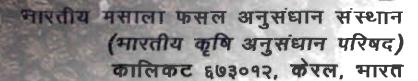
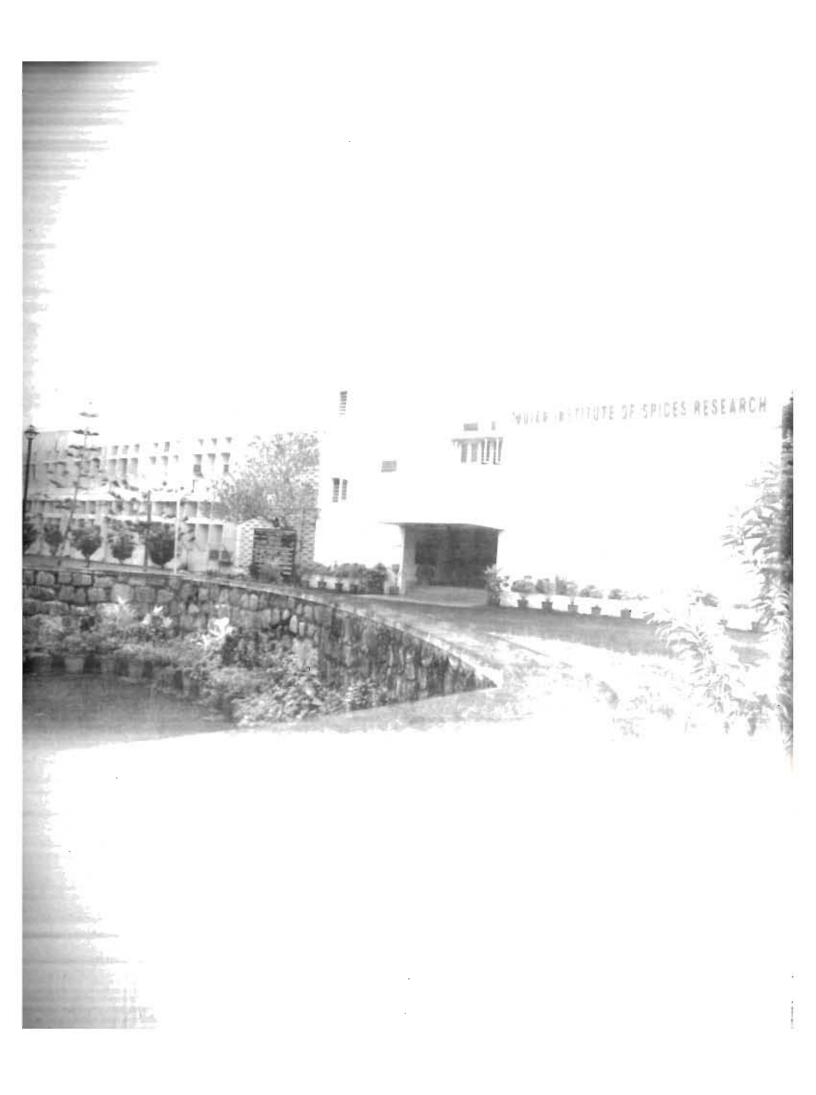
# वार्षिक प्रतिवेदन २००७-०८ Annual Report 2007-08

IISRAR-20



Indian Institute of Spices Research (Indian Council of Agricultural Research) Calicut - 673 012, Kerala, India.



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# IISR Annual Report

## 2007 - 08

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भारतीय मसाला फसल उनुसंधान संस्थान कालिकट भारतीय कृषि अनुसंधान परिषद नई दिल्ली

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Indian Institute of Spices Research Calicut



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#### Front cover

GEB 17 - Endophytic bacteria from giner effective against Pythium

#### **Back cover**

2

GEB 18 - Endophytic bacteria from ginger effective against R. solanacearum

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## Contents

Foreword (Hindi)	
Foreword	
Executive Summary (Hindi)	
Executive Summary	
Introduction	
Past Achievements	
Black pepper	
Cardamom	
Ginger	
Turmeric	
Vanilla	
Paprika	
Tree spices	
Spices - micotoxins, nutraceuticals and economics	
All India Coordinated Research Project on Spices	
Krishi Vigyan Kendra	
Agricultural Technology Information Centre	
Education and Training	
Bioinformatics Centre	
Agricultural Research Information System	
Library	
Consultancy Processing Cell	
Women Empowerment Programme	
Official Language Implementation Activities	
Institute Management Committee	
Research Advisory Committee	
Recommendations of RAC 2008	100 - 105
Quinquennial Review Team	
QRT Recommendations	
Research Publications 2007-08	
List of Projects	
Personnel	121 - 122
Weather Data 2007	

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## प्रस्तावना

आईआईएसआर के कार्यकलापों में वर्ष 2007- 08 एक महत्वपूर्ण वर्ष होता है! कई उपलब्धियों के अलावा एक उल्लेखनीय कार्य यह ई कि मसालों के लिए 'डीयुएस' (विशिष्टता, एकरूपता और स्थिरता) मार्गदर्शन विकसित करने के लिए अपने प्रस्ताव को पौध प्रजातियों के संरक्षण एवं किसानों के अधिकार प्राधिकरण द्वारा स्वीकृति प्राप्त की है और इसने डॉ. वी. ए. पार्थसारथी, निदेशक को सदस्य सचिव बनाकर टास्क फोर्स VII गठित की! हल्दी, अदरक, काली मिर्च और इलायची के लिए डीयुएस मार्गदर्शन प्रारंभ किया गया! जैव प्रौद्योगिकी प्रभाग, भारत सरकार द्वारा संख्यान को मसालों के टिशु कल्वर सुविधा के प्रत्यायन के लिए परीक्षण केन्द्र के रूप में मान्यता प्राप्त की गयी है! *फाइटोफथोरा* खुर गलन एवं मृदु पतन रोग के प्रति बयोकनसोर्टियम का मूल्यांकन करने पर रोगों को कम करने में तथा पौधों को बनाये रखने में रिज़ोबैक्टीरियल संघ की प्रभावोत्पादकता सूचित किया! यही नहीं, काली मिर्च में *फाइटोफथोरा* खुर गलन एवं मृदु पतन तथा अदरक में राइज़ोम राट रोग के प्रति जीवाणुक एन्डोफाइट्स का मूल्यांकन करने से रोग प्रबन्धन के लिए कुछ आशाजनक स्ट्रेन्स भी दिखाया! इलायची मे, एक संकरज (जीजी X एनकेई 19) घने कैप्स्यूलयुक्त एवं राइज़ोम गलन और पर्ण चित्ती के लिए खेत सहिष्णु होकर आशाजनक दिखाया! रोगमुक्त हल्दी का नमूना सूखने के लिए कम समय लेता है! लेकिन इन्हीं नमूनों में तेल एवं ओलियोरसिन प्राप्ति में बहुत कमी होती है! लौंग, दालचीनी और हल्दी का पर्ण तेल अफ्लाटोक्सिन (aflatoxin) के उत्पादन को मना किया! संरचनात्मक दृष्टि से देखें तो, उप महनिदेशक (हॉटी) दवारा पुस्तकालय, जैवनियन्त्रण अनुसंधान एवं फार्म के लिए एक एक नये मकान का उदघाटन कार्य संपन्न हआ!

तकनोलजियों को परिष्कृत करने के लिए, संस्थान में विकसित नये प्रजातियों के साथ काली मिर्च की अठारह महत्वपूर्ण प्रदर्शनियाँ काली मिर्च के खुर गलन के नियन्त्रण के लिए चालू किये पद्धतियों के साथ विकसित कीं! तीन अक्सशनों को अपनी नवीनता के लिए एनबीपीजीआर के साथ पंजीकृत किया! उपयुक्त जगह एवं उपयोगिता क्षेत्र पर पेटेंन्ट संबन्धी शोध कार्य संस्थान के सक्षम तकनोलजी प्रबन्धन समिति (आई टी एम सी) के साथ कार्यान्वित किया जा रहा है! प्रमुख हॉटीकल्वरिस्ट, डॉ एन. मोहनकुमारन सेवानिवृत्त निदेशक अनुसंधान, केरल कृषि विश्वविद्यालय की अध्यक्षता में नये शोध सलाहकार समिति गठित की और उसकी पहली बैठक हमारे शोध कार्यक्रम उत्कृष्ट बनाने के लिए संपन्न हुई! कृषि विज्ञान केन्द्र ने अपना प्रशिक्षण कार्यक्रम किसानों की आवश्यकताओं के अनुसार बढाने के अतिरिक्त स्टेट हॉर्टीकल्चर मिशन के अन्तर्गत मालियों के कौशल को विकसित करने के लिए कौशल विकसित करने का कार्यक्रम शुरू किया!

मैं इस अवसर को अपने माननीय महानिदेशक डॉ मंगला राय और हमारे आदरणीय उप महानिदेशक (हॉटी) को अपने संस्थान के प्रति विशेष रूचि रखने से उनके प्रति अपनी कृतज्ञता अर्पित करने के लिए उपयुक्त करता हूँ! हमारे पूर्व महा निदेशक (हॉटी -II) डॉ. के. वी. रमणा एवं डॉ उमेश श्रीवास्तवा, उप महानिदेशक (हॉटी -II) को उनकी मदद के लिए भी अत्यन्त कृतज्ञता प्रकट करता हूँ! मैं, आई आई एस आर, कालिकट के सभी स्टाफों को हमारा कार्यक्रम किसानों की भलाई के लिए आगे बढाने में उनके सम्मिलित समर्थन के लिए अपना आभार प्रस्तुत करता हूँ!

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(वी ए पार्थसारथी) निदेशक

कालिकट 28-06-2008

Indian Institute of Spices Research

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#### PREFACE

The year 2007-08 has been a significant one in the activities of IISR. Besides many achievements the notable one has been the acceptance of our proposal for developing the DUS (Distinctness, Uniformity and Stability) guidelines for Spices by the Protection of Plant Varieties and Farmers' Rights Authority and it constituted the Task Force VII with Dr. V.A. Parthasarathy, Director, as Member Secretary. The DUS guidelines for turmeric, ginger, black pepper and cardamom have been launched. The institute has been recognized as the test centre for accreditation of tissue culture facility for spices by Department of Biotechnology, GOI. Evaluation of bioconsortium against Phytophthora foot rot and slow decline diseases indicated the efficacy of rhizobacterial consortium in reducing the disease as well as survival of the plants. Also, evaluation of bacterial endophytes against Phytophthora foot rot and slow decline diseases in pepper and rhizome rot in ginger resulted in some promising strains for disease management. In cardamom, one hybrid (GG x NKE 19) is promising with bold capsules and field tolerance to rhizome rot and leaf blight. Cured turmeric samples took less time for drying but there was a considerable reduction in oil and oleoresin recovery in these samples. Clove, cinnamon and turmeric leaf oils inhibited the production of aflatoxins. In terms of infrastructure, a new block each for library, biocontrol research and farm have been inaugurated by the Deputy Director General (Hort.).

In order to refine the technologies, 18 front line demonstrations on black pepper have been laid out with new varieties developed at the institute along with the practices for control of foot rot of black pepper. Three accessions have been registered with NBPGR for their novelty. Patentable research on niche and utility area are underway with a strong Institute Technology Management Committee (ITMC). New Research Advisory Committee under the chairmanship of eminent horticulturist Dr. N. Mohanakumaran, Retd. Director of Research, KAU, was constituted and held its first meeting to fine tune our research programmes. The KVK has intensified its training programme to the farmers' need besides operating the programme for developing skills to develop skilled gardeners under the State Horticultural Mission.

I take this opportunity to thank our honorable Director General Dr. Mangala Rai and our respected DDG (Hort.) for their interest in our institute. The help rendered by Dr. K.V. Ramana formerly ADG (Hort.II) and Dr. Umesh Srivastava, ADG (Hort.II) is gratefully acknowledged. I am grateful to all the staff of IISR, Calicut for the united support in running our programmes for the welfare of farmers.

Calicut 28-06-2008

V.A. Parthasarathy Director

## सारांश

#### काली मिर्च जननद्रव्य संग्रहण, संरक्षण और मूल्यांकन

प्रस्तुत वर्ष 54 अक्सशनें शेनडरनी डब्लियूएलएस से, 24 बिसले घाट जंगल से और 32 कारवार जिला के *पाइपर* नाइप्रम पी. गलियटम, पी. द्राइकोस्टाच्योन, पी. अटेन्युवाटम और पी. हापनियम स्पीसीस से संचित किया, जिसमें किसानों के खेत से संचित दो साधारण कल्टिवर्स भी शामिल होता है! एक हज़ार अक्सशनों के अलावा अठानब्बे अक्सशनों को आईपीजीआरआई डिस्क्रिप्टर के आधार पर अभिलक्षणीकरण किया गया!

#### प्रजनन

पोल्लू बीटल, नेमटोड और सूखापन की प्रतिरोधकता के लिए : बुश पेप्पर प्रजाति आई आई एस आर शुभकरा को मादा पौधा के रूप में लेकर और 'पोल्लू' सहयतायुक्त लाइनों (संग्रह सं 816, 841,1084 और 1114) नेमटोड सहयता युक्त लाइनों (अक्सशन 820 और एच पी 39) और सूखापन सहयतायुक्त लाइनों (अक्सशन सं 1495, 931 और 813) को नर पौधा के रूप में लेकर एक संकरण ब्लॉक स्थापित किया! इन्हीं संकर संयोजनों से विकसित बीजपौधे खेत में रोपण करने के लिए बहुलीकरण की अवस्था पर है!

अधिक उपजता एवं अधिक कारियोफिलिन के लिए : कुल मिलाकर संकरज के 114 संततियों को उत्पादित किया, जिसमें आई आई एस आर शुभकरा (मादा) और अधिक कारियोफिलिन लाइनें (नर) शामिल होती है। श्रेष्ठ लाइनों (नर लाइन) के फलों में कारियोफिलिन संघटक की माताओं का विश्लेषण करने पर विभिन्नता सूचित किया! अम्बलवयल, पेरूवन्नामुषि, पन्नियूर और येरकाड केन्द्रों से संग्रहित नमूनों में आई आई एस आर शुभकरा के बीटा कारियोफिलिन संघटक पर स्थान प्रभाव का निरीक्षण किया!

**फाइटोफयोरा प्रतिरोधकता के लिए :** फाइटोफथोरा प्रतिरोधकता के वर्गीकरण के लिए दूसरा मानचिव्रण संख्या विकसित करने को आई आई एस आर शक्ति X आई आई एस आर शुभकरा के बीच 50 नये संकरज बनाया और संततियों को अंकुरण के लिए रोपित किया! पच्चीस नये पुटेटीव द्वानसजनिक (ओस्मोटिन) को परीक्षण के लिए कटोरीकृत बनाया!

#### पोषण

काली मिर्च की उपजता और गुणवत्ता पर सल्फेट ऑफ पोटाश : पर्णों की पोषण स्थिति, जैवरासायनिक पैरामीटर्स और उपजता संस्तृत पोटेशियम के 50% एसओपी के रूप में डालने पर पर्णों पर छिडकने और मग्नीशियम अनुपूरण करने पर या न करने पर दोनों में समान होती है! एसओपी के रूप में संस्तृत मात्रा में पोटेशियम के साथ 2% पर्ण छिडकाव भी करने पर अधिकतम उपजता अंकित की! जो एसओपी के रूप में संस्तृत पोटेशियम के 50% लगाने पर और एसओपी के रूप में संस्तुत पोटेशियम के साथ मग्नीशियम सल्फेट उपचार करने पर भी उपजता कम होती है! एसओपी उपचार में जब अधिमुल्य का विचार करें. तो संस्तुत पोटेशियम या उसका 50% एसओपी के रूप में पर्ण छिडकाव के साथ या उसके बिना उत्तम उपज प्राप्ति अंकित की और बी : सी का अनुपात (1.73 -2.10) एमओपी उपचार के समान होता है! गुणवत्ता पैरामीटर्स जैसे ओलिओरसिन और पाइपरिन माव्राएं इन्हीं उपचारों में अधिक होता है!

फोस्फरस विमोचन पर ओरगानिक खाद : फोस्फरस खंडो के पदवार आगे के प्रतिकमण विश्लेषण करने पर 0.611 के R<sup>2</sup> के साथ उपजता (= 46.25+1.034 Fe-P+0.570 Av P) के लिए उत्तम खंड के रूप में Fe-P और उपलब्ध फोस्फरस देख लिया!

#### सूखा सहिष्णु

एसओडी आईसोज़ाइम ने पांच उन्नत बैंड दिखाया, जो सहयता एवं सुग्राहय अक्सशन, दोनों के लिए सार्वजनिक होता है! कुछ अक्सशनों में अतिरिक्त एसओडी बैंड के रूप में जल बल प्रवृत्त किया। पांच दिन के बल निवेशन के बाद सहयतायुक्त अक्सशनों ने सुग्राहय की अपेक्षा अधिक प्रकाश संश्लेषण दर और अधिक रंधी चालकत्व दिखाया!

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#### उत्पादकता का दैहिक और जैवरासायनिक आधार

फलन पूर्व काल में कम उपज देनेवालों की अपेक्षा अधिक उपज देनेवालों में एनआर और कम एमडीएच कार्य देख लिया! सामान्यतया अधिक उपज देनेवाले इस काल में कम उपज देनेवालों की अपेक्षा अधिक करबोहाइड्रेट्स और स्टारच प्रदान करते देख लिया!

#### गुणवत्ता

पी. नाइग्रम में मिरिस्टिसिन : दापोली और कारवार से संचित पी. नाइग्रम के पर्ण नमूनों के पर्ण तेल संयोजन से मिरिस्टिसिन को अच्छे गाढापन में देख लिया, जो जायफल का एक सामान्य संघटक है ओर पी नाइग्रम में उसे अभी तक अंकित नहीं किया है!

स्थान एवं गुणवत्ता : काली मिर्च अनुसंधान क्षेव्र, पन्नियूर एवं सिरसी से संचित काली मिर्च नमूनों को गुणवत्ता अंगों और तेल में कारियोफिलिन माव्राओं का मूल्यांकन किया गया! कारियोफिलिन अंश में 13.9 से 18.5% तक अन्तर होता है! एच पी 1411 में 3.6% तेल, 9.8% ओलिओरसिन और 4.2% पाइपरिन होता है!

विशेष वातावरण में संभरण एवं गुणवत्ता : पन्नियूर-1 के सूखे सरस फल 120 दिनों तक तीन परतवाले लोहा आवृत पोलियस्टर कवर में, अन्दर शून्यवाले, 100% नाइद्रोजन और 90% नाइद्रोजन + 10% ओक्सिजन में संभरण किया,! आर्द्रता, एसनशियल ओयल, ओलिओरसिन और पाइपरिन की माव्राएं 60 दिनों के अन्तराल में प्रबोधन किया तो संपूर्ण अवधि में इन्हीं सरस फलों में कोई अन्तर नहीं होता है!

#### फाइटोष्योरा खुर गलन और मन्द म्लानी रोग

नेमटोट का आणविक अभिलक्षणीकरण : राडोफोलस सिमिलिस के चार संख्याओं का जो, केला (कायमकुलम, एरणकुलम और कासरगोड) और नारियल (कायमकुलम) से संचित है , आईटीएस – पीसीआर प्रयुक्त स्पीसीस विशिष्ठ प्राइमर्स द्वारा अभिलक्षणीकरण किया!

प्रतिरोधकता : इकालीस संकरज, छ: कल्टिवर्स (प्राथमिक छान बीन में साधारण सहयतायुक्त के रूप में सूची बना दी) और तीन *पाइपर* स्पीसीस को *फाइटोफथोरा काप्सीसी* का मूल निवेशन किया गया! जांच किये सभी कल्टिवर्स 13 –21 दिनों के निवेशन के बीच 100% नाश दिखाता है! वन्य अक्सशनों में अक्स. 3177 (*पी. सिल्वाटिकम*) *पी. काप्सीसी* के प्रतिरोधक दिखाया!

वर्ष 2005— 06 में तीन खुले परागित संततियों जैसे, 04— पी 24—1. 04 —एचपी1533 —2 और 04—एचपी 1533—3 का सभी रीतियों में छान बीन करने के बाद लघु सूचीबद्ध करके पेरूवन्नामुषि के रोगबाधित प्लॉट में उनके खेत में *फाइटोफथोरा* और नेमटोड प्रतिरोधकता के मूल्यांकन के लिए रोपित किये! इन्हीं पौधों में *फाइटोफथोरा* की रोग बाधा नहीं दिखाई पडी!

#### विरोधियों का मूल्यांकन

कवग और जीवाणुक विरोधियों जैसे *पोकोनिया* क्लामिडोस्पोरिया आई आई एस आर 859 और आई आई एस आर 853 का ग्रीन हाउस स्थिति में मूल्यांकन किया गयाऔर इस अध्ययन से *पी. काप्सीसी* बाधा के नियन्त्रण में आई आई एस आर 853 और आई आई एस आर 859 का प्रभाव सूचित करता है ओर यह कोप्पर आक्सिक्लोराइड के समान होता है!

#### बयोकन्सोटिंयम का खेत मूल्यांकन

पांच साल के परीक्षण के फलस्वरूप रोग आपतन कम करने तथा पौधों की उत्तरजीविता बढाने में आई आई एस आर 6, 8,13,51,151, 853 (रिज़ोबैक्टीरिया) + पीबी 21सी (पी.सोलुबिलाइसर) युक्त रिज़ोबैक्टीरियल संघ का प्रभाव स्पष्ट हआ!

कल्टिवेटड अक्सशनें सी 820 और 1041 और वन्य अक्सशनें अक्सशन 3177 (पी. सिल्वाटिकम) और अक्सशन 5225 (पी. रिबुसोयिड) आदि आई आई एस आर प्रजाति श्रीकरा के साथ कलम बॉधने के लिए मूल स्रोत के रूप में लेने के पहले फाइटोफथोरा और नेमटोड निवेशन को अधीन में लाया गया! अक्सशन 3177 दोनों नेमटोडों जैसे., राडोफोलस सिमिलिस और मेलोयिडोगिने इनकोग्निटा के लिए अत्यन्त सुग्राह्य देख लिया मगर पी. काप्सीसी के लिए प्रतिरोधक भी होता है जबकि अक्सशन 5225 केवल रोडोफोलस सिमिलिस के लिए अत्यन्त सुग्राहयता दिखायी!

साराश

#### एन्डोफाइटिक बैक्टीरिया

आशाजनक एन्डोफाइटिक बैक्टीरिया को *पी. काप्सीसी* और नेमटोड की *प्र्यूडोमोनस एरूगिनोसा* (आई आई एस आर बीपी 35), *प्र्यूडोमोनस पुटिडा* (आई आई एस आर बीपी 25), बासिलस मेगाटरियम (आई आई एस आर बीपी 17), और करटोबैक्टीरियम लुटियम (आई आई एस आर टीसी10) को बुडापेस्ट द्वीटी के अन्तर्गत आईएमटीईसीएच चंडीगढ के साथ पंजीकृत किया!

*बी. मेगाटरियम* (आई आई एस आर बीपी 17 और आई आई एस आर 522) के दो स्ट्रेनों में बीपी 17 में अधिक नेमटोड नाशक क्षमता तथा तापमान और पीएच के उत्तम अनुकूलनशीलता है! जब ग्रीन हाउस में रोग प्रबन्धन के लिए मूल्यांकन किये तो बीपी 35, बीपी 25 और बीपी 17 जैसे वियुक्तियां प्रजातियों की अपेक्षा लगभग 70% से अधिक रोग दमन अंकित किया।

#### उपनिवेशन

पी. एरूगिनोसा आई आईएस आर बीपी 35 के पी. काप्सीसी दमन के लिए आवश्यक न्यूनतम मावाएं 10<sup>12</sup> – 10<sup>13</sup> सेल / मि. लि. है! प्ररोह पर चोट के दमन के लिए जीवाणु निवेशन की न्यूनतम अवधि 15– 20 मिनुट है!

#### एंश्रेक्नोज़ रोग

तीन जगहों में काली मिर्च के एंथ्रेक्नोज़ रोग एवं पर्ण चित्ती के प्रबोधन करने पर यह प्रकट होता है कि मनसून के प्रारंभिक काल (जून) में वर्षा द्वारा मृदा गीला हाने से मृदा में रोगजनकों की व्याप्ति द्रुत गति से होते दिखाई पडी!

#### जीवाणुक रोग

पाइपर येल्लो मोटिल वाइरस बनाने की रीति : काली मिर्च में बाधित पाइपर येल्लो मोटिल वाइरस (पीवाइएमओवी) के ओपन रीडिंग फ्रेम III से ए409 कोडिंग क्षेत्र को दिवरोगवाहक, पीबीआई121 संवेदन और संवेदन रहित अनुस्थापन दोनों में पौध स्थानांतरण रोगवाहक बनाने की तैयारी के लिए प्रयुक्त किया गया!

#### पोल्लू बीटल

कल्टिवेटड अक्सशनों में अक्सशनें 4052, 1472, 4093 और 4095 कीट बाधारहित दिखाई पडी! संकरजों में, एचपी 1357 को कीट बाधारहित देख लिया!

#### इलायची

जननद्रव्य का संग्रहण

छः नये संग्रहों को जननद्रव्य संकलन में जोड दिया! अक्सशनें आई सी 547146 और आई सी 547219 को अधिक उपजता, हरे और घने कैप्स्यूल के लिए लघु सूचीबद्ध किया!

#### मूल्यांकन

पीईटी । और पीईटी ।। के एफ1 संकरज की संततियों के मूल्यांकन के फलस्वरूप 12 अधिक उपजवाले सेलक्शनों और एक संकरज (जीजी X एनकेई -19) को लघु सूची बद्ध किया, जिसके 70 प्रतिशत से अधिक का कैप्स्यूल 8 मि. मी का है और ये पौधे राइजोम गलन एवं पर्ण चित्ती के लिए खेत सह्यतायुक्त है!

सी वाई टी III और IV के अन्तर्गत मलबार सेलक्शनों के तीन अधिक उपजवाले और वाषुका सेलक्शनों के दो अधिक उपजवाले, उसके संबन्धित नियन्द्रणों (सीसीएस1 और न्जल्लानिल गोल्ड) की अपेक्षा उन्नत फसलों में उपजता 10 और 35 प्रतिशत पहचान की गयी!

#### सूखा सहिष्णु

सूखा सहिष्णु प्रकार को विकसित करने के लिए किये गये अध्ययन से आर आर 1 X 893 को दोनों गुणवत्ता और उपजता (4200 हरे कैप्स्यूल / पौधे) में और न्जल्लानिल गोल्ड X 893 का एक चयन संयुक्त पुष्पगुच्छ और हरे घने कैप्स्यूल के लिए उत्तम देखा गया! अन्य एक परीक्षण में, दो उत्तम उत्पादक (ए पी जी 497 और 498 जो प्रतिपौधे 4800 और 4906 कैप्स्यूलवाले है) जिसमें 80% से अधिक घने कैप्स्यूल होता है, अतिरिक्त मूल्यांकन के लिए सूचीबद्ध किया गया!

#### राइज़ोम गलन प्रबन्ध

इन विद्वो मूल्यांकन में, दाइकोडरमा हमाटम को इलायची के राइज़ोम गलन (पिथियम वेक्सान्स और रिजोक्टोनिया सोलानी) के प्रति अधिक प्रभावी देखा गया, जो आर. सोलानी और जिसके बाद व्यापन और परजीविता के प्रति टी. हमाटम कवग तंतु के बहुत गर्मी में उसकी गति साबित करती है!करनाटक के कोडगु, हरसन और चिकमंगलूर जिलाओं तथा वालपराई (तमिलनाडु) के इलायची के

परितंव्र से संचित इलायची, काली मिर्च, हल्दी और अन्य फसलों के रोगवाधित नमूनों से कोलेटोटाइकम ग्लोयियोस्पोरियोयिइस वियुक्तियों में उपनिवेश कोनिडियल (conidial) और. अप्रसोयिरल (appressoiral) विशेषता में विचारणीय विविधता दर्शायी! सभी वियुक्तियों को सी. क्लोयियोस्पयोरियोयिइस के रूप में पहचान की!

#### हल्दी

### संरक्षण एवं अभिलक्षणीकरण

एक्स सिटु वनस्पति रक्षागृह में 1040 कुरकुमा अक्सशनों को संरक्षण एवं अभिलक्षणीकरण किया जा रहा है!

#### इंडियन कुरकुमा स्पीसीस का चरित्रांकन

तीन इंडियन कुरकुमा स्पीसीस के 18 एसआर आरएनए के तुलनात्मक सीक्वेंस विश्लेषण से सूचित करता है कि भारत और चीन में कुरकुमा अरोमटिका, सी. लोंगा और सी. एरूगिनोसा के रूप में मान्यता प्राप्त की सत्ता समान है! भारत और जापान में सी. जेडोवरिया के रूप में मान्यता प्राप्त की सत्ता समान हो जाए जबकि चीन के सी. ज़ेडोवरिया भारत और जापान की संख्या से भिन्न होता है!

#### चरित्रांकन के लिए माइकोसाटलाइट मार्केर्स

माइको साटलाइट युक्त फाग्मन्ट विकसित पुस्तकालय से कुल 700 क्लोन का कोलनी पीसीआर द्वारा छान बीन किया और चयन किये क्लोन का सीक्वेंस किया गया और जिसके फलस्वरूप 82 मइको साटलाइट में लंबाई में विभिन्नता दोहराई गई है! इन्हीं क्षेत्रों के प्रवर्धन के लिए उपयुक्त प्राइमर्स की रूपरेखा प्रस्तुत की जा रही है!

#### कोशिका विश्लेषण

तेरह मातृ पौधों के अक्सशनों के अन्तर्गत 10 अक्सशनों में 2n = 63 की कोमसोम संख्या और तीन अक्सशनों में 2n=84 देखा गया, जबकि बीजपौधे संततियों के चार अक्सशनों में 2n=84 और एक अक्सशन में 2n=77 देख लिया!

#### पुष्पण और फल सज्जा

लगभग पचास बीजपौधे संततियों में पुष्पण अंकित किया। उनमें से 11 पौधों में बीज प्राप्ति हुई और उन्हें बीजपौधे संततियों की नवीन पीढी स्थापित करने के लिए अंकुरण हेतु रख दिया! बोने के 100 दिन बाद दो लोट्स अकुरित होने लगा!

#### पोषण

लक्षित उपज प्राप्ति : मिट्टी में नैद्रोजन, फोस्फोरस, पोटेशियम और ज़िंक के प्रारंभिक उर्वरता स्तरों के आधार पर हल्दी में प्रत्येक बेड से 15, 20, 25 कि. ग्राम के दर में लक्षित उपज प्राप्त करने के लिए उर्वरक की मावाएं कार्यान्वित करके लगा दी! उपलब्ध हल्दी राइज़ोम की उपजता 14.1, 19.4 और 20.9 कि. ग्राम / बेड है जिसमें लक्ष्य से -5-8 %, -3.0 % और -16 % व्यतिकम होता है ! आई आई एस और प्रतिभा प्रजाति में लक्षित पोषण लगाने से संस्तुत मावा से अधिक उपजता 4-54 % तक वर्षदेध हई!

#### गुणवत्ता

जैव रासायनिक चरित्नांकन : हल्दी के विभिन्न प्रजातियों (आलप्पी, राजापुरी और वयनाडन) के जैव रासायनिक अभिलक्षणीकरण से प्रकट होता है कि आलप्पी फ्रिंगर टरमरिक (ए एफ टी) में कुरकुमिन की मात्रा राजापुरी (2. 26%) और वयनाडन (2.42%) हल्दी की अपेक्षा अधिक होती है!

परिष्कृत वातावरण में संमरण : सूखे हल्दी राइज़ोम (आई आई एस आर प्रतिभा) को तीन स्तरवाले लोहा आवृत पोलीयस्टर कवर में 120 दिनों तक रखना है जो अन्तर निर्वात, 100% नाइद्रोजन और 90% नाइद्रोजन + 10% ओक्सिजन होनेवाले है! आर्द्रता, एसनशियल ओयल, ओलिओरसिन और कुरकुमिन को 60 दिनों के अन्तराल में प्रबोधन किया गया और संपूर्ण काल में कोई परिवर्तन नहीं होता है!

यांत्रिक शुष्कण और व्याधिशमन : हल्दी की प्रजातियों जैसे आई आई एस आर प्रतिभा(उपचारित और अनुपचारित) को गरम वायु युक्त ऑवन, रिवर्स फलो ड्रयर, अग्रिवेस्ट ड्रयर और सूर्य प्रकाश में सुखाया जाता है! उपचार किये नमूने सभी प्रकार के सुखाने के उपचारों में अच्छी तरह सूखने (10% तक की आर्द्रता) के लिए कम समय (40.50%) लेता है! फिर भी उपचार किये नमूनों में तेल प्राप्ति में 40% और ओलिओरसिन की प्राप्ति में 25% कमी होती है!

8

#### मब्दा में हल्दी के अन्तर्गत मृदा का आणविक समुदाय संरचना (माइकोबियल कम्यूनिटी संरचना)

आई टी एस 1 , आई टी एस2, प्रोकार्योटिक आई जी एस और 16एस आर डीएनए प्राइमर्स के लिए एकीकष्त उपचार में अधिकतम बैंडों की प्राप्ति हुई! आई टी एस कवग प्राइमर्स के लिए अधिकतम बैंडों में रासायनिक और नियन्वण उपचार पंजीकृत किया! पूरी तरह ओरगानिक उपचार के संबन्ध में प्रोकार्योटिक आईजीएस प्राइमर्स के लिए अधिकतम बैंड का निरीक्षण किया गया! आईपीएनएम और पूरी तरह ओरगानिक उपचारों में अधिकतम नेमटोड को देखा गया!

#### मृदु विगलन

दक्षिण भारत के हल्दी उगानेवाले विभिन्न क्षेद्रो से संचित पिथियम स्पीसीस के 81 वियुक्तियों से 45 वियुक्तियों में रोगजनकता देख ली! इनमें 32 वियुक्तियों को पी. अफानिडेरमाटम और 8 को पी मिरियोटिलम के रूप में पहचान किया! गुल्बरगा (करनाटक), जगतियाल (आन्ध्र प्रदेश) और चित्तूर (केरल) से संचित पी. अफानिडेरमाटम अधिक विषाक्त हो गया!

#### जीवाणुक म्लानी

मृदु विगलन रोग (पिथियम अफानिडेरमाटम के कारण) के प्रति कुरकुमा स्पीसीस का मूल्यांकन करने पर यह सूचित करता है कि कुरकुमा लोंगा, सी अरोमटिका, सी. ज़ेडोवारिया और इज़ड. ओफीशनले अधि संप्रभाव्य देख लिया जबकि सी. अमदा और सी. कैसिया में रोगबाधा नहीं दिखायी पडी!

#### प्ररोह बेधक

जैवपारिस्थितिकी : पेरूवन्नामुषि में कीट बाधा का पहला लक्षण अगस्त (नये प्ररोहों में 0.17% कीट बाधित) में देखा गया और दिसंबर में अधिक (नये परोहों में 10.9% रोग बाधित) होता है!

अपान्टलस स्पीसीस (ए. टारागम्मा) (ब्राकोनिडे) को प्ररोह बेधक के परजीवी लावें के रूप में अंकित किया, मगर परजीविता का स्तर अदरक में होनेवालों की अपेक्षा कम (2. 1 %) होता है! प्रतिरोधिता: खेत में प्ररोह बेधक के प्रति हल्दी के 891 जर्मप्लासम अक्सशनों का छान बीन करने पर सूचित करता हे कि 109 अक्सशनें कीट बाधा रहित होते हैं!

# अदरक का फिनोलजी (घटनाविज्ञान)

औसत में, अदरक के प्रजातियों जैसे आई आई एस आर वरदा, आई आई एस आर महिमा, आई आई एस आर रजता, मारन और हिमाचल आदि को 30 अप्रैल, 15 मई, 30 मई और 15 जून में रोपण करने पर अकुरण के लिए कमश: 27, 23, 17 और 12 दिन लिया है! सदृशपूर्वक डिग्री दिनों में भी अन्तर होता है जो पहले रोपण मे 412.9 डिग्री दिन होते तो अतिम रोपण के दिनों में 143.6 डिग्री दिन होती है। पहला टिलर उत्पादित करने का समय भी भिन्न होता है जो 30 अप्रैल, 15 मई, 30 मई और 15 जून में रोपण किये फसलो में कमश: 73, 61, 51 और 42 दिनों में उत्पादित होता है! प्रत्येक कार्यों के दिनों की डिगी 1029.8, 833.3, 650,1 और 501.6 होता है! अंतिम टिलर कमश: 30 अप्रैल. 15 मई 30 मई और 15 जून को रोपण किये पौधों में रोपण के 140, 144, 121 और 114 दिनों के बाद उत्पादित किया जाता है और यह 1849.5, 1854.3, 1518.7 और 1403.6 डिग्री दिनों में होता है!

#### आणविक चरित्रांकन

आरएपीडी और आईएसएसआर मार्कर्स द्वारा जीनप्रकारों का अन्तर बहुत आसानी से समझ लिया जा सकता है! अदरक के जीनप्रकारों में चार विभिन्न दलों में क्लस्टर विश्लेषण किया है जि़समें, पुटेटीव वन्य प्रकार के साथ श्रेष्ठ जीनप्रकार का ग्रूपिंग करने पर पुटेटीव वन्य प्रकार और आधुनिक कल्टिवर्स के बीच कुछ जातिवृत्तीय संबन्ध अन्तर्निहित होता है! जापान से लिये विदेशी प्रकार राइज़ोम आकृति में पुटेटीव प्रकार से समान होता है चार देशी पुटेटीव प्रकारो के साथ बहुत सदृश होता है!

#### गुणवत्ता

8

उच्च तेल प्रकार: उच्च तेल प्रकारवाले अक्सशनों में अक्सशनों 209, 50, 57, 162, 99,156 197 में 2% से अधिक तेल होता है जबकि अक्सशनें 50, 156 और 57 में 7% से अधिक ओलिओरसिन होता है! अक्सशन 57 और 162 में तेल में अधिक लिमोनेन होता है! नेपाल से संचित अदरक

संग्रह में 1.5-1.9% तेल और 3.8- 6.1% ओलिओरसिन होता है! अक्सशनें 592, 593 और 598 में 1.9% तेल है और अक्सशन 578 में 6.1% ओलिओरसिन होता है।

#### एसनशियल तेल का जीसी एमएस प्रोफाइलिंग

इंडियन (कोचिन) अदरक में कुल 20 घटकें है और चैनीस अदरक में हानेवाले 18 घटकों का जीसीएमएस द्वारा पता लगाया! पहचान किये प्रमुख घटक है कैंफने, α फिल्लानड़ेने, 1, 8 सिनोल, लिनालूल ज़िंजिबरैन, फरनसेन, और β सेसक्विफेल्लानड़ेन आदि! इंडियन उपजों (23.75%) की अपेक्षा चैनीस अदरक (27.14%) में अधिक जिंजिबरैन अकित किया !

#### उपजता का मूल्यांकन

नेपाल अदरक के चुने गये 12 अक्सशनों की उपजता का मूल्यांकन करने पर अक्सशन 581 (16.6 कि.ग्राम ⁄ 3 मीटर² बेड) को अन्य अक्सशनों से भी उत्तम साबित हुआ!

#### सूक्ष्म प्रवर्धन

एम एस बासल मीडियम होनेवाले पेट्री प्लेट तरीके 0.8% अगर, 30 ग्राम/लिटर सुक्रोस के साथ मिनिमल या अनुपयोगी वृद्धि नियामकों को विकसित किया! यह सोमाक्लोनल भिन्नता पर रोग वृद्धि और वृद्धि नियामकों के प्रभाव को कम करने पर इन विट्रो अध्ययन के लिए उत्तम होता है!

#### कयो परिरक्षण

अदरक के शूट बड्स / सोमाटिक एम्प्र्योस में परिसंपुटन और नाइद्रीकरण रीति द्वारा कयो परिरक्षण प्रोटाकॉल 70 % से अधिक सफलता के साथ मानकीकृत किया! संरक्षित सामग्रियां आणविक प्रोफाइलिंग द्वारा जननिक स्थिरता साबित की!

#### मृदु विगलन

पिथियम मिरियोटिलम अदरक एवं हल्दी के मृदु विगलन के साथ संबन्धित है! पिथियम के चार अन्य स्पीसीसों में पी अल्टिमम, पी वेक्सान्स, पी स्प्लेन्टोन्स और पी डिलेन्स भारत में अदरक के मृदु विगलन से संबन्धित है!

#### जीवाणुक म्लानी

जीवाणुक म्लानी की प्रतिरोधकता के लिए मूल्यांकन किये

ज़िंजिबरासिया के स्पीसीसों में जैसे ए. गालंगा, रलोबा स्पीसीस और सी कासिया अदरक के राल्स्टोनिया सोलानसीरम के स्ट्रेन के लिए परपोषी के रूप में देख लिया! सी आमदा, इंडियन मैंगो जिंजर जीवाणुक म्लानी के प्रतिरोधक है!

#### एन्डोफाइटिक बैक्टीरिया का मूल्यांकन

वयनाडु और कोषिक्कोड जिलाओं से संचित अदरक राइज़ोम से वियुक्त उन्नीस एन्डोफाइटिक बैक्टीरिया को रूपवैज्ञानिक कसौटी जैसे कोलनी स्वभाव, आकार, उन्नयन, मोटिलिटी और ग्राम प्रतिकिया द्वारा अभिलक्षणीकरण किया गया और सब ग्राम विरोधी दल में शामिल होता है और उसमे लघु छडी भी है! इन्हीं वियुक्तियॉ में जीईबी 7, जीईबी 9, जीईबी 13, जीईबी 17, जीईबी 18 और जीईबी 19 को जीवाणुक म्लानी एवं मृदु विगलन रोग दमन के लिए आशाजनक देख लिया!

#### पपरीका और पपरीका जैसे मिर्च

#### संग्रहण, मूल्यांकन एवं अनुरक्षण

उत्तर पूर्व से संचित अधिक तीक्ष्ण और अधिक रंगवाले एक अक्सशन 'भुट जोलोकिया' को जर्मप्लासम में जोड दिया! देशी जर्मप्लासम में, आईसीबीडी 10 में अधिकतम रंग मूल्य (302.12 एएसटीए एकक) पंजीकृत किया, आईसीबीडी 11 और आईसीबीडी 23 उसके बाद आता है! विदेशी लाइनों में, ईसी 171 में अधिकतम रंग मूल्य अंकित किया! इसमें कैप्साइसिन के अंश में 0.0071 से 0.166 तक अंतर होता है! आईसीबीडी10, केटी पीआई 19 और ईसी 71 को कम तीक्ष्णता एवं अधिक रंगमूल्ययुक्त आशाजनक देख लिया!

#### कैप्सिकम (कैप्सिकम स्पीसीस) संग्रहों का डीएनए प्रोफाइलिंग

नौ कैप्सिकम संग्रह को आई आई एस आर प्रोफाइलों ने विकसित किया और उनमें से 10 प्राइमर्स ने परीक्षण किये अक्सशनों के अन्तर समझने के लिए विश्वसनीय अंक प्रदान किया!

#### वैनिला

#### अन्तर विशिष्ट संबन्ध

*वानिला प्लानिफोलिया, वी अफिल्ला, वी. पीलीफेरा* और अन्डमान निकोबार द्वीप सें संचित *वैनिला स्पीसीसों* के

साराध

अन्तर -विशिष्ट संबन्ध के फलस्वरूप सभी संकर संयोजनों में कॉस काम्पिटिबिलिटी सूचित करके सफल फल सज्जा प्रकट हुई!

#### पराग अंकुरण

10% सुकोस होनेवाले बी और के मीडियम प्रकाश का इस्तेमाल करके बूँद तरीके 25±2° से साफ पराग कल्चरिंग करने पर *वी. अफिल्ला* (58 %) में अधिकतम अंकुरण अंकित किया और चार घटे बाद *वी. पीलीफेरा* (91.25μm) में पराग नल लंबे होते है!

#### कवग रोग

करनाटक की मंडिकेरी से संचित वैनिला पौधे जो क्लोरोसिस और पर्ण विरूपण का विशेष लक्षण दिखानेवाले है, जब इन्हें पांच विभिन्न पोटीवाइरसों के लिए आरटी – पीसीआर प्राइमर्स विशिष्ट द्वारा बीन पीला मोसाइक वाइरसस (बीवाइएमवी) के विशिष्ट प्राइमर्स के साथ प्रवर्धन किया! पीसीआर उपजों को इल्यूट किया, क्लोन किया और सीक्वन्स बनाया! न्यूक्लियोटाइड प्रतिशत और डेड्यूर्स्ड अमिनो एसिड की पहचान और फिलोजनटिक विश्लेषण के आधार पर सीक्वन्स विश्लेषण करने पर स्पष्टतया सूचित करता है कि वैनिला वियुक्तियों में बीवाईएमवी का स्ट्रेन होता है!

#### वृक्ष प्रजातियां

#### जायफल का मूल्यांकन

विभिन्न जीनप्रकारों की दक्षता का मूल्यांकन करने केलिए किये गये अध्ययन में, ए 4-17 को उत्तम और अधिक अंकुरण(80), वृदि्ध और शत प्रतिशत कलम बांधने की सफलता प्राप्त हुए देख लिया!

#### जायफल को बढाने का माध्यम

विभिन्न वृदिधदायक संयोजनों जैसे मृदा : ग्रानइट : एफवाइएम (2:1:1) लगाने पर बीजपौधों से उत्तम वृदि्ध 86% देख लिया।

#### जायफल का लिंग निर्णय

जायफल (*मिरिस्टिका फ्राग्रन्स*) के लिंग संग्रह डीएनए की नर पेड या मादा पेड की छान बीन के लिए 165 आरएपीडी प्राइमर्स और 25 आई एसएसआर प्राइमर्स का प्रयोग करने पर, सात संयोगिक प्राइमर्स ने उनके बीच पोलिमोरफिसम दिखाया और चार प्राइमर्स (ओपीएएम13, ओपीएएफ16, ओपीएएम16 और ओपीएएफ5) ने लिंग विशिष्ट बहुरूपी बैंड को दिखाया!

#### गार्सीनिया स्पीसीसों में आणविक प्रोफाइलिंग

आईएसएसआर और आरएपीडी अंकको के आधार पर डीएनए रूपरेखा में नौ गार्सीनिया स्पीसीसों को प्रयुक्त किया! कुल मिलाकर नौ आईएसएसआर प्राइमर्स और बारह आरएपीडी प्राइमर्स ने उसकी पहचान में सफलता प्राप्त की!

सिन्नमोमम एसनशियल ओयल का संयोजन

सी. वीरम और सी मलबाद्रम के फलों में होनेवाले एसनशियल ओयल का प्रमुख संघटक प्रारभिक अवस्था मे लिनालूल है! विकास की अगली अवस्था में, लिनालूल का अंश कम होता है और दोनों तेल δ-काडिनेन द्वारा (कमश: 19.6% & 24.9%) प्रभावी हो गया!

#### मसालों के सार में अन्टीओक्सिडन्ट का स्तर

फोस्फोमोलिब्डिनम रीति द्वारा विश्लेषण करने पर *गार्सीनिया* गम्मि गट्टा के एथनोल अंश और हल्दी और इमली के जल सार को सबसे अधिक ओक्सिडन्ट क्षमतायुक्त देखे गये! काली मर्च के एसनशियल ओयल, काली मर्च, कढी पत्ता और इमली के एथनोल अंश में 1, 1- डिफनिल - 2-पिक्रिलहाइड्राक्सिल राडिकल अपमार्जन क्षमता अधिक होता है! हल्दी के एथनोल सार में एफ ई III से एफ ई II कम करने की क्षमता अधिकतम होती है!

#### मसालों के एसनशियल ओयल द्वारा अफलाटोक्सिन का प्रावरोध

एस्परगिलस पर मसालों का एसनशियल ओयल और उसके अन्य प्रमुख घटकों द्वारा कवग विषाक्त और कवग नाशिकीय प्रभाव के विस्तार द्वारा अफलाटोक्सिन बी, के संबन्ध में माव्राएं देखी गयी! लौंग तेल, दालचीनी तेल और हल्दी पर्ण तेल में अफ्लाटोक्सिन उत्पादन का संपूर्ण प्रावरोध है! दोनों अफलाटोक्सिन (जी, और बी) दालचीनी छाल तेल और उसी प्रकार सिनमलडिहाइड के लिए सकिय घटक के रूप में प्रावरोध किया जा सकता हैं!

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**HIVE** 

#### मसालों में रिज़ोबैक्टीरिया

काली मिर्च (50) एवं अदरक (69) से एक सौ उन्नीस रिज़ोबैक्टीरियाओं को वियुक्त किया गया! इनमें से 53 वियुक्तियों ने आईएए का, 38 ने सोलुबिलाइज़ड फोस्फेट का, और केवल 4 वियुक्तियों ने एचसीएन का उत्पादन किया! बारह रिज़ोबैक्टीरियल वियुक्तियों को *इन विद्रो* वरीक्षण के आधार पर लघु सूची बना दी और वे *पी. काप्सीसी, पिथियम* और *फुसेरियम* के प्रति 50% से अधिक प्रतिरोधक क्षमता दर्शाई!

#### विस्तार एवं प्रशिक्षण

ग्रामीण संसाधन केन्द्र ने किसानों की भलाई के लिए वीडियो सम्मेलन आयोजित किया! प्रस्तूत काल में छ सम्मेलन सत्र संपन्न हुए जिसमें संस्थान के विशेषज्ञों के साथ कालिकट के अन्य संस्थानों के विशेषज्ञ भी भाग लिये थे! स्पाइसस बोर्ड और स्टेट हॉर्टीकल्चर मिशन के अन्तर्गत विभिन्न योजनाओं दवारा चार संस्थान केंपस प्रशिक्षण कोर्स विस्तार कर्मियों की भलाई के लिए प्रायोजित किये जिनमें 60 प्रशिक्षार्थी भाग लिये! संस्थान में 07 मई से 06 जून 2007 तक जैवरसायन, जैवप्रौदयोगिकी और जैवसूचनाओं पर ग्रीष्मकालीन प्रशिक्षण कार्यक्रम आयोजित किया जिसमें 42 एम.एससी छात्र भाग लिये! सरकारी अधिकारियों के लिए सूचना सुरक्षा पर दिनांक 12 -13 नवंबर 2007 को डीओईएसीसी केन्द्र, कालिकट के सहयोग से और एक प्रशिक्षण आयोजित किया! सेन्द्रल सेक्टर स्कीम के अन्तर्गत सिक्किम सहित उत्तर पूर्व राज्यों में एकीकृत बागवानी विकास के तकनोलजी भिशन ने 'उत्तर पूर्व राज्यों में प्रमुख मसालों का उत्पादन एवं संसाधन' पर एक राज्य स्तरीय कार्यशाला दिमापुर, नागालैंड में आयोजित की जिसमे नागालैंड के 10 जिलाओं का प्रतिनिधित्व करनेवाले 60 किसानों / विस्तार कार्मिकों ने भाग ली!

#### अखिल भारतीय समन्वित मसाला अनुसंघान परियोजना

अखिल भारतीय समन्वित मसाला अनुसंधान परियोजना के शोध कर्मियो का राष्ट्रीय दल बैठक (XIX वीं कार्यशाला) 23- 25. नवंबर, 2007 को उडीसा कृषि तकनोलजी विश्वविद्यालय (ओयुएटी) भुवनेश्वर, उडीसा में संपन्न हुई! धनिया के पर्ण प्रकार डीएच 228 का विमोचन करने का प्रस्ताव सीसीएस एचएयु, हिसार ने हरियाना स्टेट रिलीज के लिए संस्तुत किया! विभिन्न अनुसंधान परियोजनाओं से निकलते आशाजनक 11 तकनोलजियां अंगीकृत परीक्षण के लिए पहचान किये गये! विभिन्न मसाला फसलों पर बारह नये तकनीकी कार्यक्रम का संयोजन किया गया!

#### अन्य

#### जैवसूचना डॉटाबेसस

प्रस्तुत काल में रिपोर्ट के अधीन जैवसूचना केन्द्र ने दो डॉटाबेसस को विकसित किया है।

प्लासबिड: यह पौधों से संबन्धित एकीकृत बैक्टीरिया का डेटाबेस है जो कई साधनों जैसे सीक्वेंस संपादन, प्राइमर विश्लेषण आदि के साथ जुटे है!

पासकोम: मसाला संयोजनों के पूर्वानुमेय कार्यों पर विकसित डेटाबेस है! इसमें काली मिर्च का मापदंड पूरा किया गया!

पी आइ आर और फि–डिष: डाटाबेसस *फाइटोफ्थोरा* के राश्ट्रीय संग्रह के रूप में सेवा करने के लिए एकीकृत किया गया!

नई वैबसाइट्स और अन्य ई - संसाधनों का विकास: कृषि वैज्ञानिक नियुक्ति मंडल, नई दिल्ली के लिए एक नई वेबसाइट विकसित की! डॉ मंगला राय, महानिदेशक, भारतीय कृषि अनुसंधान परिषद ने दिनाक 5 नवंबर 2007 को नई दिल्ली में इस साइट को प्रक्षेपित किया! भारतीय कृषि अनुसंधान परिषद के बागवानी प्रभाग के लिए एक वेब साइट विकसित करके आधुनिक बनाया। संस्थान के पूरा किये अनुसंधान परियोजनाओं (आरपीएफ III) के लिए एक ई--बुक तैयार किया। एरिसॉफ्ट, आई0आई0एस0आर0 ऑफिस ऑटोमेशन सोफटवेर कई नई विशेषताओं के साथ परिष्कृत किया!

## Annual Report 2007-08 Executive Summary

#### **Black** pepper

#### Germplasm collection, conservation and evaluation

During the year, 54 accessions from Shendurney WLS, 24 from Bisle Ghats forests and 32 from Karwar district were collected from the species *Piper nigrum*, *P. galeatum*, *P. trichostachyon*, *P. attenuatum* and *P. hapnium* including two local cultivars from farmers' fields. Ninety eight accessions were characterized based on IPGRI descriptor in addition to 1000 accessions.

#### Breeding

For pollu beetle, nematode and drought resistance: A hybridization block was established with bush pepper var. IISR Subhakara as female parent and 'pollu' tolerant lines (Coll. No. 816, 841, 1084 and 1114), nematode tolerant lines (Acc. 820 and HP 39) and drought tolerant lines (Acc. No. 1495, 931 and 813) as male parents. Seedlings raised from these cross combinations are under multiplication for field planting.

For high yield and high caryophyllene: A total of 114 progenies of crosses involving IISR Subhakara (female) and high caryophyllene lines (male) were produced. Analysis of berry caryophyllene content of the elite lines (male lines) indicated variation. Location effect on beta caryophyllene content of IISR Subhakara was also observed in the samples collected from Ambalavayal, Peruvannamuzhi, Panniyur and Yercaud centres.

**For** *Phytophthora* **resistance:** To develop a second mapping population to tag *Phytophthora* resistance, 50 new crosses were

made between IISR Shakthi x IISR Subhakara and the progenies were planted for germination. Twenty five new putative transgenics containing osmotin were hardened for testing.

#### Nutrition

Sulphate of Potash (SOP) on yield and quality : The leaf nutrient status, biochemical parameters and yield were found to be on par in 50% of recommended K as SOP with or without foliar spray and SOP with magnesium supplementation. Recommended dose of K as SOP + 2% foliar spray recorded highest yield that was on par with 50% of the recommended K as SOP and recommended K as  $SOP + MgSO_{4}$ treatments. When a premium price for the SOP treatments is considered, the recommended K or 50% of it as SOP with or without foliar spray recorded better net returns and B:C ratio (1.73-2.10) as that of MOP treatment. Quality parameters like oleoresin and piperine contents were also high in these treatments.

**Organic manures on P release:** Step wise forward regression analysis of P fractions showed Fe-P and Available P as the best predictors for yield (= 46.25 + 1.034 Fe-P + 0.570 Av P) with a R<sup>2</sup> of 0.611\*\*.

#### Drought tolerance

SOD isozyme showed five prominent bands, which was common both for tolerant and susceptible accessions. Water stress induced an additional SOD band in some accessions. Tolerant accessions showed higher photosynthetic rate and higher stomatal conductance than the susceptible ones after five days of stress induction. Evenitive Cummen

Physiological and biochemical basis of productivity

High yielders had higher NR and lower MDH activities compared to low yielders during prebearing period. High yielders in general, had higher carbohydrates and starch during this period than low yielders.

#### Quality

**Myristicin in** *P. nigrum*: Leaf oil composition of *P. nigrum* leaf samples collected from Dapoli and Karwar, showed myristicin in good concentration, which is a general component of nutmeg and was not recorded so far in *P. nigrum*.

**Location and quality:** Black pepper samples from PRS, Panniyur and Sirsi were evaluated for quality constituents and caryophyllene in oil. Caryophyllene content varied from 13.9 to 18.5%. HP-1411 had 3.6% oil, 9.8% oleoresin and 4.2% piperine.

**Modified atmospheric storage and quality:** Dried berries of Panniyur-1 was stored in three layered metallised polyester cover under vacuum, 100% nitrogen and 90% nitrogen + 10% oxygen for a period of 120 days. Moisture, essential oil, oleoresin and piperine contents monitored at 60 days interval in the berries remained unchanged throughout the period.

## Phytophthora foot rot and slow wilt diseases

**Molecular characterization of nematodes:** Four populations of *Radopholus similis*, collected from banana (Kayamkulam, Ernakulam and Kasaragod) and coconut (Kayamkulam), were characterized through ITS-PCR using species-specific primers.

**Resistance:** Forty one hybrids, six cultivars (short-listed as moderately tolerant in the preliminary screening) and three *Piper* spp.

were subjected to root inoculation of *Phytophthora capsici*. All the cultivars tested showed 100% mortality within 13-21 days of inoculation. Among the wild accessions, Acc. 3177 (*P. sylvaticum*) showed resistance to *P. capsici*.

Three open pollinated progenies namely, 04-P24-1, 04-HP 1533-2 and 04-HP 1533-3 that were short-listed based on all methods of screening during 2005-06 were planted in a sick plot at Peruvannamuzhi for evaluating their field reaction to *Phytophthora* and nematodes. No *Phytophthora* infection could be noticed in these plants.

#### **Evaluation of antagonists**

The promising isolates of fungal and bacterial antagonists namely, *Pochonia chlamydosporia*, IISR-859 and IISR-853 were evaluated under green house conditions and the study indicated the efficacy of IISR-853 and IISR-859 in controlling *P. capsici* infection and it was on par with copper oxychloride.

#### Field evaluation of bioconsortium

The results of five years trials indicated the efficacy of the rhizobacterial consortium containing IISR-6, 8, 13, 51, 151, 853 (rhizobacteria) + PB-21C (P-solubilizer) in reducing the disease incidence as well as increasing the survival of the plants.

The cultivated accessions C 820 and 1041 and wild accessions Acc. 3177 (*P. sylvaticum*) and Acc. 5225 (*P. ribusoids*) were subjected to *Phytophthora* and nematode inoculation prior to being used as root stock for grafting with var. IISR Sreekara. Acc. 3177 was highly susceptible to both the nematodes *viz.*, *Radopholus similis* and *Meloidogyne incognita* but was resistant to *P. capsici* while Acc. 5225 showed high susceptibility to *R. similis* alone.

#### Endophytic bacteria

Evaluation: The endophytic bacteria promising against P. capsici and nematodes viz., Pseudomonas aeruginosa (IISR BP35), Pseudomonas putida (IISR BP25), Bacillus megaterium (IISR BP17) and Curtobacterium luteum (IISR TC10) have been registered with IMTECH, Chandigarh, under Budapest Treaty. Among the two strains of B. megaterium (IISR BP17 and HSR 522), BP17 possessed higher nematicidal activity and better adaptability to temperature and pH. When evaluated in the greenhouse for disease management, the isolates BP35, BP25 and BP17 recorded over 70% disease suppression irrespective of the variety.

Colonization : Minimal concentration of P. aeruginosa IISR BP35 required for suppression of P. capsici was 10<sup>12</sup> - 10<sup>13</sup> cells ml<sup>-1</sup>. The minimum duration of bacterization for suppression of lesion on shoot was 15-20 min.

#### Anthracnose disease

Monitoring of anthracnose disease of black pepper and leaf blight in three locations revealed the rapid spread of disease during early monsoon period (June) through rain splash of the pathogens in the soil.

#### Viral diseases

Preparation of Piper yellow mottle virus construct : A 409 bp coding region from open reading frame (ORF) III of Piper yellow mottle virus (PYMoV) infecting black pepper was used for the preparation of plant transformation vector construct in the binary vector, pBI 121 both in sense and antisense orientation.

#### Pollu beetle

Among the cultivated accessions, Accs. 4052, 1472, 4093 and 4095 were free from the pest attack. Among the hybrids, HP 1357 was free from pest infestation.

Indian Institute of Spices Research

#### Germplasm collection

Six new collections were added to the germplasm. Accessions, IC - 547146 and IC -547219 were short listed for high yield, green and bold capsules.

#### Evaluation

Evaluation of F1 hybrid progenies of PET I and PET II resulted in short-listing of 12 high yielding selections and one hybrid (GG x NKE - 19) with more than 70 per cent capsules having 8 mm size and field tolerance to rhizome rot and leaf blight.

Under CYT III and IV, three high yielders of Malabar selections and 2 high yielders of Vazhukka selections yielding 10 and 35 per cent higher crop than their respective checks (CCS 1 and Njallanil Gold) were identified.

#### Drought tolerance

In a study to evolve drought tolerant type, RR1 x 893 was found superior for both quality and yield (4200 green capsules/ plant) and one of the selections in Njallanil Gold x 893 for compound panicles and green bold capsules. In another trial, two best yielders (APG 497 and 498 with 4800 and 4906 capsules per plant) with more than 80% bold capsules were short listed for further evaluation.

#### Rhizome rot management

Under in vitro evaluation, Trichoderma hamatum was found more effective against rhizome rot (Pythium vexans and Rhizoctonia solani) of cardamom, which is evident from the tropical movement of T. hamatum hyphae towards R. solani followed by penetration and parasitization. Considerable diversity was observed in Colletotrichum gloeosporioides isolates with regard to colony, conidial and appressoiral characteristics from the infected **Executive Summary** 

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specimens of cardamom, black pepper, turmeric and other crops, collected from cardamom ecosystem of Kodagu, Hassan and Chickmagalur districts of Karnataka and Valparai (Tamil Nadu). All isolates were identified as C. gloeosporioides.

#### Turmerie

## Conservation and characterization

In the  $ex-sit_{ij}$  conservatory, 1040 Curcuma accessions are being conserved and characterized.

# Characterization of Indian Curcuma species

Comparative sequence analysis of the 18 S rRNA regions of three Indian Curcuma species indicated that the entities recognized as Curcuma aromatica, C. longa and C. aeruginosa in India and China are the same. The entity recognized as C. zedoaria in India and Japan may be the same, while C. zedoaria of China is distinct from the Indian and Japanese populations.

## Microsatellite markers for characterization

A total of 700 clones from the enriched libraries (microsatellite containing fragments) were screened by colony PCR and selected clones were sequenced which resulted in 82 microsatellite repeats of variable length. Suitable primers to amplify these regions are being designed.

## Cytological analysis

Chromosome number of 2n=63 in 10 and 2n=84 in 3 was observed in 13 mother plant accessions while four accessions of seedling progenies had 2n=84 and one had 2n=77.

## Flowering and seed set

Flowering was recorded in about 50 seedling progenies. Seeds were recovered from 11 of them and were put for germination to establish advanced generation of seedling progenies. Germination started in two lots after 100 days of sowing.

#### Nutrition

**Targeted yield response:** Based on the initial soil fertility levels of N, P. K and Zn, the fertilizer doses for obtaining 15, 20 and 25 kg/ bed yield targets in turmeric were worked out and applied. The achieved turmeric rhizome yield was 14.1, 19.4 and 20.9 kg/bed with a deviation of -5.8%, - 3.0% and -16% from the target. Through targeted nutrient application 4-54% increased yield over recommended dose could be achieved in IISR Prathibha.

#### Quality

**Biochemical characterization:** Biochemical characterization of different traded varieties of turmeric ('Alleppey', 'Rajapuri' and 'Wyanadan') revealed a high content of curcumin (3.11%) in Alleppey finger turmeric (AFT), as compared to Rajapuri (2.26%) and Wyanadan (2.42%) turmeric.

**Modified atmospheric storage:** Dried turmeric rhizomes (IISR Prathibha) were stored in three layered metallised polyester cover under vacuum, 100% nitrogen and 90% nitrogen + 10% oxygen for a period of 120 days. Moisture, essential oil, oleoresin and curcumin levels monitored at 60 days interval remained unchanged throughout the period.

**Mechanical drying and curing:** Turmeric var. IISR Prathibha (cured and raw) was dried in hot air oven, reverse flow drier, agriwaste drier and also sun dried. Cured sample took less time (40-50%) for completion of drying in all the drying treatments compared to raw sample. However, cured samples showed a reduction of 40% in oil recovery and 25% reduction in oleoresin recovery.

**Executive Summary** 

## Soil microbial community structure in soils under turmeric

Ginger

Maximum bands were obtained in integrated treatment for ITS1, ITS2, prokaryotic IGS and 16S rDNA primers. Chemical and control *treatments registered maximum bands for ITS* fungal primers. In case of fully organic treatment maximum bands were observed for prokaryotic IGS primers. Maximum number of nematodes was found in case of IPNM and fully organic treatments.

#### Soft rot

Forty five of the 81 isolates of *Pythium* sp. collected from different turmeric growing regions of South India showed pathogenicity. Among them 32 isolates were identified as *P. aphanidermatum*, and 8 as *P. myriotylum*. *P. aphanidermatum* collected from Gulburga (Karnataka), Jagtial (Andhra Pradesh) and Chittoor (Kerala) were the most virulent.

#### Bacterial wilt

Evaluation of *Curcuma* sp. against soft rot disease (caused by *Pythium aphanidermatum*) indicated that *Curcuma longa*, *C. aromatica*, *C. zedoaria* and *Z. officinale* were susceptible while *C. amada and C. caesia* did not take up the infection.

#### Shoot borer

**Bioecology:** The symptoms of pest infestation were first observed during August (0.17% new shoots infested) and was high during December (10.9% new shoots infested) in Peruvannamuzhi. *Apanteles* sp. (A. taragammae?) (Braconidae) was recorded to parasitize larvae of shoot borer but level of parasitism was low (2.1%) when compared to that on ginger.

**Resistance:** Screening of 891 turmeric germplasm accessions against shoot borer in the field indicated that 109 accessions remained free from pest infestation.

#### Phenology of ginger

On an average, ginger varieties viz., HSR Varada, IISR Mahima, IISR Rejatha, Maran and Himachal took 27, 23, 17 and 12 days for germination in 30 April, 15 May, 30 May and 15 June planted crops, respectively. Correspondingly degree days also varied from 412.9 degree days at first planting to 143.6 degree days for last planting. The time taken to produce first tiller was 73, 61, 51 and 42 days in 30 April, 15 May, 30 May and 15 June planted crops, respectively. The degree days for the events were in the order 1029.8, 833.3, 650.1 and 501.6. The last tiller was produced 140, 144, 121 and 114 days after planting in 30 April, 15 May, 30 May and 15 June planted crops respectively and this event happened at 1849.5, 1854.3, 1518.7 and 1403.6 degree days.

#### Molecular characterization

Genotypes could be easily discriminated using RAPD and ISSR markers. Cluster analysis placed the ginger genotypes into four separate groups, in which the grouping of elite genotypes with the putative wild types implies some phylogenetic relationship between the putative wild types and modern cultivars. The exotic type from Japan, resembling the putative types in rhizome features, shared high similarity with the four indigenous putative types.

#### Quality

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**High oil types:** Among the high oil types Acc. 209, 50, 57,162, 99, 156 and 197 had above 2% oil and Acc. 50,156 and 57 had more than 7% oleoresin. Acc. 57 and 162 contained high limonene in oil. Nepal ginger collections had 1.5-1.9% oil and 3.8-6.1% oleoresin. Acc. 592, 593 and 598 contained 1.9% oil and Acc. 578 contained 6.1% oleoresin.

**Executive Summary** 

#### GCMS profiling of essential oil

A total of 20 compounds in Indian (Cochin) ginger and 18 in Chinese ginger were detected through GCMS. The major compounds identified were camphene,  $\alpha$  phellandrene, 1, 8 cineole, linalool, zingiberene, farnesene, and  $\beta$  sesquiphellandrene. High zingiberene was recorded in Chinese ginger (27.14%) compared to the Indian produce (23.75%).

#### **Yield** evaluation

Yield evaluation of 12 selected Nepal ginger accessions revealed the superiority of Acc. 581(16.6 kg per 3 m<sup>2</sup> bed) over others.

#### Micropropagation

A method using petri plates containing MS basal medium with 0.8% agar, 30g/l sucrose with minimal or no use of growth regulators was developed. It is ideally suitable for *in vitro* studies on disease progression and reducing the effect of growth regulators on the somaclonal variation.

#### Cryo preservation

Cryo preservation protocols were standardized in ginger shoot buds/somatic embryos using encapsulation and nitrification methods with over 70% success. The conserved materials are genetically stable as evident by molecular profiling.

#### Soft rot

Pythium myriotylum was associated with soft rot of ginger. Four other species of Pythium namely, P. ultimum, P. vexans, P. splendons and P. delaines were also associated with ginger soft rot in India.

#### **Bacterial** wilt

Among the species of zingiberaceae evaluated for resistance to bacterial wilt, *A. galanga*, *Globba* sp. and *C. caesia* were found to be the hosts for ginger strain of *Ralstonia*  solanacearum. C. amada, the Indian mango ginger was resistant to bacterial wilt.

#### Evaluation of endophytic bacteria

Nineteen endophytic bacteria isolated from ginger rhizomes collected from Wayanad and Kozhikode districts were characterized using morphological criteria such as colony character, shape, elevation, motility and gram reaction, and all of them belonged to gram negative group and were short rods. Among the isolates GEB 7, GEB 9, GEB 13, GEB 17, GEB 18, and GEB 19 were promising for suppression of bacterial wilt and soft rot diseases.

#### Paprika and paprika like chillies

#### Collection, evaluation and maintenance

A high pungent and high colour accession from North East 'Bhut Jolokia' was added to the germplasm. Among the indigenous germplasm, ICBD-10 registered the highest colour value (302.12 ASTA units). The EC-171 recorded the highest colour value, among the exotic lines. The capsaicin content varied from 0.0071 to 0.166. The lines, ICBD-10, Kt-Pl-19 and EC-71 were found promising with low pungency and high colour value.

## DNA profiling of capsicum (*Capsicum* spp.) collections

ISSR profiles have been developed for nine *Capsicum* collections and 10 of the primers gave reliable markers for distinction of the accessions tested.

#### Vanilla

#### Inter-specific crosses

Inter-specific crosses performed between Vanilla planifolia, V. aphylla, V. pilifera and V. sp. from A&N islands revealed successful fruit setting in all cross combinations indicating cross compatibility.

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#### Pollen germination

Culturing fresh pollen by sitting drop method at  $25\pm2^{\circ}$ C under light using B&K medium containing 10% sucrose gave maximum germination in V. aphylla (58%) and longer pollen tube in V. pilifera (91.25 im) after 4 hours.

#### Viral disease

Vanilla plants showing typical symptoms of chlorosis and leaf deformation collected from Madikeri district of Karnataka when subjected to RT-PCR using primers specific for five different potyviruses, gave amplification with primers specific to *Bean yellow mosaic virus* (BYMV). The PCR product was eluted, cloned and sequenced. Sequence analysis based on per cent nucleotide and deduced amino acid identity, and phylogenetic analysis clearly showed that vanilla isolate is a strain of BYMV.

#### C Tree spices

#### Evaluation of nutmeg

In a study to evaluate the performance of various genotypes, A4-17 was found to be the best with high germination (80%), growth and cent per cent grafting success.

#### Growing media for nutmeg

Among the various growing media combinations, seedling growth was best in the combination of soil:granite:FYM (2:1:1) with 86% germination.

#### Sex determination in nutmeg

Out of 165 RAPD primers and 25 ISSR primers used for screening sex pooled DNA from male and female *Myristica fragrans*, seven random primers showed polymorphism between them and four primers (OPAM 13, OPAF 16, OPAM 16 and OPAF 5) showed sex specific polymorphic bands.

#### Molecular profiling in Garcinia species

Nine species of *Garcinia* were used in DNA profiling based on ISSR and RAPD markers. A total of nine ISSR primers and twelve RAPD primers could successfully distinguish them.

Composition of Cinnamomum essential oil

The chief constituent of the essential oils of fruit of C. verum and C. malabatrum was linalool during the initial stages. During the later stages of development, linalool content decreased and both the oils were dominated by  $\delta$ -cadinene (19.6% and 24.9% respectively).

#### Antioxidant levels in spice extracts

Ethanol extracts of *Garcinia gummi-gutta* and water extracts of turmeric and tamarind had the most total antioxidant capacity as analyzed by the phosphomolybdenum method. The 1,1diphenyl-2-picrylhydrazyl radical scavenging ability was greater in the essential oil of black pepper and ethanol extracts of black pepper, curry leaves and tamarind. Fe (III) to Fe (II) reducing activity was highest in the ethanol extracts of turmeric and the two *Garcinia* species.

#### Inhibition of aflatoxin production by spice essential oils

Extent of fungitoxic and fungicidal effect by spice essential oils and their major components on *Aspergillus* has been quantified in terms of aflatoxin B1. Clove oil, cinnamon oil and turmeric leaf oil had complete inhibition of aflatoxin production. Both the aflatoxins (G1 and B1) could be inhibited by cinnamon bark oil as well as the active ingredient, cinnamaldehyde.

#### Rhizobacteria in spices

One hundred and nineteen rhizobacteria have been isolated from black pepper (50) and ginger (69). Out of these, 53 produced IAA, 38 solubilized phosphate and only 4 isolates Executive Summary

produced HCN. Twelve rhizobacterial isolates were shortlisted based on *in vitro* tests and those showing more than 50% inhibition against *P. capsici, Pythium* and *Fusarium*.

#### **Extension and training**

Village Resource Centre conducts video conferencing sessions for the benefit of the farmers. During the period, six conferencing sessions were held involving experts from the institute and other institutions in Calicut. Four institute on campus training courses sponsored by various schemes under the Spices Board and State Horticulture Mission (SHM) were organized for the benefit of extension personnel in which 60 trainees participated. A summer training programme on Biochemistry, Biotechnology and Bioinformatics was organized from 07 May to 06 June 2007 in which 42 M.Sc. students participated. Another training course on "Information Security for Government Officers" was conducted during 12-13 November, 2007 in collaboration with DOEACC Centre, Calicut. Under the Central Sector Scheme, Technology Mission for Integrated Development of Horticulture in North Eastern states including Sikkim, one state level workshop on 'production and processing in major spices in NE states was organized in Dimapur, Nagaland in which 60 farmers/extension personnel representing 10 districts of Nagaland participated.

#### All India Coordinated Research Project on Spices

The National group meeting (XIX workshop) of research workers of AICRP on Spices was held at Orissa University for Agriculture and Technology (OUAT), Bhubaneswar, Orissa during 23-25, November 2007. The varietal release proposal of leafy type coriander DH – 228 from CCS HAU, Hisar was recommended for Haryana state release. The promising 11 technologies emanated from different research projects were identified for adoptive trials. Twelve new technical programmes were formulated on different spice crops.

#### Others

#### **Bioinformatics** databases

The bioinformatics centre has developed two databases during the period under report.

**PLASBID:** A database on plant associated bacteria integrated with several tools like sequence editing, primer analysis *etc*.

**PASSCOM:** A database on predicted activities of spice compounds is being developed. The module on black pepper has been completed.

The databases PIR and PHYDISH were integrated to serve as a National Repository of *Phytophthora*.

**Development of new websites and other e-resources:** A new website for Agriculture Scientists Recruitment Board, New Delhi was developed. The site was launched by Dr. Mangala Rai, DG, ICAR on 5 November 2007 at New Delhi. HortWeb, the website of Horticulture Division of ICAR was developed and updated. An e-book on completed research projects of the institute (RPF III) was prepared. ARISoft, the IISR office automation software was fine tuned and modified to incorporate several new features.

**Executive Summary** 

## Introduction

#### History

Intensive research on spices in the country was initiated with the establishment of a Regional Station of Central Plantation Crops Research Institute (CPCRI) at Calicut, Kerala, during 1975, by the Indian Council of Agricultural Research (ICAR). This Regional Station was upgraded as National Research Centre for Spices (NRCS) in 1986 by merging with it the Cardamom Research Centre of CPCRI at Appangala, Madikeri, Karnataka. The NRCS was further elevated to the present Indian Institute of Spices Research (IISR) during 1995.

#### Location

The laboratories and administrative offices of the institute are located at Chelavoor (50 m above MSL), 11 km from Calicut (Kozhikode), Kozhikode District, Kerala, on the Calicut-Kollegal road (NH 212), in an area of 14.3 ha. The research farm is located 51 km North East of Calicut at Peruvannamuzhi (60 m above MSL), on the Peruvannamuzhi-Poozhithode road in Kozhikode District, in an area of 94.08 ha. The Cardamom Research Centre, Appangala (920 m above MSL) is located at Appangala, Kodagu District, Karnataka, on the Madikeri-Bhagamandala road, 8 km from Madikeri, in an area of 17.4 ha.

#### Mandate

- To extend services and technologies to conserve genetic resources of spices as well as soil, water and air of spices agroecosystems.
- To develop high yielding and high quality spice varieties and sustainable production and protection systems using traditional and non-traditional techniques and novel biotechnological

approaches.

- To develop post harvest technologies of spices with emphasis on product development and product diversification for domestic and export purposes.
- To act as a centre for training in research methodology and technology upgradation of spices and to coordinate national research projects.
- To monitor the adoption of new and existing technologies to make sure that research is targeted to the needs of the farming community.
- To serve as a national centre for storage, retrieval and dissemination of technological information on spices.

The spice crops on which research is being conducted at the institute include black pepper (*Piper nigrum*), cardamom (*Elettaria* cardamomum), ginger (*Zingiber officinale*), turmeric (*Curcuma longa*), cinnamon (*Cinnamomum verum*), cassia (*C. cassia*), clove (*Syzygium aromaticum*), nutmeg (*Myristica* fragrans), allspice (*Pimenta dioica*), Garcinia (*Garcinia gummi-gutta* and *G. indica*), vanilla (*Vanilla planifolia*) and paprika (*Capsicum* annuum).

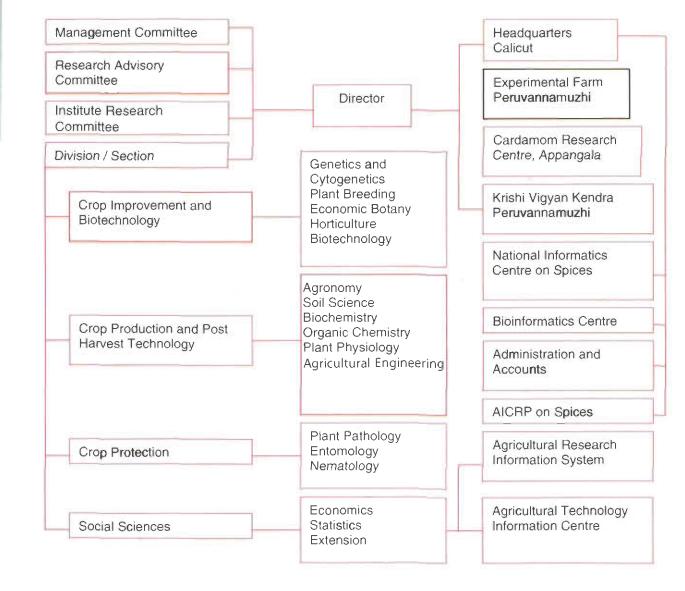
#### Organization

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The Director is the administrative head of the institute. The Institute Management Committee, Research Advisory Committee and Institute Research Committee assist the Director in matters relating to management and research activities of the institute.

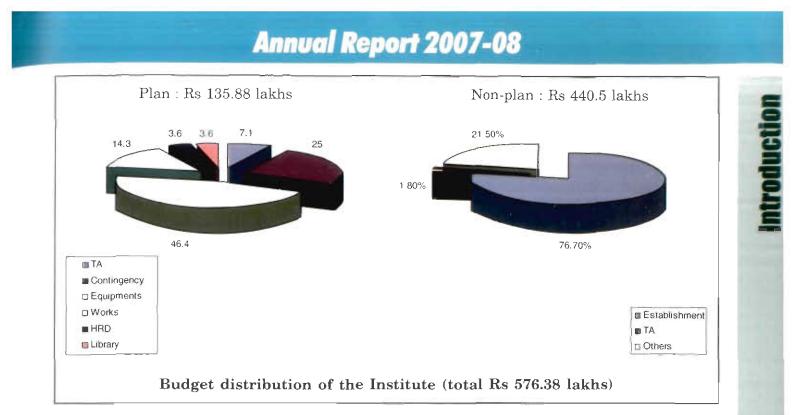
Research on various aspects of the mandate crops is conducted in three divisions, namely, Division of Crop Improvement and Biotechnology, Division of Crop Production and Post Harvest Technology and Division of Crop Protection and a Social Sciences Section. The other facilities available at the institute include Agricultural Technology Information Centre, Agricultural Research Information System, Bioinformatics Centre and Krishi Vigyan Kendra. The institute also functions as the headquarters of the All India Coordinated Research Project on Spices, and Indian Society for Spices. The institute has linkages with several universities, research institutes, and developmental agencies for collaborative research and developmental activities in spices.

**Budget:** The total budget of the institute was Rs. 576.38 lakhs during the year, which included Rs. 135.88 lakhs under Plan and Rs. 440.5 lakhs under Non Plan. In addition, Rs. 56.9 lakhs was also received as funds from other agencies.



**Organization of Indian Institute of Spices Research** 

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**Resource generation:** Institute earned a total of Rs. 2660141/- through sale of planting materials, biocontrol agents, training, publications and consultancy services.

**Staff:** The institute has a sanctioned strength of 43 scientific, 18 administrative, 36 technical and 61 supporting staff.

	In position				
Category	Sanctioned	Calicut (HQ)	Peruvannamuzhi	Appangala (Regional Center)	Vacant
Scientific	43	27	5	4	7
Technical	36	17	14	4	1
Administration	18	14	-	2	2
Supporting	61	25	13	18	5
Total	158	83	32	28	15

#### Staff position of KVK

		In position			Vacant
Category	Sanctioned	Calicut (HQ)	Peruvannamuzhi (Farm) (	Appangala Regional Center)	
Scientific	_	-		-	-
Technical	7	1	4	~	2
Administration	2	1	1	~	-
Supporting	2	-	2	-	-
Total	11	2	7	-	2

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#### New facilities

Introduction

Accreditation of Test Laboratory: The Institute test laboratories under the Division of Crop Protection and Division of Crop Improvement and Biotechnology have been accredited for virus indexing and quality testing of spices respectively for a period of two years by the Accreditation Unit of DBT established at Biotech Consortium India Limited, New Delhi as per the criteria laid down under "National Certification System for Tissue Culture Raised Plants (Registration No: TL200605; Accreditation No: TL2007001).

**Infrastructure:** New library building with 5000 sq ft. and biocontrol laboratory with 1700 sq ft. plinth area were established at Chelavoor campus. New office building with 2000 sq ft. plinth area was established at Peruvannamuzhi campus.



H.P. Singh DDG (Hort.) with Director, IISR and Peruvannamuzhi staff in front of the newly constructed office building at IISR Farm, Peruvannamuzhi after inaugurating it.
(Inset : H.P. Singh, DDG (Hort.) inaugurating the new office building at IISR Farm, Peruvannamuzhi)



Front view of the new biocontrol lab

## Annual Report 2007-08 Past Achievements

Black pepper: Germplasm collections obtained over the years through explorations are being maintained at IISR as well as in other alternate sites for developing improved varieties for yield, quality, abiotic and biotic stresses. The genetic stock has resulted in release of several improved varieties such as IISR-Sreekara, IISR-Subhakara, IISR-Panchami, IISR-Pournami, IISR-PLD-2, IISR-Thevam, IISR-Girimunda, IISR-Malabar Excel and IISR-Shakthi. Some of the unique accessions are registered with NBPGR at New Delhi. Putative transgenic black pepper plants with osmotin gene conferring resistance to drought and Phytophthora capsici has been developed. In vitro and in vivo propagation methods were standardized. Plantlets developed through micropropagation were established in farmers field in Kerala and Karnataka. Gene conferring resistance against Phytophthora capsici was isolated by targeted gene amplification using degenerate primers from Piper colubrinum. The spacing, nutrient and water requirements were standardized for different soil types of pepper growing regions. High production technologies and mixed cropping systems were developed for increasing productivity. Among different forms of potash, water-soluble and available K had significant positive correlation with berry yield, oleoresin and piperine. Organic production technology for black pepper has been standardized. Cost effective method for production of disease free rooted cuttings was developed. Mathematical models for optimum climatic factors for high production of black pepper have been developed.

Major pests, pathogens, viruses & their insect vectors and nematodes affecting pepper were characterized and documented. Morphological and molecular characterization of black pepper isolates of *Phytophthora* further revealed that isolates shared the characters of both *P. capsici* and *P. tropicalis*. A RNA virus, *Cucumber mosaic virus* (CMV) and a DNA virus, *Piper yellow mottle virus* (PYMoV) is associated with stunted disease of black pepper. A method for simultaneous isolation of RNA and DNA from infected black pepper plants and multiplex PCR for simultaneous detection of CMV and *Badna virus* in a single reaction was standardized. Phytoplasma with phyllody symptoms was most closely related to members of aster yellows group (16Sr I) of Phytoplasma.

Integrated strategies involving cultural methods, biocontrol agents, plant products and resistant varieties were developed for the management of pests and diseases including nematodes that resulted in substantial increase in yields and pesticide free produce. Largescale multiplication of biocontrol agents such as Trichoderma and Pseudomonas for distribution to farmers for management of disease was also undertaken. These organisms were deposited in the national repository of microorganisms at IMTECH, Chandigarh for future reference. Species-specific primers were developed for detection of R. similis in soil and plant samples. Black pepper accession HP-39 and Acc. 1090 were found to be resistant nematodes besides being rich in to caryophyllene. Endophytic bacteria found effective against Phytophthora capsici and R. similis in black pepper have been found. An integrated pest management schedule for management of root mealy bug has been developed. PCR based techniques were developed for identification of traded black

Past Achievements

# **Past Achievements**

## Annual Report 2007-08

pepper and to detect adulterants in commercial black pepper powder. Post harvest technologies for drying, processing, storage and production of value added product like white pepper production were standardized.

Cardamom: Four unique accessions were registered with NBPGR, New Delhi. IC numbers have been obtained for all the available 436 cardamom germplasm accessions. GC-MS study confirmed superiority of Indian cardamom over Guatemalan and Sri Lankan cardamom. Molecular profiling of Indian cardamom revealed the existence of two genetically distinct clusters such as "Kerala cluster" and "Karnataka cluster' among the germplasm collections. The improved varieties such as IISR-Vijetha, IISR-Avinash and IISR-Suvasini have been developed. Two of them have mosaic or rhizome rot resistance and have been popularized among the farmers. Coupled with production technologies, these varieties resulted in the productivity increase in cardamom. New high yielding varieties such as APG293, 398, 416 and 250 are found to be promising. Characterization of export grade cardamoms from India, Sri Lanka and Guatemala based on physical, biochemical parameters and molecular techniques revealed the superiority of Indian produce for the physical parameters such as seed to husk ratio, weight of 100 capsules, number of capsules in 100g, bulk density and moisture content. High production technology has been standardized. Drip irrigation and sprinkler irrigation once in 12 days recorded significantly higher number of tillers/clump, more number of leaves per tiller and more number of panicles per plant. Soil and water conservation measures have been standardized in cardamom based cropping system. Cardamom accessions APG 257, APG 414 and APG 434 were found to be promising for drought tolerance. High quality

(more than 40%  $\alpha$ - terpinyl acetate) cardamom such as NHY-14, MB-3, NHY-18 and OP-28 have been identified. The screening programme against leafspot and leaf blotch resulted in several moderately resistant types.

Ginger: Ginger germplasm repository at IISR is one of the largest collections with several exotic collections and high quality accessions. These accessions have been routinely used in the genetic improvement programme. An in vitro gene bank was established for conservation of germplasm. Three ginger varieties IISR Varada, IISR Rejatha and IISR Mahima were released for high yield and quality. Ginger oil components have been characterized by GC-MS. A relationship between leaf P/Zn ratio and soil P/Zn ratio to rhizome yield of ginger have been established. Post harvest technologies for processing and technologies for preparation of value added products such as salted ginger were standardized. Bacterial wilt pathogen, Ralstonia solanacearum in North Eastern states, Sikkim and Kerala were found similar in a molecular fingerprinting indicating strain migration from one place to another. Ginger strain of R. solanacearum was found to infect turmeric, cardamom, C. aromatica, C. zedoaria, Kaempferia galanga, Zingiber zerumbet and tomato. Indian Mango ginger Curcuma amada was found to be free from bacterial wilt even under inoculated conditions. The species of Pythium causing rhizome rot of ginger in Kerala, Karnataka, Uttar Pradesh and Sikkim was identified as P. myriotylum. Technique for ginger seed rhizomes treatment (for elimination of bacterial wilt pathogen) and integrated disease management strategy for soft rot & bacterial wilt diseases and shoot borer was developed. The improved varieties and technologies developed on cropping system, nutrient and

water requirement, pest and disease management and post harvest processing techniques were disseminated to farmers and other agencies through publication, training programmes and demonstrations. Large scale multiplication and distribution of elite planting material were also undertaken.

Turmeric: The germplasm collected over the years have been characterized for yield, quality, resistance to pests, diseases and drought. Molecular genetic fingerprints of sixteen Curcuma species using RAPD and Inter Simple Sequence Repeats (ISSR) technique revealed high degree of polymorphism among the accessions. Seven high curcumin and high yielding varieties. Suvarna, Sudarsana, Suguna, Prabha, Prathibha, IISR Alleppey Supreme and IISR Kedaram were released for commercial cultivation. Efficient protocol for plant regeneration through organogenesis and somatic embryogenesis was standardized. Variations in rhizome morphology were observed among calli-regenerated somaclones indicating somaclonal variation. Accessions with high curcumin and root knot nematode resistance were identified. Three different curcuminoids (curcumin, de methoxy curcumin and bis de methoxy curcumin) could be separated from oleoresin of turmeric rhizomes by employing chromatographic techniques. Turmeric oil components have been characterized by GC-MS. A PCR based method was developed to detect adulteration of turmeric powder with wild Curcuma species. The optimum spacing, nutrient and water requirement were standardized for different soils. Organic farming system was developed for turmeric. Basic data on distribution, bioecology, population dynamics of Conogethes punctiferalis & its natural enemies and crop loss due to shoot borer was generated. The improved varieties and technologies were

disseminated to farmers and other agencies through publications and demonstrations.

Tree spices: The germplasm holdings of important tree spices include nutmeg, clove, cinnamon including cassia, garcinia and allspice. IC Numbers for cinnamon, clove, nutmeg and allspice accessions were obtained from NBPGR, New Delhi. Cassia C1 (IC 370415) has been registered as INGR 05029 with NBPGR, New Delhi for its high oleoresin content (10.5%) besides a dwarf clove accessions. Two high quality cinnamon varieties, Navashree and Nithyashree and a nutmeg variety, Viswashree were released. Various improved lines with high yield and quality were developed that had a great impact in increasing the production and productivity of these crops in the country. Nutmeg accession A11/25 was found to be promising for high yield. Tissue culture protocols have been developed for nutmeg. Protocols for DNA isolation from nutmeg have been standardized. Performance of nutmeg on M. malabarica continued to be better than other rootstocks for productivity. GC-MS study revealed the presence of two chemotypes in Cinnamomum verum. Drying and processing methods for cinnamon, nutmeg and mace have been developed. Vegetative propagation techniques were standardized for nutmeg, cassia and cinnamon. Major pests and pathogens of tree spices were documented. The improved varieties and technologies developed on propagation and post harvest processing were disseminated to farming community.

Vanilla: Vanilla germplasm consisting of 93 accessions are being maintained in the repository. This included a flower colour variant collected from Andaman and Nicobar island. Comparative anatomical analysis of different vanilla species was carried out. Past **Achievements** 

**Past Achievements** 

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Interspecific hybridization was made between Vanilla planifolia and V. aphylla. Over 1000 seed progenies of V. planifolia are being fieldtested for yield and disease resistance. Protocols for micro propagation through direct shoot multiplication as well as callus regeneration were standardized. Root rot and wilting were found to be the major problems in most of the plantations. Root rot incidence ranged from 5 to 100%. Mosaic and necrosis were also observed in all the plantations and the incidence ranged from 2 to 80%. Cucumber mosaic virus (CMV) of vanilla was characterized on the basis of biological and coat protein (CP) nucleotide sequence properties, which showed that CMV infecting vanilla belongs to subgroup IB. A virus causing mild chlorotic mottle and streaks on leaves of vanilla was identified as a strain of Cymbidium mosaic

virus (CymMV) based on coat protein gene sequence comparison and phylogenetic studies. Similarly, a virus associated with necrosis and mosaic was identified as a strain of *Bean common mosaic virus* (BCMV) based on coat protein gene sequence studies.

**Paprika:** Several accessions including hybrids were added to the repository mainly through the network project on paprika. The germplasm was characterized for yield and quality such as oleoresin, pungency and colour value. Substantial amount of variability was observed in capsaicin content (pungency) of selected paprika accessions. Accessions with high yield and colour values were also identified. PCR based technique was developed to detect adulterants in commercial chilli powder.

# Black pepper

#### Genetic resources

Collection. maintenance and multiplication of germplasm : Twenty four accessions comprised of 22 Piper nigrum (wild), two accessions of *P. galeatum* and two local cultivars including an unique germplasm accession of Piper nigrum with ovate lanceolate leaves were collected from Bisle Ghats' section and farmers' field at Subramanya, Karnataka. In another exploration, P. nigrum (50). P. attenuatum (2), P. galeatum (3), P. trichostachyon (3) and P. hapnium (1) including an unusual type of P. nigrum (wild) with very dark purple colouration on the stem as well as on the leaf venation were collected from Shendurney WLS, Thenmala, Rose mala of Aryankavu range and Kattalappara of Kulathupuzha division. The forests surveyed were Kerodi, Kadra, Kaiga, Kadra- Ansi Road, Goabsitta. P. nigrum (wild)-25, P. attenuatum- 6, P. argyrophyllum-1 and three accessions of Piper species with camphylodromous venation were collected from Karwar district. Germplasm accessions were multiplied and maintained. A new germplasm block of 250 accessions for cultivated germplasm was established. Ninety

eight accessions were characterized morphologically based on IPGRI descriptor.

In addition to the fifty accessions established earlier, one hundred new accessions were planted at CPCRI, Kidu, Karnataka. IC numbers for 251 accessions were allotted recently by NBPGR, New Delhi in the new revised proforma.

**Application of GIS in black pepper:** Latitude and longitude of all the collection sites were recorded and plotted using GIS software. Biomass percentage and total phenol concentration of 148 accessions (100 from germplasm of Peruvannamuzhi farm and 48 recent collections) of *P. nigrum* collected from Karnataka and Kerala were studied. The results were plotted in SPSS software and similarity cluster was drawn using centroid method.

#### Breeding

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**For high yield and caryophyllene:** In the two different yield trials planted in 2002 and 2005, maximum fresh yield was recorded in OpKm (4.45 kg per vine), followed by HP 780 (3.6 kg)

lack Pepper

Table 1.1. Location effect on betacaryophyllene content

Location	β caryophyllene(%
Ambalavayal	12.6
Peruvannamuzhi	22.6
Panniyur	23.4
Yercaud	8.3
Dapoli	17.7

with 1.0 kg as mean yield per plot. A total of 114 progenies of crosses involving HSR Subhakara (female) and high caryophyllene lines (male) were produced. Analysis of berries of the elite lines (male) and HSR Subhakara at various locations (Table 1.1) indicated variation for caryophyllene content. However, relatively low level of caryophyllene was observed in the leaves.

For resistance to pollu beetle: A crossing block of bush pepper plants of HSR Subhakara (female parent) and pollu resistant lines (Coll. Nos 816, 841, 1114 and 1084) as male parents was established and hybridization was performed, which resulted in a total of 108 inter-cultivar hybrids for further evaluation.

For resistance to *Phytophthora*: Data on morphological characters like leaf shape and size, shoot tip colour, which are segregating in Panniyur 1 x IISR Subhakara mapping population were scored. The mapping population was screened with 5 RAPD primers and 2 ISSR primers. Twenty fresh crosses were made to develop a second mapping population between IISR Subhakara x IISR Shakthi. Fifty collections from germplasm and other progenies were collected and multiplied to develop a population for association mapping, field planting and screening to detect association between target characters. Planting of 30 more lines from mapping population 1 (1 each) was done to increase the population size. About 70 new plantlets were developed as 'transgenics' with 'osmotin' and twenty were hardened. Five putative transgenics containing osmotin were multiplied into 30 each for screening.

For resistance to *Radopholus similis*: Inter-cultivar hybridization was taken up between IISR Subhakara and Acc. 820 and HP 39. The seedling progenies raised were planted in polybags and maintained in the nursery for multiplication and planting in the field.

In DNA Profiling of resistant/suceptible parents, a total of 16 primers produced good reaction products in RAPD – PCR. Among various RAPD primers, OPA-08 (800bp), OPE – 18 (800, 250bp), S-32 (500bp), S-38 (1400bp) and among ISSR primers, ISSR 02C-16 (1000bp), and ISSR 15G/19 (1450bp) produced unique bands corresponding to either or both of the resistant cultivars. Amplification with another random primer HT/R2 resulted in a band at approximately 900 bp position to distinguish them from the susceptible cultivar, HSR Subhakara.

**For drought tolerance:** Inter-cultivar crossing and inter-specific hybridization using IISR Subhakara and drought tolerant lines and raising progenies have resulted in about 160 seedlings, which were raised in polybags for transfer to field and for further evaluation.

#### On-farm evaluation of tissue cultured plants of black pepper

Twenty five thousand tissue cultured plants (TCP) of black pepper were multiplied and fortified with *Trichoderma harzianum* and VAM while hardening and planted in 25 ha of area in farmers field in Kerala and Karnataka.

TCP had better root growth and establishment (86%) while, it was 70% for the conventional plants. Twenty percent of the TCP showed early flowering. In general, TCP showed better growth, early flowering and better yield compared to control. Field trials in farmers field helped to demonstrate the superiority of TC plants over conventionally propagated black pepper plants. DNA profiling of TCP demonstrated that the TC plants are genetically stable.

#### Nutrition and cropping system

The efficacy of organic amendments in nutrient release in soil and its crop uptake: The efficiency of organic amendments in reducing the P fixation capacity of Peruvannamuzhi soil series was studied under lab conditions. Addition of organic manures like FYM, vermicompost, leaf compost and neem cake increased the availability of organic and residual P fraction in pepper soil and the order of availability of different P fractions was residual/ organic P > Ca-bound P ~ reductant soluble P > Fe/Al P > loosely bound P. Amongthe sources, vermicompost and FYM applications recorded higher buildup of available P in surface and sub surface layers of Panniyur and Karimunda varieties of bush pepper which was on par with that of chemical fertilizers. Neem cake recorded the lowest P buildup. Available P content showed highly significant positive correlation with Al-P  $(0.97^{\ast\ast})$  followed by residual P  $(0.712^{\ast\ast})$  and Ca-P (0.649\*\*). Significantly higher yield was recorded in vermicompost application in both Panniyur and Karimunda varieties of bush pepper. Al-P showed significant positive correlation with yield  $(0.62^{**})$  and P uptake (0.45\*\*). Step wise forward regression analysis showed that Fe-P and available P as the best predictors for yield (= 46.25 + 1.034 Fe-P + 0.570 Av. P) with  $R^2$  value of  $0.611^{**}$ .

Sulphate of Potash (SOP) as potassium source on growth, yield and quality: The effect of muriate of potash (MOP) and SOP on the yield of pepper was analyzed by comparing the fresh yield of pepper. In bush pepper, the treatment receiving 50% recommended K as SOP recorded the highest yield of 295 g/pot followed by recommended K as MOP and 125% of recommended K as SOP. Under field condition, maximum yield (3.16 kg/vine) was obtained in recommended K as SOP + MgSO, which was on par with recommended dose of K as SOP and was significantly higher than control. Among different forms of K, water soluble and available K had significant positive correlation with berry yield, oleoresin and piperine contents. The nitrate reductase activity was highest in leaf and berry during berry maturity and harvesting stages in SOP + MgSO, application. Other biochemical parameters like carbohydrate, reducing sugars and starch in different plant parts of bush pepper like leaf, stem and berry did not show any definite trend with the application of sources and doses of potassium.

**Organic farming:** Black pepper was grown organically by applying FYM, vermicompost, ash and rock phosphate, azospirillum and phosphobacteria and *Trichoderma* and *Pseudomonas* sp. (IISR-6 & 853) as biocontrol agents for disease control. Highest average fresh yield of 2.06 kg vine<sup>-1</sup> was observed in integrated system which was on par with chemical system. The organic system recorded the lowest yield of 1.39 kg/vine. Mortality of black pepper vines due to disease incidence was high under organic system.

**Intercropping:** Cowpea, black gram, horse gram, amorphophallus, coleus, yam, turmeric and ginger were planted in replicated trial as intercrops. Medicinal plants such as

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**Black Peppe** 

Desmodium gangeticum, Pseudarthria viscida, Adathoda vasica, Plumbago rosea, Piper longum, Gymnema sylvestre, Hemidesmus indica and fodder crops such as congo signal grass, guinea grass, hybrid napier grass-CO3 were also planted. In the evaluation of legumes, the maximum shoot mass noted in Mimosa (25.81g/plant) whereas root mass was more in Crotalaria striata (11.3g/plant) after six months of growth. Nitrogen content was maximum in Clitoria spp (2.44 %).

Yield recorded for ginger, turmeric, coleus, amorphophallus and tapioca in 5 year old black pepper garden was 5kg/bed, 7kg/bed, 3kg/bed, 1kg/plant, and 1kg/plant respectively. In the case of vegetables, yield obtained was, amaranthus green 200g/plant, amaranthus red 80g/plant, Brinjal 100g/plant and chillies 50g/ plant under rain fed condition planted during January. Average yield recorded for black pepper at RARS Ambalavayal was 5kg/plant.

In juvenile black pepper garden, yield recorded was higher than that in 5 year old garden. Recorded yields in juvenile garden were 7kg/ bed, 8kg/bed, 4kg/bed, 1.5kg/plant, 1.5kg/plant and 1.7kg/plant respectively for ginger, turnieric, amorphophallus, tapioca, coleus and yam. In the case of vegetables, yield obtained was amaranthus green 500g/plant, amaranthus red 300g/plant, brinjal 750g/plant, chillies 200g/ plant and bhendi 150g/plant for the crop planted during December.

#### Physiological and biochemical basis of productivity

NR activity ( $\mu$  moles NO<sub>2</sub> g<sup>-1</sup> h<sup>-1</sup>) during preflowering period: High yielders maintained higher NR activity than that of low yielders during both preflowering and bearing (at harvest) periods. The mean activity of high yielders was 819.1 and 270.1 $\mu$  moles NO<sub>2</sub> g<sup>-1</sup> h<sup>-1</sup> while that of low yielders was 556.3 and 89.8  $\mu$  moles NO<sub>2</sub> g<sup>+</sup> h<sup>+</sup> during pre-flowering and bearing periods respectively.

**MDH activity (dA/min) during preflowering period:** MDH activity was also higher during pre-flowering period (April-May) compared to bearing period (Dec-Jan) in both high and low yielders. Though the mean activity was higher in high yielders (0.42 and 0.33) than that of low yielders (0.38 and 0.26), the difference was negligible and there was lot of inconsistency. Some of the high yielders exhibited very low activity while some low yielders exhibited very high activity. Hence, MDH activity may not be useful in distinguishing high and low yielders.

**Starch and carbohydrates:** Pre-flowering leaf and stem carbohydrates and starch content were analysed in both high and low yielders. Both leaf and stem carbohydrates and starch were higher in high yielders compared to low yielders indicating the usefulness of this trait in distinguishing high and low yielding black pepper accessions.

#### Mechanism of drought tolerance in black pepper

**Isozymes of SOD:** Native PAGE (10 % gel) was conducted to investigate if there exists variation in isozyme pattern between tolerant and susceptible accessions. Electrophoresis was conducted at a constant current of 45 mA for 3 hrs in a few selected accessions (20 tolerant and 20 susceptible accessions). Five major bands were obtained in all the samples (Em values: 0.19, 0.48, 0.53, 0.70, 0.74). An extra band with Em value of 0.6 was obtained in a few accessions under stress conditions. No significant change in banding pattern between tolerant and susceptible accessions could be seen.

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**Black Pepper** 

**Protein profile:** SDS PAGE (10 % gel, carried out at a constant current of 45 mA) of the samples yielded eight common bands and a maximum of 14 bands (Em values : 0.10, 0.14, 0.20, 0.27, 0.35, 0.58, 0.65, 0.78). However, tolerant and susceptible accessions could not be distinguished based on the electrophoretic pattern.

**Photosynthetic and gas exchange parameters:** Mean photosynthetic rate was higher in tolerant accessions compared to susceptible accessions under stress condition though the difference was very less under control condition. There was no significant difference in transpiration rate and stomatal conductance between tolerant and susceptible accessions both under control and stress conditions while leaf temperatures were slightly lower in susceptible accessions compared to tolerant accessions.

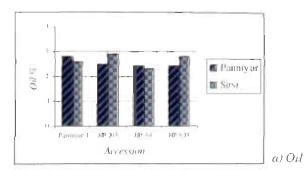
#### Quality

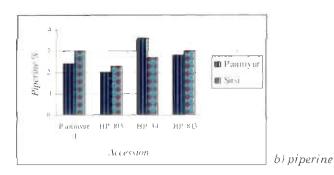
Controlled atmospheric storage and quality: Black pepper variety Panniyur-1 with a moisture content of 10.5% was stored for 120 days in three layered metallised polyester cover under vacuum, 100% N, and 90% N, + 10% O<sub>2</sub> atmosphere and the sample was compared with that of black pepper stored in ordinary gunny bag. Samples collected at 60 days interval were powdered and evaluated for oil, oleoresin and piperine. Oil content was 2.0%, oleoresin 6.7% and piperine 2.2%. Moisture content varied between 10-11%. Quality parameters were found to be on par with control values. The appearance of the sample was cleaner in metallised containers compared to normal gunny bag packing.

**Location and quality:** Chemical quality of cultivars and hybrids grown at Panniyur as well as at Sirsi was compared. Chemical

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quality of Panniyur-1 was comparable both at Sirsi and at PRS, Panniyur (Fig. 1.1).





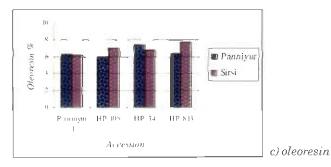


Fig. 1.1. Oil, piperine and oleoresin content of pepper varieties grown at Panniyur and Sirsi

#### Studies on market samples and post harvest technology

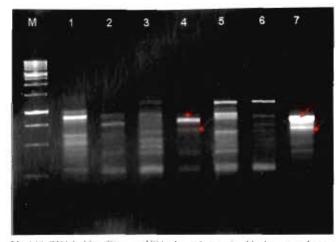
**Determination of purity of powdered market samples of black pepper:** A viable protocol for isolation of DNA (modified CTAB) from the recalcitrant materials was developed and amplified. Out of the five branded commercial samples of black pepper powder analyzed for detection of adulteration using RAPD/ISSR markers, market sample III amplified two papaya specific bands of approximately 830bp and 470bp with RAPD



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primer OPA 18 (5'-AGGTGACCGT- 3') (Fig. 1.2.). All the five market samples tested were free from *Piper attenuatum* and *Piper galeatum* specific bands.



M - 1 kb DNA indder (Biogene, USA), Lane 1- genuine black pepper, Lanes 2 to 6- market samples, Lane 7 - papaya seed. Arrows indicate the papaya specific band.

Fig. 1.2. RAPD profile of genuine black pepper, commercial samples of black pepper powder and papaya seed amplified using OPA 18.

GC MS analysis of essential oil of traded black pepper: GC MS profiling showed a total of 18 compounds in Indian, 16 in Vietnam, 17 in Indonesian and 15 in Malaysian peppers of which 14 are common for all the four produces. The main compounds identified were  $\alpha$ -thujene,  $\alpha$ -pinene, sabinene, myrcene,  $\alpha$ -phellandrene,  $\beta$ -caryophyllene,  $\beta$ -selinene, and  $\alpha$ -humulene.

Value addition in black pepper through microbiological process: The bacteria used as a principal active ingredient in the white production pepper process viz., Microbacterium barkeri **IISRWP25** (MTCC5404), Bacillus subtilis IISRWP33 (MTCC5405), Bacillus subtilis IISRWP34 (MTCC5406), Bacillus subtilis IISRWP38 (MTCC5407) and Bacillus licheniformis IISRWP43 (MTCC5408) were deposited in IMTECH, Chandigarh. Bacterial fermentation process for production of white pepper did not leave significant population of bacterial cells on the finished product (Fig. 1.3). The persistence of microorganisms ranged between  $1x \ 10^1$  to  $5 \ x \ 10^1$  cells per gram of white pepper which is equivalent to 0.5-2.5 cell per white pepper seed (1g of white pepper~20 white pepper seeds). A PCR based assay system was devised to detect the bacterium on the finished product which is based on specific nucleotide sequence isolated from the above said bacteria. Primers specific for M. barkeri HSR WP 25 (FP 5' GGT CTG TGG TGG GAA AGA T'TT3', RP 5'ATC CCA TGA GTT CCC ACC ATA ACG 3') produced an amplicon of size 940; specific primers for B. subtilis IISR WP 33 (FP 5' GGT GGC TTC GGC TAC CAC TTA CAG 3' RP 5' GGA TTG GCT TAA CCT CGC GGT TTC 3') produced an amplicon of size 1100; specific primers for B. subtilis IISR WP 34 (FP 5-ATA CAT GCA AGT CGT ATC GGA-3 RP 5-TTC GCT CCT CAG CGT CAG TTA-3) produced an amplicon of size 725; specific primers for B. subtilis HSR WP 38 (FP 5-TGC CTA TAC ATG CAA GTC GAG-3 RP 5-TTC AGA CCA GAG AGT CGC CTT-3) produced an amplicon of size 708 and specific primers for B. licheniformis HSR WP43 (FP 5' GTC CAC GCC GTA AAC GAT CGA GTG 3' RP 5'GTG GGA TTG GCT TAG CCT CGC GGC 3') produced an amplicon of size 480.

**Production of white pepper through fermentation technology:** The following are the salient achievements of the study:

• Eight of the 52 microorganisms isolated were found to decorticate 60-90% of black pepper into white pepper in 5 days. More than 95% conversion of green pepper to white pepper was achieved with eight bacteria within five days. The white berries were creamy in colour but sizeable quantities of wrinkled berries were found

**Black Pepper** 

36

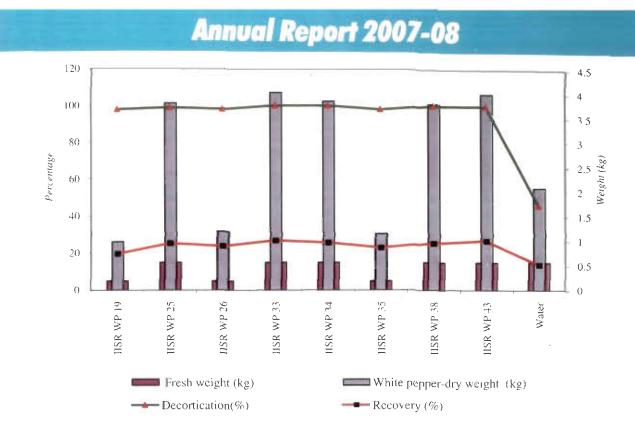


Fig. 1.3. Production of white pepper from green pepper by bacterial fermentation

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among the white pepper indicating their immaturity.

- Optimum concentration of microbes for conversion of green pepper to white pepper was found 10<sup>9</sup> cfu ml<sup>-1</sup>.
- Morphological and molecular profiling indicated that the isolates were of different species.
- Bulk conversion of green pepper in to white pepper using short listed microorganisms yielded 98 to 100% conversion with good quality such as colour and appearance.
- Quality evaluation of white pepper produced by microbial fermentation of matured green pepper reveals that there is no difference in the quality parameters of white pepper.
- The persistence of microorganisms in dried white pepper ranged between 10 to 50 cfu per gram of white pepper.
- Among the six varieties evaluated, Panniyur 1 was found to be most suitable followed by Karimunda and IISR Subhakara for white pepper production.

 Short listed eight bacteria were identified using 16s rDNA amplification and sequencing.

#### Phytopthora foot rot

**Characterization:** Forty five *Phytophthora* isolates collected from various hosts representing different geographical locations were added to the repository thus making the total collection to 447. These isolates were maintained at room temperature in agar slants overlaid with liquid paraffin and sterile distilled water kept in small screw cap bottles.

Colony morphology, growth rate on agar, caducity, mating type determination, pathogenicity test, *in-vitro* screening against the biocontrol agent and fungicide resistance were studied for different *Phytophthora* isolates in order to understand the intra specific and inter specific diversity. Carrot agar was used to study colony morphology, growth rate at different temperatures, sporangialcharacters and mating type determination. The



phenotypic characters of these isolates varied intra specifically and inter specifically. The LB ratio of the isolates ranged from 1.7- 2.7 and mean pedicel length ranged from 38.2-209mm. Mating type determination of *Phytophthora* isolates was done by the single unknown isolate method in which a pure culture of each isolate was paired with known A1 and A2 tester on carrot agar plates containing b-sitosterol (30mg/l) 30 mm from each other.

Pathogenicity test was conducted to study the intra specific variability of the isolates in their virulence. Stem inoculation technique was used for evaluating the aggressiveness of 78 isolates on a susceptible black pepper variety IISR Subhakara in green house condition. Disease severity was rated after 72h of incubation, as lesion length in millimeter and depth of penetration as index in a scale of 0-4. Based on the disease severity, the isolates were grouped into three, viz., less virulent, moderately virulent and highly virulent. Lesion length of the isolates ranged from 3.7 to 55.0mm and depth of penetration from 1 to 4. In vitro screening of black pepper isolates against the biocontrol agent, fluorescent pseudomonads, IISR6 indicated that there was no correlation of virulence variability of isolates with their reaction to biocontrol agents.

Molecular characterization of *Phytophthora* involves characterization of the species to reveal the intra and inter specific variations among the isolates. The ITS region of the isolates was amplified using the primers ITS 6 (5'-GAAGGTGAAGTCGTAACAAGG-3') and ITS 4 (5'-TCCTCCGCTTATTGATATG-3'). The PCR amplified a product of ~800bp to 1200bp, from the isolates as routinely observed in the amplification of ITS region using ITS 4 and 6 universal primers. The ITS amplicons were

digested with restriction enzymes to reveal the species identity and the intra and inter specific variations among the isolates. High levels of intraspecific and interspecific variability in the ITS regions were observed among all the isolates. RFLP fingerprints clearly showed the existence of four genetic groups on black pepper and supporting the existence of subgroups within P. capsici from black pepper. Genetic distances and cluster analysis revealed high level of intraspecific variation among these isolates. Results indicated that disease of black pepper is caused by numerous and distinct strains of P. capsici that are genetically variable. Variation occurs within and between different geographical locations. For the confirmation of the species identity, ITS amplicons were sequenced. The entire sequence of ITS 1, ITS 2 and 5.8S partial sequences of 28 S and 18S regions were obtained for P. capsici, P. palmivora and P. meadii. These sequences were deposited with Genbank (Accession Numbers: AM422703, AM422704, AM422705, EU515167, EU515168, EU515169, EU515170, EU515171, EU515172, EU515173, EU515174, EU515175 and EU515176).

**Cloning of** *Phytophthora* resistance and **defense genes from** *Piper colubrinum:* Ligation mediated walking on cDNAs from plants challenged with *Phytophthora* resulted in the amplification of fragement of size of approximately 400 which was cloned.

#### Host resistance

Forty one hybrids and six cultivars short-listed as moderately tolerant in the preliminary screening were subjected to root inoculation with *P. capsici*. The mortality of the plants ranged from 20-100% in 100 days. Hybrid HP 449 showed the least mortality (20%) followed by HP 1375 (30%) which indicated the

8

**Black Pepper** 

tolerance of the hybrids towards *P. capsici* infection. Six cultivars tested showed 100% mortality in 13-21 days of inoculation showing the highly susceptible nature.

Screening wild and related species of *Piper:* Three wild species viz., *P. ribusoids* (Acc. 5225), *P. sylvaticum* (Acc. 3177) and *P. sarmentosa* (Acc. 4381) were evaluated for *P. capsici* resistance. Of these, *P. sylvaticum* (Acc. 3177) showed resistance against the pathogen (Fig. 1.4).

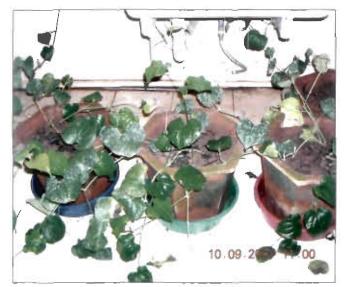


Fig. 1.4. Acc 3177 – a wild accession resistant to *P. capsici* 

**Selection from open pollinated seedlings:** Seedlings raised from 27 accessions were subjected to preliminary evaluation for *P. capsici* resistance. None of the seedlings survived infection.

#### Multiple resistance

Three open pollinated progenies viz. 04- P24 - 1, 04- HP 1533 -2 and 04-HP 1533 -3 that were short-listed from all the methods of screening during 2004-2006 were planted in a sick plot at IISR Farm, Peruvannamuzhi for evaluating their field reaction to *P. capsici* and nematodes. The progeny 04-P24-1 (Fig. 1.5) showed a



Fig. 1.5. Open pollinated progeny 04- P24 -1

survival rate of 77.78% and no *P. capsici* infection was noticed in these plants.

Screening of root stocks and their grafts against P. capsici and nematodes: Accessions, C 820, 1041 and, wild accessions 3177 (P. sylvaticum) and 5225 (P. ribusoids) were subjected to P. capsici and nematode (R. similis and M. incognita) inoculation prior to being used as root stock for grafting with IISR Sreekara. Of these, the Acc C 820 was found resistant to R. similis and susceptible to M. incognita while its reaction to P. capsici is under evaluation. The Acc. 1041 (IISR Thevam) was found susceptible for both the nematodes and P. capsici. The Acc. 3177 (P. sylvaticum) was found susceptible to both the nematodes while it was resistant to P. capsici. Acc. 5225 showed high susceptibility to R. similis but was resistant to M. incognita.

#### Disease management Biocontrol

**Black Pepper** 

**Evaluation of promising antagonists:** The promising isolates of fungal and bacterial antagonists viz. Pochonia chlamydosporia and Pseudomonas fluorescens (IISR-859, IISR-853) were evaluated under green house conditions in comparison with recommended fungicides viz. copper oxychloride (0.25%), potassium phosphonate (0.3%)) and biocontrol agents (T. harzianum + P. fluorescens) against P. capsici. The study showed that IISR-853 and IISR-859 were effective in checking P. capsici infection and was on par with copper oxychloride (Table 1.2).

Field evaluation of bioconsortia: A field trial to evaluate the effect of five different combinations of rhizobacteria, in the form of consortia, against foot rot and slow decline diseases in two varieties of black pepper, was conducted for five years from 2003-2007 in comparison with *T. harzianum* alone and copper oxychloride. The pooled data after five years indicated that the rhizobacterial consortium containing IISR-6, 8, 13, 51, 151, 853 (rhizobacteria) + PB-21C (P-solubilizer) was effective in reducing the disease incidence and

enhancing the survival of the plants in IISR-Subhakara (Table 1.3). Among the eight treatments, the mortality of vines was lower in this treatment. But no significant difference was observed among the treatments in the variety Panniyur 1.

characterization Isolation and of rhizobacteria : Fifty rhizobacterial strains were isolated from healthy black pepper vines from different geographical regions in Kerala and Karnataka. Five cultivars of black pepper were used for isolation of rhizobacteria. The samples were serially diluted upto 10<sup>-10</sup> and isolations were made using Tryptic Soy Agar (TSA), Nutrient Agar (NA) and basal medium amended (glucose, mannitol, sorbitol, inositol and sucrose). Maximum diversity and abundance of rhizobacteria were obtained in TSA followed by NA. The individual bacterial colonies were selected and subcultured on NA and cryopreserved at -80°C in 20% glycerol. Isolates were tentatively grouped based on phenotypic characteristics such as colour, form, elevation, margin, diameter, surface, opacity and texture. Motility, cell morphology, size, gram reaction and spore formation were also recorded using standard procedures.

#### Table 1.2. Evaluation of promising antagonists against P. capsici

Treatment	Mortality (%)	% control
Copper oxychloride, 0.25%	0.00	100.00
Potassium phosphonate, 0.3%	14.81	81.82
T. harzianum (P26), 50g/pot	44.44	45.46
P. chlamidosporia, 50g/pot	11.11	86.78
P. fluorescens IISR 853, 50g/pot	0.00	100.00
P. fluorescens IISR 859, 50g/pot	7.41	90.91
T. harzianum (P26) + P. fluorescens	18.52	77.27
(IISR 6), each @ 50g/pot		
Control (P. capsici alone)	81.48	-
LSD 0.05%	12.71	

Table 1.3. Evaluation of bioconsortia against foot rot and slow decline diseases

Treatment	Disease incidence					
AND REAL PROPERTY AND A	IISR-Subhkara	Panniyur1	Mean			
IISR 6, 8, 13, 51, 151, PB21CI	27.16*	41.87	34.51			
ISR 6, 8, 13, 51, 151, P1AR6	35.14	41.22	40.18			
IISR 6, 8, 13, 51, 151, 853	14.98	60.57	37.78			
IISR 6, 8, 13, 51, 151, 859	33.03	54.20	43.62			
IISR 6, 8, 13, 51, 151, 853, 859, PB21C, PIAR6	33.97	43.24	39.11			
T. harzianum	40.79	33.98	37.38			
Copper oxychloride	36.29	24.56	30.43			
Control	36.75	32.71	31.73			
Gen. Mean	32.39	42.05**	NS			
LSD 0.05	Variety x Treatment = 22.51					

8

Rhizobacterial isolates were screened using dual plate culture technique against *Phytophthora, Pythium* and *Fusarium*. Isolates showing more than 50% inhibition were shortlisted (Table 1.4). Five isolates inhibited all the three pathogens. These isolates were also tested for their ability to promote plant growth, production of indole acetic acid, HCN, nitrogen fixation, and phosphate solubilization.

#### **Bacterial endophytes**

**Identification:** Promising endophytic bacteria against foot rot were identified as *Pseudomonas aeruginosa* IISR BP35 (MTCC 5410), *P. putida* IISR BP25 (MTCC 5411), *Bacillus megaterium* IISR BP17 (MTCC 5412) based on 16s rDNA analysis. The percentage nucleotide identity was more than 99% for both the species. *Pseudomonas aeruginosa* IISR BP35 (MTCC 5410) is deposited under Budapest Treaty at IMTECH, Chandigarh.

Field trials: A field trial is in progress on the performance of endophytic bacteria for foot rot

management. The experiment consists of seven treatments such as P. aeruginosa IISR BP35, P. putida IISR BP25, B. megaterium IISR BP17, Curtobacterium luteum IISR TC10, metalaxyl-mz (1.25g/l), phorate 10g/vine and a check. Three varieties (IISR-Thevam, OPKM and IISR-Sreekara) are being evaluated. The bacterized rooted cuttings were planted and observations on establishment and disease incidence are being monitored at bimonthly intervals. The initial data revealed that the bacterial endophytes could enhance the establishment of the vines. The percentage establishment recorded for the endophytic bacteria are: B. megaterium IISR BP17 (82%), P. putida IISR BP25 (72%), P. aeruginosa IISR BP35 (82%) and C. luteum IISR TC10 (88%). The chemical check recorded 85% and the untreated check recorded 78% establishment. The trial is in progress for observations on disease incidence and yield.

Endophytic colonization of *P. aeruginosa* IISR BP35 in cut shoots and rooted cutting: A green house trial is in progress on the

Ð	lis
	Isolate Phylo
5	BRB-5
	BRB-21
-	BRB-24
	BRB-28
	BRB-35
	BRB-49
	BRB-50
	BRB-7

**BRB-13** 

**BRB-15** 

**BRB-18** 

**BRB-19** 

BRB-22

**BRB-34** 

**BRB-36** 

**BRB-39** 

**BRB-41** 

# Annual Report 2007-08

Table 1.4. Antagonistic activity of short-listed rhizobacterial isolates

phthora

50.73

74.80

83.67

84.40

83.67

81.33

70.73

5.60

51.87

22.20

83.67

60.00

76.30

50.00

50.00

51.47

51.47

aeruginosa IISR BP35 for P.

performance of endophytic bacteria P.

management. When evaluated in the green house

for disease management, the isolates endophytic

Pathogen suppression (%)

Fusarium

84.40

75.57

80.37

71.83

80.73

42.20

51.10

57.43

42.57

50.00

10.73

17.43

5.20

17.80

5.57

36.70

30.73

capsici

Pythium

75.17

71.83

69.63

67.43

71.70

78.90

0.00

11.10

11.10

34.07

0.00

0.00

0.00

0.00

0.00

0.00

0.00

found to be  $10^{12}$ - $10^{13}$  cells ml<sup>-1</sup>. At this concentration the endogenous population of the bacterium was  $10^5$ - $10^6$  cells g<sup>-1</sup> of tissue. Duration of bacterization vis-à-vis endogenous population of bacteria and suppression of *P*. *capsici* was optimized. The minimum duration of bacterization for suppression of lesion on shoot was 15-20 min. At this duration endogenous population of *P*. *aeruginosa* was  $10^6$  cells g<sup>-1</sup> of tissue with significant lesion inhibition (96-100%) on the excised shoots.

Ascending movement of *Pseudomonas aeruginosa* IISR BP 35 in rooted cuttings of black pepper was confirmed by plate assay and PCR assay using specific primers. The population size of the bacterium was more in root (5-6 log units  $g^{-1}$  of tissue when compared to stem (4-5 log cfu  $g^{-1}$  of tissue) and leaf (3-4 log cfu  $g^{-1}$  of tissue). Duration of bacterization positively influenced the endogenous population size (Figs. 1.6 & 1.7).

Mode of action of P. aeruginosa: P. aeruginosa was found to secrete a biosurfactant belong to cyclic lipopeptide in Kings B and Pseudomonas agar medium (88µg ml<sup>-1</sup>). The cyclic lipopeptide showed biocidal activity zoospores and mycelium against of Phytophthora capsici in vitro. The critical concentration for the inhibitory activity was 500µg ml<sup>-1</sup>. The biosurfactant inhibited the lesion development on stem cutting of black pepper cultivar IISR Panchami (Fig. 1.8). RP-HPLC analysis of the biosurfactant established the identity of the compound as Massetolide A.

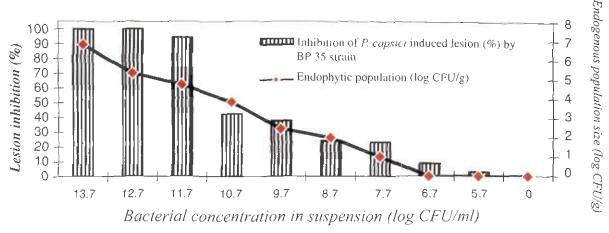
#### Anthracnose

**Characterization of** *Colletotrichum* **spp. infecting black pepper and other spices:** Thirty six isolates of *Colletotrichum gloeosporioides* and four isolates of *C. capsici* 

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BP35, BP25 and BP17 recorded over 70% disease suppression. Though performed well in Karimunda and Panniyur-1 varieties of black pepper, disease suppression was marginally better in Panniyur-1 (80%) when compared to Karimunda (60-70%). The endophytes could offer protection even with the pathogen population size of 6-7 log cfu g<sup>-1</sup> of soil which showed the role of endogenous population size for protecting the plants. The endogenous population and spatiotemporal colonization of the bacterium in bacterized shoot was analysed. Minimal concentration of bacterium required for suppression of *P. capsici* was



Endophytic colonization: y = 0.7968x+7.0013 (R<sup>2</sup>=0.9576) Lesion inhibition: y = -12.596x+112.55 (R<sup>2</sup>=0.9002)

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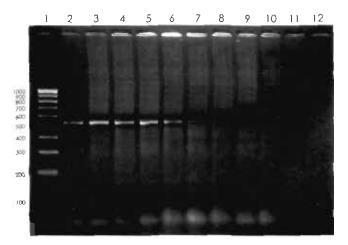
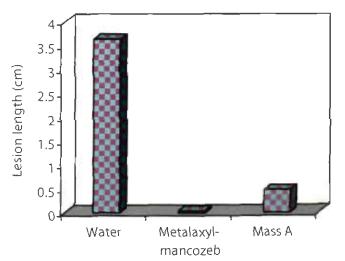


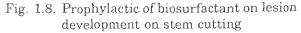
Fig. 1.7. PCR based optimization of concentration of bacterial cells for endophytic colonization of *Pseudomonas aeruginosa* 

were obtained from the infected specimens of black pepper, cardamom, turmeric and other crops of cardamom ecosystem collected from Kodagu, Hassan and Chickmagalur districts of Karnataka and Valparai (Tamil Nadu). C. capsici was isolated from turmeric, chilli, bird's eye chilli and bell pepper. All isolates from cardamom and black pepper were identified as C. gloeosporioides. Considerable diversity was observed in C. gloeosporioides isolates with regard to colony (Fig. 1.9a), conidial (Fig. 1.9b) and appressoiral characteristics (Fig. 1.9c&d). The study of 48 isolates comprising 13 isolates from black pepper, 10 isolates from cardamom and 25 isolates from other hosts, revealed wide variation in appressoiral character which ranged from simple to 8-lobed structure (Table 1.5). The colonies of different isolates appeared light grey, white and dark in purified culture on potato dextrose agar medium. There was considerable variation in the size and shape of conidía.

#### Pathogenicity

Pathogenicity trials were conducted under controlled conditions with six predominant





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Fig. 1.6. Density dependent endophytic movement by *Pseudomonas aeruginosa* in stem cutting of black pepper vis-à-vis lesion inhibition by *P. capsici*.

 Table 1.5. Colony, conidial and appressorial characteristics of black pepper isolates

 of Colletotrichum gloeosporioides

		Co	nidial o			
Place of collection	Colony colour	Length (mm)	Width (mm)	Shape	Appressoria character	
Nelliahu- dikeri	Grey, with orange slime, reverse side black	12.18	4.12	Cylindrical with obtuse ends	Multi lobed (2-3 lobes)	
Magalu	White and dark grey mycelium	17.81	4.12	Cylindrical with slightly tapered base and obtuse apex	Unlobed	
Ave Maria	White and light grey with orange slime. Reverse side orange centre region surrounded by black region	14.75	3.6	Cylindrical	Multi lobed (3-4 lobes)	
Appangala	Dark grey puffy mycelium, reverse side black and dark grey	17.6	3.75	Cylindrical with obtuse ends	Multi lobed (2-3 lobes)	
Hakathur	Dark grey mycelia with a narrow white margin, reverse side black and dark grey	14.6	4.5	Cylindrical with slightly tapered base and obtuse apex		
Cooverkolly	White mycelium with orange slime, reverse side creamy with light grey spots	10.31	3.75	Cylindrical with slightly tapered base and obtuse apex		
Kodlipet	White and light grey	10.68	3	Cylindrical with obtuse ends	Multi lobed (3-4 lobes)	
Haleri Grey with a narrow creamy margin, reverse side black		14.12	3.75	Cylindrical with a slightly tapered base and obtuse apex	Multi lobed (2-3 lobes)	
Byakar- avally	Dark grey, reverse side black	17.81	3.93	Cylindrical with Obtuse ends	Multi lobed (3-4 lobes)	
Sakleshpur	White and orange	17.43	3.75	Cylindrical with obtuse ends	Slightly lobed	
Thithimathi	Grey, reverse side	15.75	4.13	Cylindrical with obtuse ends	Multi lobed (2-3 lobes)	

44

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**Black Pepper** 

	Annual	меры	1 200		
Pollibetta	Light grey, reverse side orange	15.37	4	Cylindrical with obtuse ends	Slightly lobed
Chettalli	White, grey and orange, reverse side grey	15	3.75	Cylindrical with slightly tapered base and obtuse apex	Slightly lobed

isolates of C. gloeosporioides from black pepper and cardamom and C. capsici from turmeric. A set of susceptible varieties of black pepper, cardamom and turmeric were used for pathogenicity and cross inoculation studies. All the seven test isolates infected black pepper whereas, cardamom and turmeric were selectively infective. Out of four C. gloeosporioides black pepper isolates tested, three infected turmeric.

Monitoring of anthracnose disease of black pepper and leaf blight in three locations indicated the rapid spread of disease during early monsoon period (June). Exposure of black pepper rooted cuttings in polybags to rain splash with 8 month old crop residue indicated that the pathogen survives in soil and spreads through splash during early monsoon period.

#### Stunted disease

Indexing through PCR and establishment of virus-free mother vines: PCR based method was used in indexing 702 plants representing 14 popular varieties of black pepper showing no visible external symptoms (apparently healthy) for Piper yellow mottle virus (PYMoV). Total DNA isolated from 50 mg leaf tissue (from first fully opened leaf) was used as template in the PCR. Primers designed for the amplification of either 450 bp region of ORF I or 400 bp region of ORF III were used. PCR was performed using two template volumes (0.5 and 1.0 µl). A plant was declared infected if it showed expected size amplicon in the PCR using either 0.5 or/and 1.0 µl template (Fig. 1.10). Results showed that of these, 567 plants were positive (81%) for the virus. The per cent infected plants ranged from 59-100% in different varieties. Many of the indexed PCR positive plants exhibited visible symptoms such as mild chlorosis, yellow specs and mottling within 1-3 months. Results showed that indexing by PCR successfully detected PYMoV in infected plants showing no visible symptoms and hence can be used in nursery certification programme. The identified virus-free plants of all released



a. Colletotrichum colony



b. Colletotrichum conidia



c. Colletotrichum bilobed appressorium.

Fig. 1.9. Diversity in C. gloeosporioides isolates



d. Colletotrichumun lobed appressorium

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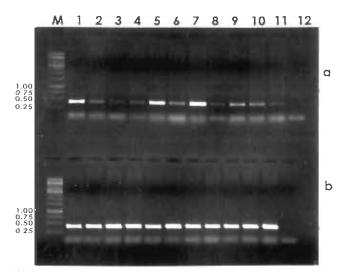
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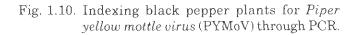
# **Black Pepper**

# Annual Report 2007-08

varieties served as mother vines for further propagation and distribution.



Lane M 1 kb ladder. Lane 1 Positive control (known infected plant). Lanes 2-11: Test plants of var. HSR Sreekara, Lane 12 Negative control (known healthy black pepper plant) (a): PCR performed with 1.0 µl template. (b): PCR performed with 0.5 µl template



Preparation of Piper yellow mottle virus (PYMoV) ORF III construct in sense and antisense orientation in a binary vector: A 600 bp region corresponding to open reading frame (ORF) III of PYMoV infecting black pepper was amplified through PCR using badnavirus specific primers and total DNA isolated from infected black pepper as template. The amplicon was cloned in a TA vector and its nucleotide sequence determined. From this, a 409 bp region was subcloned at Bam H1 site in the binary vector, pBI 121. Plasmid DNA isolated from positive recombinants were then subjected to PCR analysis to identify PYMoV sense and antisense oriented clones. Each of the identified sense and antisense clones were then mobilized into A. tumefaciens strain EHA 105 by triparental mating with the help of helper plasmid pRK 2014. The presence of recombinant pBI 121 with PYMoV ORF III insert was further confirmed by isolating plasmid from *A. tumefaciens* and subjecting them to restriction analysis and PCR. The sense and antisense constructs were designated as pBI 121-PYMoVS and pBI 121-PYMoVAS respectively.

Transformation of black pepper explants with Cucumber mosaic virus (CMV) coat protein (CP) gene construct: Explants harvested from fully grown in vitro plants of Panniyur I and IISR Subhakara were used in transformation. Explants (leaf, internode and petiole) were treated in Agrobacterium harbouring CMV CP construct (pBI 121-CMVAS) for 15 min, blot dried and kept in basal MS medium at 28°C for 48 h. Explants were washed in cephataxime (50  $\mu$ g ml<sup>-1</sup>) and kept in basal MS containing cephataxime for 2-4 days to kill Agrobacterium. Then, the explants were transferred to regeneration medium (SH and MS) containing different concentrations of BAP, NAA and TDZ along with selective marker, kanamycin. Among the two media, SH with BAP 3mg l<sup>-1</sup> and NAA 1mg l<sup>-1</sup> gave better response.

#### Nematodes

#### Molecular characterization

Four populations of *Radopholus similis*, collected from banana (Kayamkulam, Ernakulam and Kasaragod) and coconut (Kayamkulam), were characterized through ITS-PCR using speciesspecific primers. Extraction of nematode genomic DNA from formalin fixed specimens preserved for more than 10 years was standardized (Fig. 1.11).

#### Host resistance

Screening of root stocks and their grafts against nematodes: Seven germplasm accessions, to be used as root stocks, were screened against *Meloidogyne incognita* and *Radopholus similis*. Acc. 692 and Acc. 3177

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Line 1:100 bp marker, Lane 2, 4 & 5: Samples from Wayanad, Lane 3 & 6: Sumples from Peruvannamuzhi, Calicut, Lane 7: Negative control and Lane 8: Positive control

Fig. 1.11. Amplification of rDNA from formalin fixed nematode samples collected during 2000-01.

were susceptible to both R. similis and M. incognita while Acc. 3357 and Acc. 5525 showed high susceptibility to R. similis alone. Six OP progenies of the polyploid were screened against nematodes, out of which C5/12 was resistant to R. similis. The material is being multiplied for confirming the results obtained. Field testing of nematode resistant black pepper lines: The ongoing field evaluation using nine short-listed black pepper accessions was continued for the fifth year. Out of the nine accessions only one accession, HP 39, continued to show resistance to *R. similis* (Table 1.6). Similarly Acc. 1090 too persisted against *M. incognita* infection even after four years of planting.

#### Biocontrol

Annual Report 2007-08

**Studies on Bacillus megaterium:** Two strains of *B. megaterium* (BP17 and IISR 522) having nematicidal properties were studied for the pH and temperature requirements for their optimum growth. It was found that the optimum pH ranged between 5 to 9 for both BP17 and IISR 522. BP 17 had a better multiplication rate across a pH range of 5 to 9. Highly acidic or alkaline pH was detrimental to both bacterial strains. Similarly, the optimum temperature for both strains was 25° C. In general BP 17 had a better multiplication rate than IISR 522.

Accession		Nematode le	evel / g root	
	R. similis	Incidence	M. incognita	Incidence
HP 39	0	0/4	0	0/4
HP 60	68.75	1/4	0	0/4
HP 290	200	2/4	1393.7	2/4
C 812	0	0/4	1687.5	1/4
C 820	62.5	1/4	31.2	1/4
C 1047	12.5	1/4	0	0/4
C 1090	. 0	0/4	0	0/4
C 1204	56.2	1/4	0	0/4
C 4103	62.5	2/4	1031.2	2/4
Panniyur I	43.7	2/4	237.5	1/4
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#### Table 1.6. Incidence of plant parasitic nematodes in short listed accessions

**Black Pepper** 

Culture filtrates of both isolates were toxic to R. similis and caused >60% mortality. However, BP17 was slightly more efficient when compared with IISR 522. Similarly, the volatile metabolites produced by both isolates too were toxic to R. similis and the mortality was significantly high in the case of IISR 522. Both the isolates moderately caused repellence of R. similis.

BP17 and IISR 522 have the ability to decrease the length of primary roots of tomato plants. There was significant difference in root hair production and root architecture of tomato plants indicating the crucial role of endophytes in growth promotion.

#### Pest management

Pollu beetle (Lanka ramakrishnai)

**Field screening of germplasm:** Sixty-one black pepper hybrids and 57 cultivars were field screened against pollu beetle at IISR Farm, Peruvannamuzhi. Among the hybrids, the maximum incidence of 33.33% was recorded on Acc. 946; Acc. 1357 remained free from pollu beetle attack. Among the cultivars, Acc. 1449 recorded the highest incidence (34.4%). Acc. 1472, 4052, 4093 and 4095 were free from pest attack.

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**Black Pepper** 



#### Genetic resources

Collection, characterization and conservation: Six new landraces namely Pulikal Elam-1, Pulikal Elam-2, Madhunivas, Cheruparambil, Panikulangara and PNS-Vaigai were added to the germplasm. At present ex-situ gene bank consists 442 collections; hybrids and disease resistant selections which have been maintained in field germplasm repository. Fifty accessions characterized for morphological and yield characters based on IPGRI descriptor revealed wide variability for capsule wet weight (71.84 %) followed by number of panicles (56.69 %). Three accessions viz., IC 547146, IC 547161 and IC 349631 for yield attributing characters and two accessions viz., IC 349633 and IC 349627 for boldness are shortlisted.

**Characterization of germplasm for biotic stress:** Of the 58 accessions screened against natural incidence of leaf blight, none of the accessions exhibited highly resistant reaction. However, one accession (IC 349646) and *Amomum subulatum* were found to be resistant to leaf blight infection.

#### **Crop** improvement

Large numbers of hybrids (high yielding x disease resistant selection) have been evaluated under various comparative yield trials (CYT) and preliminary yield evaluation trial (PET) for yield and disease resistance, to exploit the heterosis for yield and disease resistance.

**CYT III** (2005-07): The mean yield performances of Malabar accessions (MA 28 and MA 29) were on par with checks in the trial laid out during 2003.

**CYT IV** (2005-07): In vazhukka types, among the farmers varieties, maximum yield was recorded in Palakuzhi selection with mean dry yield of 899.17 kg ha<sup>-1</sup>, while AMB 2 and VA 1 recorded the highest yield of 1140 and 1100 kg ha<sup>-1</sup>, respectively, among the other promising selections (Table 2.1).

**PET I:** Among the hybrids, highest yield per hectare was recorded in the hybrid combinations, NKE 12 x MB 5 and MB 5 x NKE 19 with 1499 and 1461 kg ha<sup>-1</sup>, respectively in the trial laid out during 2004.

**PET II:** Among the crosses, NKE 12 (mosaic resistant selection)  $\mathbf{x}$  GG (Green gold) recorded significantly higher yield of 1746 kg ha<sup>+</sup> in the trial laid out during 2005.

**Somaclonal variation:** A simple technology for micropropagation using petri plate system was developed without the use of growth regulators.

**Evaluation of bold capsule types :** Five bold capsule plants were selected from farmers field and also from NATP trials. The seedlings were raised in polybags and planted in replicated trial in the field. Growth and yield parameters

were recorded and two plants were selected (Fig. 2.1) based on growth and yield parameters with 80 % bold capsules



Fig. 2.1. APG 498, an accession with 80% bold capsules

Table 2.1. Comparative performance of farmers' varieties

Entry		Dry yield per he	ctare (kg)	
	2005	2006	2007	Mean
VA 1	1487.68	872.2	941.1	1100.33
VA 8	1013.46	769.64	484.6	755.9
NHY 10	1283.59	525.78	744.48	851.28
NHY 24	652.91	388.81	531.34	524.35
CP 3	1029.99	535.58	366.5	644.02
CP 10	1250.79	472.37	491.74	738.3
CP 11	1206.48	357.44	480.61	681.51
CP 12	948.75	441	566.7	652.15
AMB 2	1770.75	823.94	826.94	1140.54
CP 4	1152.58	603.14	583.82	779.85
IISR Suvasini	1085.84	610.62	767.37	821.28
Green gold	1076.11	649.62	826.47	850.73
Vander cardamom	1165.69	492.13	530.17	729.33
Palakuzhi selection	1500.95	671.05	525.5	899.17
Mean	1187.54	586.67	619.1	
SEd	438.54	89.5	63.31	
CV (%)	46.71	. 21.14	18.05	
CD (p=0.05)	901.44	149.54	130.14	

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Cardamom accessions with high quality Cardamom accessions were analysed for oil content and the percentage of 1, 8 cineole and  $\alpha$ - terpinyl acetate in oil. Accessions with high oil of 6.0% (including husk) are Acc. NHY- 3, 14, 15, 35, RR-1 x MB-3, VA-1, AMB-2, MA-7 and CCS-1(OP). Accessions with high terpinyl acetate of above 45% are Acc. RR1 x OP, RR-1 (self), CCS-1 (self), RR-1 x CCS-1 and VA-1. High 1,8 cineole accessions are CP-10, MA-7, NHY-3, CCS-1 x 893, RR-1 x GG and RR-1 x 893.

#### Drought tolerance

Enzyme activity in selected drought tolerant and susceptible genotypes: Enzymes such as catalase, peroxidase and super oxide dismutase were assayed in control and moisture stress treatments. There was no consistent pattern for catalase and peroxidase activities between tolerant and susceptible genotypes during stress. Super oxide dismutase activity decreased under stress. Susceptible genotypes had more decrease than the tolerant ones. CL-893 recommended for registration recorded increased activity of catalase, peroxidase and super oxide dismutase during stress compared to other genotypes.

**Photosynthesis and gas exchange parameters:** Photosynthetic rate, stomatal conductance and transpiration rate reduced under water stress. Tolerant and susceptible genotypes had similar values for these parameters (Table 2.2).

**Evaluation of four cross combinations for drought tolerance in field:** Three genotypes (RR1, CL-893 and Green gold) relatively tolerant to moisture stress and CCS -1, a susceptible genotype were crossed in an attempt to develop drought tolerant variety with good yield and quality characters. Crosses

Genotype	i lei ri	C	ontrol		A AL	Stre	SS	
	Е	g,	A	C,	E	g,	А	C
Tolerant								
CL-893	0.45	0.14	1.64	232.2	0.31	0.07	1.08	338.6
Green gold	0.33	0.08	2.45	279.5	0.36	0.07	0.82	279.6
LR 1	0.27	0.07	2.26	293.6	0.12	0.02	1.26	259.6
APG 149	0.34	0.09	1.46	325.6	0.2	0.05	1.845	280.6
Susceptible								
CCS1	0.54	0.19	5.9	257.2	0.45	0.16	1.31	269.3
M1	0.4	0.11	3.14	275.4	0.15	0.03	0.3	280.6
NKE 19	0.37	0.09	1.83	265.4	0.3	0.07	1.09	329.6

#### Table 2.2. Photosynthesis and gas exchange parameters

A= Photosynthetic rate ( $\mu$  moles), E = Transpiration (m moles),  $g_s$  = Stomatal conductance (cm s<sup>-1</sup>), Ci = Internal CO<sub>2</sub> concentration (ppm)

Cardamom

Cardamom

were evaluated for growth and yield parameters after withholding two irrigations compared to control. Growth parameters such as plant height, number of tillers per clump, yield parameters such as number of panicles per clump, panicle length (cm), no of capsules per panicle, number of capsules per clump and dry capsule yield (kg/ha) were recorded (Table 2.3). CL-893 and its cross combinations recorded better growth and yield characters. CCS1 x RR1 recorded maximum yield (790.2kg/ ha) followed by CCS-1 x 893 (699.4kg/ha). RR1 x 893 was found superior for both quality and yield (4200 green capsules/ plant) and one of the selections in Njallanil Gold x 893 for compound panicles and green bold capsules (Fig. 2.2).

Leaf rolling test: Fourth and fifth cardamom leaves from about twenty crosses were collected and exposed to open sunlight on concrete ground to find out time taken by different genotypes for leaf folding. Genotypes 893 OP, RR1 X 893, 893 x GG, CCS1 x RR1, CCS 1 x 893 and CCS1 x GG took longer time to fold than other genotypes, indicating their drought tolerance ability.

#### Diseases

#### Characterization of Colletotrichum spp.

The study of ten isolates of *Colletotrichum* gloeosporioides obtained from cardamom and other isolates obtained from other crops of cardamom ecosystem collected from Kodagu, Hassan and Chickmagalur districts of Karnataka and Valpari (Tamil Nadu) showed wide variation in appressorial character, colony morphology such as shape, colour and size (Table 2.4).

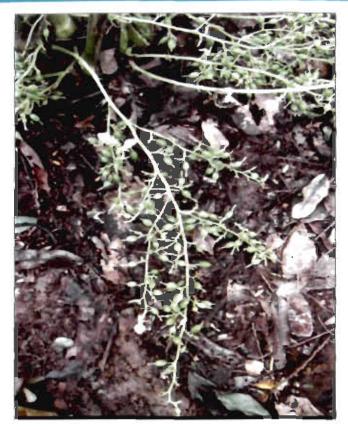


Fig. 2.2. GG x 893, hybrid having compound panicles

#### Development of consortium of bioinoculants for management of C. gloeosporioides

Bioassays with two each of promising fungal and bacterial antagonists viz., Trichoderma harzianum, T. hamatum and Pseudomonas fluorescens (IISR-6 and IISR-853) indicated that, the C. gloeosporioides isolates were sensitive to all four-test antagonists. The isolates differed in their interaction with antagonists. Marked differences were observed in their inhibition and sensitivity towards antagonists. Among the four antagonists tested, T. hamatum was more effective in suppressing the growth of all 14 predominant pathogenic isolates (Fig. 2.3).

### Table 2.3. Growth and yield parameters of selected cross combinations

Sl. No.	Genotype/ Crosses	Plant height (cm)	Tillers/ clump	Panicles/ clump	length	Capsu- les per panicle	Oil content (%)	Capsules per clump	Capsule dry yield (kg/ha)	Specific leaf weight (mg/cm <sup>2</sup> )
1	RR1 (OP)	265.2	28.9	40.0	58.2	51.4	5.0	1366.3	576.8	5.31
2	893 (OP)	268.8	25.8	46.6	67.3	46.7	5.0	1427.3	663.8	5.03
3	GG (OP)	255.6	23.5	27.4	45.2	48.2	6.0	905.6	397.4	5.44
4	CCS 1 (OP)	235.4	31.6	48.4	53.6	40.7	6.5	1472.7	679.9	4.70
5	RR1 (self)	275.0	32.1	31.8	56.3	48	6.0	1437.3	632.3	5.24
6	893 (self)	278.3	23.5	39.5	67.8	37.9	5.0	1111.3	452.8	4.69
7	GG (self)	255.9	28.5	29.8	45.4	57.5	5.0	1327.2	565.6	5.44
8	CCS1 (self)	221.1	25.3	44.9	48.4	36.6	5.5	1405.4	612.8	4.28
9	RR1 x 893	244.2	28.9	31.1	47.7	50.4	4.5	1454.8	647.2	4.79
10	RR1 x GG	281.6	27.8	40.7	51.3	49.4	5.5	1522.8	673.4	4.83
11	RR1 x CCS1	280.0	28.4	30.0	57.6	43.3	4.25	1357	604.7	5.50
12	893 x RR1	254.6	26.2	40.5	51.7	41.6	5.0	1011.7	451.9	5.42
13	893 x GG	253.2	27.4	44.2	56.8	33.4	5.75	1443.4	617.2	5.42
14	893 x CCS1	230.0	25.5	46.1	60.7	37.2	6.0	1331.7	575.9	5.87
15	GG x RR1	253.1	26.8	31.6	58.1	56.9	5.5	1282.9	574.4	6.07
16	GG x 893	279.8	23.0	29.7	55.5	63.2	5.5	1324.7	555.5	4.85
17	GG x CCS1	254. <b>9</b>	28.3	31.7	50.1	49.9	6.0	1535	653.3	5.42
18	CCS1 x RR1	235.1	31.8	53.4	53.4	43.9	6.5	1802.2	790.2	5.94
19	CCS1 x 893	243.5	25.2	48.9	62.1	57.7	6.5	1604	699.4	4.86
20	CCS1 x GG	224.6	25.6	43.5	47.8	33.3	6.25	945.7	421.2	5.80
	S.E	21.3	4.47	6.5	9.7	8.7		156.1	354.6	0.15
	C.V %	10.25	20.5	20.7	21.8	22.7		32.2	32.07	6.04
	CD at 5%	NS	NS	13.4	NS	17.5		NS	NS	0.31

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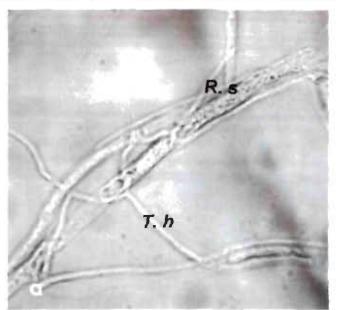
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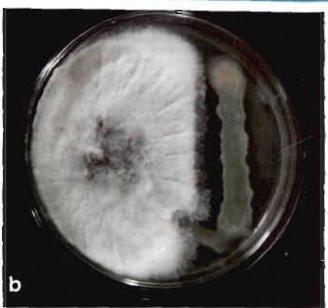
Cardamom

Table 2.4. Colony, conidial and appressorial characteristics of cardamom isolates of

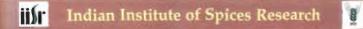
Cardamom

		Conidial character					
Place of collection	Colony colour	Length (µm)	Width (µm)	Shape	Appressorial character		
Appangala	White, light grey and orange slime, reverse side cream	16.12	4	Cylindrical with obtuse ends	Multi lobed (3-4 lobes)		
Sakleshpur	Dark grey with white margin with pink slime	15.75	3.75	Cylindrical with obtuse ends with narrowing of the center (Pea nut pod shape)	Unlobed		
Pollibetta	White and grey, reverse side cream and orange	15.18	4.12	Cylindrical with obtuse ends	Multi lobed (2- 3 lobes)		
Siddapur	White and black with orange slime, reverse is black	12.75	4.38	Ovoid	Multi lobed (3-4 lobes)		
Chettalli	Dark grey and orange, reverse side black	15.75	4.28	Cylindrical with obtuse ends	Multi lobed (2-3 lobes)		
Nelliahudikeri	Grey and orange slime	12.56	4.5	Cylindrical with a slightly tapered base and obtuse apex	Multi lobed (3-4 lobes)		
Hakathur	White and light grey mycelia with light orange slime in the center	16.13	3.75	Cylindrical	Multi lobed (3-4 lobes)		
Byakaravally	White puffy mycelia with light grey spots	17.25	3.75	Ovoid	Unlobed		
Ave Maria	Gray and orange	15.75	4.06	Cylindrical with obtuse ends	Multi lobed (3-4 lobes)		
Thithimathi	White and dark grey and reverse side dark grey and cream	15	3.3	Cylindrical with a slightly tapered base and obtuse apex	Unlobed		





a). Rhizoctonia solani x Trichoderma hamatum b). Rhizoctonia solani x Pseudomonas fluorescens Fig. 2.3. Trichoderma hamatum, an effective organism against rhizome rot



Cardamom



#### Genetic resources

**Conservation of germplasm:** In the ex-situ conservatory, 700 Zingiber accessions are conserved.

**Performance of Nepal selections and high oil types:** Twelve selected Nepal ginger selections along with control (Varada) were evaluated in replicated trial (RBD). Yield evaluation of high oil type revealed variation in oil content due to season.

Molecular characterization of primitive, elite and exotic genotypes: Molecular fingerprints of elite (Varada), exotic ('Kintoki') and primitive/ putative wild types ('Ellakallan', 'Kakkakalan', 'Kozhikkalan', 'Sabarimala' and 'Pink ginger') were developed using RAPD and ISSR markers to characterize and protect the accessions (Figs. 3.1 & 3.2). Among the 30 molecular markers studied, 13 could easily discriminate the genotypes.

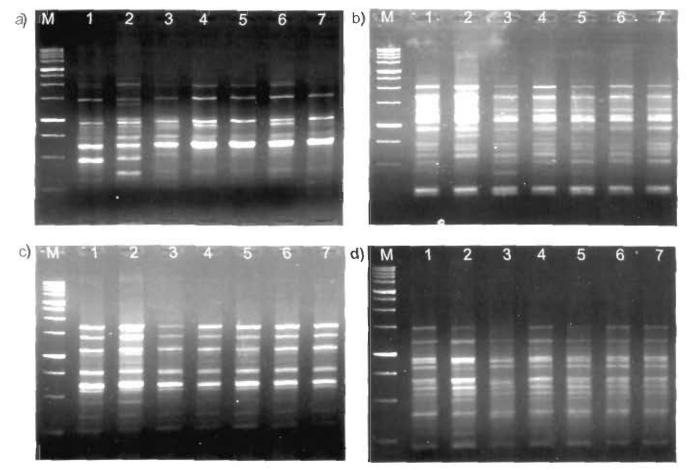
Cluster analysis of data using UPGMA dendrogram implied some phylogenetic relationship between the putative wild types and modern cultivars. The exotic type from Japan, resembling the putative types in rhizome features, shared high similarity with the four indigenous putative types.

Screening germplasm for pollen fertility and pollination: Pollen fertility based on stainability, *in vitro* and *in vivo* germination in 12 accessions revealed that there was no *in vivo* germination of pollen on self pollination.

#### Crop improvement

**Performance of M1 and M2 generations derived from irradiated (gamma) buds:** In M1 generation of three ginger varieties irradiated with five doses (0.5, 0.6, 0.7, 0.8 and 0.9 kr) of gamma irradiation, maximum sprouting (100%) was recorded in variety IISR Varada @ 0.5 and 0.8 kr, while maximum percent chlorophyll mutants were noticed in IISR Rejatha @ 0.7 kr. Variation in plant height and number of tillers was evident among M2 plants treated with specific dose of radiation.

**Variation in the lobes of floral labellum:** Of the 12 cultivars examined, Acc nos. 53, 50



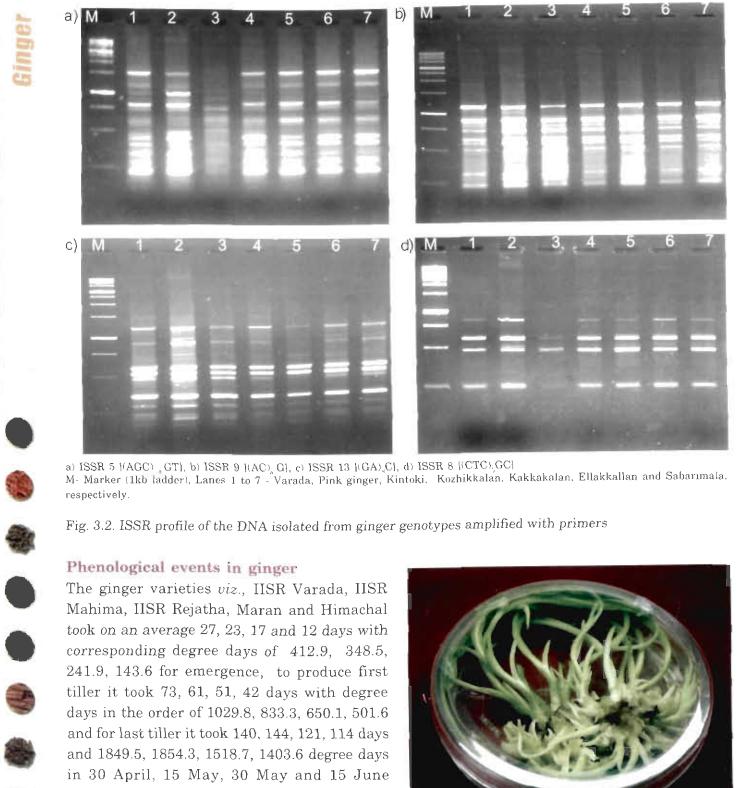
a) OPJ-07 (5'CCTCTCGACA3'), b) OPA-08 (5'GTGACGTAGG3'), c) OPD-08 (5'GTGTGCCCCA3'), d) OPD-07 (5'TTGGCACGGG3') respectively. M-Marker (1kb ladder), Lanes 1 to 7 - Varada, Pink ginger, Kintoki, Kozhikkalan, Kakkakalan, Ellakkallan and Sabarimala, respectively.

Fig.3.1. RAPD profile of the DNA isolated from ginger genotypes amplified with primers

(PGS-33), 45 (PGS-26) and 165 showed unilobate labellum while Acc nos. 26, 38, 31, 91, 12, 147, 125 and Jamaica showed trilobate labellum.

**Somaclonal variation:** An improved efficient technology was developed for micro rhizome induction in ginger for production of diseasefree planting materials and these microrhizomes can directly be planted in the field without hardening. A simple technology for micropropagation without the use of growth regulators using petri plate system was developed (Fig. 3.3). The micropropagated

plants of ginger through direct and indirect organogenesis as well as microrhizome pathway did show variations in varying degrees in morphological, cytological, biochemical and molecular characters. In general, the highest variations were observed among callus regenerated plants and minimum variations among microrhizome derived plants. One promising polyploid somaclone with bold rhizomes was identified. The resultant somaclonal variation can effectively be used to compensate the lack of variability through sexual reproduction and recombination in ginger.



a) ISSR 5 [(AGC) , GT], b) ISSR 9 [(AC), G], c) ISSR 13 [(GA),C], d) ISSR 8 [(CTC),GC] M- Marker (1kb ladder), Lanes 1 to 7 - Varada, Pink ginger, Kintoki, Kozhikkalan, Kakkakalan, Ellakkallan and Sabarımala, respectively.

Fig. 3.2. ISSR profile of the DNA isolated from ginger genotypes amplified with primers

#### Phenological events in ginger

The ginger varieties viz., IISR Varada, IISR Mahima, IISR Rejatha, Maran and Himachal took on an average 27, 23, 17 and 12 days with corresponding degree days of 412.9, 348.5, 241.9, 143.6 for emergence, to produce first tiller it took 73, 61, 51, 42 days with degree days in the order of 1029.8, 833.3, 650.1, 501.6 and for last tiller it took 140, 144, 121, 114 days and 1849.5, 1854.3, 1518.7, 1403.6 degree days in 30 April, 15 May, 30 May and 15 June planted crops, respectively (Table 3.1). The variations among varieties for emergence, first tiller production and last tiller production in terms of days were 27, 7, 11, 6; 12, 9, 16, 9; 8, 27, 34 and 9 respectively for 30 April, 15 May,



Fig. 3.3. Micropropagation of ginger using petri plate system

Gingel

Table 3.1. Days and degree days required for different events in ginger

	E	limachal		arada	1	Mahima	1	Rejatha	м	aran
Time of planting	Days	GDD	Days	GDD	Ei Days	nergend GDD	e Days	GDD	Days	GDD
30-April	18	288.8	17	267.4	25	389.1	29	456.8	44	662.3
15-May	27	406.0	22	344.9	20	308.8	24	364.7	21	318.3
30-May	13	199.3	15	224.8	23	318.2	12	174.4	21	292.8
15-June	8	103.0	14	167.3	14	166.8	10	122.3	13	158.8
				F	First til	ller app	earanc	e		
30-April	79	1089.7	79	1094.8	69	980.7	73	1026.3	67	957.5
15-May	62	846.9	60	825.4	60	824.4	66	889.9	57	780.0
30-May	57	716.8	56	705.3	55	701.4	46	592.2	41	534.9
15-June	37	436.5	40	475.3	42	502.3	46	548.7	46	545.1
				I	ast till	ler appe	earance			
30-April	141	1857.8	137	1805.6	144	1896.1	135	1778.8	145	1909.3
15-May	136	1755.6	149	1929.3	148	1913.7	129	1657.5	156	2015.3
30-May	126	1583.7	114	1421.7	117	1463.6	107	1340.9	141	1783.8
15-June	117	1444.7	115	1411.7	116	1430.0	115	1411.8	108	1320.0

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30 May and 15 June planted crops respectively. There exists a positive association between degree day's requirement for different events and yield (emergence r=0.66, first tiller appearance r=0.017, last tiller appearance r=0.34).

#### Nutrition

Validating the optimum P/Zn ratio requirement for increasing the yield in ginger: The threshold value of leaf P/Zn ratio was confirmed to be 90 through second order response function ( $R^2=0.499^{**}$ ). On validating the same under field conditions, the initial leaf P/Zn ratio at 60 DAP in the range of 145-223 could be brought down to the threshold range of 69-150 after foliar spray of Zn (0.5%) twice during August and October. By lowering the ratio below 108, increased rhizome yield up to 20% could be achieved (Table 3.2).

#### Table 3.2. Relation between leaf P/Zn, soil P/Zn and yield

Leaf P/Zn	Soil P/Zn	Yield (kg/bed)
69.7	4.9	8.4
77	5.7	9.1
101	4.4	9.5
108	4.0	10.6
136	5.4	7.8

Ginger

Nutrient requirement for targeted production of ginger: Based on the fixed yield targets and initial soil availability of major nutrients, nutrient required for berry production (NR), contribution of nutrient from soil (CS) and contribution from fertilizer (CF) were calculated for ginger from the earlier data and fertilizer doses to obtain yield targets of 10, 15, 20 kg/bed were calculated and applied. Zinc was also supplemented as foliar spray to bring the leaf P/Zn ratio in the threshold range. The achieved ginger rhizome yield was 10.8, 12.4 and 11.4 kg/ bed with a deviation of +0.5%, -17% and -43% from the target. Through targeted nutrient application 14-26% increased yield over recommended dose could be achieved in IISR Varada with reduced fertilizer application.

Organic farming: Ginger was grown organically by applying FYM, vermicompost, ash and rock phosphate, azospirillum and phosphobacteria. Trichodermaand Pseudomonas sp. (IISR-6 & 853) were applied as biocontrol agents for disease control. The mean yield (10.2 kg/3 m<sup>2</sup>) recorded (var. IISR Varada and IISR Mahima) under integrated system was 29.7% and 14.5% higher over chemical and organic farming, respectively. Organic farming resulted in a mean yield of 8.8 kg/ bed recording 13% higher yield over chemical practices. Soil nutrient buildup and microbial biomass carbon were observed to be high in organic and integrated managements as compared to conventional system. FYM + neem cake + vermi/coir pith compost application recorded higher yield on par with FYM application alone. All the PGPR combinations applied were equally effective in controlling rhizome rot of ginger and produced yield on par with chemical control.

#### Quality

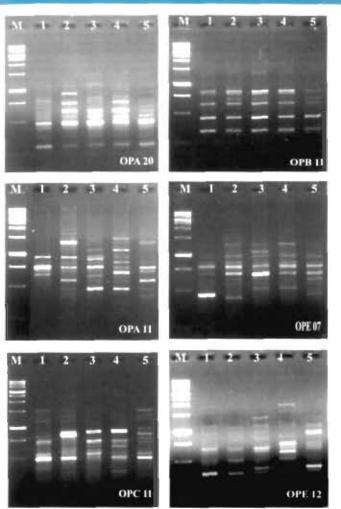
Ginger accessions with high quality: Acc nos. 209, 50, 57,162, 99, 156 and 197 recorded high oil (above 2%). Acc 57 and 162 contained high limonene in oil compared to other accessions. Among the Nepal collections, oil content ranged from 1.5 to 1.9% and oleoresin from 3.8 to 6.1%. Acc. 592, 593 and 598 contained 1.9% oil and 578 contained 6.1% oleoresin. Among the Sikkim collections, Bhaise contained 1.5% oil, 6.2% oleoresin and 12 % crude fibre with 16% dry recovery. Gorubathani contained 1.5% oil, 4.7% oleoresin and 5.2% fibre.

GC MS analysis of essential oil of traded ginger: GC MS profiling has shown a total of 20 compounds in Indian ginger and 18 in Chinese ginger, which were common for both the produces. The main compounds identified are  $\alpha$  -pinene, camphene,  $\beta$ -myrcene,  $\alpha$  phellandrene, geranyl acetate and zingiberene. Zingiberene (27.14%) and  $\alpha$  phellandrene (10.89%) are high in Chinese ginger.

Molecular characterization of traded ginger from India and China: Out of 80 random decamer primers screened, 30 primers which gave consistent amplification pattern were selected for RAPD analysis. Amplication pattern for some of the primers is given in Fig. 3.4.

#### Etiology of soft rot

Pythium myriotylum and P. aphanidermatum were found associated with soft rot. Four other species of Pythium such as P. ultimum, P. vexans, P. splendons and P. deliense were also found associated with soft rot. Among them, P. myriotylum was predominant and PCR based method was found suitable for identification of P. myriotylum.



a) OPA 20, b) OPB11, c) OPA11, d) OPE 07, e) OPC11 and OPE 12 respectively. M-Marker (1kb ladder), Lanes 1 to 5- Cochin ginger, Chinese ginger, Varada, Rejatha and Himachal, respectively.

Fig. 3.4. RAPD profile of the DNA isolated from traded ginger amplified with primers

#### Host resistance

Screening for soft rot resistance: Five accessions viz. Acc nos. 2, 97, 98, 554 and 558 that showed resistance against *P. myriotylum* under green house conditions were evaluated in the field for resistance. Soft rot incidence was less in these accessions as compared to susceptible check Himachal.

Resistance against bacterial wilt in related genera: Among the species of Zingiberaceae such as *Globba* sp, *Alpinia galanga*, *Maranda* spp, *Costus* sp, *Curcuma caesia*, *Amomum*  subulatum and an unidentified Curcuma species, only three of them viz., Alpinia galanga, Globba sp, Curcuma caesia etc. were found to be the host for ginger strain of Ralstonia solanacearum (Fig. 3.5). Curcuma amada, the Indian mango ginger was found to be resistant to bacterial wilt. This is in addition to other susceptible hosts such as Curcuma longa, Curcuma aromatica, Curcuma zedoaria and Kaempferia galanga.

#### Biocontrol

Annual Report 2007-08

Isolation of endophytic bacteria from ginger rhizomes: Nineteen endophytic bacteria were isolated from ginger rhizomes collected from Wayanad and Calicut in Kerala using modified protocols for isolation of endophytes. An enrichment method was adopted where the rhizome was incubated in the nutrient rich medium. The necessary sterility check and other confirmatory assays were adopted in order to isolate true These bacteria endophytes. were characterized using morphological criteria such as colony character, shape, elevation, motility and gram reaction. All of them belonged to gram negative group and were short rods. Five of them were highly motile. These isolates are being evaluated in the green house for their potential against soft rot and bacterial wilt disease of ginger. Among the isolates GEB 7, GEB 9, GEB 13, GEB 17, GEB 18, and GEB 19 showed promise for suppression of wilt and soft rot under green house conditions (Fig. 3.6).

Isolation of rhizobacteria from ginger roots: Rhizosphere soils were collected from different varieties of healthy ginger plants. Six varieties/accessions of ginger were used in this study. They were IISR Varada, IISR Mahima, Himachal, Suprabha, Maran and IISR Rejatha. Rhizobacteria of ginger isolates that showed

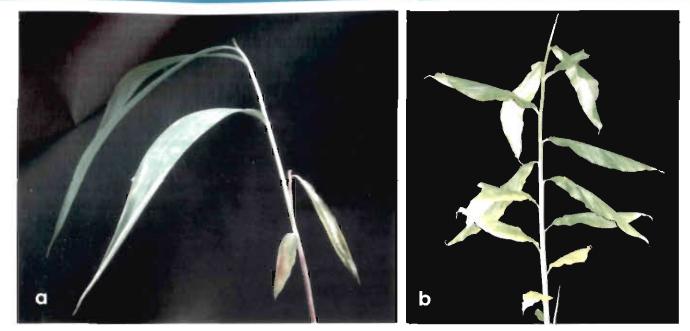


Fig. 3.5. Alpinia galanga (a), Globba spp (b) - hosts for ginger strain of Ralstonia solanacearum Biovar 3

more than 50% inhibition against any of the three pathogens namely *P. capsici*, *Pythium* and *Fusarium* were short-listed. Six isolates namely GRB 25, GRB 35, GRB 36, GRB 57, GRB 58 and GRB 68 inhibited all the three pathogens by more than 50% (Table 3.3).

# Integrated management strategies for control of diseases

**Chemical control:** A disease management trial consisting of copper oxychloride (0.25%), cheshunt compound (0.3%), Bordeaux mixture (1%), potassium phosphonate (0.3%) and an immunomodulator (bromo propane diol) was laid out at IISR Farm, Peruvannamuzhi. The disease incidence ranged from 6.0-23.5% among the treatments and minimum incidence was recorded in cheshunt compound (0.3%).

**Biocontrol trial:** In a trial on management of soft rot using biocontrol agents *Pseudomonas aeruginosa* (IISR BP35), *P. fluorescens* IISR51, *Trichoderma harzianum* (GTH7) and *P. fluorescens* IISR6 were compared with the chemicals such as mancozeb or metalaxyl

# Table. 3.3.Antagonistic activity of short-<br/>listed rhizobacterial isolates<br/>from ginger against pathogens

Isolate	Patho	ogen suppres	sion (%)
	Pythium	Phytophthora	Fusarium
GRB-25	78.53	79.63	83.30
GRB-35	64.83	72.93	72.20
GRB-36	80.00	64.07	50.00
GRB-57	85.97	81.01	70.73
GRB-58	85.93	78.53	70.37
GRB-68	84.03	77.80	70.00
GRB-63	75.93	0.00	74.03
GRB-67	5.60	72.20	54.47
GRB-21	0.00	35.17	51.47
GRB-22	0.00	45.17	54.07
GRB-38	25.53	51.10	43.67
GRB-64	7.80	41.10	62.97

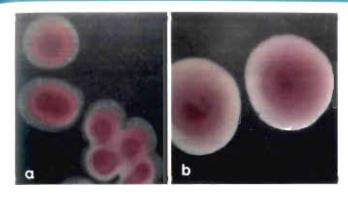


Fig. 3.6. Endophytic bacteria from ginger rhizome found effective against Pythium. (a) GEB17; (b) GEB 18

mancozeb. The treatment was given on 0, 30, 60 and 90<sup>th</sup> day of planting as seed treatment as well as soil application. The rot incidence ranged from 5-19% under various treatments. The results indicated that the mancozeb or *Pseudomonas* application performed better than other treatments.

#### GIS studies on land suitability

Secondary data of area, production and productivity of ginger have consistently shown an increasing trend during the last 3 decades. Increase in area is not always in proportion with the increase in production. Thirty years area and production curves of the important ginger growing states were compared with the suitability model which indicated Eco-crop that suitability has direct impact on production. Orissa, West Bengal, Mizoram and Kerala are very highly suitable while North western states like Gujrat, Rajasthan, Uttar Pradesh and Madhya Pradesh are marginally suitable or unsuitable. North eastern and South western states are ideal and have high suitability for

ginger cultivation. Future prediction of Ecocrop model shows that if the temperature increases by 1.5 to 2°C, the suitability of Orissa and West Bengal will reduce drastically from high suitability to marginal suitability, indicating the effect of climate change (Fig. 3.7).

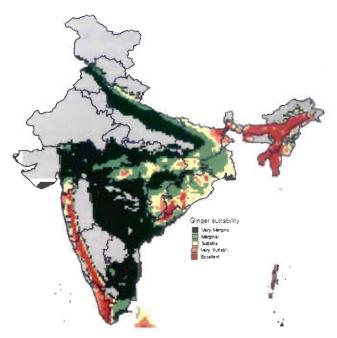


Fig. 3.7. Future prediction of land suitability changes (as per Eco crop model) if temperature increases by 1.5°C to 2°C

#### Growth and instability of ginger production in India

Change in area has contributed a maximum to change in production in period-II except in few states (Table 3.4). The recorded highest instability (towards the negative side) in major producing states like Kerala is the cause for poor export performance by the country.

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Table 3.4.Changes in average production and components of change in average<br/>production of ginger (1985/86 - 1994-95 to 1995/96 - 2002/03)

The Lotter	Average production (t)			Con	ponents	Contribution to change in		
State	First period	Second period	Change (%)	Change in mean area	Change in mean yield	Intera- ction effect	Change in area - yield co-variance	average national production
Kerala	45733	43738	-4.4	470.1	-473.7	96.9	6.8	-1.7
Meghalaya	33434	46526	39.2	78.8	19.6	6.1	-4.4	10.9
Orissa	13969	24673	76.6	69.8	16.4	8.6	5.2	8.9
Sikkim	12028	17824	48.2	124.6	-40.6	-23.2	39.2	4.8
West Bengal	10395	18522	78.2	60.3	27.7	12.4	-0.3	6.5
Mizoram	8815	26805	204.1	109.1	-2.9	-5.9	-0.3	15.0
Andhra Pradesh	7968	7065	-11.3	47.1	46.0	-2.4	9.3	-0.7
Arunachal Pradesh	7563	30376	301.7	67.6	10.9	23.7	-2.2	19.0
Tamil Nadu	7311	12272	67.9	-73.0	222.9	-88.4	38.5	4.0
Madhya Pradesh	3602	5492	52.5	115.2	-7.2	-4.3	-3.7	1.6
Karnataka	3199	8707	172.2	99.9	0.1	0.1	-0.1	5.3
Tripura	1336	2256	68.9	77.2	19.3	10.5	-7.0	0.8
Himachal Pradesh	760	5256	592.0	1,1	81.3	5.0	12.6	3.7
Others	7987	34950	337.6	34.6	41.0	50.1	-25.7	22.4
India (total)	164099	284462	73.4	59.8	27.5	12.1	0.5	100.0

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#### Genetic resources

**Conservation of germplasm:** In the *ex situ* conservatory, 1040 *Curcuma* accessions are conserved.

#### **Crop** improvement

**Performance of seedling progenies and their mother plants:** One hundred and seventy six seedling progenies and 14 mother plants were multiplied in pots, in addition to the maintenance of 84 seedling progenies in poly bags. Morphological characters and yield were recorded for the plants maintained in pots. The yield level varied from less than 50 g to more than one kg. Twenty eight progenies produced more than one kg rhizome yield.

**Cytological analysis of seedling progenies and their mother plants:** Cytological analysis of 14 mother plants indicated a chromosome number of 2n=63 in 11 and 2n=84 in 3 of them. Cytological analysis of five seedling progenies showed 2n=84 in four of them and 2n=77 in one (Fig. 4.1). Flowering and seed set in seedling progenies: Flowering was recorded in 50 seedling progenies. Seeds were recovered from 11 of them and put for germination to develop

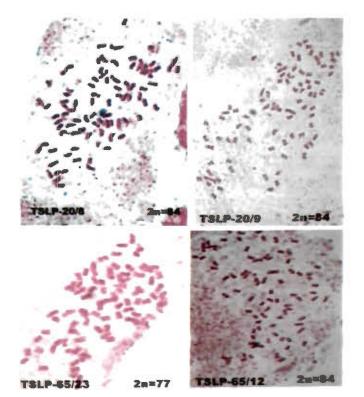


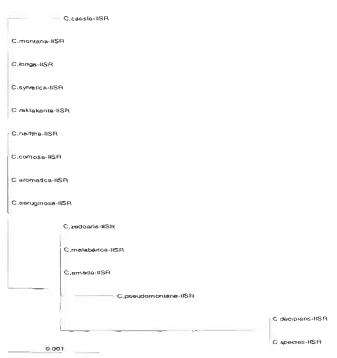
Fig. 4.1. Chromosome number variation among turmeric seedling progenies

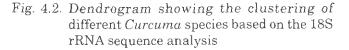
advanced generation of seedling progenies. Germination started in two lots after 100 days of sowing. 52 advanced generation seedling progenies were recovered so far. Maximum number of seeds (94 and 92) were recovered from 138/23 and 138/74 seedling progenies. The germination of seeds has already started.

**Somacional variation:** An improved efficient technology was developed for micro rhizome induction in turmeric for production of diseasefree planting materials. These micro rhizomes can directly be planted in the field without hardening. One turmeric somacione with very close nodes was also identified.

Single nucleotide polymorphism (SNP) in Indian Curcuma species: The single nucleotide polymorphism in 18S rRNA gene sequence in Indian Curcuma species was analysed and the sequence data was compared with the nucleotide sequences of 18S rRNA gene from the different species of Curcuma deposited at the NCBI GenBank. The single nucleotide polymorphism at the 234<sup>th</sup> position of partial 18S rRNA gene of C. aromatica, C. longa and C. aeruginosa agreed with the already reported sequences of these species from China (cytosine residue at 234th position of 18S rRNA gene), while the sequence of Indian C. zedoaria showed the similarity with Japanese populations of C. zedoaria (thymine nucleotide at 234<sup>th</sup> position) rather than Chinese population. The Indian C. zedoaria showed similarity with the C. xanthorrhiza reported from China. The rest of the species studied are the first time reports and the species viz., C. raktakanta, C sylvatica, C. montana, C. haritha and C. comosa showed cytosine as the nucleotide at 234<sup>th</sup> position while C. malabarica and C. amada showed thymine as the nucleotide at 234<sup>th</sup> position. The bootstrap analysis of the nucleotide sequences

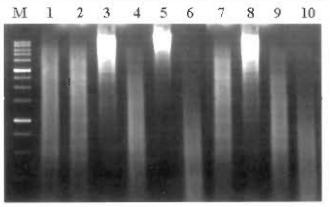
of 18S rRNA gene placed the fifteen Indian *Curcuma* species studied in five different groups (Fig.4.2).





**Development of microsatellite markers** and characterisation of Curcuma spp.: Good quality genomic DNA was isolated and purified from 15 Curcuma species available in the field genebank and the yield ranged from 63 -110 µg per gram fresh leaf tissue. Among the ten restriction enzymes screened, Rsa I produced a smear ranging from 2.5 kb to 100 bp with more fragments centered at around 500 bp, which was selected for further study. The optimized reaction mix for digesting 500ng DNA contained 1X assay buffer, 20 Units enzyme (Rsa I) in a reaction volume of 20  $\mu$ l with 4 hour incubation at 37°C. A smear of fragments (1.0kb – 250bp) centered at ~500 bp was obtained as successful linker ligation (Fig 4.3). A total of 700 clones from the enriched libraries (microsatellite containing fragments) were screened by colony PCR and selected

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Lane M: 1,0 kb DNA Ladder, Lane 1: Mse I, Lane 2: Hae III, Lane 3: Hind III, Lane 4: Sau 3AI, Lane 5: Nco I, Lane 6: Rsa I, Lane 7: Eco RI, Lane 8. Bsr Bl, Lane 9. Bst UI, Lane 10: Alu I

Fig. 4.3. Restriction digestion profiles of 10 different restriction enzymes.

clones were sequenced which resulted in 82 microsatellite repeats of variable length.

Suitable primers to amplify these regions are being designed.

#### Phenological events in turmeric

The turmeric varieties *viz.*, IISR Prabha, IISR Prathibha, IISR Alleppey Supreme, IISR Kedaram and Suguna took on an average 31.9, 32.2, 23.5 and 24.2 days with corresponding degree days of 498.5, 480.1, 326.7, 295.6 for emergence, to produce first tiller it took 80.5, 71.3, 62.8, 53.7 days with degree days in the order of 1115.1, 947.1, 787.3, 578.0 and for last tiller it took 156.4, 139.4, 122.3, 109.7 days and 2054.5, 1794.3, 1528.1, 1342.1 degree days in 30 April, 15 May, 30 May and 15 June planted crops, respectively (Table 4.1). The variations among varieties for emergence, first tiller

Table 4.1. Days ar	d degree	days	required	for	different	events in	turmeric
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Time of		100	11.00	and the second	Alle	ppey	1.0					
planting	Prabha		Prat	hibha	Sup	reme	Kedaram		Suguna			
Emergence												
	Days	GDD	Days	GDD	Days	GDD	Days	GDD	Days	GDD		
30-April	26.7	420.1	29.3	461.7	32.3	503.5	34.7	537. <del>9</del>	36.7	569.3		
15-May	21.0	324.1	33.0	<b>49</b> 3.3	31.7	477.7	38.3	561.0	37.0	544.5		
30-May	23.7	329.5	23.0	322.3	26.0	356.1	21.7	304.5	23.0	320.8		
15-June	26.3	320.8	26.0	316.9	22.7	277.6	22.7	277.4	23.3	285.4		
			H	First till	er app	earance	e					
30-April	73.7	1036.3	84.0	1155.4	84.0	1155.4	81.7	1128.7	79.3	1099.7		
15-May	70.3	936.3	73.0	967.3	71.7	951.9	72.0	955.4	69.3	924.5		
30-May	59.0	743.7	62.7	786.3	65.7	821.1	63.3	792.8	63.3	792.6		
15-June	50.7	589.8	61.3	667	59.3	567.8	52.0	532.8	45.3	532.8		
			ĵ	Last till	er app	earance						
30-April	155.0	2036.2	156	2049.7	157.7	2071	157.0	2061.8	156.3	2053.7		
15-May	131.3	1691.4	137.3	1767.8	142.0	1826.7	143.3	1845.6	143.0	1839.8		
30-May	121.3	1497.1	124.3	1558.3	119.3	1494.7	123.3	1545.4	123.3	1545.2		
15-June	110.3	1349.5	112.7	1379.9	109.0	1332.8	108.3	1324.7	108.3	1323.7		

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9
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production and last tiller production in terms of days were 10.0, 17.3, 4.3, 3.0; 10.3, 3.7, 6.7, 16.0; 2.7, 12, 5, 4.4 respectively for 30 April, 15 May, 30 May and 15 June planted crops.

#### Nutrition

Nutrient budgeting: Based on the fixed yield targets and initial soil availability of major nutrients, nutrient required for berry production (NR), contribution of nutrient from soil (CS) and contribution from fertilizer (CF) were calculated for turmeric from the earlier data and fertilizer doses to obtain yield targets of 15, 20 and 25 kg/bed was calculated and applied. The achieved turmeric rhizome yield in IISR Prathibha was 14.8, 15.8 and 16.3 kg/ bed with a deviation of -1%, -20% and -34% from the fixed target. Through targeted nutrient application 12 - 20% increased yield over recommended dose could be achieved with reduced fertilizer application by considering the FYM's nutrient contribution also.

Organic farming: Turmeric was grown organically by applying FYM, vermi compost, ash and rock phosphate, azospirillum and phosphobacteria. Trichodermaand Pseudomonas sp. (IISR-6 and 853) were applied as biocontrol agents for disease control. In turmeric (IISR Alleppey Supreme and IISR Prathibha), integrated system recorded a higher mean yield of 15 kg/3 m<sup>2</sup> which was 12.8% higher compared to the organic system. Organic system showed on par yield as that of conventional (chemical) farming. Soil nutrient buildup and microbial biomass carbon were observed to be high in organic and integrated managements as compared to conventional system. FYM + neem cake + vermi/coir pith compost application recorded higher yield on par with FYM application alone. All the PGPR combinations applied were equally effective in controlling rhizome rot of turmeric and

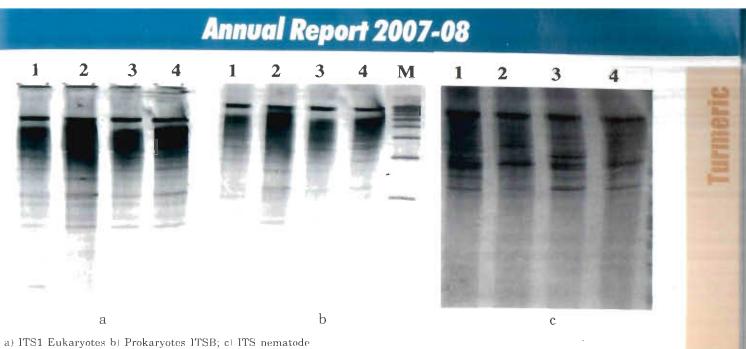
produced yield on par to chemical control measures.

#### Assessment of quality of soils under spice based cropping systems involving turmeric

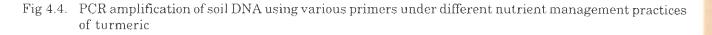
The organic and INM treatments favored the accumulation of nutrients especially organic C, dissolved organic C (DOC) and N (DON) in soil. The data on parameters indicating microbial activity in soils indicated that microbial biomass-C (C $_{\rm MIC}$ ), -N (N $_{\rm MIC}$ ), –P (P $_{\rm MIC}$ ) and CO $_2$ production levels under the integrated and organic treatments were markedly higher than the level under the chemical and control treatments. The qCO<sub>2</sub> (metabolic quotient) levels were greater in the chemical and control treatments. The activities of dehydrogenase, phosphodiesterase casein-protease, urease, arylsulphatase and  $\beta$ -glucosidase were markedly higher in the integrated and/or organic treatments.

The results further revealed diversity of microbial composition in soils under various management regimes using PCR RFLP and denaturing PAGE (Fig. 4.4). The community profiles resolved better under denaturing conditions and the touch down protocol improved the resolution and number of bands detected. When all the primers are taken together, maximum number of bands was obtained in case of integrated (60) in case of denaturing PAGE. Maximum bands were obtained in integrated for ITS1, ITS2, prokaryotic IGS and 16S rDNA primers. Chemical treatment and control registered maximum for ITS fungal primers. In case of fully organic treatment, maximum bands were observed for prokaryotic IGS primers. The control registered the lowest population in all the cases. Maximum number of nematodes was found in case of integrated and fully organic

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Lane 1- Control, 2- Integrated, 3- Chemical, 4- Fully organic, M- Marker (100bp ladder)

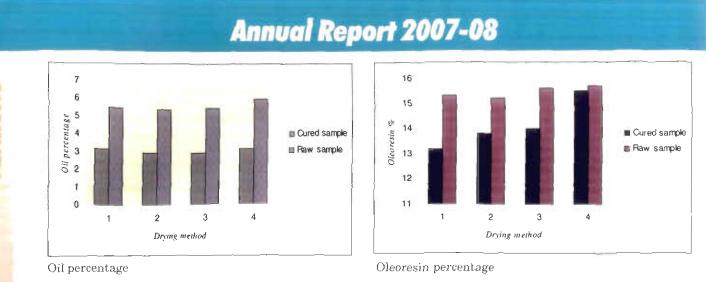


treatments. In general prokaryotic population was highest under integrated treatment. Maximum eukaryotic population was observed under integrated and fully organic treatments. The banding profiles when analyzed showed that based on the presence or absence of bands maximum similarity of 66% was observed among chemical and control treatments. The banding profiles of fully organic and IPNM treatments showed a close similarity with each other and clustered together showing a similarity of 60%.

#### Post harvest technology and quality

Quality under controlled atmospheric storage: Turmeric variety IISR Prathibha was stored for 120 days in three layered metalised polyester cover under vacuum, 100%  $N_2$  and 90%  $N_2$  + 10%  $O_2$ . The turmeric samples were analysed for oil, oleoresin and curcumin content. No significant variation was observed in the quality constituents compared to control which was stored in ordinary gunny bag. Mechanical drying and quality changes: Turmeric is traditionally dried in sun after boiling for 45 to 90 minutes. In the present study, one set of turmeric sample was cured normally and another was not cured. Both were kept for drying under drying systems such as hot air oven (1), RRL-T reverse flow drier (2), agri waste (CPCRI) drier (3) and sun drying (4). In all drying systems, drying was conducted for 6 hrs every day. The drying time varied between 18 hrs (3days) to 84 hrs (14days). Sun drying of raw sample took 14 days of six hour duration while it took only 10 days for cured sample. Cured samples took less time in all systems. Hot air oven took minimum time for both cured and raw samples. Curing resulted in a reduction of 40 % in oil and 15% in oleoresin content compared to raw samples (Fig. 4.5). The curcumin content was not affected due to curing.

Analysis of curcuminoids in germplasm: HPLC analysis of the curcuminoids in 200 accessions of turmeric revealed that in low





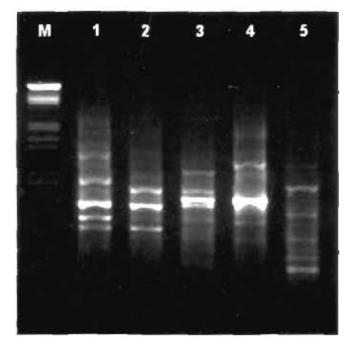
curcumin accessions, curcumin III was at lower concentration (0.027-0.38%). Average ratio of curcuminoids I, II and III was found to be 43.5: 45.5:11.0. In high curcumin accessions, curcumin III ranged from 17.1 to 27.9 % while curcumin II showed lower levels as compared to low curcumin accessions. Ratio of curcumin I: II: III was 43.4: 32.8: 23.8 respectively.

**Downstream enzymes in curcumin biosynthetic pathway:** Enzyme assay for major biosynthetic enzymes viz., cinnamate hydroxylase in high and low curcumin accessions revealed that low curcumin accessions had higher activities towards maturity (244-351units) than high curcumin accessions (110-154 units).

**Determination of purity of powdered market samples:** A viable protocol for isolation of DNA (modified CTAB) from the recalcitrant inaterials was developed (Fig. 4.6). This DNA could be successfully amplified using RAPD markers (Fig. 4.6).

#### Diseases and Pests Etiology of soft rot

*P. aphanidermatum* was found associated with soft rot. Forty five of the 81 isolates of *Pythium* sp. collected from different turmeric growing regions of South India showed pathogenicity. Among them, 32 isolates were identified as *P.aphanidermatum* and eight belonged to *P. myriotylum*. *P. aphanidermatum* from Gulburga (Karnataka), Jagtial (AP) and Chittoor (Kerala) were found to be most virulent.



M- Marker (Eco RI Hind III double digest), Lane 1-Curcuma longa, Lane 2- Curcuma zedoaria, Lane 3-Market sample 1, Lane 4-Market sample 2, Lane 5-Market sample 3

Fig. 4.6. PCR amplified products of DNA isolated from turmeric powder / market samples of turmeric using RAPD primer.

#### Host resistance

Screening for soft rot resistance : Fifty one turmeric accessions were screened for soft rot resistance caused by *Pythium aphanidermatum*. Twenty two accessions *viz.*, 77, 79, 81, 85, 87, 93, 94, 100, 102, 103, 106, 114, 116, 117, 118, 125, 127, 129, 132, 133, 134 and 139 did not show any disease.

**Resistance against soft rot in related genera:** Curcuma longa, C. zedoaria, C. amada, C. aromatica, Z. officinale and C. caesia were inoculated with a turmeric isolate of Pythium aphanidermatum. Curcuma longa, C. aromatica, C. zedoaria and Z. officinale succumbled to infection. C. amada and C. caesia remained unaffected.

#### Integrated management strategies for control of diseases

#### Chemical control

**Tamil Nadu:** A disease management trial was conducted at Settiputhur in Coimbatore district of Tamil Nadu where the disease pressure was high on turmeric. The results of the trial after 90 days showed that the disease incidence was comparatively less in copper oxychloride (7.2%) which was on par with metalaxyl- mancozeb (10.8%) when compared to control which recorded an incidence of 24.4%.

**Kerala:** A disease management trial consisting of copper oxychloride (0.25%), cheshunt compound (0.3%), Bordeaux mixture (1%), potassium phosphonate (0.3%), and an immunomodulator (bromo propane diol) was laid out at IISR Farm, Peruvannamuzhi. There was very negligible disease incidence during the season.

## Shoot borer (Conogethes punctiferalis)

Seasonal population of shoot borer: Studies on seasonal population dynamics of shoot borer on turmeric were conducted in the field at Peruvannamuzhi. The symptoms of pest infestation were observed through out the crop season (July to December). The symptoms were first observed during August (0.2% shoots infested) and maximum new infestations occurred during December (10.9%).

**Natural enemies of shoot borer:** Apanteles taragamme ? (Braconidae) was observed to parasitize the larvae of the shoot borer for the first time but the level of parasitization was low (2.1% during December).

**Evaluation of plant products and insecticides against shoot borer:** Four plant products namely, neem oil 0.5%, Neemgold 0.5%, Neemazal 0.25%, Nimbicidine 0.5% and four insecticides namely, malathion 0.075%, dimethoate 0.075%, carbosulfan 0.075%, imidacloprid 0.0125% and lamda cyhalothrin 0.0125% were evaluated against the pest under green house conditions. The plant products were sprayed at 2 week intervals during July to November and the insecticides at 3 week intervals during July to November. The trials indicated that among the plant products and insecticides, imidacloprid 0.0125% and carbosulfan 0.075% were promising.

#### Field screening of germplasm of turmeric:

Eight hundred and ninety accessions of turmeric were screened in the field against the shoot borer, *Conogethes punctiferalis*. Acc. No. 733 recorded maximum incidence of shoot borer (75%). One hundred accessions remained free from the pest attack.

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#### **Genetic** resources

Collection and maintenance of germplasm and seedling progenies: One cultivar was collected from Anakkampoyil of Wynad district of Kerala. Ninety four accessions of vanilla including 81 cultivars and 13 wild species were maintained at Peruvannamuzhi. Alternate germplasm site is also being established at HQ Chelavoor One hundred seedling progenies were maintained and 180 seedling progenies were planted in the field for preliminary evaluation. 350 cultures of seedling progenies and hybrids were maintained. One Hundred seedling progenies and 50 hybrids/colchicine treated plants were transferred *ex vitro*.

#### Crop improvement

Inter-specific crosses: Inter-specific crosses were performed between Vanilla planifolia, V. aphylla, V. pilifera and Vanilla sp. from A&N islands. Successful fruit setting was observed in all cross combinations. Out of 53 flowers pollinated, fruit set was observed in 51 flowers. The seeds were put under *in vitro* cultures for germination. Pollen germination: Pollen germination studies have been initiated in V. aphylla, V. pilifera and Vanilla sp. from A&N islands. Culture of fresh pollen by sitting drop method at  $25\pm2^{\circ}$ C under light using B&K medium containing 10% sucrose resulted in 58% germination in V. aphylla with 62µm pollen tube length after 4 hours. However, maximum pollen tube length of 91 µm was recorded in V. pilifera.

#### Viral diseases

**Production of antiserum against** *Potyvirus* **associated with vanilla:** *Potyvirus* associated with vanilla identified based on particle morphology through electron microscope propagated on cowpea under insect proof conditions were used for purification. Purification involved extraction with borate buffer, clarification (using organic solvents) and precipitation of the virus particles by ultracentrifugation. Further purification was achieved through 10-40% sucrose density gradient centrifugation. The light scattering virus layer was identified by passing light vertically from the top of the tube and virus

fraction was collected from the centrifuge tube with the help of a syringe. Purified virus preparation was injected into New Zealand white rabbit for polyclonal antiserum production. Five intramuscular injections at 10 days interval were given and serum containing antibodies were collected after 15 days from the last injection. Serum was centrifuged after overnight incubation and stored in equal volume of glycerol at -20 °C.

Preparation of IgG-enzyme conjugate and development of ELISA method for detection of the virus: From the crude polyclonal antiserum, immunoglobulin G (IgG) was purified through affinity chromatography. The affinity column contained protein A linked to cyanogen bromide. The crude antiserum was passed through the column and washed with wash buffer to remove all contaminants. The IgG bound to the column was then eluted using elution buffer. IgG was quantified by taking spectral reading at 280 nm and 1mg of this IgG was then used for the preparation of IgGalkaline phosphatase conjugate by one step glutaraldehyde method. After conjugation, the unused glutaraldehyde was removed by dialysis. Double antibody sandwich (DAS) ELISA was standardized using IgG as coating antibody and conjugate as detecting antibody for the reliable detection of the virus in diseased vanilla samples collected from different regions.

#### Occurrence of Bean yellow mosaic virus

Vanilla plants showing typical symptoms of chlorosis and leaf deformation collected from Madikeri district of Karnataka when subjected to RT-PCR using primers specific for five different potyviruses, gave specific amplification with primers specific to Bean yellow mosaic virus (BYMV). The PCR product was eluted, cloned in a TA vector and sequenced. The cloned region contained portion of NIb and coat protein gene. The sequenced region of coat protein contained 778 bases potentially coding for 259 amino acids. Sequence analysis based on per cent nucleotide and deduced amino acid identity and phylogenetic analysis clearly showed that vanilla isolate is a strain of BYMV (Fig. 5.1).



Fig. 5.1. Occurrence of *Bean yellow mosaic virus* on vanilla

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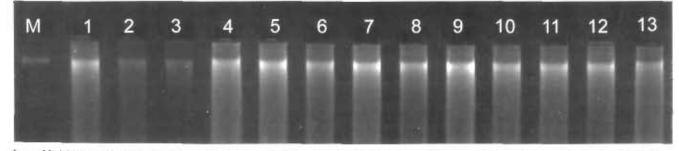


### Genetic resources

**Collection, evaluation and maintenance:** A high pungent and high colour type from North East '*Bhut Jolokia*' was added germplasm. Fifty four germplasm accessions including 21 Bydagi collections and six exotic collections were raised and multiplied. Twenty seven new germplasm accessions were purified by single plant selection and by selfing. Moderate to high variability was recorded for plant characters. Among the indigenous germplasm, ICBD-10 registered the highest color value (302.12 ASTA units), followed by ICBD-11 and ICBD-23. The EC-171 recorded the highest color

value, followed by EC-71 and EC-490, among the exotic lines. The capsaicin content among germplasm accessions varied from 0.0071 to 0.166 (Table 6.1). The lines ICBD-10, Kt-pl-19 and EC-71 were promising with low pungency and high colour value.

**Determination of purity of powdered market samples:** A viable protocol for isolation of DNA (modified CTAB method) from the recalcitrant materials was developed (Fig. 6.1) and the DNA could be successfully amplified using RAPD markers (Fig. 6.2).



Lane M -Marker (Human genomic DNA, Lane 1 -Genuine chilli powder, Lane 2-Red beet pulp, Lane 3- Almond shell dust, Lane 4- Ziziphus nummularia, Lane 5 -Simulated sample A, Lane 6- Simulated sample B, Lane 7 - Simulated sample C, Lanes 8 to 13 - Market samples.

Fig. 6.1. Genomic DNA isolated from dried chilli powder, market samples of chilli powder, simulated samples and prohable adulterants.

Table 6.1. Variability for morphological and quality characters in paprika accessions

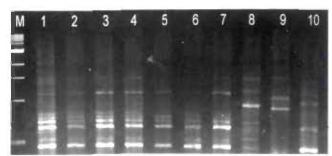
Carls Concerning Series		-	-	
Character	Range	Mean	CV (%)	Promising lines
Plant height (cm)	71, 63 – 184, 83	112.10	21.46	EC-1, ICBD-18, EC-2, ICBD-15
Fruit length (cm)	5. 12 - 16. 45	12.01	17.5	ICBD-20, ICBD-2, EC-2, ICBD-3, ICBD-7
Fruit girth (cm)	3. 38 - 8. 45	7.35	19	ICBD-13, LCA 427, ICBD-2, EC 1, ICBD-11
Wt. of pericarp (g)	3.11-21.33	6.4	31.26	ICBD-20, Kt-pl-19, ICBD-10, ICBD-8
Seeds/ fruit	39.00 - 104.0	78.80	21.82	ICBD-2, ICBD-7, Sel.2, ICBD-11, ICBD-1
Yield/plant (g)	176.45 - 890.95	455.65	32.86	Sel.2, ICBD-20, ICBD - 8, ICBD-10, Kt-pl-19
Color(ASTA units)	86.20 - 306.50	185.2	24.65	ICBD-10, EC-1, ICBD-11, Kt-pl-19, ICBD-18
Capsaicin (%)	0.0061 - 0.153	0.0845	63.13	ICBD-3, ICBD-19, ICBD-14, ICBD-15, ICBD-4, ICBD-20, EC 1

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**PCR based detection of adulterants in the commercial samples of chilli powder:** Using RAPD/ISSR markers, Ziziphus nummularia ('Jhatber' or 'Choti ber') was identified as adulterant in chilli powder with a molecular size of approximately 400 bp using the primer OPA10 (5'-GTGATCGCAG- 3').

**Characterization:** Eighteen ICBD (Bydagi collections) lines and four exotic collections have been characterized using twenty RAPD primers. The three primers (OPA 4, OPA 5 and OPA 11) generated 21 bands, of which 14 were polymorphic. The number of fragments per primer varied from 6 (primer OPA 4) to 10 (primer OPA 11) with an average of 7 bands per primer. The grouping analysis allowed the separation of 22 germplasm accessions into two major groups.

**Protein profiling of chilli powder using SDS PAGE analysis:** A representative gel picture of the SDS PAGE analysis of protein isolated from different market samples of chilli powder indicated beet root as a possible adulterant.



M - Marker 1 kb ladder. Lane 1- Genuine chilli powder, Lanes 2 to 7 -Market samples, Lane 8 -Zwiphus nummularia, Lane 9 -Red beet pulp, Lane 10- Almond shell powder.

Fig. 6.2. PCR amplified products of DNA isolated from chilli powder (*Capsicum annuum*), market samples of chilli powder and their probable adulterants using RAPD primer OPA15.





Genetic resources Chinese cassia

**Germplasm registration:** Chinese cassia accession A1 (IC370400) has been approved for registration at NBPGR, New Delhi (Registration no. INGR08045) for high cinnamaldehyde content in bark oil (81.5%) and leaf oil (80.5%). Already two more Chinese cassia accessions C1 (IC370415) and D3 (IC370425) have been registered.

### Crop improvement Nutmeg

Field evaluation of elite lines: Among the seven lines, A9/185 recorded the maximum height (438 cm), canopy (459 cm) and girth (37.8 cm) and A9/25, A9/150 and A4/22 were found to be better in bearing.

**Evaluation of grafts for productivity:** Among the grafts on three rootstocks, the performance of nutmeg on *M. malabarica* continued to be better than other rootstocks. Physiological parameters like carbohydrates, starch, amino acids, NR activity, *etc.* did not show any definite trend among different rootstocks. However, the values were higher for A9-69 scions on all the rootstocks when compared to A9-4 scion.

**Evaluation of grafts (plagiotropic v/s orthotropic grafts):** The growth was better in orthotropic grafts but early flowering was observed (for the second year) in plagiotropic grafts, followed by that of seedlings.

**Pruning:** Shoots of nutmeg trees were pruned at monthly intervals from August to January retaining 25%, 50%, 75% and 100% shoots to study the effect of pruning on nutmeg growth and productivity. New shoots were produced from all the pruned shoots but no flowering on any of the newly produced shoots was observed.

*In vitro* multiplication of clonal rootstocks: Shoot tips of *Myristica* malabarica were cultured on SH media supplemented with various hormones for shoot proliferation. Cent percent bud break was observed on SH + 1% TDZ. Callus formation was also observed from stem explants.

Effect of media on germination and seedling growth: Maximum germination percentage was recorded with the treatment combination, soil:granite:FYM (2:1:1), followed by soil:granite:FYM (1:1:1) and granite alone indicating that sand can be substituted with granite powder for raising seedlings of nutmeg. Early germination and initial growth was also highest in the same media combination (soil:granite:FYM at 2:1:1).

Effect of genotype on germination, seedling growth and graft uptake: A4-17 was found to be the best with highest germination (80%), growth and graft success (100%), among the various accessions studied on its own rootstock. Dipping the seeds in GA increased their germination. In another experiment, the graft uptake ranged from 90-76% on the wild rootstock, *Myristica malabarica* and the accessions, A4-22, A9-71 and A9-95 recorded 90% success.

#### Chinese cassia

**Progeny evaluation:** At Peruvannamuzhi, no significant difference was observed for morphological and yield characters, among the four elite lines (A2, C1, D1, D3) evaluated. However, C4 recorded the maximum bark(dry) yield (1000g/plant) followed by D5 (717g/plant) at Appangala (Table 7.1).

#### Cinnamon

### Chemical characterization of cinnamomum germplasm

Eighty two accessions of Cinnamomum verum, collected from IISR Experimental Farm, Peruvannamuzhi, were evaluated for bark oil and oleoresin contents and oil composition. The oil content varied from 0.3% to 3.0%, whereas oleoresin content ranged from 3.3 -15.23%. Maximum oil content was obtained from the accession No. 290 (3.0%) which was followed by accession No 309 (2.3%). Accession nos. 265, 290, 301, 309, 363, 376, 378, 490, 492 and 507 contained more than 10% oleoresin.

The chief constituent of the bark oil was cinnamaldehyde, which varied from 4.5% (Accn No 210) to 59.3% (Accn No. 311). Other major constituents were eugenol (0.99 - 15.26%), benzyl benzoate (1.7-48.7%), t-caryophyllene (2.7-27.7%) and linalool (1.2-26.1%). Accn Nos. 290 and 309 contained 48.4% and 51.6% cinnamaldehyde respectively.

Eighty seven accessions of *Cinnamomum* germplasm were analysed for leaf oil composition by GC-MS analysis. The major constituents of the oil were eugenol (75-98%) and eugenyl acetate (0.11-35.6%) in all the accessions except Acc. no 405. Accession no. 405 was identified as the benzyl benzoate chemotype of *C. verum*, which contained about 76.8% benzyl benzoate.

Essential oil yield and composition of fruits of C. verum and C. malabatrum indicated that both the oils were dominated by linalool during the initial stages of fruit development. However, the concentration of linalool was higher in C. malabatrum (60.5%) compared to that of C. verum (25%). Subsequently the concentration of linalool declined with the advancement of fruit maturity. The essential oils from ripened fruits were rich in cadinene and its analogues (43% in C. verum and 54% in C. malabatrum).

#### Garcinia

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#### DNA profiling of Garcinia spp

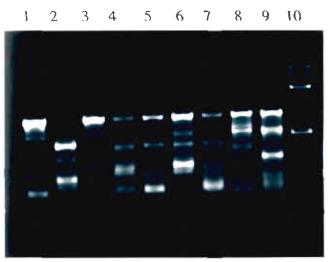
About 20 species of garcinia are found in India of which some important spp. like Garcinia gummi-gutta, Garcinia indica, Garcinia mangostana, Garcinia tinctoria, Garcinia hombroniana etc. are maintained in the tree ree spices

SL. No.	Acc. No	Plant height (cm)	No. of bran- ches	Girth (em)	leaf length (cm)	leaf breadth (cm)	Can- opy (sq.m)	Fresh weight (g)	Dry weight (g)
1	A2	444.3	22	19.25	20.13	5.65	6.26	1268	512.5
2	A6	392.5	16.33	14.5	23.53	6.03	4.73	1216.6	416.67
3	A7	410	16.5	17.58	22.93	5.76	3.84	2380	348.3
4	A8	411.4	18.42	16.42	20.77	6.26	5	1285	450
5	B2	465.0	21	17.71	20.34	5.95	6.85	2082.5	850
6	B4	407.14	20.85	16.57	20.23	5.74	3.39	1613.8	623.75
7	B8	387	19.8	15.2	19.62	5.68	4.24	1076.3	450
8	C1	416.25	19.36	18	20.6	6.08	5.73	2040	608.34
9	C4	407.14	15.86	15.8	19.63	5.73	4.24	1971.6	1000
10	D1	395	20.17	19.5	21.77	5.9	4.47	1233.3	450
11	D2	340.71	13.43	14.86	19.63	5.82	4.31	1438.8	583.3
12	D3	347.14	16.43	14.43	20.4	5.91	3.7	1163.3	400
13	D5	399.3	21.83	18	20.47	6.02	3.98	2150	716.67
14	D6	375.7	16.29	17.86	20.23	5.99	5.14	2583	960
15	D7	369.38	18.63	16.88	21.27	6.19	3.75	1405	750
	Mean	398.12	18.46	16.84	20.7	5.89	4.67	1645.1	626.25
	SD	57.23	3.01	2.66	1.85	0.68	1.25	499.51	163.89
	CV (%)	28.75	32.71	31.61	17.88	23.20	53.73	60.73	52.3
	CD (P=0.1)	NS	NS	NS	NS	NS	NS	1312.2	430.5

Table 7.1. Yield and its attributes of promising cassia lines at Appangala

spices germplasm at IISR. Nine of these species of *Garcinia* were used in DNA profiling based on ISSR and RAPD (Fig. 7.1) markers. A total of nine ISSR primers and twelve RAPD primers could efficiently amplify all the genotypes used in the experiments. Among the nine primers used in the profiling the following primers ISSR-02T, ISSR-3G, ISSR-20T/16, ISSR-17/17, ISSR-20/15, ISSR-24 were able to produce unique bands in PCR. Out of 19 RAPD primers tested 12 gave polymorphic bands in all the species. The study based on RAPD or inter-simple sequence repeat (ISSR) variations revealed close relationship between *G. gummi-gutta* and *G. indica*.

**Isozyme studies in** *Garcinia gummi-gutta* Twenty four natural populations of *G. gummi*-



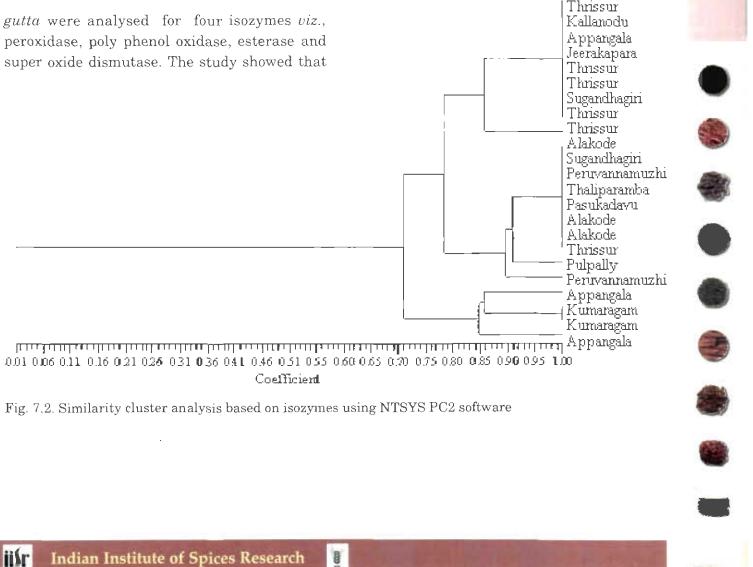
Lane 1- G. malabarica, Lane 2- G. mangostana, Lane 3-G hombroniana(1), Lane 4- G, hombroniana(2), Lane 5- G. tinctoria, Lane 6- Collection AC11a, Lane 7- Collection GM51, Lane 8- G. induca, Lane 9- G. gummigutta, Lane 10- Marker.

Fig. 7.1. RAPD profiling of Garcinia species

the mean percentage of polymorphic loci was 52.5%, which indicate that genetic variability within the population of G. gummi-gutta was not much higher than out crossing wind pollinated woody plants which is 53%, and the result is consistent with the average of tropical tree species which is 60.9%.

**Free spices** 

Cluster analysis revealed low level of genetic separation between the studied populations. There was no significant pattern related to geographic distance, although a tendency was clearly seen. The dendrogram showed that most of populations from the similar geographic locations were the first ones which grouped themselves together (Fig. 7.2).



### Spices - micotoxins, nutraceuticals and economics

### Management of mycotoxin contamination in spices

Storage of spices : Variations in the biochemical parameters associated with aflatoxin contamination in black pepper, ginger and turmeric in relation to existing drying and storage practices were studied. Among the packing materials, minimal fungal population was noticed in 300 gauge polypropylene bags and 3 layer metallised polypropylene bags stored under 100% N, and 100% vacuum (Table 8.1). No significant changes were noticed in the essential oil, oleoresin and moisture contents between the storage treatments, indicating their effectiveness.

Fungi toxic effects of spice essential oils: Extent of inhibition by various plant essential oils, their major components and aqueous fractions of spices on the growth of Aspergillus

Table 8.1. Effective treatments in different spice crops that reduce aflatoxin contamination

Crop	Treatment							
	Packing material	Packing atmosphere	Plant product	Plant materia				
Black pepper	Pet Jar 300 g PPB	2L MP 100% V 3L MP 100% N <sub>2</sub>	Allspice oil Cinnamon oil Clove oil	Curry leaves Cinnamon Garlic				
Turmeric	Pet jar 100 g & 300g PPB 2L&3L MP	2L MP 100% V 3L MP 100% N <sub>2</sub> 300 PPB	Allspice oil Cinnamon oil Turmeric leaf oil	Curry leaves Allspice Garlic Cassia				
Ginger	300 g PPB (upto 6m) 3L MPB (upto 6m)	2L MP 100% V 3L MP 100% N <sub>2</sub> 300 PPB (100% V, 100% N <sub>2</sub> )	Clove oil Turmeric leaf oil	Curry leaves Neem leaves Garlic				
Nutmeg	Pet jar 2L & 3L MP	2L MP 100% V 3L MP 100% N <sub>2</sub> 300 PPB (100% V, 100% N <sub>2</sub> )	No treatment	No treatment				
Mace	300 g PP 2L MP	2L MP 100% V 3L MP 100% N <sub>2</sub>	No treatment	No treatment				

ibr Indian Institute of Spices Research

was quantified in terms of aflatoxin  $B_1$ . Clove oil, cinnamon oil and turmeric leaf oil had complete inhibition of aflatoxin production.

**Isolation and characterisation of aflatoxigenic microorganisms:** Bioassay using chillies and groundnut could identify toxigenic and non toxigenic strains using ammonia exposure method. Out of 70 cultures of *Aspergillus* sp. collected so far, 36 were found to be non-toxigenic and 34 toxigenic. ITS primers, ITS-1 and ITS-2 were used to identify different species of *Aspergillus*. Amplicon sizes ranging from 500 – 1000 bp were obtained.

# Nutraceutical properties of bioactive compounds in a few spices

Black pepper (IISR Thevam), ginger (IISR Rejatha), turmeric (IISR Alleppey Supreme), curry leaves (Sengambu), Garcinia indica, Garcinia gummi-gutta and tamarind (PKM 1) were extracted for their water and ethanol fractions. Only black pepper, ginger, turmeric and curry leaves yielded essential oil. The parameters to assay the antioxidant property of essential oil, water and ethanol extracts of spices in vitro have been standardized and the analysis was carried out. The total antioxidant capacity as analyzed by the Phospho molybdenum method was maximum in the ethanol extracts of Garcinia gummi-gutta and water extracts of turmeric and tamarind. The 1, 1-diphenyl-2-picrylhydrazyl radical scavenging ability was greater in the essential oil of black pepper and ethanol extracts of black pepper, curry leaves and tamarind. Fe (III) to Fe (II) reducing activity was highest in the ethanol extracts of turmeric and the two garcinia species, much more than in the food additives BHA and BHT; black pepper essential oil exhibited the most Fe (II) chelation activity. Ethanol extracts of Garcinia gummi-gutta and turmeric had higher phenol

content. The essential oil was also studied for its composition by GC-MS and no significant difference was found in the quality and quantity of the major constituents. A database of black pepper compounds titled 'PassCom' (Predicted Activity Spectrum of Spice Compounds) has been created for accessing data on structures, MOL formats, activities, effects and mechanisms of compounds.

### Spices as natural food colours

Rinds of Garcinia cowa, G. gummi-gutta, G. hombroniana, G. indica and nutmeg and mace of nutmeg (0.1 g) were used to extract color in two solvents – acetone and ethanol (100 ml). The absorption maxima were read (330 – 346) in a spectrophotometer for acetone extract. The two extractions were subjected to pH (by addition of 1N HCl and 1N NaOH) and temperature (heat), where the heat did not alter the color of the extracts.

### Impact of socio-ecological changes on spices production

Analysis of transition in agricultural landscape in Wayanad district of Kerala reveals that, the traditional crops like black pepper, coffee, cardamom, ginger, rice *etc.* have last their area to other non-traditional crops like coconut, arecanut, banana *etc.* (Table 8.2).

Owing to the changes in socio-economic factors, there is a marked change in the ownership of agri-land and farm size. The average size of holding and share in the total land has come down in the case of large (>10 acres) and medium (4-10 acres) farms. Though there is no change in the size of holding, marginal (<1acre), small (1-2 acre) and semi-medium (2-4 acre) farm categories have improved their share in total number of holdings and total land occupied (Fig. 8.1).

1

Table 8.2. Percent share of crop area during different periods

Period	Pepper	Ginger	Turmerie	Vanilla	Cardamom	Arecanut	Coffee	Rubber	Others'
As per stat	istics:								
1990-91	18.9	4.6	0.1	-	3.1	0	42.4	3.4	24.4
2003-04	22.8	2.4	0.1		2.2	4.7	36.4	3.6	21.8
2005-06	21.5	2.5	na	na	1.97	2.89	32.3	3.1	27
As per surv	vey data	a:							
I (Past)	31.6	8.6	1.1	0.1	0.1	0.5	33.5	7	17.5
II (Present)	28.9	13.1	2.8	2.2	1.7	2.2	24.8	3.9	20.5
III (Future)	21.6	11.2	3.4	3.9	2.8	15.7	16.8	4.6	19.9

\* Includes paddy, banana, tapioca etc.

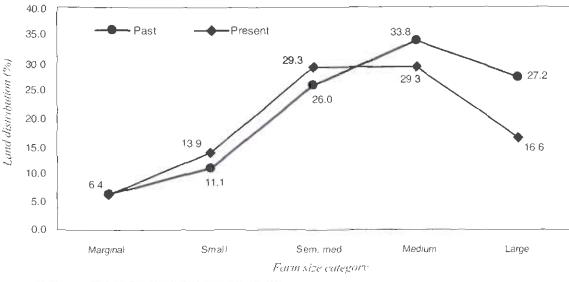
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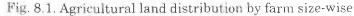
- Income through agriculture is highest in medium size farms in both the periods, while the per capita expenditure is maximum in small farms.
- The partial productivity of land (output per acre) is highest in marginal farms followed by small farms and is lowest in large farms.
- Irrespective of farm size, there is a decline in partial productivity of land in all category of farms between the periods.
- The partial productivity of labour (ouput per worker) is lowest in marginal farms and

is highest in large farms because of more number of workers per unit area.

### Production of nucleus planting materials of spices

During the year, the institute produced 80,000 rooted black pepper cuttings, 5000 cardamom seedlings,1000 cardamom suckers, 11 tonnes turmeric seed rhizomes, 10 tonnes ginger seed rhizomes and 6000 nutmeg grafts. These were distributed to farmers and other developmental agencies.





### All India Coordinated Research Project on Spices

#### Genetic resources

The AICRPS centers are maintaining 6831 accessions of spices germplasm (major spices -2926; seed spices -3841 and other spices - 64) assembled from indigenous and exotic sources in *ex-situ* gene bank.

#### Black pepper

A total of 691 black pepper germplasm accessions have been maintained under different AICRPS centres. Dapoli and Pundibari centres have collected two and seven new germplasm accessions from Ratnagiri and Sub-Himalayan region, respectively. Trials at Panniyur and Sirsi revealed maximum yield per vine with the application of biofertilizers (Azospirillum @ 50g/Phosphobacteria @ 50g) + FYM 10 kg along with recommended dose of NPK.

#### Cardamom

Pampadumpara and Mudigere centers together maintain 305 germplasm accessions of cardamom. Biofertilizer trial conducted in cardamom revealed superior performance of the treatment, inorganic N (100%) + Azospirillum (50 g) + 5 kg FYM, for yield. Application of neem cake @ 0.5 or 1.0 kg/plant significantly reduced the shoot and capsule borer damage and resulted in significant yield increase both at Pampadumpara and Mudigere.

#### Ginger

A total of 660 ginger germplasm accessions are maintained under AICRPS centers. The CVT trial at Pundibari showed maximum yield of 20.34 t/ha in Gorubáthan compared to the check Acc. 117 (12.95 t/ha). The experimental results at various centers revealed that soil and foliar application of Zn, Bo and Fe recorded significant increase in yield and quality parameters. At Pundibari, the treatment combination of soil application of 10 kg borax, 25 kg zinc sulphate and 10 kg ferrous sulphate per hectare recorded highest fresh rhizome yield of 35.17 t/ha, 1.33% essential oil and 6.52% oleoresin. Study on rhizome rot of ginger revealed the effectiveness of seed treatment (mancozeb 3 g/l + carbendazim 1g/l + chlorpyriphos 2 ml/l for 30 min) and soil application of thimet 10G (1 kg ai/ha) for highest fresh rhizome yield (17.96 t/ha).

#### Turmeric

Turmeric germplasm (2280 accessions) is maintained by eight centers under AICRPS. The biofertilizer experiment at Coimbatore revealed inorganic N (50%) + Azospirillum (5 kg/ha) + 5 t FYM recorded the highest yield. Kumarganj, application of 50%At. recommended dose of inorganic fertilizer (60:40:40 kg/ha NPK) + 50% FYM (10t/ha) + 5 kg/ha Azospirillum + seed treatment and soil application of Pseudomonas fluorescens and Trichoderma @ 50 g/m<sup>2</sup> recorded maximum fresh rhizome yield (34.01 t/ha). Recommended dose of NPK + FYM + seed treatment and soil application of consortia of Trichoderma viride and Pseudomonas fluorescens @ 4g/kg and 12.5 kg/ha and 25 kg/ha as basal and top dressing respectively was found to be effective for control of rhizome rot of turmeric with cost : benefit ratio of 1:2.8 at Coimbatore.

#### Tree spices

8

A total of 37 clove, 119 nutmeg, 39 cinnamon and 6 cassia germplasm accessions are maintained under AICRPS centres. Dapoli and Peechiparai. ICRPS

AIGRPS

National group meeting (XIX workshop) of research workers of AICRPS on spices The National group meeting (XIX workshop) of research workers of AICRP on spices was held at Orissa University for Agriculture and Technology (OUAT), Bhubaneswar, Orissa during 23-25 November 2007. It was inaugurated by Prof. D. P. Ray, Vice Chancellor, OUAT, Bhubaneswar. The group meeting was attended by over 100 scientists from 19 AICRPS centers. Dr K.V. Ramana, ADG (Hort. II), ICAR, New Delhi, Dr V.A. Parthasarathy, Director, IISR, Dr B.B. Vashistha, Director, NRCSS and Dr M. Anandaraj, Project Coordinator, Spices led the three day delebrations on genetic resources, crop improvement, crop production and crop protection of different mandate spice crops. The varietal release proposal of leafy type coriander variety DH – 228 from CCS HAU, Hisar was recommended for Haryana state release. The plenary session was chaired by Dr H.P. Singh, DDG (Hort.), ICAR, New Delhi on 25 November 2007 and highlighted the roadmap for future spices research in the country.

### Krishi Vigyan Kendra

#### Training programmes

The KVK conducted 68 training programmes on various subjects during the period under report. A total of 2056 persons have benefited from the programmes. The details of the training programmes are furnished in Table 10.1 & 10.2.

In addition, training programmes were organised for women SHG members on vermicompost production and utilisations and also on mushroom cultivation.

### Vocational training programmes

**Repair of farm implements:** The Kendra conducted one long duration (three months) vocational training programme for rural youth on 'repair and maintenance of farm implements'. The programme was organised in collaboration with the Government Community Polytechnic College, West Hill, Calicut. A total of 20 unemployed rural youth were benefited from the training.

**Gardeners' training programme:** A Gardeners training programme of six month

85

#### Table 10.1. Training programmes conducted during the year 2007-'08

SI.	Category	No. of	No	No. of SC/ST		
No.		courses	Male	Female	Total	participants
1	Practising farmers, farm women and rural youth	60	1348	641	1989	192
2	Extension functionaries	8	127	66	193	0
	Total	68	1475	707	2182	192

#### Table 10.2. Discipline-wise training programmes conducted

Sl. No.	Discipline	No. of courses	No. Male	of Particip Female	oants Total	No. of SC/ST participants
1	Crop Production	32	749	185	934	59
2	Horticulture	3	40	26	66	11
3	Animal Science	23	<b>49</b> 0	410	9.00	86
4	Agricultural Engineering	10	196	86	282	36
	Total	68	1475	707	2182	192
iiśr	Indian Institute of Spi	ces Research		A Participant		51 JA 2

duration was started with the assistance of State Horticulture Mission in two batches of 25 each.

### Coconut Development Board scheme on integrated farming in coconut holdings on cluster basis

The KVK is associating with Coconut Development Board to implement a scheme on integrated farming in coconut holdings on a cluster basis at Koothali panchayat. Twenty five hectare area was selected under the scheme covering 56 farmers.

#### Revolving fund programme

The Kendra has a revolving fund programme to generate income for productive uses. Under this programme, quality planting materials of various crops are produced and made available to public at affordable rates. Presently, bush pepper, allspice seedlings, garcinia grafts, mango grafts, guava layers, arecanut seedlings *etc.* are produced under this programme. The closing balance of revolving fund as on 31.3.2008 was Rs.1.34 lakh. During the period an amount of Rs. 2.58 lakh has been realised through sale of planting materials, chicks and the activities of Plant & Animal Health Centre.

#### Other extension activities

**Kisan mela and exhibitions:** KVK is regularly participating in exhibitions and seminars within the district. During the period under report, KVK participated in the following exhibitions; a) Dist. Agrl. Meet, Dept. of Agriculture, Koduvally, Calicut. b) Agri-Fair 2007, Gandhiji Study Centre, Thodupuzha. c) Exhibition at Govt. VHS, Vadakara. d) Calicut Flower show 2008, Agri-Horti Society, Calicut. e) National Biodiversity Exhibition, Malabar Botanical Garden, Olavanna.

Farmers' study tours: A team of 30

progressive farmers of volunteers vikas vahini (VVV) club, Chakkittapara visited Assam, Meghalaya, Tripura and Nagaland during 3.6.2007 to 18.6.2007 under financial assistance from National Horticulture Board.

**Seminars:** The KVK conducted 4 seminars during the period. The topics covered were organic farming, value added products from coconut, diseases of cattle and control measures and infertility management.

### Maintenance of demonstration blocks/ units

The KVK maintained model demonstration hlocks/units in its farm for 'seeing and learning' by the farmers. Some of the demonstration blocks maintained are medicinal plant unit, model homestead garden, demonstration plot of improved varieties of black pepper, model arecanut seed garden, nutmeg and mango scion banks, dairy, poultry and quail demonstration units *etc*.

#### Human resources development

HRD activities undertaken by KVK is listed in Table 10.3.

#### **Technologies** transferred

FLD programme: This is a new concept of field demonstration and the main objective is demonstrate newly released crop to production and protection technologies and its management practices in farmer's fields under different agro-climatic regions and farming situations. In this programme, technologies are demonstrated for the first time by the scientists themselves, before being fed into the main extension system of State Department of Agriculture. These programmes are carried out the co-operation with and complete participation of progressive farmers under the direct supervision of KVK scientists. A part of

### Table 10.3. Staff deputed for training/ workshop

Name of the person	Title of the programme	Period of training	Place of training
K.M. Prakash and S. Shanmugavel	Action plan meeting of KVKs	12.4.07 to 13.4.07	KAU, Trichur
K.M. Prakash and S. Shanmugavel	One day workshop on documenting achievements of KVK	31.5.07	TNAU, Coimbatore
K.M. Prakash	Training cum workshop on Agmark network	20.11.07 to 22.11.07	KAU, Trichur
K.M. Prakash	Seminar on medicinal plants-strategies for conservation	4.12.07	Aryavaidyasala, Kottakkal
S. Shanmugavel	Hygienic meat handling system	12.3.08 to 14.3.08	TANUVAS, Chennai
K.M. Prakash	Workshop on zingiberaceous spices	19.3.08 to 20.3.08	IISR, Calicut

1

the expenditure of this demonstration is met by KVK.



Bush pepper production technique-training to women SHGs

**OFT programmes:** The major OFT programmes carried out during the period are listed below:

- 1. Effect of *Pseudomonas fluorescens* and vermicompost in foot rot management of black pepper.
- 2. Management of sigatoka leafspot of banana using *Pseudomonas fluorescens*.
- 3. Broiler goat production.
- 4. The effect of treatment with a GnRH analogue on conception rate in repeat breeder cows.
- 5. Fertility management in repeat breeder cows following double PGF2 alpha injection.

### **Agricultural Technology Information Centre**

The Agricultural Technology Information Centre (ATIC) is coordinating the extension and training functions of the institute. It is involved in technology dissemination functions through a single window, coordinating with various divisions of IISR. The major activities carried out by the centre are

- Production and distribution of quality planting material and printed literature
- Farm advisory services including crop diagnostic services
- Information dissemination through multimedia, and interactive databases
- Providing audio visual aid support to the institute activities
- Organising technology dissemination services like exhibitions and seminars to farmers and other users

#### **Milestones and Achievements**

**Technology inputs and Advisory services:** The technology inputs distributed from the centre include quality planting material of improved varieties of spices, bio control agents, vermi compost, fresh whole spices and scientific publications including extension literature. During the year 2007-08, planting material worth Rs 51415 was distributed from

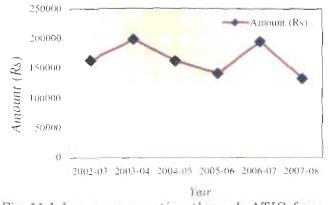
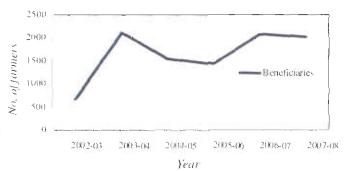


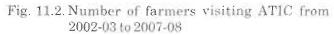
Fig. 11.1. Income generation through ATIC from 2002-203 to 2007-08

the centre. The proceeds from sale of publications amounts to Rs 36843. The details of income generation from the centre over the years is furnished in Fig. 11.1. Eight hundred and twenty seven farmers who visited the centre were benefited from advisory services. This includes 273 farmers from within Calicut district, 170 farmers from outside Calicut district and 384 farmers from outside the state. Pattern of advisory services is given in the Table 11.1. The total visits recorded in the centre over the years is furnished in Fig. 11.2. One thousand and one hundred and forty four students from various educational institutions visited the centre. An amount of Rs 5750 was realized through soil and manure sample analysis. Trichoderma and other bio agents formulations for Rs 16478 were distributed to farmers.

### Table 11.1. Pattern of advisory services

Purpose of visit	Frequency score
Planting material	192
Scientific information	514
Bio inputs	49
Diagnostic services	13
Publications	54





### **Education** and **Training**

#### Post graduate studies

#### Ph.D

**P. Sumathi,** Studies on somaclonal variations in zingiberaceous crops. University of Calicut.

**A.K. Lincy,** Investigations on direct *in vitro* shoot regeneration from pseudostem explants of ginger and its field evaluation. University of Calicut.

#### M.Phil

**V. Vinod**, Biochemical and Molecular characterization of black pepper (*Piper nigrum* L.) pericarp degrading bacteria. Acharya Nagarjuna University.

#### M.Sc projects

Twenty two students from various universities undertook their M.Sc project work in Biotechnology, Biochemistry, Microbiology and Plant Pathology under the guidance of the scientists of the institute.

#### Post M.Sc Training

Four post M.Sc candidates under went hands



Trainces listening to lecture by IISR scientists at Nagaland

on training in various techniques of Microbiology and Biotechnology.

#### M.Phil projects

Two students undertook their M.Phil project work in Biotechnology under the guidance of the scientists of the institute.

#### Summer training for M.Sc students

Twenty nine students from various universities attended the summer training programme on Techniques in Biochemistry and Biotechnology. Thirteen students from various universities attended the summer training programme on Techniques in Bioinformatics.

# Trainings / Meetings organized by the Institute

Summer training programme on Techniques in Biochemistry and Biotechnology for M.Sc students, May 07 to June 06, 2007.

Summer training programme on Bioinformatics for M.Sc students, May 07 to June 06, 2007.



Demonstration of ginger planting by IISR scientists at Nagaland

**Education & Training** 

Training programmes attended by staff

Name of the staff	Name of the training	Duration	Organized by
T J Zachariah	IPR and WTO related issues	July 30- Aug 03, 2007	ASCI, Hyderabad
D Prasath	Intellectual property management and technology transfer	May 19-22, 2007	ISTM, Goa
T E Sheeja	First training cum workshop on intellectual Property and technology	May 28-30, 2007	NAARM, Hyderabad
	management in ICAR system		
Senthil Kumar	Capacity building for risk analysis and modeling to promote trade	July 23 to Aug 02, 2007	ANGRAU, Hyderabad
C N Biju	Capacity building for risk analysis and modeling to promote trade	July 23 to Aug 02, 2007	ANGRAU, Hyderabad
M Anandaraj	Management development programme in agricultural research	Aug 24-30, 2007	NAARM, Hyderabad
K Jayarajan	Networking essentials for information management in agriculture	Oct 16-25, 2007	NAARM, Hyderabad
Utpala Parthasarathy	Climate change, crop yield and sustainability	Feb 15 to March 07, 2008	IARI, New Delhi
T E Sheeja	IPR and WTO related issues	Feb 18- 22, 2008	ASCI, Hyderaba
B Chempakam	Capacity building for IP protection and technology licensing in agriculture	Feb 18-20, 2008	KAU, Thrissur
D Prasath	Advances in biometrics	Feb 8 -22, 2008	IASRI, New Dell
A I Bhat K N Babu	Protection of transgenic crops under PPVFRA with a focus on Bt cotton	13-Feb-08	IISc, Bangalore
A I Bhat	Management and monitoring of field trials of genetically modified crops	29-Mar-08	EPTRI, Hyderabad

**Education & Training** 

Internet security in Government offices, 12-13 November 2007.

XIX National group meeting of research workers of AICRP on spices held at OUAT, Bhubaneswar, 23-25 November 2007.

National Symposium on Spices, Medicinal and Aromatic Crops (SYMSAC IV) held at OUAT, Bhubaneswar, 25-26 November 2007.

First meeting of the Technical Advisory Group: "Task Force 7/2007" to finalize the draft of DUS test guidelines for black pepper, cardamom, ginger and turmeric, 29-30 October 2007.

Second meeting of the Technical Advisory Group: "Task Force 7/2007" for preparation of DUS test guidelines for spice crops – cardamom, black pepper, turmeric and ginger, 5-6 December 2007.

National Workshop on Zingiberaceous Spices: Meeting the growing demand through sustainable production, 19-20 March 2008

Workshop on Spices Production Technology at Nagaland under technology mission for integrated development of spices in North Eastern States from 25-27 March 2008



Inauguration of DUS Test Centre for Spices at IISR

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### Visits Abroad S. Devasahayam

First Meeting on Integrated Pest Management for Pepper, Jakarta, 26–27 April 2007

Second Meeting on Good Agricultural Practices for Pepper, Jakarta, 24–25 April 2007

#### Awards

Dr. H.S. Mehta Memorial Award for Best Research Paper- awarded to Utpala Parthasarathy, Jayarajan K., Johny, A.K. and Parthasarathy, V.A. for best Oral presentation entitled " Identification of Sutiable areas and effect of climate change on ginger-A GIS study" presented at National Symposium on Spices, Medicinal and Aromatic Crops on 'Threats and solutions to spices and aromatic crops industry', held OUAT, Bhubaneswar, 25-26 November 2007.

Alapati Prasada Rao Award 2007 was awarded to Bhadramurthy, V., Bhat, A.I., Jasmine George, Thankamani, C.K. and Mathew, P.A. for the best poster entitled 'Variation in the concentration and indexing black pepper plants for viruses through DAS-ELISA' presented at National Symposium on Spices, Medicinal and Aromatic Crops on 'Threats and solutions to spices and aromatic crops industry', held OUAT, Bhubaneswar, 25-26 November 2007.

**Dr J S Pruthi Award 2007** was awarded to Venugopal, M.N., Prasath, D. and Ravindra Mulge for the best research paper entitled "IISR Avinash – a rhizome rot resistant and high yielding cardamom (*Elettaria cardamomum* Maton)" in the Journal of Spices and Aromatic Crops 15:14-18 (2006). **Education & Training** 

# Annual Report 2007-08 Bioinformatics Centre

#### **Research** activities

### Development of database of plant associated bacteria and software

**PLASBID:** A database on plant associated bacteria integrated with several tools like sequence editing, primer analysis *etc.* was developed and launched. This database is available online (www.spices.res.in/plasbid).

Druggability of spice compounds :

Biological properties and druggability of compounds in black pepper essential oil were described using *in silico* tools. PASSCOM, a database on predicted activities of spice compounds is being developed. The module on black pepper has been completed.

**PLASBID** 

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**EST analysis of ginger and turmeric :** Expressed Sequence Tags (ESTs) available in public databases were compiled, clustered and their functional annotation was attempted using *in silico* tools. Microsatellites and SNPs available in both the crops were also found out using the EST clusters. Quality primers were designed for identifying microsatellites in ginger and turmeric.

#### Training activities

**Summer training :** A summer training programme in Bioinformatics was organized from 07 May to 06 June 2007 in which 13 M.Sc. students participated.

Bacterna associated with plants are diverse in their ability to affect plant health, their genotypic and phenotypic characteristics and their phylogeny. These bacteria are typically members of complex microbial communities, with only a few establishing pure clonal populations within a plant. Majority of research on plant associated bacteria has focused on phytopathogens and diazotrophic photosymbions. Increased interest in the diversity of organism associated with plant has induced developement of several tools to assess their diversity. It is clear that many plant associated microbes, even those that comprise only a small proportion of a community, can have functions that are of agricultural or environmental importance. Technical advances in microbial ecology and genomics have been paralleled by advances in our understanding of the structure and dynamics of these

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PLASBID is an attempt to consolidate information available on bacteria reported to be associated with plants. It contains nucleotide sequences belonging to 165 rRNA. The sequence information, G + C content of each sequence are given, and also a link to NCBI database for additional information. You can also see the bacterial taxonomy and their salient features in brief

microbial communities and in the molecular basis of plant microbe and microbe- microbe interactions

The sequence level identification of the bacterial species can be done with the Bacterial Identification Tool It does a sequence similarity search using BLAST and outputs the results based on the percentage of similarity The sequence provided by the user is searched against PLASBID 16S rRNA nucleotide database and identifies

Home page of PLASBID, the database on plant associated bacteria

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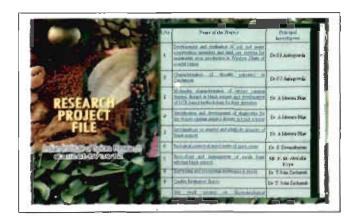
## Agricultural Research Information System (ARIS)

As per the mandate given to ARIS cell, the cell at this Institute has routinely attended to the proper maintenance of the computers and local area network. It has established a new wireless/optic fiber connectivity to the silver jubilee hall, bio control lab, scientist hostel and guest house. It serves as a service centre for various purposes like internet browsing, statistical analysis of data and documentation. It also extended its technical support for conducting video conferencing in satellite enabled village resource expert centre.

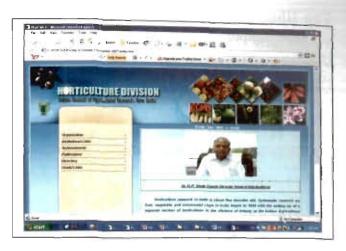
ARIS maintained and updated on regular basis information on all the staffs of the institute by accessing and editing PERMISnet of ICAR and given training to all the dealing officials for using the IRS software and regularly updating the intelligent reporting system of ICAR.

Besides ARIS has developed the following.

'Research Project File ': An e-book for completed research projects of the Institute (RPF III).



Website: Developed the Horticulture Division (ICAR, New Delhi) website.



Engagement diary: Prepared an engagement diary for Dr. H.P. Singh, DDG (Hort.)



Software: As per the instruction of RAC members, a software providing information about Technological inputs and services from ATIC was developed and installed it in the kiosk.



8

**IRIS** 

Library

The library of the institute was set up to provide support to research activities of the institute and to function as a national retrieval information storage, and dissemination system for spices and related areas. At present, it has a collection of 4311 books, 3451 bound volumes, 2305 reprints, 906 technical reports, 183 CDs, 127 theses and 153 project reports. The library is subscribing to 36 foreign journals and 84 Indian journals in addition to two CAB CD databases (Horticultural database and Plant protection database) and J Gate (Agricultural and biological sciences), the e journal portal. The library provides bibliographic services published in Journal of spices and Aromatic Crops and database services apart from publishing "Agri-Science Tit Bits" at quarterly intervals. The additions during the year include 133 books, 8 technical reports, three

theses, 15 project reports and six reprints.

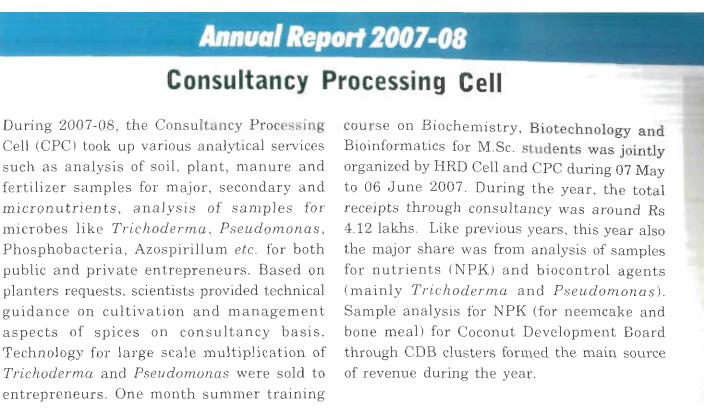
Sharing of resources between the libraries of Central Plantation Crops Research Institute, Kasaragod, National Research Center for Cashew, Puttur and IISR, Calicut, was continued. Access to online journals and content page service was provided through library website (SpicE) to the respective users. Links are also provided to online journals and open access journals in the web site. Internet facility is provided in the library for literature search, web search, checking e-mails etc. During the year National Agricultural Innovation Project, ICAR provided free access of three databases SCOPUS, Science Direct and Annual Reviews. The institute library was shifted to the new library building and was inaugurated by H.P. Singh, DDG (Horticulture) on 18th March 2008.



Annual Report 2007-08

Library

H.P. Singh, DDG (Hort.) inaugurating the new library building



### Women Empowerment Programme

International women's day was observed on 14<sup>th</sup> March 2008. Mr.Vinish, Art of Living Course was the chief guest. His speech focused on tension free life. Women's cell also joined hands with recreation club during annual day celebration by arranging food stall and fun games. During Onam celebration organized in the month of September 2007, a get together was organized by the women's cell.



International women's day celebrations

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Women Empowerment / CPC

### **Official Language Implementation Activities**

Quarterly meetings of official language implementation committee (OLIC) were held four times, during the year 2007-08. During this period, four hindi workshops were conducted on noting and drafting. Four quarterly progress reports and one annual report on official language implementation were prepared.

Dr. V. A. Parthasarathy, Director attended the orientation programme in hindi for the heads of member offices of TOLIC, Kozhikode on 30.05.2007. Sri. B. Krishnamoorthy, and Ms. N. Prasannakumari attended TOLIC meetings held on 31.05.2007 and 17.12.2007.

Hindi Day/Hindi Week was inaugurated on 14.09.2007. Various hindi competitions (hindi song, extempore speech, fun games, memory test, paragraph reading, noting and drafting, calligraphy, anthakshari, quiz *etc.*) were conducted during the Hindi Week. Prizes and certificates were distributed during the valedictory function on 24.09.2007. Dr. K. Balakrishnan, Deputy Director (Impl.) Regional Implementation Office, Cochin visited the institute on 02.11.2008 and inspected the official language implementation activities. IISR Experimental Farm and Krishi Vigyan Kendra, Peruvannamuzhi, also have been taken under the scheme of official language implementation.

During the year, nine staff members passed Pragya examination in May 2007 conducted by Hindi Teaching Scheme, Calicut. Hindi versions of the half yearly publication of Spices News volume 18(1) January - June 2007 and 18(2) July-December 2007 were published as Masala Samachar. Executive summary of project coordinator's report and executive summary & preface of annual report were translated in hindi and incorporated in annual reports of AICRPS and IISR, Calicut. Research Highlights 2006-07 was translated and published in hindi separately. Hindi word /phrase and the equivalent english word/ phrase were displayed both on white board and in the IISR website.

### Institute Management Committee

- Dr. V A Parthasarathy Director IISR, Calicut – 673 012
- 2. Dr. S Devasahayam Head, Division of Crop Protection IISR, Calicut – 673 012
- Sri. P A Mathew Scientist-in-Charge IISR Farm, Peruvannamuzhi
- Sri. Sanjay Mariwala 26/2802,Vidya Vihar Road Thevara, Cochin – 682 013
- The Associate Director of Research Kerala Agricultural University Regional Agrl. Research Station Mele Pattambi (Post) Palakkad – 679 306, Kerala
- Sri. K Mukundan Kadaly House, Kadalamad P O Vaduvanchal, Wayanad – 673 581
- Dr. George V Thomas Director, CPCRI Kasaragod – 671 124, Kerala

- Dr. K V Nagaraja Principal Scientist NRC for Cashew, South Kanara Puttur – 574 202, Karnataka
- The Additional Director of Agriculture (CP) Directorate of Agriculture Government of Kerala Thiruvananthapuram - 33
- The Finance & Accounts Officer Central Rice Research Institute Cuttack – 753 006, Orissa
- 11. The Commissioner of Horticulture & Plantation Crops Government of Tamil Nadu Chennai – 600 009
- 12. Dr. Umesh Srivastava Asst. Director General (Hort. II) ICAR, Krishi Anusandhan Bhavan – II Pusa, New Delhi – 110 012
- 13. Sri A P Sankaran,Asst. Administrative OfficerIISR, Calicut 673 012

# **Research Advisory Committee (2007-10)**

RAG

98

<b>Dr. N. Mohanakumaran</b> (Formerly Director of Research, KAU) Chirakkara Palace Pappanamcode Thiruvananthapuram – 695 018	Chairman
<b>Dr. Umesh Srivastava</b> Asst. Director General (Hort. II) ICAR, Krishi Anusandhan Bhavan – II New Delhi – 110 012	Member
Dr. I. Irulappan, Formerly Dean Tamil Nadu Agricultural University 4-1-121A, Rammunni Nagar T-Kallupatti – 625702 (Tamil Nadu)	Member
Dr. M. Udaya Kumar Formerly Professor & Head Department of Crop Physiology University of Agricultural Sciences G.K.V.K. Campus Bangalore – 560065 (Karnataka)	Member
Dr. K.U.K. Nampoothiri Project Director M.S. Swaminathan Research Foundation (Formerly Director, CPCRI) Community Agro-Biodiversity Centre Puthoovayal P.O. Kalpetta – 673 121	Member
<b>Dr. N.S. Rao (Formerly Principal Scientist)</b> N-109, Innovative Natura Vinayaka Layout, Puttanahalli Yelahanka, Bangalore – 560 064	Member
Dr V.A. Parthasarathy Director Indian Institute of Spices Research Calicut - 673012, Kerala	Member

iiir Indian Institute of Spices Research

### Members of IMC (Two)

Members

**Member Secretary** 

RAC

Dr B. Chempakam

Head, Division of Crop production & PHT Indian Institute of Spices Research Calicut 673012, Kerala



RAC meeting in progress

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#### Annual Report 2007-08 **Recommendations of RAC 2008 RAC Recommendations Action initiated** SL No. Recommendation Areas from which collection of spices germplasm Unexplored areas for black pepper: 1. are to be specified and carried out after making For Piper species: Goodrickal forest range of 1. a gap analysis. Time frame for completing Pathanamthitta district, Periyar WLS and Western Ghats forest of Kasaragod district (Subject to cataloguing work need to be indicated. approval from the forest department, Govt. of Kerala). 2. For cultivated black pepper: Coastal regions of Alleppey district, Perumbavur, Kunnathunadu, Vellidippara, Arakulam. 3. For wild species: Karwar, Bababudin hills, Yellapur, Bandipur, Bhadravathi and Kudhremukh (Karnataka), subject to approval from forest department). Tamil Nadu: Kolli hills, Kodaikanal, Yercaud and 4. Sheveroy hills (Subject to approval from forest department). 2. Characterization of existing spices germplasm This will be taken up and completed in three years. Range (2007-08) has to be completed in three years, and notables will be given. using minimal descriptor. While presenting, range and notables may be given. The range of ginger oil content has to be The genotypes will be grouped into Low: < 1%, Medium: 3. indicated into three categories (low, medium, 1.1-2%, High: >2%. high) and then the genotypes are to be grouped The work so far done has identified 13 ginger accessions (Acc. 50, 57, 95, 99, 156, 162, 197, 209, 217, 225, 228, 411 and 420) as having above 2% oil. An elite accession 246 is registered as high oil type (above 3%). The released variety IISR Rejatha is also having >2% oil. Other collections like Kakkakalan, Kozhikalan and Ellakallan are also rich in oil (>2%). However year to year fluctuation in oil content is also noticed. 4. Germplasm for spices may be established at Bush peppers of important accessions will be established alternate site in IISR- main campus, as bush from bearing vines at main campus. pepper. Donors for specific characters from the 5. Accessions 4216, 4226, 1228, 1343, Kottanadan 1495 1. germplasm are to be listed for further and 1315 were found to be relatively tolerant to utilization. drought during initial screening. 2. Four cultivated (Acc. C 4163, 4175, 1090 and 334) and 4 wild (Acc. 3219, 3286, 3287 and 3311) accessions to root knot nematode and 5 wild (3141, 3200, 3283, 3291 and 3299) and 1 hybrid (HP 309) were resistant to Radopholus similis in preliminary screening. P24, hybrid lines HP-1, HP-3, HP-780, Coll. 339 and 3 P-1534 showed a tolerant reaction to P. capsici. Four accessions of cultivars (Acc. 816, 841, 1084,1114), six wild Piper species and one wild P. nigrum (Acc.270) were resistant to pollu beetle. ibr Indian Institute of Spices Research 100

- 6. A list of elite germplasm material in each crop which needs to be procured from other countries may be prepared and sent to NBPGR for further action.
- 7. A status report may be prepared to ascertain the availability of germplasm materials in each of the mandate spice crops along with place, status of collection and gaps in collection. Characterization/evaluation and conservation may also be indicated in the status report.
- Mega Project II: Breeding improved varieties of spice crops for yield, quality, drought and resistance to pests and diseases. The project title may be modified as "Breeding improved varieties of spices for yield, quality and tolerance to biotic and abiotic stresses".
- 9. Target yield should be specified for breeding high yielding spice varieties. Also indicate the expected yield from the first year of bearing to the yield stabilization in the case of perennial spices.
- 10. The exact nature of sexual incompatibility between *P. nigrum* x *P. colubrinum* needs to be defined and reasons for the same may be investigated.
- 11. The performance of released varieties in high altitude conditions in Wayanad and Idukki districts may be evaluated with the help of willing NGOs in these two districts.
- The possibility of defining ideotypes for black pepper and cardamom may be presented as a status paper in the next RAC.
- Reasons for alternate bearing in black pepper may be worked out.
- 14. Screening of ginger and turmeric for disease resistance should be taken up in "sick plots".

4. CLTP 122, CLTP 07, CLTP 24 and CLTP 123 are lines having high caryophyllene content.

The list of various spices materials to be procured from different countries, along with the addresses from where these can be procured has been sent to NBPGR, New Delhi.

A status report will be prepared regarding the genetic resources of spices, including status of collections and gaps in collection, conservation, characterization and evaluation of the mandate spice crops.

The title will be modified as suggested.



Recommendation

Black pepper:

First year of bearing (400g- 1kg, fresh)

Yield at stabilization (fifth year of bearing) (4-5 kg, fresh). Cardamom:

Yield starts from second year after planting (500 kg/ha) and stabilizes on fourth year (1000 kg/ha). *Clove:* 

The tree starts bearing from  $6^{th}$  year after planting (300g dry) and stabilizes at  $15^{th}$  year (8kg dry).

*Nutmeg:* Starts bearing from 6<sup>th</sup> year after planting (100 fruits) and stabilizes at 20 years (2000 fruits/tree).

Reasons for incompatibility between *P. nigrum* and *P. colubrinum* would be investigated.

Suitable NGOs will be tracked and the performance of released varieties in high altitude conditions in Wayanad and Idukki districts would be evaluated with the willing NGOs.

The available data on black pepper and cardamom will be compiled to identify crop and environmental factors influencing yield for defining ideotypes for these crops.

This will be included as one of the technical programme under the new project "Investigations on factors controlling spiking in black pepper".

A 'sick plot' would be established during the current year for screening germplasm of ginger and turmeric for disease resistance.

- 15. The project on Rootstock scion interaction in tree spices may be shifted under the Mega Project VI: Propagation.
- 16. Efforts may be made to redefine the genus *Curcuma*.

**RAC Recommendations** 

102

17. The performance of released varieties in high altitude conditions in Wayanad and Idukki districts may be evaluated with the help of willing NGOs in these two districts.

 Cost benefit ratio of organic farming in spices has to be worked out. Will be shifted to Mega Project VI: "Propagation studies in spice crops".

Molecular genetic fingerprints of sixteen Curcuma species were developed using Inter Simple Sequence Repeats (ISSR) and Random Amplified Polymorphic DNA (RAPD) markers to elucidate the genetic diversity/relatedness among the species. Cluster analysis of data using UPGMA algorithm placed the sixteen species in seven groups that are somewhat congruent with classification based on morphological characters proposed by earlier workers. However, the study also warrants the limitations of the conventional taxonomic tools for resolving the taxonomic confusion prevailing in the genus and suggests the need of molecular marker in conjunction with morpho-taxonomic, cytologic studies while revising the genus, which is currently in progress. The maximum molecular similarity observed between two of the Curcuma species namely C. raktakanta and C. montana is suggestive of the need for relooking the separate status given to these two species. Further, the status of C. montana and C. pseudomontana, the two species mainly discriminated based on the presence of sessile tubers also needs to be reassessed. More studies using 18 S rRNA is in progress.

The data generated at AICRPS centre Pampadumpara will be used for this purpose for Idukki. For Wayanad, a separate study has to be taken up.

The cost benefit of different management systems were worked out. The total costs of cultivation for ginger are Rs. 163996, Rs. 125031, Rs. 148564 under organic, inorganic and integrated systems respectively. The integrated system recorded the highest B/C ratio of 2.05 with a profit of Rs. 156236 equal to that of chemical system that recorded a B/C ratio of 2.02 with a profit margin of Rs. 126969, when the fresh produce is considered with a price of Rs. 15 per kg. The organic system recorded the lowest B/ C ratio of 1.67. If a 20% premium price is considered for the organically grown produce, the B/C ratio of organic system will be 2.01, comparable to other two systems. When the returns and profit based on dried produce are calculated, the returns are worked out to be very less as the prevailing product price is very low (Rs. 60.00 per kg of dried ginger) and hence the B:C ratio was low under all the three systems like 1.34, 1.61 and 1.64 for organic, chemical and integrated systems, respectively. Similarly, for turmeric the cost benefit ratios were 1.6, 1.87 and 1.83 under organic, inorganic and integrated systems respectively on fresh basis (with a price of Rs. 10 per kg). If a premium of 20% was considered for the organic system, it showed a benefit up to 1.92. The returns and profit based on dried produce are calculated and the B: C ratio was low under all the three systems like 0.96, 1.12 and 1.10 for

- 19. Studies on rhizosphere competition, nutrient removal and recycling of different standards in black pepper may be carried out. Allelopathic effects may also be investigated.
- 20. Develop sustainable production system with minimum cultural interventions for maximum returns.
- 21. The number of accessions to be screened for drought tolerance in the next year may be specified.
- 22. Collect information on the value added products of spices with commercial value.
- 23. Formulate research projects for developing machinery for post harvest handling and processing of spices in collaboration with CIPHET, CIAE & Spices Board.
- 24. Develop dryers that do not lead to reduction in oleoresin and oil content.
- 25. Modify project title of Mega Project VI as "Propagation studies in spice crops".
- 26. Explore the possibility of establishing a progeny pepper plantations among those farmers who have taken the nucleus planting materials from IISR.
- 27. Collect data about distribution of IISR varieties and their performance from those who have taken more than 500 cuttings.
- 28. Survey for viral diseases of black pepper should be undertaken in farmers' fields who took virus-

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Indian Institute of Spices Research

8

organic, chemical and integrated systems, respectively (Rs. 30.00 per kg of dried turmeric).

This has been included as a technical programme for the current year in the project on "Nutrient budgeting for improved varieties of spices".

The microbial diversity and structure in the rhizosphere of standards like erythrina, gliricidia, ailanthus, garuga and dead standard (RCC pole) have been investigated. Rhizosphere competence and studies on allelopathy have been included as a technical programme for the current year in the project "Assessment of soil quality in spices based cropping systems".

Experiments on sustainable production system have been initiated under the ongoing Mega Project III: "System approach for sustainable production of spices". Vegetahles, pulses, tuber crops, medicinal plants and fodder crops are under test in the inter-space of pepper garden. Annual legumes (cowpea, horse gram, green gram and black

gram) and a perennial legume *Mucuna* are included to enhance soil productivity in the pepper garden.

About 200 accessions of black pepper will be screened for drought tolerance next year.

Major products with commercial potential are white pepper, dehydrated green pepper, green pepper in brine, sterilized black pepper, pink pepper in brine, dry ginger, ginger powder, dry turmeric, turmeric powder, curcumin, cinnamon powder *etc.* Market potential of these products as well as other additional products will be collected.

A project will be initiated as soon as the Agricultural Engineer returns from study leave. Discussions are on with IIHR, Bangalore to collaborate with the project on mechanization in horticulture.

Detailed evaluation of the available dryers will be taken up in the coming season.

Mega Project title will be modified as "Propagation studies in spice crops".

Farmers will be contacted for the above purpose during February 2008.

The plantations of farmers who have taken more than 100 black pepper cuttings from the institute during 2006 and

**RAC Recommendations** 

free planting material from IISR in order to understand the status of the diseases.

- 29. Life cycle of shoot borer may be studied in highly resistant and susceptible accessions of ginger for comparison.
- 30. All the accessions have to be screened against all key pests.

**RAC Recommendation** 

104

- 31. A compiled table showing accessions resistant to different pests need to be prepared.
- 32. For screening of ginger and turmeric against *Pythium*, identified sick plot may be used to ensure enough pathogen load.
- 33. Use of phorate in trials may be discontinued.
- 34. Status report on the *Erytihrina* gall wasp infestation may be prepared before May 2008 and sent to Director, PDBC and ADG (Plant Protection) for necessary action.
- 35. Impact analysis on the usage of biocontrol agents may be taken on priority basis with the collaboration from Extension/Economics scientists.
- 36. Mass multiplication of quality biocontrol agents may be taken up by KVK for commercial purpose and if necessary one or two persons may be trained at PDBC, Bangalore.
- 37. Studies on the incidence of pests and diseases in relation to climate change may be initiated in existing projects.
- Cost of cultivation and marketing of major spices have to be updated.
- 39. Economic evaluation of technologies developed and transferred by the institute has to be completed.
- 40. Development and maintenance of data bases of spices is recommended.
- 41. The survey data should be analysed and impact on spread of IISR varieties and technologies must be submitted as an Interim Report by May, 2008 jointly by Drs. M.S. Madan and P. Rajeev as advised by earlier RAC.

2007 would be visited to check the incidence of the disease.

The life cycle of shoot borer would be studied in highly resistant and highly susceptible accessions during the current season.

All the accessions of black pepper are being screened against *Phytophthora*, nematodes and pollu beetle.

A compiled table of black pepper accessions resistant/ tolerant to *Phytophthora*, nematodes and pollu beetle would be prepared by April 2008.

A 'sick plot' would be established during the current year for screening germplasm of ginger and turmeric for disease resistance.

Phorate would be replaced with carbofuran in the existing trials.

The survey for the incidence of *Erythrina* gall wasp has been initiated in Wayanad district in Kerala and the status report of the pest in major black pepper areas would be prepared by May 2008.

This programme will be taken up in the new project to be taken up by scientists of social science section.

The KVK is already multiplying and selling the biocontrol agents identified by the institute. Newer organisms also would be taken up.

The incidence of *Phytophthora*, viruses, nematodes and pollu beetle on black pepper in Kozhikode district would be studied from the current year onwards to study the influence of climate change.

This work will be taken up in the Mega Project X: "Economics, statistics and modeling".

The evaluation has been done for production of Trichoderma by entrepreneurs. The same will be done for other technologies during 2008-09.

Data bases on marketing, export and prices will be developed and maintained by the Extension scientist.

Another set of study with the following scientists will be undertaken to see the impact of our technologies on ginger, turmeric and cardamom.

Ginger:V. Srinivasan and A. KumarTurmeric:K. Kandiannan and K.S. KrishnamurthyCardamom:S.J. Ankegowda and C.N. Biju

- Extension bulletins may be updated and published immediately.
- 43. Touch screen facility may be upgraded with more details like available facilities to farmers, price list and more user friendly information systems to farmers.
- 44. The areas where training that the institute could offer have to be indicated and given wide publicity.
- 45. Varietal variation of the antioxidant potential may be studied as an offshoot of this project in future.
- 46. While assessing the antioxidant potential the parameters may be restricted to the minimum.
- 47. RAC made the following suggestions to make the "computerized information dissemination system" more effective:
  - (i) Inclusion of FAQ (questions that would be frequently asked by the farmers and the answers there of)
  - (ii) Maps indicating the areas of cultivation of different spices; hot-spots of diseases, pests, etc.
  - (iii) Data on markets, price levels, etc.
- 48. RAC was very appreciative of the efforts put up for office automation system. The committee felt to highlight this effort and bring this to the notice of other Heads of Institutions, SAUs, ICAR, etc. so that "paper-less office functioning" can become a reality in a wider area.
- 49. RAC felt that it will be highly desirable to adopt a wider area and enthuse the farmers in the area to adopt IISR technologies so that farmer groups from other parts of the state and other states can visit and get first hand information.

The process has been initiated and would be completed in about 3 months.

The information on facilities available has already been included in the touch screen kiosk. Periodic up dates will be added.

Training schedule will be prepared and placed in the website.

Based on the results of the current programme, studies on varietal variation will be taken up in subsequent years.

This recommendation will be incorporated in the current technical programme.

- (i) This is included in the website. The same will be done for touch screen.
- (ii) This would be done with the help of the scientists concerned.

This may be taken up after obtaining permission from ICAR.

The technologies developed by IISR will be demonstrated through KVK.

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**RAC Recommendations** 

### Quinquennial Review Team (For the period ending 31.12.06)

- Dr. G. Kalloo Chairman
   Vice Chancellor
   Jawaharlal Nehru Krishi
   Vishwavidyalaya
   Jabalpur 482004
- Dr. I. Irulappan Member Former Dean, College of Horticulture Tamil Nadu Agricultural University 3-1-51, Ram Nagar T. Kallupatti - 625702 Madurai District, Tamil Nadu
- 3. Dr. K. C. Bansal Member Professor (Plant Biotechnology) National Research Centre for Plant Biotechnology Indian Agricultural Research Institute New Delhi - 110012.
- 4. Dr. R. Samiyappan Member Director Centre for Plant Protection Studies

106

Tamil Nadu Agricultural University Coimbatore - 641003

- 5. Dr. B. N. Choudhary Member Former Assistant Director General (Extension) Indian Council of Agricultural Research Flat No.154, Keshav Apartments Plot No. 15A, Sector-22, Dwarka New Delhi - 110075
- 6. Dr. A. G. Appu Rao Member Head, Department of Protein Chemistry & Technology Central Food Technological Research Institute Mysore - 570020
- 7. Dr. S. Devasahayam Secretary Acting Head, Division of Crop Protection Indian Institute of Spices Research Calicut - 673012



QRT members visiting the labs

### **QRT** Recommendations

#### SCIENTIFIC Crop Improvement and Biotechnology

- 1 IISR has the largest germplasm collection of black pepper, ginger and turmeric. It is also identified as the nodal agency for germplasm exchange in spices. In order to function effectively as the national centre for collection, conservation, documentation and exchange of germplasm, the National Repository of Spices Germplasm may be strengthened with the following infrastructure facilities : net-houses with ejet irrigation (for multiplication and conservation of collections), green house with humidity control (for conserving unique collections and high elevation species), and electric fencing of germplasm conservatories (to protect from wild animal attack).
- 2 Most of the black pepper growing areas and forests in South India have been explored so far. However, explorations may be carried out in the following areas that are yet to be explored : *Kerala: Shendurney Wildlife Sanctuary, parts of* Kulathupuzha, Goodrical Forest Range, coastal regions of Alleppey District, Perumbavur, Kunnathunadu, Vellidippara, Arakulam in Ernakulam District, and parts of Kasaragod District. Karnataka: Karwar, Bababudin hills, Yellapur, Bandipur, Bhadravathi, Kudhremukh. Tamil Nadu: Kolli hills, Kodaikanal, Yercaud and Shevaroy hills.
- 3 Conservation of germplasm in *in vitro* repositories is an effective and safe way of conserving vegetatively propagated spices. The *in vitro* gene bank needs to be strengthened with sufficient manpower on a priority. A technical assistant (T3) may be provided on permanent or on contract basis with a contingent funding of Rs 50,000/- per year. The feasibility of *in situ* conservation of germplasm should also be explored.
- 4 A large collection of spices germplasm is maintained at a huge cost and work load. A good beginning on molecular characterisation has been made and molecular profiling of about 100 collections each in black pepper, cardamom, ginger, turmeric and about 50 lines of vanilla has been completed. This has to be extended to all the genotypes (about 3000 in black pepper, 400 in cardamom, 1000 in turmeric and 800 in ginger). The molecular data for the remaining genotypes at least in black pepper and cardamom, should be generated and this coupled with the morphological data already available can be used to short list core collections. This may be undertaken

with external/contingent funding and the core collections should be safely conserved for future utilisation.

- 5 All the released and promising varieties and genotypes with unique characteristics must be finger printed and registered. Attempts may be made to barcode important and unique genotypes and species. The crop-wise priority for undertaking this work can be in the following order : black pepper, cardamom, ginger, turmeric, nutmeg, garcinia, clove, cinnamon and vanilla.
- 6 GIS has tremendous potential for genetic diversity estimation and identifying ecological niches for *in situ* conservation. GIS coupled with remote sensing data also help in predicting newer areas suitable for spices cultivation. The good beginning made in this direction may be intensified.
- Gene tagging, especially for QTLs, major biotic and abiotic stresses, will help in marker assisted selection (MAS). Molecular profiling of about 100 collections each in black pepper, cardamom, ginger, turmeric and about 50 lines of vanilla, 10-20 lines of cinnamon, garcinia, nutmeg and clove has already been done. A mapping population has been developed for preparing molecular maps in black pepper and about 200 polymorphic markers scored. Populations have been assembled for tagging genes by association mapping in black pepper and cardamom. The additional data required may be generated and MAS technology made available for black pepper and cardamom by the end of the XI Plan period.
- 8 Developing black pepper varieties resistant to *Phytophthora*, nematodes, *pollu* beetle and drought, *katte* and drought resistance in cardamom, rhizome rot resistance and low fibre content in ginger and high curcumin content in turmeric should be given a high priority.
- 9 Black pepper cultivars that mature early like Arakulam munda may be used as parents in breeding programmes to develop varieties to reduce the period and time of harvest in black pepper. The cardamom genotypes with synchrony in flowering may also be used in breeding programmes to synchronize the period of flowering and harvesting.
- 10 In breeding and developing new varieties, survey and selection should be given a high priority and a core group comprising of breeders, horticulturists, physiologists and plant protection scientists should

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**ORT Recommendations** 

be constituted to undertake the work.

**ORT Recommendations** 

108

- 11 Developing novel genotypes especially for pest, disease and drought resistance may be given priority. Chloroplast transformation approach may be ideal to avoid gene flow, as spices are native to India.
- 12 Biotechnology has an important role in spices research and it is necessary to strengthen this unit considerably. An auto radiographic facility and green house to maintain transgenic plants and virus free mother plants may be provided. Though most of the instruments needed are available many of them are over 10 years old and need to be replaced. The feasibility of providing a one time grant of about Rs. 50 lakh may be examined for upgrading the equipments, periodic calibration of all analytical equipments and maintaining them with service contract for next 5 years. This is all the more important to get the laboratories accredited with ISO standards.
- 13 Sufficient linkages should be built up with other national/international organizations in biotechnology. Though reasonable linkages with a few national laboratories have been established, the institute is handicapped by restrictions in collaboration especially with the best laboratories abroad. This may be relaxed for improving the exposure, calibre and standards of scientists.
- 14 More emphasis may be given to develop short duration varieties in ginger and turmeric. Studies on lack of seed set in the crop may be intensified. Specific recombinant breeding programme in turmeric may be initiated to bring the characters of interest together which was not attempted so far.
- 15 Tree spices need introduction of germplasm from centres of origin. Introducing sweet nutmeg (low myristicin) from West Indies and their evaluation will help the food industry.
- 16 Indian nutmegs are rich in myristicin, an anticarcinogenic compound. This property may be further verified and exploited in collaboration with CSIR laboratories to give value addition to Indian nutmeg.
- 17 Garcinia is the source of important anti-obesity compound, hydroxycitric acid (HCA). Efforts should be made to identify genotypes with higher HCA content and the possibility of isolation and purification of this compound as a major nutraceutical product should be explored.
- 18 Vegetative propagation remains the most effective method of multiplying elite genotypes of tree spices. The efficiency of this method may be enhanced using rootstocks at seedling stage without damaging their

tap root system and root stocks with known drought tolerance.

19 The effect of growth regulators in increasing yield may be studied in tree spices. Pruning studies may be undertaken to remove unproductive branches, doctor the canopy and increase flowering.

### Crop Production and Post Harvest Technology

- 20 The demand for organic products is increasing in the western markets at about 20% every year. IISR may strengthen research in organic farming in black pepper, cardamom, ginger and turmeric. Use of vermicompost and FYM/compost/oil cakes/green manure in INM may be encouraged. Linkages may be developed with Spices Board in this aspect since it is the nodal agency to promote organic farming in spices in the country.
- 21 IISR has done good work in developing management schedules against black pepper diseases incorporating soil amendments. These studies may be advanced further to determine critical soil pH, soil profiling and soil health levels (including rhizosphere) in black pepper and cardamom especially in organic farming systems.
- 22 Programmes on biological nutrient management in black pepper may be intensified, especially on microbial diversity and community structure in the rhizosphere and soils of black pepper. This would help in identifying strains of microbes beneficial for enhancing soil nutrient availability for black pepper. It is also important to understand the effects of applied biocontrol agents on populations of indigenous beneficial bacteria, which commonly occur in the rhizosphere. Comprehensive studies on these aspects are possible through molecular approaches. This will aid in formulating INM package utilizing beneficial microbes.
- 23 The crop-weather relationship for black pepper established by IISR based on the weather and yield data from Regional Agricultural Research Station (KAU), Ambalavayal, cannot be extrapolated to other regions due to inherent limitations in the statistical model. Hence, similar relationships for different black pepper producing centers may be obtained with the help of SAUs/AICRPS to arrive location-specific conclusions.
- 24 Some of the popular standards of black pepper like *Erythrina sp. are* susceptible to new insect pests resulting in their mortality. Efforts should be made to identify more suitable standards for black pepper. New agro-forestry species need to be evaluated before popularization.

- 25 Climatic change is drastically altering the environment including rainfall patterns in all spice growing regions. Some spice crops like black pepper and cardamom grown primarily in forest ecosystems are greatly affected by this climate change. The effect of these changes on performance of genotypes may be monitored with an objective of identifying better performers in changed conditions. Facilities such as open top chamber/greenhouse with light, temperature and  $CO_2$  control may be provided to undertake such studies.
- 26 In view of the changing climate, water will be a limiting factor in future. Studies on drought resistance and tolerance may be given high priority in black pepper and cardamom. Various agronomic practices to mitigate water shortage may be tried to identify cost effective technologies for drought management. Basic studies and evaluation of drought resistance has to be taken up on priority.
- 27 Studies on crop phenology will help in planning crop management strategies so as to schedule the field operations to enhance the yield with available resources. A programme may be initiated to see the relationship of flowering with yield and curcumin content in turmeric. Leaf area of turmeric may be correlated with rhizome yield in the on-going crop phenology study.
- 28 Quality of produce is one of the important factors for spices export. Breeding programmes must concentrate on genotypes with high quality parameters, especially high  $\beta$ -caryophyllene in black pepper, high zingiberene and shagoals in ginger, high  $\alpha$ -terpinyl acetate in cardamom and high cinnamaldehyde in cinnamon, in addition to yield trait.
- 29 A majority of the spices and their active principles are excellent antioxidant nutraceuticals which play significant role in the prevention of a number of agerelated discases. IISR may initiate studies on the identification and characterization of these bioactive components in spices, with a long term objective of inclusion of these constituents in various drug formulations.
- 30 Exploitation of natural food colours in spices is an area worth exploring. Programmes may be initiated to isolate the natural colour components in potential spice crops.
- 31 Research on post harvest technology and processing needs immediate attention. Cost effective primary processing technologies for important on-farm operations like harvesting, grading, packaging and storage which can improve the quality of the produce may be developed in collaboration with CSIR

laboratories. Studies on mechanization of farm operations may also be initiated. Processing and product diversification are extremely important in spices. Equally important is indigenous technology to develop new products. Establishment of an Agroprocessing unit at IISR is to be given priority, taking into consideration the recent trends in the field of value addition.

- 32 Studies on use of molecular markers for detecting adulteration in spice products that is being undertaken at present may be intensified.
- 33 Studies on various factors involved in improving vanillin content through processing and identifying efficient microbes involved in fermentation and conversion should be initiated.
- 34 Pesticide residue analysis is mandatory in the area of quality improvement and safety assessment of spices. The QRT recommends that the institute build up the required expertise and infrastructure facilities for the same. The Spices Board's ISO 9002 certified Quality Laboratory may be utilized for pesticide residue and microbiological analysis (particularly for aflatoxin), till such facilities are built up at the institute, as the certificates issued by them are accepted by the international trade agencies.

### **Crop Protection**

- 35 The institute has large collections of *Phytophthora* isolates from different hosts including black pepper. Morphological and molecular studies on these isolates have been initiated. The work must be continued to understand the variability existing in different *Phytophthora* species and for the identification of races. Molecular diagnostics to differentiate species also need to be taken up.
- 36 Phytophthora capsici and nematodes (Meloidogyne spp and Radopholus similis) are the predominant organisms associated with foot rot and slow decline disease of black pepper. However, in certain areas especially at higher altitudes occurrence of Fusarium sp. is also reported. Hence the role of Fusarium in the slow wilt of black pepper needs to be established and suitable control measures evolved.
- 37 The institute has a large collection of biocontrol agents and a few of them which have shown promise against *Phytophthora capsici* and *Meloidogyne* spp. have already been commercialized. Besides, several endophytes also have shown promising results against fungi and nematodes. In depth studies on the ecology of these organisms, their efficacy especially multiple efficacy in controlling more than one pathogen and their commercialization needs to be done.

T Recommendations

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- 38 Major spread of the pathogens (viruses, bacteria, fungi and nematodes) takes place through seeds/rhizomes and nursery planting material. Hence it is important to develop and employ sensitive techniques to detect all the pathogens early in the soil/root system. Though the institute has developed ELISA and PCR based diagnostics for certain viruses, bacteria and fungi, they need to be extended to all pathogens infecting spice crops. Wherever possible multiplexing should also be attempted.
- 39 Efforts should be intensified to characterize fungi associated with rhizome rot of ginger and turmeric in various regions and develop strategies for management including development of resistant varieties through conventional and biotechnological approaches.
- 40 Unlike cyst and root-knot nematodes, the genes associated with the parasitism/resistance of the endoparasitic nematode *Radopholus similus* are not clearly understood. There are reports on involvement of phenolics and phenyl propanoid pathway contributing to *R. similis* resistance in banana. Studies on mode of parasitism may be undertaken to develop resistant black pepper lines.
- 41 Changes in nematode community in soil reflect soil and ecological processes. Nematode diversity tends to be greatest in ecosystems with least disturbance, and bacterial-feeding nematodes make the greatest contribution to the decomposer food web in more intensively managed ecosystems. Studies on the role of nematodes in these processes will help in understanding the relationship between plants and soil nematode communities especially in perennial crops.
- 42 Infestations of new insect pests such as root mealyhugs on hlack pepper and Erythrina gall wasp on *Erythrina* sp. have reached alarming proportions in some hilly districts of Kerala and Karnataka. Since adequate manpower is not available in the Entomology Section, research work on these insect pests may be initiated through external funding.
- 43 Little information is available on the microhial pathogens affecting insect pests of spice crops. Systematic efforts may be made to document, evaluate and develop mass production technologies for utilizing microhial pathogens for the management of insect pests.

### Social Sciences

44 Agro-ecosystem analysis of spices production in different farming situations, problem prioritization, technology intervention and participatory on-farm assessment of newly developed technologies should be planned and undertaken by the Social Sciences Section of the institute.

45 A study of the present level of demand and supply of spices, its projection both for domestic and export, cost of production vs import cost is required to be made on priority.

#### Extension

- 46 The north eastern India provides excellent opportunities for expanding and developing spices cultivation in the region. Production, processing, adoption and marketing constraints of spices in north eastern hill states should be studied critically and promotion programmes need to be prepared and executed in the next Plan. Collaboration with Spices Board, ICAR Research Complex at Shillong, CPCRI Research Centers at Mohitnagar (West Bengal) and Kahikuchi (Assam) may help in bridging the gap. Improved varieties of ginger and turmeric may be introduced in larger scale in the region. Arecanut and tea-based cropping systems may be used to introduce black pepper in this region.
- 47 The research efforts of IISR must be complimented with the efforts of AICRPS, SAUs, DASD and Spices Board. It is also important to utilize the expertise and resources at RRLs, APPEDA, NABARD, NHM and state agricultural/horticultural departments in a synergistic effort for aggressive production, product development, market and trade strategies. Hence partnership arrangement with these agencies are to be encouraged.
- 48 Production and distribution of high quality and disease free planting material is the most important input in increasing production and productivity in spices. The facility at IISR may be strengthened and developed as a separate unit with funds from Technology Mission on Horticulture. Protected mother gardens of improved varieties need to be built in black pepper and cardamom and scion banks in tree spices. The planting materials should be fortified with efficient bio control agents at the nursery level. This programme may be supplemented by encouraging certified nurseries and biocontrol units in farmer's fields in different regions. Micro propagation techniques may be used where ever possible for initial bulking up of disease-free stocks.
- 49 Dissemination of information and technologies developed at IISR must be aggressive and effectively involve ATIC, KVK, Spices Board, DASD, state agriculture/horticulture departments and farmers. This should be enterprising, and conducted using modern audiovisual techniques. The institute should also lay front line demonstrations (FLDs) in major spice growing regions with the help of KVK to

110

**ORT Recommendations** 

demonstrate the feasibility of the technologies developed at the institute.

- 50 Impact assessment of technologies transferred is an important input to develop future strategies for the institute. Utilizing the in house ATIC and KVK facilities, inputs from the Department of Agriculture/ Horticulture may be periodically obtained, documented and analysed for deciding future strategies of the institute.
- 51 ITKs available in spices need to be verified, refined and documented so as to promote them through FLDs in farmers fields for increasing the sproduction.
- 52 The Institute Village Linkage Programme (IVLP) of ICAR implemented by many institutes has been successful in testing and modifying technologies in order to make them most suitable for local conditions of the farmers in different regions. This programme has now been discontinued. The QRT recommends to start the IVLP again during the XI Plan and the institute should propose adequate budget for the same.

### KVK

- 53 The availability of KVK staff is not according to norms and all the posts should be filled up on priority. The functioning of the KVK is affected due to the vacant posts of Programme Coordinator and SMSs in Plant Protection, Fisheries and Home Science.
- 54 The SMS (Home Science) should be encouraged to perform the twin role as support personnel for women programmes in adoption of improved farming technologies such as dairying and animal husbandry, production of vermicompost and post harvest management of spices, food grains, fruits and vegetables.
- 55 The Scientific Advisory Committee meeting of KVK should be held twice in a year regularly. Half yearly achievements of KVK should be reviewed critically and suggestions be made so as to enable KVK to make mid-term corrections in the programme and reorient the same to make it demand driven.
- 56 In the KVK programme emphasis was found tilted towards training and FLDs ignoring the importance of on-farm trials. The annual and five yearly programme of KVK should be reoriented giving equal emphasis on all the four mandates namely. training of farmers, on-farm trials, front-line demonstrations and training of field level extension functionaries.
- 57 The process of collection, documentation and use of field feed-back for reorientation of research and extension programmes in the KVK was found to be

weak. This needs to be given due importance in the KVK activities.

- 58 The KVK Farm and its demonstration units should be maintained scientifically to have the desired 'demonstration effect' on the visitors. Each demonstration unit should be labeled giving full technological information about the demonstration. On farm trial plots and FLDs should also be well labeled to give full information about the treatments/ interventions.
- 59 All important and critical agricultural operations in the FLDs need to be made in presence of KVK /IISR scientists in order to fully realize the production potential of technologies included as interventions. Only newly released technologies or the technologies which are likely to be released in near future should be the interventions in all FLDs.
- 60 The Dr. Mohan Singh Mehta Committee, while conceptualizing the KVK, suggested to identify innovative/progressive farmers including farm women who should be given comprehensive training and designated as 'key communicators' in the villages. Their services should be utilized for transfer of technologies in the villages. Further, the Committee suggested that school drop-outs should be trained in technical knowledge and skills in agriculture and allied fields by them under the close supervision of KVK scientists. This needs to be implemented in selected areas by the KVK.
- 61 Training need analysis should be done by the extension scientists of KVK and IISR in order to uncover training needs of different target groups which should form the basis to develop need-driven training courses for different target groups. Emphasis in KVK/IISR training should shift from quantity to quality. Development of 'lesson plan' for each course is sinequa-non for imparting quality training. Impact points in 'lesson plan' helps in objective evaluation of training. Every training course needs to be evaluated against the specific objectives of the course.
- 62 The programme for capacity building of the SMSs of KVK and scientists of IISR specially in the field of participatory training methods and extension tochniques requires special attention. Their knowledge and skills should be progressively improved in these areas. They need training in training need analysis techniques, training evaluation, audio visual aids preparation and use of socio-metric techniques.
- 63 The need and urgency of forging linkages and collaborative functioning of KVK with development departments of the state, SAUs and nearby ICAR institutes need to be recognized and followed by well

**DRT Recommendations** 

planned concrete action. The Director, HSR in consultation with the Coordinator of Zone VIII need to do strategic thinking and work out details for establishing working linkages and also guide and assist the KVK to achieve them.

- 64 The following facilities may be provided to the KVK so that it can effectively fulfil all its mandates biocontrol production unit; Information and Communication Technology laboratory; mobile-agroveterinary clinic.
- 65 Sufficient funds may also be provided to the KVK for development of KVK farm and demonstration units, conducting kisan melas and field days and POL charges for vehicles.

### HRD

- 66 Excellence requires highest level of HRD at all levelsadministrative, technical, and scientific. Phased training programmes need to be imparted in improving the quality of personnel who understand and meet the requirement of globalised and technology savvy world. The list of institutions where training programmes can be undertaken by scientists have been listed in Chapter 6. The opportunities available with various agencies including international institutes may be utilized for HRD. HSR may encourage utilization of sabbatical leave by scientists for continued education and upgrading of knowledge.
- 67 Access to information is an important input in any activity of excellence. The bioinformatics and the library facilities of the institute may be integrated into National Informatics Centre for Spices with state of art facilities Emphasis may he given for development of a digital library including electronic collection and digitising the most consulted reference sources for which appropriate softwares and CD-DVD mirror servers may be procured. The library website has to be further developed as a gateway to information resources available in the library. A 'nation wide open access movement' should be developed and linkages strengthened with similar institutes in sharing their institute repositories.

### Infrastructure

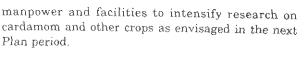
68 As post harvest technology is an emerging area of research, it has to be strengthened. The institute has also identified several microorganisms for the management of pests and diseases of various spice crops. However, adequate facilities are not available for their mass multiplication, quality evaluation and distribution. Various research laboratories of molecular biology and hiotechnology are scattered and congested and have to be integrated into a common unit. The institute also draws considerable visitors such as students, researchers and farmers and adequate space is not available in the ATIC for housing various live and preserved specimens of spices and also exhibits of significant achievements and technologies transferred from the institute Hence the QRT recommends the construction of the following:

- Post Harvest Technology Block
- Biocontrol Production Unit
- Molecular Biology and Biotechnology Block
- Spice Museum
- 69 At present, the Project Coordinator's office is housed in the main building which is very congested and insufficient to keep records and samples received from centres for analysis. Hence the QRT strongly feels the need for a separate Project Coordinator's Office at Calicut.
- 70 The institute has built up excellent instrumentation facilities over the years to cater to the needs of research. However, many of the existing equipments are more than 10 years old and need replacement and some new equipments are needed to carry out the envisaged programmes. Hence, the QRT recommends the purchase of the following equipments during the next Plan period:
  - Spice Museum
  - Herbarium facility
  - Autoradiography and radioactive facility
  - Automated protein purification unit
  - Liquid chromatography mass spectrometer
  - Gas chromatograph
  - Atomic absorption spectrophotometer with graphite analyser
  - Infra red spectrophotometer
  - Spectrofluorimeter
  - CHNS analyzer
  - 💌 🛛 Anion analyzer
  - Stereomicroscope
  - High and ultra speed refrigerated centrifuge-Floor model
  - Deep freezer
  - PCR / Real Time PCR units
  - Microwave digester
  - Denaturing gradient gel electrophoretic unit
  - BIOLOG System
  - Internet connectivity and wide area networking of Calicut, Peruvannamuzhi and Appangala
  - Computers/servers for office automation at Calicut, Peruvannamuzhi and Appangala
  - Pre-cleaners, separators and drivers for post harvest processing
  - Cryo grinding and super critical extraction unit
  - Solar water heating system
- 71 Adequate supply of water is essential for the laboratories throughout the year and the experimental fields during the post monsoon and summer seasons. Since the rains are becoming errant

112

year-by-year it is imperative to develop water harvesting structures and water sheds to meet this growing need. Hence the QRT suggest the development of watersheds at Calicut, Peruvannamuzhi and Appangala.

- 72 Since the entire Kodagu District is of tourist interest, getting satisfactory accommodation is very difficult especially during the peak tourist season at Appangala, for scientists and trainees who visit the centre. Hence, the QRT feels the necessity of construction of a Training cum Scientists Hostel at CRC, Appangala.
- 73 Posting of adequate number of scientists is crucial for the overall development of the Experimental Farm at Peruvannamuzhi. Hence the QRT proposes that additional facilities such as residential quarters and scientists hostel may be provided at Peruvannamuzhi for the same.
- 74 The minibus, jeep and staff car available at the headquarters are more than 10 years old leading to frequent breakdowns and repairs and demand high expenditure for their maintenance. The QRT feels that all the existing vehicles should be replaced. A new bus/minibus is essential at the headquarters for comfortable transportation of staff to the farm and also at KVK, Peruvannamuzhi for transportation of farmers for training programmes.
- 75 The present area of CRC, Appangala is only 17.4 ha. This is inadequate to meet the present requirements since cardamom is raised as an under storey crop under forests and only a fraction of the gross area is available for cultivation. Moreover a duplicate set of germplasm available at Peruvannamuzhi may have to be maintained at Appangala. The QRT suggests that the ICAR/IISR may approach the Karnataka Government to make available at least 25 ha additional land preferably contiguous to the existing farm to augment the land facilities at Appangala to meet the increased demand.
- 76 The present area of the headquarters of the institute at Chelavoor (Calicut) is only 17.4 ha. The institute is facing acute shortage of land for further expansion of physical facilities (laboratory, administrative and other buildings) and for essential field experimentation. The QRT suggests that the ICAR/ IISR may approach the Kerala Government to make available at least 25 ha additional land preferably contiguous to the existing land to meet the increased demand.
- 77 The CRC at Appangala under IISR mainly conducts research on cardamom and also to a limited extent on black pepper and tree spices. The centre may be upgraded as a Regional Station with necessary



- 78 Spice crops such as black pepper, ginger, turmeric and tree spices are grown to a large extent in the north eastern region. There is also great potential to increase the area and the productivity of these crops in this region. Hence there is an urgent need to start a Regional Station of IISR in the north eastern region to overcome the problems in cultivation of these crops and to tap the potential available in the region.
- The institute has proposed numerous programmes to  $79^{-}$ tackle issues related to spices research and development. However, sufficient scientific manpower is needed to fulfil the objectives and to achieve the set goals. The sanctioned scientific cadre strength of the institute has remained unchanged at 43 both during IX and X Plan periods. There is an urgent need to create a post of Head, Division of Crop Improvement and Biotechnology at Calicut. Additional manpower is also required in Entomology, Plant Pathology, Computer Applications, Bioinformatics and Food Technology/Post Harvest Technology in the grade of Senior Scientist. A post of Instrumentation Engineer in the grade of Senior Scientist is also required at Calicut for maintenance and repair of sophisticated equipments.
- 80 A Principal Scientist position in Plant Breeding/ Horticulture is required at Cardamom Research Centre, Appangala, to carry out various research activities envisaged in the next Plan period and also to head the station when it is upgraded as a Regional Station. Similarly, another Principal Scientist position in Plant Breeding/Horticulture would be required when a new Regional Station of IISR is sanctioned in the north eastern region.

### Administration

- 81 The institute has made tremendous progress in automation of office and is perhaps the only one of its kind in the entire ICAR. Sufficient funds may be sanctioned to provide networking of the headquarters at Calicut with its Experimental Farm at Peruvannamuzhi and Research Centre at Appangala.
- 82 The institute handles a large number of externally funded projects besides regular institute projects. The administration is headed by a lone Assistant Administrative Officer and there are no senior positions in administration such as Administrative Officer/Senior Administrative Officer. Hence the QRT recommends sanctioning of two administrative positions, one at Calicut and another at CRC, Appangala, in the rank of at least Administrative Officer.

Indian Institute of Spices Research

**IRT** Recommendations

### **Research Publications 2007-08**

- 1. Balaji, S., Bhat, A.I. and Eapen, S.J. (2008) A phylogenetic reexamination of *Cucumber mosaic* virus isolates based on 1a, 2a 3a and 3b proteins. *Indian Journal of Virology* 19: 27-33
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114

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### List of Projects

### I. Institute projects

Mega Project I: Collection, conservation, characterization and cataloguing of germplasm of spice crops for yield and other economically important characters [Project Leader: P.A. Mathew]

- 1 Gen. I (813): Collection, conservation, cataloguing and evaluation of black pepper germplasm (1972-2008) [K.V. Saji and Utpala Parthasarathy]
- 2 Gen. XIX (813): Conservation, characterisation, evaluation and improvement of Zingiber and Curcuma Spp (2007-2012) [B. Sasikumar and K. Abirami]
- 3 Gen. IX (813): Conservation and characterization of cardamom germplasm (2007-2012) [R. Senthil Kumar and C.N.Biju]
- 4 Gen. XIV (813): Characterization of turmeric germplasm for curcuminoids (2004-2008) [B. Chempakam and A. Shamina]
- 5 Gen. XXVI (813): Evolving high yielding and high quality nutmeg clones by selection (2007-2011) [B. Krishnamoorthy and J. Rema]
- 6 Gen. XXVII (813): Improvement of cassia by selection (2007-2010) [B. Krishnamoorthy and R. Senthil Kumar]
- Gen. XVI (813): Maintenance, enhancement and characterization of genetic variability in vanilla
   (Vanilla planifolia Andrews) (2005-2010) [R. Ramakrishnan Nair and P.A. Mathew]
- 8 Hort. III (813): Collection, characterization, evaluation and maintenance of paprika and paprika alike chilies (2004-2009) [D. Prasath and K.N. Shiva]

Mega Project II: Breeding improved varieties of spice crops for yield, quality, drought and resistance to pests and diseases [Project Leader: B. Krishnamoorthy]

- Gen. XVII (813): Breeding black pepper for high yield, quality, biotic and abiotic stress. (2006-2011) [V.A. Parthasarathy, B. Sasikumar, T. John Zachariah, K. Nirmal Babu, R. Suseela Bhai, Johnson K. George, Santhosh J. Eapen, K.V. Saji, S. Devasahayam, K.S. Krishnamurthy, R. Ramakrishnan Nair and T.E. Sheeja]
- 1 Gen. XVII (813): Breeding black pepper for high yield and caryophyllene (2007-2010) [B. Sasikumar and T. John Zachariah]
- 2 Gen. XVII (813): Breeding black pepper for *Phytophthora* resistance (2007-2010) [K. Nirmal Babu and T.E. Sheeja]
- 3 Gen. XXI (813): Breeding black pepper for resistance to "pollu" beetle (2006-2010) [K.V. Saji and S. Devasahayam]

8

List of Projects

- 4 Gen. XXII (813): Breeding black pepper for tolerance to drought (2006-2010) [T.E. Sheeja and K.S. Krishnamurthy]
- 5 Gen. XXIII (813): Breeding black pepper for developing resistance to *Radopholus similis* and its molecular genetic analysis (2007-2010) [Johnson K. George and B. Krishnamoorthy]
- Gen. X (813): Breeding cardamom for high yield, disease resistance and drought (2007-2012)
   [R. Senthil Kumar, M.N. Venugopal and D. Prasath]
- 7 Gen. XV (813): Investigations on the reasons and solutions for the absence of seed set in ginger (Zingiber officinale Rosc.) (2005-2010) [R. Ramakrishnan Nair and D. Prasath]
- 8 Biotech. VII (813): ISSR markers for black pepper improvement (2004-2007) [Johnson K. George and B. Sasikumar]
- 9 Hort. IV (813): Rootstock-scion interactions in tree spices (1998-2008) [J. Rema and K. Abirami]
- 10 Biotech. IX (813): Development of transgenic for resistance to *Phytophthora* and drought in black pepper (2006-2011) [K. Nirmal Babu and T.E. Sheeja]
- Gen. XXV (813): Genetics of seedling progenies of turmeric (Curcuma longa L.) (2007-2011)
   [R. Ramakrishnan Nair and K.N. Shiva]

## Mega Project III: System approach for sustainable production of spices [Project Leader: K. Kandiannan]

- SSC. IV (813): Nutrient budgeting for improved varieties of spices (2005-2010) [V. Srinivasan,
   R. Dinesh, C.K. Thankamani, K. Kandiannan, S.J. Ankegowda and S. Hamza]
- 2 SSC. III (813): Assessment of quality of soils under spices based cropping systems (2005-2008)
   [R. Dinesh, V. Srinivasan and S. Hamza]
- 3 Agr. XXIV (813): Phenology of ginger and turmeric (2006-2009) [K. Kandiannan and Utpala Parthasarathy]
- 4 Agr. XXVII (813): Enhancing the productivity in black pepper by intercropping (2006-2009) [C.K. Thankamani and K. Kandiannan]
- 5 Agr. XXVIII (813): Input use efficiency in turmeric in relation to quality (2007-2010) [K. Kandiannan and S. Hamza]

Mega Project IV: Production physiology of spice crops [Project Leader: B. Chempakam]

- 1 Phy. VII (813): Physiological and biochemical basis for productivity in black pepper (2003-2008) [K.S. Krishnamurthy and B. Chempakain]
- Phy. VIII (813): Mechanism of drought tolerance in cardamom and black pepper (2005-2008)
   [S.J. Ankegowda and K.S. Krishnamurthy]

8

117

List of Projects

	Annual Report 2007-08
	ga Project V: Value addition and post harvest processing of spices [Project Leader: nn Zachariah]
1	PHT. III (813): Studies on drying and storage parameters in black pepper, ginger, turme and nutmeg (2004-2009) [E. Jayashree, T. John Zachariah and K.N. Shiva]
2	PHT. IV (813): Evaluation for physical and biochemical quality of spices (2005-2009) [T. Jo Zachariah, K.N. Shiva and N.K. Leela]
	ga Project VI: Production of nucleus planting materials of improved varieties of spi ps [Project Leader: C.K. Thankamani]
	ga Project XIII: Investigations on nutraceutical and pharmacokinetic aspects of spic oject Leader: A. Shamina]
1	Biochem. III (813): Studies on the nutraceutical properties of bioactive compounds in few spic (2007-2010) [A. Shamina and K. Abirami]
2	Biochem. IV (813): Exploration of spices for natural food colours and pigments (2007-201 [K.N. Shiva and A. Shamina]
	ga Project VII: Identification, characterization and development of diagnostics again its, pathogens, nematodes of spice crops [Project Leader: M.N. Venugopal]
1	Path. XV (813): Investigations on diseases of vanilla (2003-2008) [A. Ishwara Bhat and C. Biju]
	ga Project VIII: Conventional and molecular approaches for developing per hogen and nematode resistance in spice crops [Project Leader: R. Suseela Bhai]
1	Crop Prot 1.4 (813) Identification of black pepper genotypes with multiple resistance again Phytophthora and nematodes (2006-2009) {R. Suseela Bhai and Santhosh J. Eapen}
2	Ent. XIII (813): Screening of germplasm accessions of spices and evaluation of antibios resistance to major insect pests (2006-2011) [T.K. Jacob and S. Devasahayam]
	ga Project IX: Developing integrated pest and disease management strategies in spi ops [Project Leader: S. Devasahayam]
1	Path. XVI (813): Etiology and management of rhizome rot complex in ginger and turmer (2004-2009) [A. Kumar and R. Suseela Bhai]
2	Org. Chem. II (813): Characterization of bioactive compounds with pesticide properties (200 2008) [N.K. Leela]
3	Biocontrol II. (813): Development of consortium of bioinoculants for management of pes diseases and nematodes in spices (2004-2008) [M.N. Venugopal, Santhosh J. Eapen and C. Biju]

- 4 Ent. XII (813): Bioecology and integrated management of shoot borer *Conogethes punctiferalis* Guen. infesting turmeric (2005-2009) [S. Devasahayam and T.K. Jacob]
- 5 Hort. V (813): Rootstock intervention to manage root infection of *Phytophthora* and nematodes in black pepper (2006-09) [P.A. Mathew and K. Abirami]
- 6 Path. XVII (813): Characterization, epidemiology and management of *Colletotrichum* spp. infecting black pepper, cardamom and turmeric (2006 2009) [M.N. Venugopal and C.N. Biju]

Mega Project X: Economics, statistics and modeling [Project Leader: M.S. Madan]

- 1 Econ. III (813): Remote sensing and GIS in evaluating the impact on socio-ecological changes on spices production in Western Ghats region (2003-2008) [M.S. Madan and A.K. Johny]
- Econ. IV (813): Assessing sustainability of cropping systems involving spices (2007-2010)
   [M.S. Madan, A.K. Johny and K. Jayarajan]

Mega Project XI: Extension and training [Project Leader: P. Rajeev]

- 1 Ext. IV (813): Training of research and extension personnel (2005-2010) [P. Rajeev and T.K. Jacob]
- 2 Ext. V (813): A study on diffusion, adoption and impact of varieties released from IISR and scientific crop management practices (2006-2009) [P. Rajeev and M.S. Madan]

Mega Project XII: Developing customized software and expert systems on spices [Project Leader: Santhosh J. Eapen]

1 Stat. I (813): Development of databases and software (2004-2007) [P. Rajeev and K. Jayarajan]

### **II. Externally aided projects**

### i) Department of Biotechnology, New Delhi

- 1 DBT-CIB-3: Development of microsatellite markers and characterization of curcuma spp. [B. Sasikumar and T.E. Sheeja]
- 2 DBT-CP 3: Genetic transformation of black pepper to confer resistance against viruses (2006-2009) [A. Ishwara Bhat and R. Suseela Bhai]
- 3 DBT-CP 2: Endophytic bacteria for biological system management of *Radopholus similis*, the key nematode pest of black pepper (*Piper nigrum* L.) (2003-2008) [Santhosh J. Eapen and A. Kumar]
- 4 DBT-SS1: Distributed Information Sub-Centre (Bioinformatics Centre) (2000-2008) [Santhosh J. Eapen]

ii)	Indian Council of Agricultural Research, New Delhi	
1	ICAR-CIB 2: Cloning of <i>Phytophthora</i> resistance and defense genes from <i>Piper colubrinum</i> (2004-2007) [Johnson K. George and M. Anandaraj]	
2	ICAR-CIB 1: Strengthening the cause of geographical indication of major spices using molecular morphological and quality profiling techniques (2004-2007) [B. Sasikumar and T. John Zachariah]	
3	ICAR-CPPHT-4: Chemical characterization of <i>Cinnamomum</i> germplasm (2005-2008) [N.K Leela and J. Rema]	
4	ICAR-CPPHT-1: Network project on organic farming (2007-2012) [V. Srinivasan, C. K Thankamani, A. Kumar and T. John Zachariah]	
5	ICAR-CPPHT-5: Prevention and management of mycotoxin contamination in commercially important agricultural commodities (2004-2007) [B. Chempakam, M. Anandaraj and N.K. Leela	
6	ICAR-CP 2: Molecular characterization and maintenance of National Repository o Phytophthora (2004-2007) [M. Anandaraj, R. Suseela Bhai and A. Ishwara Bhat]	
7	ICAR-CP 4: Application of microorganisms for Agriculture and Allied Sectors: Nutrient management, PGPR and biocontrol (2006-2008) [M. Anandaraj, R. Dinesh and A. Kumar]	
iii)	Ministry of Environment and Forests, New Delhi	
1	MOEF. I (813): Biodiversity in <i>Piper</i> and <i>Garcinia</i> and identification of spots of species richness in Western Ghats (using GIS and molecular markers) (2006-2009) [P.A. Mathew, Utpala Parthasarathy, Johnson K. George and K.V. Saji]	
iv)	Kerala State Council for Science, Technology and Environment, Trivandrum	
1	KSCSTE-CPPHT-1: Kerala State Council for Science Technology and Environment Project Production of white pepper through fermentation technology (2005-2008) [T. John Zacharial and A. Kumar]	
v)	National Horticultural Mission, New Delhi	
	NHM-CPPHT-1: Production of nucleus planting materials of improved varieties of spice cro (2005-2010) [C.K. Thankamani and S.J. Ankegowda]	

120

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## Personnel

Sl.	Name	Designation
No.		

### 1. Headquarters

Scie	atific	NAME AND POST OFFICE ADDRESS OF TAXABLE PARTY.
1.	Dr. V.A. Parthasarathy	Director
2.	Dr. M. Anandaraj	Project coordinator (Spices)
3.	Dr. B. Chempakam	Head, Crop Production Div.
4.	Dr. S. Devasahayam	Principal Scientist (Entomology)
5.	Mr. B. Krishnamoorthy	Principal Scientist (Plant Breeding)
6.	Dr. K. Nirmal Babu	Principal Scientist (Plant Breeding)
7.	Dr. M.S. Madan	Principal Scientist (Agri. Economics)
8.	Dr. T. John Zachariah	Principal Scientist (Biochemistry)
9.	Dr. B. Sasikumar	Principal Scientist (Plant Breeding)
10.	Dr. T.K. Jacob	Principal Scientist (Entomology)
11.	Dr. Johnson K. George	Sr. Scientist (Gen. & Cytogenetics)
12.	Dr. C.K. Thankamani	Sr. Scientist (Agronomy)
13.	Dr. R. Dinesh	Sr. Scientist (Soil Science)
14.	Dr. A. Ishwara Bhat	Sr. Scientist (Plant Pathology)
15.	Dr. R. Ramakrishnan Nair	Sr. Scientist (Gen. & Cytogenetics)
16.	Dr. P. Rajeev	Sr. Scientist (Agril. Extension)
17.	Dr. K.S. Krishnamurthy	Sr. Scientist (Plant Physiology)
18.	Dr. Santhosh J. Eapen	Sr. Scientist (Nematology)
19.	Dr. N.K. Leela	Sr. Scientist (Org. Chemistry)
20.	Dr. A. Kumar	Sr. Scientist (Plant Pathology)
21.	Dr. V. Srinivasan	Sr. Scientist (Soil Science)
22.	Dr. A. Shamina	Sr. Scientist (Bio chemistry-PS)
23.	Dr. K.V. Saji	Sr. Scientist (Economic Botany)
24.	Dr. K.N. Shiva	Sr. Scientist (Horticulture)
25.	Dr. T.E. Sheeja	Scientist (Sr. Scale) (Biotechnology)
26.	Dr. D. Prasath	Scientist (Sr. Scale) (Horticulture)
27.	Dr. K. Abirami	Scientist (Horticulture-FS)
Tech	nical Officers	
1.	Dr. Johny A. Kallupurackal	Technical Officer (T9)
2.	Dr. Hamza Srambikkal	Technical Officer (Lab) (T7-8)
3.	Mr. P. Azgar Sheriff	Technical Officer (Lib.) (T7-8)
4.	Dr. Utpala Parthasarathy	Technical Officer (T7-8)

1.	Dr. Johny A. Kallupurackal	Technical Officer (T9)
2.	Dr. Hamza Srambikkal	Technical Officer (Lab) (T7-8)
3.	Mr. P. Azgar Sheriff	Technical Officer (Lib.) (T7-8)
4.	Dr. Utpala Parthasarathy	Technical Officer (T7-8)
5.	Mr. M.M. Augusthy	Technical Officer (T6)

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6. Mr. K. Jayarajan

- 8. Mr. K.T. Muhammed
- 9. Mr. V. Sivaraman
- 10. Dr. C.K. Sushama Devi

### Administration

- 1. Sri V.L. Jacob
- 2. Sri A.P. Sankaran
- 3. Smt P.V. Sali

Technical Officer (Stat.) (T5) Technical Officer (T5) (Workshop) Technical Officer (T5) (Farm) Technical Officer (T5) (Farm) Technical Officer (T5) (Lib.)

Asst. Fin. & Accts. Officer Asst. Admn. Officer Private Secretary

### 2. IISR Experimental Farm, Peruvanamuzhi

Scie	ntific	
1.	Mr. P.A. Mathew	Principal Scientist (Horticulture)
2.	Dr. J. Rema	Principal Scientist (Horticulture)
3.	Dr. R. Suseela Bhai	Sr. Scientist (Plant Pathology)
4.	Dr. K. Kandiannan	Sr. Scientist (Agronomy)
5.	Ms. E. Jayashree	Scientist (Sr. Scale)(AS&PE)
		(on study leave)
Tech	nical Officers	
1.	Mr. V.K. Aboobacker Koya	Farm Supdt. (T9)
2.	Mr. N.A. Madhavan	Technical Officer (T5) (Farm)
3.	Mr. K. Kumaran	Technical Officer (T5) (Farm)

### 3. Krishi Vigyan Kendra

Tech	nnical Officers
1.	Mr. P.S. Manoj
2.	Dr. S. Shanmugavel
3.	Mr. K.M. Prakash

(T7) (Hort.) (on study leave) (T7-8) (Veterinary Science) (T7-8) (Agronomy)

### 4. IISR Cardamom Research Centre, Appangala

2. 3.	Dr. S.J. Ankegowda Dr. R. Senthil Kumar	Sr. Scientist (Plant Physiology) Sr. Scientist (Horticulture)
	Dr. C.N. Biju	Scientist (Plant Pathology)
dm	inistration	
•	Sri B. Sathish	Asst. Admn. Officer (on deputation

## Weather Data - 2007

Month	TMax.	TMin.	Rainfall (mm)	Rainy days (no.)
January	33.46	18.38	4.0	01
February	34.87	18.80	3.0	01
March	35.77	22.35	10.0	01
April	35.63	23.15	282.0	11
May	33.25	23.61	313.2	12
June	29.95	23.13	986.6	25
July	26.77	22.38	1987.2	31
August	28.17	22.22	1059.0	26
September	28.93	22.73	1060.4	26
October	30.46	22.19	644.8	22
November	32.63	20.50	61.8	06
December	33.54	20.25	1.0	00
Mean /Total	31.95	21.64	6413.0	162

### IISR Experimental Farm, Peruvannamuzhi

### Cardamom Research Centre, Appangala, Madikeri

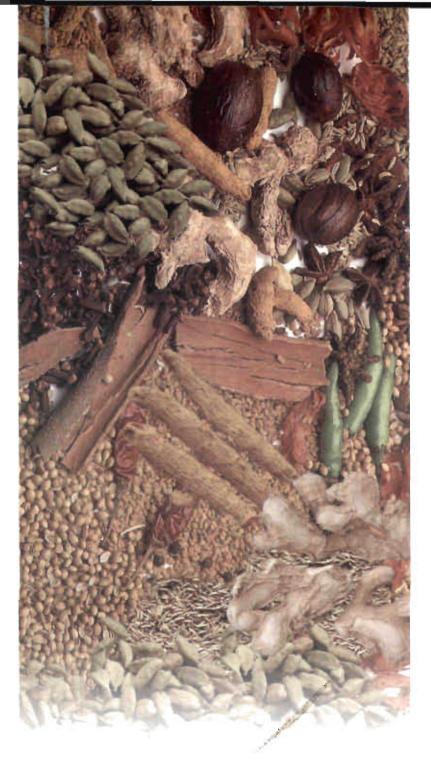
Month	TMax.	TMin.	Rainfall (mm)	Rainy days (no.)
January	29.8	NR	0.0	00
February	31.2	NR	0.0	00
March	32.2	15.9	2.1	01
April	32.1	17.0	22.2	07
May	29.4	16.4	65.9	06
June	24.2	16.6	794.4	20
July	24.4	16.3	1083.3	25
August	24.8	16.5	862.8	22
September	25.0	15.7	454.4	26
October	27.1	15.7	162.4	13
November	27.7	11.4	63.2	02
December	27.2	10.3	0.0	00
Mean/Total	27.9		3510.7	122

NR - Not recorded

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Weather





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