2007

Exhibitions: NRCB participated in the exhibitions held during the following Seminars, Trainings, Field Days, Kisan Melas, etc.

Sl. No.	Title	Organised by/ Place/ State	No. farmers/ visitors participated	Date
1.	Coconut Festival	Coconut Development Board, Thiruvananthapuram, Kerala	6000	03-06.05.07
2.	Horticulture Summit - 2007	CIH & CISH, Lucknow, U. P.	6000	16-19.06.07
3.	Workshop - cum - Exhibition on Agriculture	Tamil Nadu State Agriculture and Horticulture Department, Trichy, T. N.	2000	30.06.07
4.	IIVR Foundation Day	IIVR, Varanasi, U. P.	1500	28.08.07
5.	Banana Farmers Training	NHM & Tamil Nadu State Hort. Dept., Salem, T.N.	1500	29.09.07
6.	National Conference on Banana	AIPUB and NRCB, Trichy, T. N.	5000	25-27.10.07
7.	Farmers and Scientist Interaction Programme	Nagar Uzhavar Mandram, NRCB and Lalgudi Uzhavar Mandram, Lalgudi, T. N.	600	11.05.07
8.	Kisan Mela-2007	Sugarcane Breeding Institute, Coimbatore, T. N.	5300	11-13.12.07
9.	South Indian Agricultural Fair-2007	Kerala Agriculture University, Thrissur, Kerala	5600	27-30.12.07
10.	International Hort. Expo- 2008	NAS, New Delhi	1500	31.01.08 to 02.02.08
11.	National Vegetable Growers Mela	IIVR, Varanasi, U. P.	1000	09.02.08
12.	Kisan Mela - cum - Exhibition	NHM & Bihar State Horticulture Dept., Kathihare, Bihar	5000	09-10.03.08



Dr.M.M. Mustaffa's participation in the valedictory function of Kisan Mela, SBI, Coimbatore

7 Education and Training

Guidance of Students

Name of the student	Title of the thesis	Name of the guide
G. Anusuya	Interaction between entomopathogenic nematode and root-knot nematode (<i>Meloidogyne incognita</i>) on banana in cv. Robusta	P. Sundararaju
N. Jeenathunisa	Efficacy of biofertilizers against root-lesion nematode (<i>Pratylenchus coffeae</i>) infesting banana in cv. Rasthali	
T. Sangeetha	Isolation, identification and evaluation of <i>Pseudomonas fluorescens</i> strain against root-lesion nematode in banana	
K. Sheeba Sahaya Mary	Biological suppression of root-knot nematode in banana using two antagonistic fungi <i>Trichoderma viride</i> and <i>T. harzianum</i> .	
P. Vanitha	Evaluation of the Cyanobacterium for its toxic effects against banana nematode	
C. Saranya	Evaluation of biocontrol agents against citrus mealy bug, <i>Plannococcus citri</i>	B. Padmanaban
R. Sivapriya	Compatibility of agriculturally important microbials against commonly used fertilizers	
V. Rohini	Evaluation of biocontrol agents against banana aphid, <i>Pentalonia nigronervosa</i>	
N. Dhivya Ruba	Survey for microbial entomopathogens for banana weevil management	
M. Raziya Sulthana	Isolation of active principles of <i>Dodonaea angustifolia</i> for the management of banana stem weevil, <i>Odoiporus longicollis</i>	
G. Shakitha Parveen	Screening of semiochemicals and banana leaf sheath volatiles against banana stem weevil, <i>Odoiporus longicollis</i>	
S. Rasina Nilofer	Investigations on banana leaf sheath volatiles (Cultivars: Nendran and Poovan) for the management of banana weevil	
M. Banu	Isolation and characterization and evaluation of isolates of entomopathogenic fungi (Non-pathogenic <i>Fusarium oxysporum</i>)	
T. Karthika	Isolation and characterization and evaluation of isolates of entomopathogenic fungi (<i>Verticillium lecanii</i>)	
R. Brinda	Comparative study of microbial, enzymatic and chemical retting for the extraction of banana fibre	C. K. Narayana
B. S. Basanth Kumar	Studies on banana stem and stem juice powder	
N. Viswasundar	Standardization of parameters for the bulk production of banana wine	



P. Abirami	Preliminary studies on the molecular markers associated with (<i>M. musicola</i>) Sigatoka disease resistance in banana	S. Uma
B. Mahalakshmi	Effect of biofertilizers and sucker decortications on macropropagation of banana	
F. Stanley Rosarin	Genotypic expression of the phenotypic trait ashyness in banana (<i>Musa</i> spp) using DNA markers	
B. Kiruthika	Preliminary studies on <i>in vitro</i> screening for salt stress in banana (<i>Musa</i> spp) varieties	
M. Muhilan	Studies on biochemical changes of low temperature stored green banana fruits and amplification of <i>BMV8</i> gene from genomic DNA of different banana cultivars	I. Ravi
N. Madhurambika	Studies on biochemical changes of drought affected banana cultivars and amplification of <i>DREB</i> genes from genomic DNA of different banana cultivars	
C. Anitha	Studies on exogenous application of salicylic acid on plant growth and activities of antioxidative enzymes in NaCl treated banana plants	
J. Ramachandran	Evaluation of drought component traits under soil moisture deficit conditions in different banana genotypes	
L. Nancy	Amplification of drought and salt tolerant genes from the genomic DNA of banana plants	
R. Sudha Parimala	Studies on NaCl salinity induced biochemical changes in cultivated bananas	
Y. Lukka Thuyavan	Cloning of coat protein gene of banana bract mosaic virus infecting plantain	R. Selvarajan
M. Mary Sheeba	Cloning and expression of coat protein gene of Cucumber Mosaic Virus-Banana isolate	
K. Santhanalakshmi	Molecular analysis of BSV infected banana by using ISSR markers	
A. Bhuvana	Amplification, cloning and sequence analysis of partial fragment of Banana Streak Mysore Virus <i>Musa accuminata</i> spp. <i>zebrina</i> collected from Bangalore	
S. Deepthi	Cloning and sequence analysis of replicase gene of Banana Bunchy Top Virus- Andaman isolate	
T. Selvi	Cloning and sequence analysis of replicase gene of Banana Bunchy Top Virus - Cumbum isolate	
N. Shabana	Cloning and sequence analysis of replicase gene of Banana Bunchy Top Virus - Solapur isolate	
N. Shalini	Cloning and sequence analysis of partial coat protein gene of Banana Bract Mosaic Virus - Coimbatore isolate	



G. Thamaraiselvi	Cloning and sequencing of cucumber mosaic virus infecting banana collected from Jalgaon district of Maharashtra	
G. Shanmugapriya	Cloning and sequence analysis of coat protein gene of Cucumber Mosaic Virus infecting banana- (Tiruchirapalli isolate)	
S. Padmapriya	Cloning and sequence analysis of a partial fragment of ORF-III of Banana Streak Mysore Virus (BSMysV) - Coimbatore isolate	
P. Murugananthan	Survey, isolation and molecular identification of rhizome rot causing bacteria in banana	
S. Venkat Kumar	Molecular characterization of isolates of <i>Fusarium oxysporium</i> f. sp. <i>cubense</i> , the wilt pathogen of banana using ISSR markers	
G. Priyalakshmi	Detection of episomal and integrated banana streak virus (BSV) genomic fragments in various commercial cultivars of <i>Musa</i>	
M. Kalaiselvi	Studies on soil microbial population and nutritional changes as influenced by spacing, plant density and fertigation in the rhizosphere of banana cv. Grand Nain	V. Kumar
H. Divya	Changes in phenol content and disease defense enzymes in banana inoculated with <i>Pratylenchus coffeae</i> (Zimmermann) Goodeyi	M. Mayilvaganan
K. Valli	Molecular and biochemical characterization of polyphenol oxidase in bananas in relation to oxidative browning	
M. Sowmiya	Standardization of RNA isolation and cDNA synthesis from banana roots and confirmation through RAPD markers	S. Backiyarani
K. Prasanya Selvam	Determination of the best marker system for diversity analysis within the Cavendish subgroup of banana (<i>Musa</i> spp)	M.S. Saraswathi
G. Karunya	Preliminary studies on induced mutation using EMS in banana cv. Rasthali	

8 Awards and Recognitions

Dr. M. M. Mustaffa, Director, NRCB was awarded 'Kadali Purashkar' for his outstanding contribution to banana research and development in India by the Association for the Improvement in Production and Utilization of Banana (AIPUB) at National Conference on 'Production and Utilisation of Banana for economic livelihood and Nutritional Security' during 25-28, October 2007 at Tiruchirapalli.

Dr. P. Sundararaju was awarded 'Kadali Purashkar' for his outstanding contribution to banana research and development in India by the

Association for the Improvement in Production and Utilization of Banana (AIPUB) at National Conference on 'Production and Utilisation of Banana for economic livelihood and Nutritional Security' during 25-28, October 2007 at Tiruchirapalli.

Dr. P. Sundararaju was appointed as a subject matter expert for setting up of question papers for Ph. D. Nematology during the academic year 2007-08 in Tamil Nadu Agricultural University, Coimbatore.

Dr. P. Sundararaju was the Member Secretary of Second Quinquennial Review Team (QRT) constituted by the Indian Council of Agricultural Council (ICAR), New Delhi, to review the five years (2002-2007) progress of the Centre.



Dr.M.M. Mustafa receives *Kadali Purashkar* award



Dr.P. Sundararaju receives *Kadali Purashkar* award



Dr.S. Uma receives Fellow of AIPUB award



Dr.C.K. Narayana receives *Rashtriya Sanman* award

Dr. P. Sundararaju was a selection committee member for recruiting Scientific and Training Associates for KVKs of Thanjavur and Thenkasi on 12th November 2007.

Dr. P. Sundararaju has been elected as the General Secretary-AIPUB in the General Body Meeting held on 27. 10. 2007.

Dr. C.K. Narayana received '*Rashtriya Sanman*' Award for his outstanding contribution in the field of Agriculture from Economic Growth Association of India, Mumbai.

Dr. S. Uma was recognized as 'Fellow of AIPUB' for her outstanding contribution to banana research and development in India by the Association for the Improvement in Production and Utilization of Banana (AIPUB) at National Conference on "Production and Utilisation of Banana for economic livelihood and Nutritional Security" during 25-28, October 2007 at Tiruchirapalli.

Dr. S. Uma was invited to convene a session on 'Role of crop improvement in pest and disease management' in the International Symposium on 'Recent Advances in Banana Crop Protection for Sustainable Production and Improved Livelihoods' during 10-15 September, 2007 at White River, South Africa.

Dr. S. Uma was invited to present a lead paper on "Crop improvement strategies for pest and disease management in banana" in the International Symposium on 'Recent Advances in Banana Crop Protection for Sustainable Production and Improved Livelihoods' during 10-15, September 2007 at White River, South Africa.

Dr. S. Uma was invited by FAO/IAEA, Vienna, Austria as the National Observer in the Second Research Co-ordination Meeting (RCM) of the co-ordinated Research project on 'Molecular tools for quality improvement in vegetative propagated crops including banana and cassava' held at Thiruvananthapuram during 5-9, April 2007.

Dr. I. Ravi represented as ICAR nominee on 12th June 2007 in the selection committee for recruiting a Subject Matter Specialist and Farm Manager for the KVK-SCAD, Vagaikulam, Tuticorin.

Dr. R. Selvarajan was the external examiner for conducting qualifying *viva voce* for five M. Sc. students of Department of Plant Pathology, CPPS, TNAU on 13th Oct. 2007.

Dr. R. Selvarajan acted as convener for the technical session on 'Badnavirus and Nanovirus' in International Conference on Emerging and

Reemerging Viral Diseases of the Tropics and Sub-tropics held at IARI, New Delhi, during 11-14, December 2007.

Drs. R. Selvarajan and M. M. Mustafa received best poster presentation award for the paper on "Erwinia head rot: An emerging problem in tissue culture banana in India" in National Conference on Banana: "Production and utilization of banana for economic livelihood and nutritional security" held at Trichy, Tamil Nadu, during 25 – 28, October 2007. The award was received from Mr Jairam Ramesh, Hon'ble minister for state for commerce, Govt of India.

Dr. R. Selvarajan was recognized as guide for M. Phil. scholars for the project work in Biotechnology and Botany by Vinayaka Mission Research Foundation Deemed University, Salem, Tamil Nadu.

Dr. R. Selvarajan is re-nominated as member in the executive council of Association for Improvement in Production and Utilization of Banana (AIPUB), Trichy.

9 Linkages and Collaboration in India and Abroad

Developed linkages with CICY, Mexico, NARI, Laloki, Papua New Guinea, ARC-ITSE, Nelspruit, South Africa through INIBAP, France and developed a project on 'Evaluating Banana germplasm for drought tolerance for the benefit of resource poor farmers' for funding under Global Crop Diversity Trust, FAO, Rome, Italy.

A new project entitled 'Induced mutation-A crop improvement strategy for developing dwarf and Sigatoka leaf spot resistant banana cv. Grand Nain' has been initiated in collaboration with BRNS, DAE, GOI, Mumbai.

10 Publications

Research Papers

Jeyabaskaran, K. J. and Pandey, S. D. 2008. Effect of foliar spray of micronutrients in banana under high soil pH condition. *Ind. J. Hort.*, **65** (1):102-105.

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Book Chapter

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Technical Bulletins

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Narayana, C.K., Kamaraju, K. and Priyalakshmi, G. 2007. Banana recipe (in Tamil). *Association for Improvement in Production and Utilization of Banana*, NRCB, Tiruchirapalli.

Narayana, C. K. and Priyalakshmi, G. 2007. Banana-Nature's Marvel-Therapeutical and Medicinal Values. *Association for Improvement in Production and Utilization of Banana*, NRCB, Tiruchirapalli.

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11 Consultancy Services and Commercialization of Technologies

Resource generation through contract research

M/s Indian Oil Corporation Limited, Chennai sanctioned Rs. 2,43,200/- for the research project entitled 'Evaluation of Servo agrospray against selected insect pests of banana'.

M/s Indian Oil Corporation Limited, Chennai provided Rs. 1,80,000 for undertaking the contract research project on evaluating Agrospray of IOC against sigatoka leaf spot disease of banana during the year 2007-'08.

M/s Tropical Agro, Chennai granted Rs. 1,50,000 for the project for testing their product, Fungaflor (Imazalil) against post-harvest (crown

rot) disease of banana in cultivar Ney Poovan and Grand Nain.

Resource generation through contract service-virus testing in banana

Tissue culture banana plants and mother plant suckers from different TC industries such as SPIC-ABC, Coimbatore; Godrej Agrovvet, Ranga Reddy Dist, AP; Ramco Biotech, Bangalore; Reliance Bio, Mumbai; Jain irrigation system Ltd., Jalgaon; Gogle Biotech, Pune; Blossom Tissue Culture Nursery, Hosur; Arcadia Agro, Vadodara; Rise'n Shine Biotech, Pune and PK KVK, Puduchery were indexed with PCR and NASH technology. This financial year 4,525 samples have been tested for BBTV (2,821), BBrMV (239), BSV (1,402) and CMV (and approximately an amount of Rs. 9 lakhs was generated by the Centre under this contract service.

Resource generation through training

An amount of Rs 40,000 resource was generated by conducting the paid training on 'Molecular characterization and diagnostic techniques for plant viral and fungal pathogens' conducted during 21-28, May 2007.

Resource generation through sales

Sales of farm produce - Rs.4.5 lakhs.

12 RAC, IMC, QRT and IRC

RAC Meeting

The 9th Research Advisory Committee meeting commenced with the field visit to Research Farm in the morning of 27th November, 2007. The Chairman and all the members reviewed the ongoing experiments in the field. All the experiments under various projects were explained to the RAC Chairman and members by the Director and the scientists concerned. The Chairman and members of the RAC appreciated the well maintenance of the research farm and the research work under different programmes. After the field visit, the RAC visited to all the laboratories, inspected the infrastructure



RAC Chairman and members with Director and scientists of the Centre



facilities available and interacted with scientists. In the afternoon, the 9th Research Advisory Committee Meeting was held and Dr. R. M. Pandey, Chairman, chaired the session and conducted the proceedings.

Dr. M. M. Mustaffa, Director, National Research Centre for Banana, Trichy after welcoming the Chairman and members of RAC, gave a brief account of the salient research achievements made by the NRCB during the last one year. The Action Taken Report on the recommendations of 8th RAC was presented by Dr. P. Sundararaju, Member Secretary.

RAC Members

1.	Dr. R. M. Pandey	: Chairman
2.	Dr. S.J. Singh	: Member
3.	Dr. R.C. Tiwari	: Member
4.	Dr. (Mrs.) Lalitha Anand	: Member
5.	Dr. N. Kumar	: Member
6.	Dr. B. Bandyopadhyay	: Member
7.	Shri . V.L. Mahajan	: Member
8.	Shri . S.S. Kadam	: Member
9.	Dr. M.M. Mustaffa	: Member
10.	Dr. P. Sundararaju	: Member Secretary

The Chairman and the members were fully satisfied about the action taken on the recommendation of previous RAC meeting and approved the minutes of 8th RAC. This was followed by presentations of individual scientists on the various research activities on banana. After detailed discussions on the research progress of the on going projects, the RAC suggested the future programmes to be carried out in various areas of research in banana.

The Chairman and other RAC Members appreciated the work carried out by the scientists and also the infrastructure facilities developed at the Centre. The RAC was of the opinion that the research programmes are meeting the demands of the farming community.

The RAC after reviewing the research activities of the NRCB suggested the following recommendations:

Crop Improvement

- While evaluating the hybrids population on banana, non-parthenocarpic hybrids may also be evaluated for desirable economic and biotic resistance traits as it is likely that some of the non-parthenocarpic hybrids may get reverted

to parthenocarpic hybrids in the subsequent generation.

- Efforts should be made to develop a new variety/ hybrids of banana with the help of biotechnological approaches against biotic and abiotic stresses.

Crop Production

- Since *Sespania* sp. is a good source of organic matter, it should be tried as an inter-crop in banana garden. Glyricidia may also be grown in the border of the farm as nitrogen fixing green leaf manure plant.
- Awareness should be created among the public for not to use the calcium-carbide chemicals for ripening banana as this chemical has been banned and cause health hazard.
- A series of cottage type processing houses should be established with single or cluster of villages. They will prepare the intermediary products and transfer to big processing houses, who in turn prepare the final product for domestic purpose and also for export.

Crop Protection

- Effective strains of bio-control agents have to be identified to impart resistance against nematodes and wilt diseases.

General

- A proposal for establishing an ATIC Centre at NRCB may be sent to Council for approval.

Institute Management Committee

The 11th IMC meeting was held on 28th July 2007 under the chairmanship of Dr.M.M. Mustaffa, Director, NRCB, Trichirapalli. He welcomed the management committee members and briefed on the research achievements of NRCB. Shri.B. Vijayakumar, AAO, NRCB, Trichy and Member Secretary presented the action taken report on the proceedings of the sixth management committee meeting. After the detailed discussion, the IMC members approved the proceedings. During the meeting various policy decisions were taken for the overall development of the Centre.

Chairman

Dr.M.M. Mustaffa

Director, NRCB, Trichy

Members

Dr.V.R. Thatham

Dean, TNAU, Coimbatore



Dr.E.R. Suresh
Principle Scientist, I.I.H.R, Bangalore

Dr.G.S. Prakash
Principle Scientist, I.I.H.R, Bangalore

Dr.P. Sundararaju
Principle Scientist, NRCB, Trichy

Members (Nom.)
Shri.V.L. Mahajan
Secretary, B.G.A.I, Maharashtra

Shri.S.S. Kadam
Nanded, Maharashtra

Member Secretary
Shri.B. Vijayakumar
AAO, NRCB, Trichy

Quinquennial Review Team

The QRT constituted by the Indian Council of Agricultural Research, New Delhi had the following members:

Chairman
Dr.P. Rethinam
Plantation Crops Management Specialist
'Bhagireth'
18, Srilakshmi Nagar, S.N.Palayam, SBI Post
Coimbatore-641 007

Members
Dr.Y. Narayana Reddy
Keerthi Residency,
Flat No.203, Balaji Nagar,
Kukut Palli, Hyderabad - 500 072

Dr.V.A. Parthasarathy
Director, Indian Instt. of Spices Research,
Marikunnu Post, Kozhikode - 673 012

Dr. Samarjit Rai
N-8/236-A-I, Nwade Sunderpur, Varanasi -
221 005

Dr.Y.S. Ahlawat
Emeritus Professor,
Advanced Centre for Plant Virology,
Divn. of Plant Pathology, IARI,
Pusa, New Delhi-I 10 012

Member Secretary
Dr.P. Sundararaju
Principal Scientist,
NRC for Banana, Trichy - 620 102

The QRT Chairman and members visited the National Research Centre for Banana, Trichy during 25-29.06.2007 and 25-28.07.2007 and on 08.08.2007 and reviewed the progress of work during the period 2002-'07. The Chairman, Dr.P. Rethinam and other eminent members Dr. Samarjit Rai, Dr.Y.S. Ahlawat, Dr. V.A. Parthasarathy and Dr.Y. Narayana Reddy interacted with scientists and submitted the final report to ICAR.



QRT Chairman and members visiting Centre's experimental farm

Some of the major recommendations of the QRT are:

- The Centre may be elevated to the level of Project Directorate of Banana Research
- The centre should coordinate all the nine banana Centres under AICRP (TF), which are now being coordinated by IIHR, Bangalore
- The QRT feels that the impact assessment of the technologies provided by NRCB may be studied. For this purpose, the Council may be requested to provide a post of Social Scientist
- It is suggested to have a building with a plinth area of 2000 sq. m. for providing space for containment facility, glass-houses, net-houses, greenhouses, insectory and an animal house may be kept in XI Plan of the Centre for funding
- A training hostel with proper boarding and lodging arrangements and a mobile mini-bus have to be provided under Transfer of Technology programmes.

IRC Meeting

Institute Research Council meeting was held on 20.11.2007. Project wise presentations were made by respective scientists. The salient achievements along with the activities to be taken up for next year were presented. The Director, Chairman of IRC, gave critical inputs for the experiments to be conducted in the field as well as in lab. He asked the scientists



to propose new projects for external funding in the identified area of research in banana. Two new institute projects were proposed, which were approved by the house. The member secretary proposed a vote of thanks at the end of meeting.



Dr.M.M. Mustafa, Director, reviewing progress of research works in IRC meeting

The significant decisions of IRC are:

- NRCB selections (Saba and FHIA-03) to be evaluated under multilocational trials under AICRP (TF).

- *In vitro* studies on various parameters of ECS and screening for salt stress to be carried out.
- Dr. S. Uma will provide parents and progenies of plants resistant to Sigatoka to Dr. R. Selvarajan for evaluation.
- A new experiment on Standardizing the spacing and nutritional requirement for NRCB released banana cv. Udhayam may be initiated.
- Udhayam variety may be evaluated for its nutritional qualities and processing purposes.
- CIRCOT collaboration may be sought for analysis of quality of fiber.
- Observation on photosynthetic parameters for two plants per hill and three plants per hill should be repeated.
- Nitrogen metabolism may be studied in the high density and fertigation project.
- Drs. M. Mayilvaganan, Sr. Scientist and S. Backiyarani, Sr. Scientist, may be included as Co-PI in the project 'Host-Virus Interactions in Banana'.

13. Trainings/ Workshops/ Seminars/ Conferences/ Winter & Summer Courses/ Meetings Attended by Scientists

Scientist	Seminar/ Symposium/ Conference/ Workshop/ Meeting/ Summer & Winter Course	Date(s)
M.M. Mustafa	CPMB-TNAU, NRCB Collaborative Research Project Discussion meeting, TNAU, Coimbatore	11.04.2007
	Indian Horticulture Congress – 2007, ICAR Research Complex for NRH Region, Barapani	18-21.04.2007
	ICAR Horticulture Division Directors' Meeting, ICAR, New Delhi	27-28.04.2007
	Banana Farmers Interaction Meeting, Farmers Training Centre, Lalgudi, Trichy	11.05.2007
	NRCB-QRT Preliminary Meeting, ICAR, New Delhi	15.05.2007
	Hill Banana Farmers' Meet – TN Hill Banana Growers Association, HRS, Thadiyankudisai	02.06.2007
	Seminar on Banana Cultivation – Theni Distt. Banana Growers Association, Chinnamanur	13.06.2007
	Horticulture Seminar, CISH, Lucknow	16-19.06.2007
	AGRI MEET, Dept. of Agril. Engineering, Govt. of Tamil Nadu, Trichy	30.06.2007
	Scientific Advisory Committee Meeting – KVK, Sandhiyur, Salem	05.07.2007



	ICAR Institute Directors' Conference, ICAR, New Delhi	16-18.07.2007
	11 th FYP EFC Review Meeting, IIHR, Bangalore	18-19.08.2007
	Banana Development – Workshop, Banana Cultivators Association, Lalgudi, Trichy	18.09.2007
	Regional Workshop on Planting Material and Rejuvenation of Horticulture Crops, Dept. of Hort., Govt. of Karnataka	21.09.2007
	INIBAP Collaborative Project discussion meeting with Dr. Molina-DDG (Hort.), ICAR, New Delhi	10.10.2007
	National Consultation Meeting on International Treaty on Plant Genetic Resources, NBPGR, New Delhi	07.11.2007
	Directors' Conclave on Motivation Techniques, NAARM, Hyderabad	14-17.11.2007
	Brain Storming Session on Long term Hort. Policy and Action Plan, CARI, Port Blair, AN Islands	02-03.11.2007
	Kissan Mela – Valedictory Function, SBI, Coimbatore	13.12.2007
	NHB Board of Directors Meet / EFC Document Discussion Meeting with DDG(H), Bangalore	17-18.12.2007
	NHM Meeting, TNAU, Coimbatore	08.01.2008
	Chief Guest – Rapinet Herbrrium, St. Joseph College, Trichy	06.01.2008
	ICAR Regional Committee Meeting, CTCRI, Trivandrum	11-12.01.2008
	Agri. Conference – CII, Karur / Saraswathi KVK Inaugural Function	22.02.2008
	Selection Committee Meeting, Saraswathi KVK, Karur	07.03.2008
	Seed and Planting Material – Discussion Meeting, CISH, Lucknow	10-12.03.2008
	Banana Cultivation – Workshop, BCKVV, Kalyani	24-25.03.2008
P. Sundararaju	Delivered a talk on Nematode management in hill bananas Field Day, Thadiyankudisai, TN	02.06.2007
	Delivered a lecture on Pest Management in Horticultural Crops, Cauvery College for Women, Tiruchirapalli	07.08.2007
	XV Biennial Group Meeting of AICRP on Nematodes, AAU, Jorhat, Assam	20-21.11.2007



	National Symposium on Nematology in 21 st Century : Emerging Paradigms at Assam Agricultural University, Jorhat, Assam	22-23.11.2007
	National Seminar /Kissan Mela, Kathihar, Bihar	9-10.03. 2008
	Divisional Project Evaluation and Monitoring meeting, IIHR, Bangalore.	18-19.08.2007
B. Padmanaban	Training on Advanced instrumentation and analytical techniques in natural products, CIMAP, Lucknow	03-12.09. 2007
	Training Course on Arthropod Endocrine Research, Centre for Arthropod Bioresources and Biotechnology, University of Kerala, Thiruvananthapuram	17-31.03.2008
	Delivered lecture on the Management of Insect Pests of Banana, Central Integrated Pest Management Centre, Tiruchirapalli	23.08.2007
	Delivered a lecture on Crop protection in banana, organized by the Department of Horticulture, Govt. of Tamil Nadu under National Horticulture Mission programme	20.09.2007
	Delivered a lecture on Integrated pest management in banana ADAC& RC, TNAU, Tiruchirapalli.	24.09.2007
	Delivered a lecture on Eco-friendly management of banana weevils on field day, Thadiyankudisai, TN	02.06.2007
	Delivered a lecture on Eco-friendly Management of Banana weevils in the NHM Farmers training organized KVK, TNAU, Sirugamani	23.11.2007
C. K. Narayana	Training programme on Capacity building for Intellectual Property Protection and Technology Licensing in Agriculture, KAU, Thrissur	18-20.02.2008
	ICAR foundation day meeting, New Delhi	16-17.07.2007
	Meeting for addressing SHG members of Myrada KVK, Kallipatti , Gobichettipalayam.	30.08.2007
S. Uma	International Symposium on 'Recent Advances in Banana Crop Protection for Sustainable Production and Improved Livelihoods,' White River, South Africa	10-15.09.2007
	PROMUSA meeting of Crop Improvement Working Group, White River, South Africa	15.09.2007
	Consultative meeting for developing a collaborative project for funding under Global Diversity Trust, NBPGR, New Delhi	5-6.10.2007



I. Ravi	Delivered a lecture on Molecular markers: an overview in the training of Molecular characterization and diagnostic technique for plant viral and fungal pathogens, NRCB, Trichy	21-28.05.2007
	National Seminar on Plant Physiology, Dr. Balasaheb Sawant Konkan Krishi Vidhyapeeth, Dapoli (Ratnagiri Dt.), Maharashtra.	29-30.11&01.12.2007
R. Selvarajan	National Seminar on Horticulture biotechnology in India: Present status and future action plan, IIHR, Bangalore	06.12.2007
	International conference of emerging and re-emerging viral disease of the tropics and sub-tropics, IARI, New Delhi	11-14.12.2007
	Delivered a lecture on Plant viruses, ADAC & RI, Tiruchirappalli	10.05.2007
	Delivered lecture on banana disease Management, KVK, Sirugamani	05&16.09.2007
	Delivered a lecture on PCR detection of BBTV St. Joseph College, Tiruchirappalli	16.11.2008
	Delivered lead lectures in National training programme on immunological and molecular diagnostics of plant virus at NBAIM, Mau, U.P.	24-26.11.2008
	Project review meeting on Network project- Diagnostics of emerging plant viruses at IARI, New Delhi	3&4.07.2007
	DBT project monitoring committee meeting on Development of transgenic Hill banana resistant to BBTV (replicase mediated) Delhi University, New Delhi	3&4.07.2007
	Meeting on Network project on diagnosis and management strategies for virus and virus like diseases in field and horticultural crops, New Delhi	06.11.2007
	Meeting on National network project on <i>Phytophthora</i> , <i>Fusarium</i> and <i>Ralstonia</i> diseases of Horticultural and field crops, New Delhi	16.01.2008
V. Kumar	Delivered a talk on Bunchy top viral disease management at Field day, Thadiyankudisai, T.N.	02.06.2007
	Delivered a talk on banana disease management, Lalgudi, Trichy	11.05.2007
	Delivered a lecture on Banana Cultivation Technologies in the training programme for the SHG's Thanjavur, T.N.	19.04.2007
	Banana Seminar Paramathy Velur, Namakkal, T.N.	23.05.2007



	Farmer's Training Programme on Advanced Production Technologies for Precision Farming Erode and Chennampatty, T.N.	13.08.2007
	Delivered a lecture on Advanced Banana Production Technologies, Salem, T.N.	20.09.2007
	35 th Scientific Advisory Committee meeting of KVK, Sirugamani, Tiruchirapalli	15.10.2007
	Interaction Meeting on Tools and Machinery for Development of Horticulture CISH, Lucknow	18.01.2008
	International Horti Expo- 2008, Pragati Maidan, New Delhi	31.01.-02.02.2008
	National Vegetable Growers Mela, IIVR, Varanasi, U.P.	09-10.02.2008
	Delivered lectures in the Training Workshop under the Water Management in Command Area Programme, Tiruchirapalli and Karur Districts	13.02.2008 19.02.2008 26.02.2008 14.03.2008
M. Mayilvaganan	National level group discussion on the diseases of complex etiology of coconut and arecanut, CPCRI - RS, Kayangulam, Kerala	28.01.2008
K. J. Jeyabaskaran KVK, Sirugamani	Delivered a lecture on Macro and Micro nutrient management in banana,	13-15.11.2007
	Delivered a lecture on Banana Nutrition KVK, Sirugamani	23.11.2007
	Delivered a lecture on Macro and Micro nutrient deficiencies and remedies in banana, TN Agrl. Engineering Dept., Trichy	03.02.2008
	Delivered a lecture on Soil and nutrient management in banana, KVK, Sirugamani	18.2.2008
S. Backiyarani	National Seminar on Horticultural Biotechnology in India: Present status and future action plan held at IIHR, Bangalore	08.12.2007
	Delivered a lecture on <i>In-vitro</i> germplasm conservation technique, AC & RI, Madurai	26.03.2008
	Delivered a lecture on Identification of transgenic plants against disease resistance in short course advanced molecular diagnostic techniques for viruses and other sucker borne pathogens of banana, NRCB, Trichy	19-28.09.2007
M. S. Saraswathi	National Seminar on Horticultural Biotechnology in India: Present status and future action plan, IIHR, Bangalore	08.12.2007
	The Director and all the scientists of the Centre participated and presented lead and/or contributory papers in National Conference on Banana - Production and utilisation of banana for economic livelihood and nutritional security, Tiruchirapalli, Tamil Nadu	25-28.10.2007



14 Seminars/ Meetings/ Workshops/ Conferences/ Summer Institutes and Farmers Training Organized at the Centre

Conducted a short term training on “Processing and Production of Value Added Products of Banana” from 13 to 20, Dec. 2007.

A seven days training programme on “Molecular Characterization and Diagnostic Techniques for Plant Viral and Fungal Pathogens” was conducted during 21-28, May 2007.

ICAR sponsored short course on “Advanced molecular diagnostic techniques for viruses and other sucker borne pathogens of banana” was conducted from 19 to 28, Sept. 2007.



Participants and faculties of ICAR sponsored short course on 'Advanced molecular diagnostic techniques for viruses and other sucker borne pathogens of banana' has been conducted from 19 to 28, Sept. 2007

National Conference on Banana

The Association for the Improvement in Production and Utilization of Banana (AIPUB) and National Research Centre for Banana (NRCB), Tiruchirapalli, in collaboration with Confederation of Indian Horticulture and Banana Growers' Association of India organized a National Conference on Banana with a theme on 'Production and Utilization of Banana for Economic Livelihood and Nutritional Security' during 25-28 October, 2007 at Tiruchirapalli, Tamil Nadu. The Conference was attended by over 500 delegates representing research scientists, entrepreneurs, extension workers, farmers, government officials and industrial representatives from all over the country and had participation from 15 banana growing states. Thiru. Veerapandi S. Arumugam, Hon'ble Minister for Agriculture, Tamil Nadu was the Chief Guest in the inaugural session and Dr.H.P. Singh,

Deputy Director General (Horticulture) delivered key note address. Dr. Singh attributed the tremendous growth of production and productivity of banana to adoption of improved technologies and emphasizing the need for genetic enhancement, precision farming and plant health management while cautioning about the likely threat of *Fusarium* race - 4.

The Chief Guest gave away the AIPUB award 'Kadali Purashkar' to Dr.M.M. Mustafa, Director and Dr. P. Sundararaju, Principal Scientist, NRCB and Dr. (Mrs.) Rema Menon, Professor, KAU in recognition of their contribution to research and development of banana. On the occasion, two technical booklets on 'Banana Recipes' and 'Therapeutic and Medicinal Values of Banana' were released. A banana exhibition was also arranged participated by different input organizations, entrepreneurs and ICAR institutes, tissue culture companies and developmental and promotional agencies. Nine technical sessions were held during the three days conference : 1. National and International scenario of banana research, 2. Gene safeguard and management, 3. Biotechnological approaches in banana production and improvement, 4. Breeding strategies, 5. Quality production of planting materials, 6. Production strategy in banana and plantain, 7. Post-harvest management, marketing and trade and 9. Recent development in pest and disease management and in all sessions together over 220 research papers were presented and discussed.

The plenary session of the Conference was chaired by Dr.H.P. Singh, DDG (Hort.) and Shri Jairam Ramesh, Hon'ble Minister of State for Commerce, Govt. of India was the Chief Guest of the function, who emphasized the need for increasing the production and productivity of banana and also reduction of post-harvest losses. A field visit was arranged to the farmers and delegates to visit the experimental farm of National Research Centre for Banana on 28th October 2007.



Dr.H.P. Singh, DDG (Hort.), ICAR, New Delhi delivering keynote address in the National Conference on Banana



15 Distinguished Visitors

List of VIP - Visitors

Sl. No.	Date	Name & Address
1.	22.07.2007	QRT Members
2.	28.07.2007	IMC Members
3.	21.08.2007	Dr. Ponnaivaiko, V. C., Bharathidasan University, Trichy
4.	06.10.2007	Dr. H. P. Singh, DDG (Horticulture)
5.	27.10.2007	Shri. Jairam Ramesh, Minister of State for Commerce, GOI
6.	31.10.2007	A. V. N. S. Sastry, Ministry of HRD, New Delhi
7.	31.10.2007	A. K. Singh, Desk Officer, HRD, New Delhi
8.	31.10.2007	K. Natarajan, Ministry of Commerce, New Delhi
9.	27.11.2007	RAC Members
10.	25.02.2008	Mr. Venkatesan, Chief Manager, EXIM Bank, Mumbai

List of other Dignitary - Visitors

Sl. No.	Date	Name & Address
1	03.04.2007	S. P. Ramanathan, KVK, Sirugamani
2.	19.04.2007	C. Elangovan, Asst. Director of Hort., Lalgudi
3.	29.08.2007	C. T. Mariyappan, Madipakkam
4.	13.12.2007	Mr. Narasimman, Perunthalaivar Kamarajar KVK, Puduchery
5.	11.02.2008	D. Vinoth Kumar, Trichy
6.	15.02.2008	S. John Britto, St. Joseph's College, Trichy
7.	16.02.2008	Dr. K. Angappan, TNAU, Coimbatore
8.	19.02.2008	M. Veeraswamy, Dept. of Horticulture, Lalgudi
9.	26.02.2008	B. J. Benny, Horticulture Officer, Andhra Pradesh
10.	12.03.2008	Mahesh Singh Chandran, Busharpur, M.P.



Shri. Jairam Ramesh Hon. Minister of State for Commerce, GOI and Dr.H.P. Singh, DDG (Hort.), ICAR, New Delhi visiting NRCB on 27.10.2008



List of Farmer - Visitors to the Centre

Sl. No.	Date	Name & Address	Purpose of visit	No. of Farmers
1.	18.04.2007	Farmers of Kanchipuram dt.	About NRCB	100
2.	29.06.2008	K. V. Aravind, Gobichettipalayam	Banana fibre extraction	2
3.	03.07.2007	S. Karthikeyan, Srirangam, Trichy	About NRCB	1
4.	14.08.2007	Farmers of Cuddalore dt.	Banana Cultivation	7
5.	27.08.2007	Farmers with P. Pachiappan, SMS-Horticulture, MYRADA KVK, Gobichettipalayam	Training visit	17
6.	06.09.2007	Farmers with Dr. S. Easwaran, KVK, Sirugamani	Field visit	70
7.	03.10.2007	Farmers with M. Vijayakumar, Hort. Officer, Dindigul	Banana Cultivation	53
8.	08.10.2007	N. Mahesh, Trichy	Banana products	2
9.	09.10.2007	P. Ramadoss, Salem	About NRCB	1
10.	06.11.2007	C. Muthuvel, Erode	Banana fibre extraction	2.3
11.	11.11.2007	Farmers with Dr.S. Easwaran, KVK, Sirugamani	Field visit	32
12.	14.11.2007	Farmers with Dr. S. Easwaran, KVK, Sirugamani	Training visit	35
13.	16.11.2007	Farmers with Dr .S. Easwaran, KVK, Sirugamani	Field Visit	32
14.	23.11.2007	P. Subramaniam, Agri-Clinic, Madurai	Trainees visit	24
15.	03.12.2007	Mr. Vinod Patil, Burhanpur, M.P.	Banana Shakthi Conference	2
16.	07.12.2007	Mr. Sathiya Moorthi	Banana Experience	3
17.	04.01.2008	G. Mohanraj, Nagapattinam	About NRCB	1
18.	22.01.2008	Trainees from Agriculture Eng. Training Centre, Trichy	Field visit	24
19.	04.02.2008	Farmers with R. Anitha, Saraswathi KVK, Pulutheri	Exposure visit	25
20.	04.02.2008	Farmers with S. Mathivanan, Snehithi Trust, V. Pudur	Exposure visit	25
21.	06.02.2008	Farmers with S. J. Vijayalalitha, KVK, Sirugamani.	Exposure visit	73
22.	11.02.2008	Farmers with V. Krishnamoorthy, Agri. College, Trichy	Exposure visit	59
23.	12.02.2008	Farmers with Mr. Sainathan, Salem	About NRCB	48
24.	14.02.2008	Farmers with S. J. Vijayalalitha, KVK, Agri. College, Trichy	Exposure visit	43
25.	04.03.2008	Farmers with S. Nagarajan, Agri. Engineer, Trichy	Visit and Training	19
26.	14.03.2008	Farmers with V. Devan, Jr. Asst., Trichy.	Exposure visit	23
27.	15.03.2008	Farmers with Mr. Balakrishnan, Horticulture Officer, Nagarcoil	Exposure visit	51
28.	25.03.2008	A. Murugesan	About NRCB	2
29.	29.03.2008	Farmers with Dr. Q. Rajukannan	Field visit	24



List of Student - Visitors to the Centre

Sl. No	Date	Name & Address	Purpose of visit	No. Students
1.	20.07.2007	Students with Dr. V. Kumaresan, Thanthai Hans Roever College, Perambalur	Biotechnology related work	59
2.	02.08.2007	Students of A. R. K. Vidyajothi Vikhas Matriculation School, Vengur, Trichy	Project	9
3.	23.08.2007	Students with Dr. S. Senthil Kumar, St. Joshep's College, Trichy	Exposure visit	21
4.	01.09.2007	Students with Dr. K. Manonmani Lady Doak College, Madurai	Lab. visit	63
5.	09.10.2007	Mrs. Kalaivani Murugan, Tenkasi	Project	1
6.	05.11.2007	R. Kolandaisamy, Thanjavur	Virus consulting	3
7.	24.11.2007	Trainees with E. Paramsivam, National Plant Protection Training Institute, Hyderabad - 30	Study tour	10
8.	22.01.2008	Students of Carmel's Matriculation School, Trichy	Educational tour	47
9.	16.02.2008	Students with Dr. K. Angappan, TNAU, Coimbatore	Exposure visit	32
10.	18.03.2008	Students with Dr. Manisekaran, Professor, H. C. & R. I., Periyakulam	Educational visit	56

16 Empowerment of Women

More than 70 women entrepreneurs were trained on small scale mass multiplication of banana through tissue culture during the year. Scientists of the Centre explained about the importance of use of disease-free clean planting material, source of loan availability, insurance and the production feasibilities.

Special lectures were delivered by Drs. S. Uma, M. S. Saraswathi and S. Backiyarani in Cavery College of Women, Trichy, Mother Theresa Women's University, Kodaikanal and Bishop Heber College Ladies club respectively, where girl students were encouraged to take up biotechnology course and research. They also enlightened on multiple avenues available for career development.

Above 250 girl students visited the Centre from the different Academic Arts and Science Colleges, Home Science Colleges and Horticultural Colleges of TNAU. The NRCB Scientists explained the technologies/ research activities to the visiting students.

17 Personnel

Appointment

Dr. S. Backiyarani, Senior Scientist, was appointed as Senior Scientist (Plant Biotechnology) w. e. f. 29.08.2007.

Transfer

Dr. M. Mayilvaganan, Senior Scientist (Plant Biochemistry), joined on 05.09.2007 on transfer from Central Plantation Crops Research Institute-Regional Station, Kayangulam.

Probation clearance

Mrs. C. Gomathi, Assistant Finance & Accounts Officer, w.e.f. 08.07.2007.

Resumed duty

Mr. R. Natarajan, Scientist (Economic Botany) has resumed duty after study leave on 21.08.07.

Dr. R. Thangavelu, Senior Scientist, has resumed duty after his postdoc training from Australia on 28.01.08.

Retirement

Mr. Raghuraman, T-5 Junior Garden Superintendent, retired from Council's service on VRS on 31.08.2007.

Demise

Mr.M. Shanmugavel, T4 Refrigeration mechanic, passed away on 27.08.2007.



Scientific Staffs

Name	Designation
Dr.M.M. Mustaffa	Director
Dr.P. Sundararaju	Principal Scientist (Nema.)
Dr.B. Padmanaban	Senior Scientist (Ent.)
Dr.C. K.Narayana	Senior Scientist (Hort.)
Dr.S. Uma	Senior Scientist (Hort.)
Dr.I. Ravi	Senior Scientist (Pl.Phy.)
Dr.R. Thangavelu	Senior Scientist (Pl.Path.)
Dr.R. Selvarajan	Senior Scientist (Pl.Path.)
Dr.V. Kumar	Senior Scientist (Hort.)
Dr.K.J. Jeyabaskaran	Senior Scientist (Soil Sci.)
Dr.M.S. Saraswathi	Scientist (Sr.Scale) (Hort.)
Mr.R. Natarajan	Scientist (Eco.Bot.)

Technical Staffs

Name	Designation
Mr. Raghuraman	T-5 Technical officer
Mr.S. Palanichamy	T-5 Technical officer
Mr.P. Durai	T-4 Lab Technician
Mr.P. Ravichamy	T-4 Technical Asst. (Journalism)
Mrs.T. Anitha Shree	T-4 Lab Technician
Mr.D. Ramachandramoorthy	T-3 Tech. Asst (Civil Overseer)
Mrs.C.S. Jacqueline	T-3 Tech.Asst. (Computer Prog.)
Mr.R. Pitchaimuthu	T-2 Field Technician
Mr.N. Marimuthu	T-2 Field Technician
Mr.V. Selvaraj	T-2 Lab Technician
Mr.T. Sekar	T-2 Lab Technician
Mr.K. Kamaraju	T-2 Lab Technician
Mr.A. Subramanian	T-2 Driver
Mr.P. Mohan	T-2 Tractor Driver
Mr.V. Manoharan	T-2 Driver

Administrative, Audits & Accounts and Supporting Staffs

Name	Designation
Administration	
Mr.B. Vijaykumar	AAO
Mr.M. Krishnamoorthy	PA to Director
Mr.R. Krishnamurthy	Upper Division Clerk
Mrs.S. Durgavathy	Lower Division Clerk
Mr.R. Sridhar	Stenographer Gr.III
Mr.M. Devarajan	Lower Division Clerk
Audit and Accounts	
Smt.C. Gomathi	AFAO
Mr.M. Balu	Assistant
Mr.R.N.M.S. Kannan	Stenographer Gr.III
Supporting	
Mr.R. Mohanraj	Mali SSG-III
Mr.V. Pandiyan	Mali SSG-III
Mr.V. Thangaraju	Messenger SSG-II
Mr.P. Kamaraj	Mali SSG-II
Mr.V. Ganesan	Mali SSG-I
Mr.C. Kumaran	Mazdoor SSG-I
Mrs.K. Mariammal	Safaiwali SSG-I



18 Other Informations

New Projects

Induced Mutation - A crop improvement strategy for developing dwarf and Sigatoka leaf spot resistant banana cv. Grand Nain sanctioned by BRNS, DAE (GOI), Mumbai for 3 years with Rs.15 lakhs.

Accredited Test Laboratory (ATL) under the National Certification System for Tissue Culture raised Plants (NCS-TCP) sanctioned by DBT, New Delhi for a period of three years.

NRCB Foundation Day

The 14th Foundation day of the NRCB was celebrated on 21.08.2007. Dr. M. Ponnavaiko, Vice-Chancellor, Bharathidasan University, Tiruchirapalli was the Chief Guest and Dr. M.M. Mustaffa, Director, NRCB presided over the function. During the foundation day celebration, Dr. Ponnavaiko observed that technology that did not reach the common man serves no purpose and emphasized on need - based research, which would

not only enhance productivity and living standards of the farmers, but also expand the ambit of funding.

Club Day

The Annual Day of the NRCB Recreation club was also celebrated on the day of NRCB Foundation day. On the occasion, the Chief Guest, Dr.M. Ponnavaiko distributed the prizes to the winners of various sports and cultural activities. The staff members along with their family participated in the function.



Dr. Ponnavaiko, H'ble. Vice-Chancellor, Bharathidasan University, Trichirapalli visiting NRCB - Exhibition



ANNEXURE – I

List of On-going Institute Projects

I. Crop Improvement

1. 2000711002-Crop improvement of banana through conventional breeding
Project Leader : **M. M. Mustaffa**
Project Associates : S. Uma, S. Backiyarani and R. Natarajan
2. 2000711003-Crop improvement of banana through non-conventional breeding
Project Leader : **S. Uma**
Project Associate : M. S. Saraswathi
3. 2000711004-Improvement and management of banana genetic resources in Indian subcontinent
Project Leader : **S.Uma**
Project Associates : M. S. Saraswathi, R.Thangavelu, P. Sundararaju, B. Padmanaban and C. K. Narayana
4. 2000711005-Identification and characterization of nematode resistance gene(s) in banana
Project Leader : **S. Backiyarani**
Project Associates : S. Uma, M. S. Saraswathi, P. Sundararaju and M. Mayilvaganan
5. 2000711006-Improvement of Rasthali through induced mutagenesis
Project Leader : **M. S. Saraswathi**
Project Associates : S. Uma, S. Backiyarani and R.Thangavelu

II. Crop Production and Post-Harvest Technology

6. 2000713001-Standardisation of agro-techniques for banana production and productivity
Project Leader : **V. Kumar**
Project Associates : M. M. Mustaffa and K. J. Jeyabaskaran
7. 2000713004-Studies on micronutrients in banana
Project Leader : **K. J. Jeyabaskaran**
Project Associate : V. Kumar
8. 2000713006-Fertilizer tailoring for targeted banana yield and sustainable soil health
Project Leader : **K. J. Jeyabaskaran**
Project Associate : V. Kumar
9. 2000717001-Studies on handling, storage and processing of banana
Project Leader : **C. K. Narayana**
Project Associates : M. M. Mustaffa and R. Selvarajan
10. 2000717002-Standardization of storage conditions for banana
Project Leader : **C. K. Narayana**
Project Associates : I. Ravi
11. 2000716001-Studies on physiology of flowering and fruit development in banana
Project Leader : **I. Ravi**
Project Associates : C. K. Narayana and K. J. Jeyabaskaran



12. 2000716002-Drought stress tolerance in banana: Understanding the physiological, biochemical and molecular mechanism of drought tolerance

Project Leader : **I. Ravi**

Project Associates : M. M. Mustaffa, C. K. Narayana, M. Mayilvaganan and S. Uma

13. 2000716003-Salt stress tolerance in banana: Understanding the physiological, biochemical and molecular mechanism of salt tolerance

Project Leader : **I. Ravi**

Project Associates : M. M. Mustaffa, C. K. Narayana, M. Mayilvaganan and K. J. Jeyabaskaran

14. 2000716004-Physiological and biochemical mechanism of nematodes and pseudostem weevil resistance and identification of 'biomarker metabolites' in banana

Project Leader : **M. Mayilvaganan**

Project Associates : M. M. Mustaffa, I. Ravi, P. Sundararaju and B. Padmanaban

III. Crop Protection

15. 2000715002-Studies on banana nematodes and their management

Project Leader : **P. Sundararaju**

Project Associates : B. Padmanaban and R. Thangavelu

16. 2000715006-Management of banana weevils

Project Leader : **B. Padmanaban**

Project Associates : P. Sundararaju and R. Thangavelu

17. 2000715003-Investigation on fungal and bacterial diseases of banana and their management

Project Leader : **R. Thangavelu** (on deputation)

Project Leader (i/c) : R. Selvarajan

18. 2000715005-Studies on viral diseases of banana and their management

Project Leader : **R. Selvarajan**

19. 2000715007-Host-virus interactions in Banana: Molecular mechanisms of resistance and susceptibility, latency, integration and episomal expression of EPRV's

Project Leader : **R. Selvarajan**

Project Associates : I. Ravi, M. Mayilvaganan, S. Backiyarani and S. Uma

List of On-going Externally-Funded Projects

1. Developing dwarf and Sigatoka resistant Grand Nain bananas through induced mutation
2. Development of virus resistant transgenic crops-Development of transgenic hill banana resistant to banana bunchy top virus (replicase gene mediated)
3. Accredited Test Laboratory for virus testing under National Certification System for Tissue Culture raised Plants (NCS-TCP)



ANNEXURE - II

Meteorological Data

Month	Max. Temp. (°C)	Min. Temp. (°C)	Relative Humidity (%)	Rain Fall (mm)
April 2007	26.8	38.0	87.1	5.1
May 2007	27.6	39.0	78.2	2.2
June 2007	26.6	36.9	74.8	0.8
July 2007	26.4	36.9	72.9	17.5
August 2007	28.2	34.9	80.7	24.9
September 2007	25.9	36.6	79.1	-
October 2007	24.9	33.5	83.4	28.3
November 2007	23.1	31.4	88.9	8.0
December 2007	21.4	29.4	91.7	205.0
January 2008	20.7	30.6	90.7	-
February 2008	22.1	30.8	89.8	11.4
March 2008	23.0	32.3	91.6	64.2

