



वार्षिक प्रतिवेदन Annual Report 2014/15



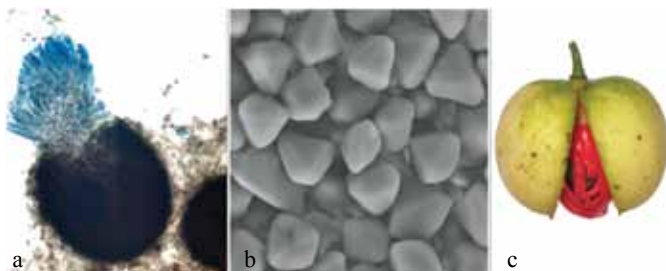
भा. कृ. अनु. प.-भारतीय मसाला फसल अनुसंधान संस्थान
ICAR-Indian Institute of Spices Research
कोषिकोड - 673012 केरल, भारत Kozhikode - 673012, Kerala, India



ABOUT THE INSTITUTE

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, Karnataka. The NRCS was further elevated to the present Indian Institute of Spices Research (IISR) during 1995.

The laboratories and administrative offices of the institute are located at Chelavoor, 11 km from Kozhikode, Kozhikode District, Kerala, on the Kozhikode-Kollegal road (NH 766), in an area of 14.3 ha. The research farm is located 55 km North East of Kozhikode at Peruvannamuzhi, on the Peruvannamuzhi-Poozhithode road in Kozhikode District, in an area of 94.08 ha. The IISR Regional Station, Appangala is located in Hervanad Village of Madikeri Taluk, Kodagu District, Karnataka in an area of 17.4 ha. Madikeri is the headquarters of Kodagu or Coorg District.



- a. Globose perithecia of *Colletotrichum gloeosporioides*
b. Scanning electron microscope image of *Spilarctia obliqua* NPV
c. A nutmeg collection with bold nut, thick and entire mace

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ANNUAL REPORT
2014/15



भाकृ अनुप
ICAR

भा. कृ. अनु. प. - भारतीय मसाला
फसल अनुसंधान संस्थान
(आईएसओ ISO 9001:2008 प्रमाणित संस्थान)
कोषिकोड - 673012



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Spices Research**

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PREFACE

The research achievements of the institute during 2014/15 are presented here as Annual Report. During this year, 243 accessions of black pepper were collected which includes 158 cultivars and 85 accessions of related taxa. A population of *Piper barberi*, considered to be an endangered species, was located in the evergreen forests of Anakulam forest range. In a farmer participatory germplasm collection, 31 nutmeg germplasm including few farmers' varieties and few unique germplasm were collected and conserved. A cardamom variety 'Appangala-2' developed through heterosis breeding has been recommended for release by AICRPS.

The fertilizer recommendations for cardamom were made for fixed target yield levels based on the soil test values for Appangala 1 and Green gold varieties. *In vitro* antioxidant activity and cytotoxicity of sequential extracts from selected black pepper varieties and *Piper* species indicated highest antioxidant activity in methanol extract of Malabar Excel followed by methanol extract of *P. colubrinum*. *In vitro* cytotoxicity indicated that chloroform extract of all the samples and hexane extract of *P. colubrinum* showed high cytotoxicity.

Screening of natural products and newer insecticides against cardamom thrips indicated that Spinosad, a natural product derived from *Saccharopolyspora spinosa* can be used for the effective management of cardamom thrips. Also, the field trials with the promising entomopathogenic fungus *Lecanicillium psalliotae* for the control of cardamom thrips indicated that combined application of *L. psalliotae* as spray and basal application gave better control.

The institute conducted 17 training programmes of various durations for effective technology transfer to diverse stakeholder groups like farmers, youth, tribal beneficiaries and students. We have also embarked on empowering the tribal farmers under the aegis of the Tribal Sub-Plan of ICAR. In KVK, about 131 training programmes for practicing farmers and farm women, rural youth and extension functionaries were conducted and 4215 trainees were benefitted. Ten front line demonstrations and five on farm trials on technology assessment and refinement were carried out. Non-exclusive license for commercializing designer micronutrient formulations have been given to four agencies through Business Planning and Development (BPD) unit. IISR has entered into a Memorandum of Understanding with Kerala Industrial and Technical Consultancy Organization Ltd. in an effort to jointly promote entrepreneurship development

We have launched two new ICAR funded network programmes viz., High Value Compounds, and Organic Horticulture. I consider it a privilege to place on record the encouragement given by Dr. S. Ayyappan, Secretary, DARE and Director General, ICAR. We are also grateful for the strong support and guidance received from Dr. N.K. Krishnakumar, Deputy Director General (Horticulture), Dr. S.K. Malhotra, former ADG (Hort. II) and Dr. Janakiraman, ADG (Hort.). I appreciate the efforts and zeal shown by all the project investigators in executing various programmes. The financial support for the projects received from ICAR is gratefully acknowledged. I also commend the editors for having compiled and brought out this publication.



(M. Anandaraj)

Kozhikode,
15.06.2015



EXECUTIVE SUMMARY

BLACK PEPPER

Genetic resources

A total of 243 accessions were collected during the survey (Kerala and Karnataka) including 158 cultivars and 85 accessions of related taxa. The present status of germplasm holding at the NAGS is 3181 (1669 cultivars, 1503 accessions of related taxa and 9 exotic species).

Drought stress

Ubiquitin gene, the most stable reference gene, was identified using three different accessions of *Piper nigrum* under water deficit stress. Among the genes tested for expression analysis in different accessions, *Myb* and *NAC* protein genes were found to be expressed three fold and above in susceptible cv. Sreevara under water deficit compared to control. The increase in expression of these genes was low in drought tolerant line, Acc. 4216. *Dehydrin* gene was again found to be expressed many fold in Acc. 4216 compared to low expression in Sreevara.

Characterization of mentor grafted progeny

Seedling progenies grafted onto *Piper colubrinum* showing characteristics of the root stock are being investigated for the presence of sequences from *P. colubrinum* using dd-RAD sequencing. Of the 1186604 million IlluminaHiSeq reads, 2000 reads of about 100bp in length were recovered.

Phytophthora capsici – *Piper colubrinum* interaction

Quantitative RT-PCR was employed to assess the level of expression of pathogenicity genes of *P. capsici* viz., *Glycoside hydrolase*, *NPP1*, *RCLR* and *pectate lyase* during *P. capsici* - *P. colubrinum* interaction.

In planta expression and docking studies of a glucanase inhibitor gene from *Phytophthora capsici* and beta 1, 3 glucanase gene from *P. colubrinum*

The *in planta* expression of *GIP* gene from *P. capsici* was at its peak during initial hours of challenge

inoculation and the expression of *pcEGase* gene was at its peak at 16 hpi (hours post inoculation). Molecular docking studies between *pcEGase* gene and *GIP* revealed that substrate inhibition is obtained by recognizing arginine and isoleucine residues in substrate molecule.

Identification and characterization of miRNAs in *P. colubrinum*

The *de novo* assembled *P. colubrinum* transcripts were analyzed for lncRNAs (long non-coding primary RNAs), microRNAs (miRNAs) and their corresponding mRNA targets. Of the 4542 targets, 881 transcripts were predicted with putative functions.

Phytophthora – *P. nigrum* interaction

The R genes showed early expression in resistant variety than in susceptible. The *glucanase* gene showed constitutive expression in both the genotypes with the upregulation only in resistant variety upon infection with *P. capsici*.

In vitro antioxidant activity and cytotoxicity of sequential extracts from selected black pepper varieties and *Piper* species

Antioxidant activity and cytotoxicity of four medicinally valued *Piper* species viz., *Piper nigrum*, *P. chaba*, *P. longum* and *P. colubrinum* were examined. Among all extracts investigated, methanol extracts showed highest antioxidant activity for all the four assays. Methanol extract of cv. Malabar Excel was found to be highest for all the assays. *In vitro* cytotoxicity on cervical cancer cell line CaSki by MTT assay indicated that chloroform extract of all the samples and hexane extract of *P. colubrinum* showed high cytotoxicity. Cytotoxicity increased with increase in the amount of extract as well as time of exposure of extract with CaSki.

Integrated management of *Phytophthora* foot rot and slow decline diseases

Field evaluation of bioagents integrated with chemicals effective against *Phytophthora* and

nematodes showed that metalaxyl-mancozeb 0.125% + carbosulfan 0.1% + *Trichoderma harzianum* + *Pochonia chlamydosporia* was effective in reducing yellowing and decline of vines.

Evaluation of actinomycetes against nematodes

An experiment to study effect of actinomycetes on nematodes *in planta* showed that combined application of IISR Act 2 (*Ketosatospora setae*) with IISR Act 5 (*Streptomyces sp.*) or IISR Act 9 (*S. tauricus*) were effective in reducing the nematode population in the soil by an extent of 58 - 75%.

Biological control

In a pot experiment with 15 *Trichoderma* isolates, highest growth promotion was observed in PhytoFuRa-3 followed by PhytoFuRa-14 and highest biomass production was in PhytoFuRa-10, which also consistently showed significantly higher biocontrol potential against *Phytophthora* foot rot.

Comparative genomics of *Phytophthora* species

Secretome analyses of *Phytophthora* species (*P. capsici* (05-06 and 98-93), *P. sojae*, *P. infestans*, *P. ramorum*) were done using different softwares like SignalP, TMHMM and TargetP.

Artificial induction of perfect stage of *C. gloeosporioides* infecting black pepper

The perfect stage (perithecia) was artificially induced under *in vitro* conditions based on mating-test model, in which sterilized toothpicks, dried leaves and twigs of black pepper as well as split, unsplit twigs of silky oak placed between confronting inoculum sources (pathogen culture, infected young and dried leaves of black pepper) served as inert platforms for the induction of perithecia. Under *in vitro* conditions, production of perithecia was observed in all the combinations, while formation of ascospores (indication of fertile perithecia) was observed only in the combination of dried black pepper twig + infected young and dried leaves. Exudate embedded with ascospores produced from fertile perithecia was observed in the combination;

black pepper twig + infected young leaf even three months after inoculation. The twigs with exudate, partly or wholly, when tested for infectivity on variety Panniyur-1 under lab and field conditions resulted in the development of characteristic anthracnose symptoms 4-6 days after inoculation.

Sequential events in the colonization and proliferation of *C. gloeosporioides*

Conidial germination was observed 4h after inoculation. The germinating conidia were found congregating more towards stomatal region and 75% of conidia germinated either with one (most cases) or two germ tubes after 10-12h. Higher percentage of germination was noticed, when the conidia were in disaggregated condition which later produced melanized appressoria. The infection hyphae originating from appressoria entered through stomata and subsequent intra/intercellular invasion was observed. Invading hyphae in the mesophyll cells and localized tissue death were noticed after 48h. Acervulus initials were formed and mature acervuli with prominent setae were observed after 48 and 72h, respectively. Several localized necrotic spots manifested on leaf surface after 72h and the invaded epidermal cells turned brown, resulting in rapid collapse and death, 72h after inoculation.

New target genes in *Radopholus similis*

Potential target genes of *R. similis* involved in parasitism such as *FMR* Famide-like peptides (nematode FLPs), β -1, 4, endoglucanase, *trans-thyretin-like protein 3 precursor*, *serine-threonine phosphatases* and survival such as *glutathione-S-transferase(s)*, *acetylcholinesterase*, *tetratricopeptide TPR-1*, *superoxide-dismutase* and *actin* were amplified and sequenced.

Viral disease

Rapid identification of transgenic black pepper using loop-mediated isothermal amplification (LAMP) and real-time LAMP assays

A loop-mediated isothermal amplification (LAMP) and real-time LAMP based assays were developed for quick and sensitive detection of transgenic black



pepper plants. The assays were validated by testing putative transformants of black pepper. The results clearly showed that LAMP and real-time LAMP assays developed in this study can provide a rapid and simple approach for screening transgenic black pepper and other plants transformed by using target gene sequences.

Sequencing of *RNA2* and *RNA3* of *Cucumber mosaic virus* infecting black pepper

Cloning and sequencing of *2a*, *2b*, *3a* and *3b* gene of black pepper isolate of CMV showed that it consists of 2573, 337, 840 and 657 nucleotides respectively potentially encoding proteins with 857, 111, 279 and 218 amino acids, respectively. In the phylogeny all the four genes (*2a*, *2b*, *3a* and *3b*) showed close clustering with CMV subgroup I strains and distant relationship with subgroup II strains. Among the four genes, *3b* showed high level of sequence conservation while *2b* showed the least with other members in the sub group.

Screening against *Piper yellow mottle virus* (PYMoV)

Out of 2437 germplasm accessions screened for resistance against *Piper yellow mottle virus*, four accessions showed resistance in the preliminary test.

CARDAMOM

Genetic resources and breeding

A total of 618 accessions are being maintained at NAGS and about 117 accessions were characterized for morphological and yield characters. FGB-13 and FGB-82 recorded maximum yield and more number of capsules per plant.

The accession, IC 547167 (Appangala 1 x NKE 19) with potential yield of 1393.12 kg ha⁻¹ (three years after planting), with mosaic resistance and good quality characters has been recommended for release in Karnataka as new variety under the name Appangala 2 by XXV AICRPS meeting held at UBKV, Pundibari, West Bengal in September 2014.

Standardizing the parameters for target yield

In Green gold, recorded yield levels per plant basis was 0.7, 0.9 and 0.9 kg plant⁻¹ for the targets 0.4, 0.6 and 0.8 kg plant⁻¹ with a positive mean deviation of 72, 55 and 15%, respectively. Similarly, in Appangala 1 yield per plant has shown a positive mean deviation of 83, 76 and 14% for the fixed target levels.

Soil carbon pools under cropping systems

The total and particulate organic carbon (POC) and nitrogen pools were quantified under different spice based cropping systems and high density multiple cropping system. The POC and PON pools were higher in coffee + black pepper system (56.7 & 16.8 mg ha⁻¹) with highest total organic C & N (TOC & TON) pools (90.1 & 33.4 mg ha⁻¹). POC constituted 63% of TOC in this system. The non particulate carbon and nitrogen (NPOC & NPON) pools were higher under cardamom alone and coffee + black pepper + cardamom cropping system (67.3 and 58.3 mg ha⁻¹) constituting 73-78% of TOC pools.

Among different management systems in black pepper, organic management has resulted in higher POC, NPOC and TOC pools as compared to integrated and conventional management systems. In HDMCS, black pepper basin has accumulated highest TOC, NPOC and POC pools (106.8, 71.6, 35.2 mg ha⁻¹, respectively) and coconut and nutmeg systems had higher NPON and PON (7 & 0.8 mg ha⁻¹) as compared to other component crops.

Quality evaluation of cardamom varieties

Different varieties *viz.*, Njallani, Pannikulangara-1, Pannikulangara-2, Thiruthali, Elarajan and Wonder cardamom collected from Idukki district were analyzed for essential oil profile. The essential oil content ranged between 5.8-7.4% on capsule weight basis. Pannikulangara-2 recorded highest essential oil content. Among the 21 components identified, the chief constituents of the oil, 1,8-cineole and α -terpinyl acetate varied between 18.1-32.7% and 36.9-48.5%, respectively. Concentration of pinene, sabinene, myrcene, α -terpineol, 4-terpineol, nerol, neryl acetate and nerolidol ranged from 1-5%.

Quality evaluation using E-nose

Hand-held electronic nose was modified with suitable sensor array for determining quality. Samples were analyzed using the modified hand-held electronic nose for essential oil content and could be graded into low (<4.0%), medium (4.0-6.0%) and high (>6%).

Management of thrips (*Sciothrips cardamomi*)

Screening of natural products and newer insecticides for three years indicated that Spinosad, a natural product derived from *Saccharopolyspora spinosa* can be used for the effective management of thrips. The product can also be used in organic system. Standardization of spray schedule of promising insecticides is in progress.

Evaluation of entomopathogenic fungus

Field trials with the promising entomopathogenic fungus *Lecanicillium psalliotae* for the control of thrips were conducted at Kodagu, Wayanad and Idukki districts. The trials indicated that combined application of *L. psalliotae* as spray and basal application gave better control than other treatments at Wayanad.

Studies on *Wolbachia*

Studies on removal of the endosymbiont *Wolbachia* from thrips as a strategy for its management indicated that when the thrips were fed with tetracycline treated leaves, *Wolbachia* was completely eliminated from the insect system which was confirmed by molecular studies.

Root grub (*Basilepta fulvicorne*)

Infectivity of entomopathogenic nematodes (EPNs) against root grub, *Basilepta fulvicorne* was tested *in vitro*. Among the test EPNs, *Heterorhabditis* sp. (IISR-EPN 01) and *O. gingeri* (IISR-EPN 07) were more pathogenic as they caused 100% mortality to the insect within 72h post exposure, followed by *Steinernema* sp. (IISR-EPN 03), *S. carpocapsae* (IISR-EPN 06) and *Oscheius* sp. (IISR-EPN 08). *Steinernema* sp. (IISR-EPN 02) and *Oscheius* sp. (IISR-EPN 04 and 05) took 120h to kill the test insect.

Occurrence of perfect stage of *C. gloeosporioides*

Surveys carried out in cardamom plantations revealed manifestation of different types of foliar symptoms *viz.*, spot, blight and shredding. The cultures isolated from these samples exhibited variations in colony morphology and colour. Among the cultures, greyish white culture appeared puffy with a faster growth rate (14 mm day⁻¹) and produced dark brown-black, globose perithecia, four weeks after incubation.

Differential reaction of *C. gloeosporioides* isolates on varieties

Differential reaction of 20 *C. gloeosporioides* isolates was studied on cardamom varieties *viz.*, Appangala 1, IISR Vijetha and IISR Avinash. The isolates exhibited differential reaction as indicated by prominence and non-prominence of yellow halo and streak. The area of lesions developed on young leaves varied between 4.91 – 40.82, 7.85 – 60.45 and 11.78 – 38.47 mm² in IISR Avinash, IISR Vijetha and Appangala 1, respectively.

Evaluation of microbes for antagonistic potential against rhizome and root rot pathogens under *in vitro* conditions

The endophytic fungal isolates from varieties IISR Vijetha, IISR Avinash and Appangala 1 were evaluated *in vitro* for antagonistic efficacy against *Fusarium oxysporum*, *Rhizoctonia solani* and *Pythium vexans*. Among the isolates tested, Va 4-2 (IISR Vijetha), Cb 4-1, Cb 6-2 (Appangala 1) and Aa 1-1 (IISR Avinash) were found promising against *F. oxysporum*. While, Cb 4-1, Cb 6-2 (Appangala 1) and Ab 6 (IISR Avinash) were effective against *P. vexans* and Cb 2 (Appangala 1) was inhibitory to *R. solani*.

GINGER

Genetic resources and breeding

Six hundred and sixty eight *Zingiber* accessions have been maintained in the field gene bank. Germplasm conservatory was enriched with 17 ginger accessions including extra bold local ginger accessions from Kerala and West Bengal.

Among 13 extra bold ginger accessions evaluated, maximum fresh and bold rhizomes were recorded in Acc. 723, Acc. 247 followed by Acc. 713. Four genotypes (IISR Varada, IISR Mahima, Acc. 182 and Acc. 247) were subjected to γ - irradiation (900 buds each) at different doses of 0.80, 1.00 and 1.20 kR. Differential response was observed for sprouting.

Thirty accessions of *C. amada* have been screened for resistance to race 4 strain of *R. solanacearum* by soil and pseudostem inoculation methods. Two accessions were found to be resistant by both soil and pseudostem inoculation. Bright field and fluorescence microscopic work was carried out with inoculated and uninoculated samples of *C. amada* and *Z. officinale*. It was noticed that the stelar portion of *C. amada* had extensively thick cell walls compared to *Z. officinale*. The casparian thickenings were clear and thick compared with the endodermal cells of *Z. officinale*.

Tissue specific expression analysis of short-listed genes/ESTs using qPCR

Among the candidate genes, *LRR-NBS*, *ABC transporters*, *4-coumarate: coenzyme A ligase (4-CL)*, *WRKY transcription factor 8* and *callose synthase* were studied for their expression level in ginger and mango ginger at different time intervals (0, 1, 4, 8, 16, 24, 48, 72, 96 and 120 hpi) in leaves and pseudostem. In general the expression patterns of the genes were higher in *C. amada* compared to *Z. officinale*.

Fertigation scheduling

Fertigation schedule is being standardized under soil less ginger production using coir pith and farm yard manure (1:1). Five treatments with varying doses of fertilizers (75-125%) and 75% recommended dose + PGPR was laid out. Sampling at 120 DAP showed that the maximum dry matter was partitioned into stem (43-50%) followed by rhizome (25-32%) and control (solid fertilizer at monthly interval) followed by recommended dose had the maximum rhizome fresh weight.

Whole genome sequencing of *Ralstonia solanacearum*

Two strains of *R. solanacearum* (GRs-SIK and GRs-

MEP) were Illumina sequenced and the raw data has been assembled using A5-miseq. Both the strains have been annotated using Prokka (a software tool for the rapid annotation of prokaryotic genomes). In GRs-MEP there are 5120 CDS, 80 tRNA, and 1 tmRNA while, GRs-SIK possesses 5080 CDS, 63 tRNA and 1 tmRNA. For better classification of the predicted proteins from Prokka, a refined annotation has been done using Blast2GO with $1.0E^{-3}$ as e-value cut off and 33 as HSP cut off length. The genomes were mined for various effector proteins and other virulence factors.

Hairy Caterpillar (*Spilarctia obliqua*)

A new tetrahedral shaped, multiple nucleocapsid nucleopolyhedrovirus (IISR-NPV-02) isolated from *S. obliqua* was characterized based on sequencing of conserved baculovirus genes and restriction endonuclease analysis. *Polyhedrin* and *lef-9* gene sequencing and phylogenetic analyses revealed that SpobNPV is a new addition to the group I NPVs and is very closely related to other NPVs infecting Arctiidae.

TURMERIC

Genetic resources and breeding

One thousand four hundred and four *Curcuma* accessions are being maintained in the field gene bank. Germplasm conservatory was enriched with nine *Curcuma* accessions.

A multi-locational trial with three promising accessions (Acc. 48, Acc. 79 and Acc. 849) along IISR Prathiba and local check was laid out in Kerala (Peruvannamuzhi), Andhra Pradesh (Vijayawada), Tamil Nadu (Erode) and Karnataka (Chamrajanagar and Chettali). The short duration genotypes *viz.*, Acc. 48 and Acc. 79 performed well under different locations.

Amplification of full length cDNA

A simple protocol for cloning of full length gene was optimized by inverse PCR combined with SMART system using gene specific primers. Full length cDNA of *curcumin synthase 3 (curs3)* with 137 bp

of 5' UTR and 299 bp of 3' UTR was amplified from normalized cDNA library constructed from pooled tissues of turmeric using *curs3* specific outward primers.

Cloning of specific miRNAs

Four miRNAs viz., miR156, miR167, miR172 and miR396 were cloned and sequenced by stem loop RT-PCR method. Among these, targets of two miRNAs viz., miR156 and miR172 were predicted and identified as squamosa promoter binding like genes and floral homeotic protein *AETALA 2* like isoform *XI*, respectively. Targets were also identified for miRNAs identified through deep sequencing which mainly included conserved transcription factors. Important targets identified were growth regulating factors (GRFs), *NAC* domain containing proteins, F-box family proteins, *GAMYB* transcription factor like proteins, homeobox leucine zipper proteins, TCP transcription factors and three auxin response factors were targeted by miR396, miR164, miR394, miR319, miR166, miR171 and miR160, respectively.

Mining for genomic SSRs

MultiNA analysis of 10 polymorphic genomic SSR primers in 96 accessions was performed. MultiNA is a microchip based electrophoresis system with high sensitivity detection that uses LED excited fluorescence detector. Although major differences among the released varieties could not be detected, the varieties Suvarna, Suguna and Sudarshana could be distinguished from other released varieties and also Suvarna showed a distinguishable pattern from the rest.

Influence of coloured shade nets on ginger and turmeric production

Ginger and turmeric were grown under red, green, white and black shade nets with conventional planting as control. Black, red and white recorded similar rhizome fresh weight at harvest & least in open. Not much variation in photosynthetic and transpiration rate was noticed among treatments. Quality parameters viz. oil and oleoresin were slightly higher under red and black shade nets compared to other treatments.

Studies on symbiotic bacteria of EPNs

The symbiotic bacterium associated with *Heterorhabditis* sp. (IISR-EPN 01), promising against shoot borer of ginger and turmeric, was identified as *Photorhabdus luminescens* (IISR-EPN BC 09) on the basis of morphological, biochemical and molecular characterization.

Evaluation of EPNs

The efficacy of four promising EPNs such as *Heterorhabditis* sp. (IISR-EPN 01), *Steinernema* sp. (IISR-EPN 02), *O. gingeri* (IISR-EPN 07) and *Oscheius* sp. (IISR-EPN 08) was tested against shoot borer infesting ginger and turmeric under field conditions. Among the test EPNs, *O. gingeri* (IISR-EPN 07) treated plants showed minimum shoot damage in ginger and turmeric (19.4 and 28.4 %, respectively) in comparison to control (36.9 and 51.9%, respectively) which was on par with malathion treatment (18.4 and 24.6%, respectively).

TREE SPICES

Genetic resources

A farmer participatory nutmeg germplasm collection was made in Idukki, Kottayam, and Malappuram districts of Kerala and 31 nutmeg germplasm including few farmers varieties were collected and conserved. The unique germplasm collected include a nutmeg with rudimentary sterile seed; nutmeg with bold nut; thick and entire mace type; high yielding monoecious nutmeg and Punnathanam Jathi, a farmer's variety which had very bold nut and thick mace.

Isolation and amplification of genomic DNA from nutmeg mace

A protocol was developed to isolate high quality DNA from nutmeg mace. The purity of the DNA was checked by qualitative and quantitative estimation, restriction digestion, RAPD and amplification of the barcoding loci *rbcl* and *ITS*.

Standardisation of barcoding loci *rbcL* and *ITS* for *Myristica* species (*M. fragrans*, *M. malabarica*, *M. andamanica*, *M. fatua*, *M. beddomei*, *M. amygdalina*)

The PCR temperature profiles were optimized with annealing temperatures of 52.5°C and 56.0°C for *rbcL* and *ITS* loci respectively. The *rbcL* and *ITS* amplicons yielded products of 600bp and 500bp respectively.

Generation of barcode sequences

The *matK* barcode sequences for some *Cinnamomum* species (*C. verum*, *C. glaucescens*, *C. sulphuratum*) were generated and submitted to GenBank nucleotide database of NCBI.

Phytochemical analysis of *Myristica* species

Essential oil profile of nut, mace and pericarp of *M. fragrans* was studied. Nut and mace had similar composition, the chief components being sabinene, pinenes, myrcene, γ -terpinene, 4-terpineol, saffrole, myristicin and elemicin. The pericarp of *M. fragrans* was dominated by 4-terpineol, α -terpineol, γ -terpinene, α -terpinene, pinenes and myrcene. Seeds of *M. prainii* and *M. fragrans* yielded 40% and 32% butter, respectively. Fatty acid profile of nuts of *M. prainii* and *M. fragrans* indicated that both were dominated by myristic acid (>80%).

Antioxidant activity of volatiles of nutmeg

Antioxidant activity of major essential oil constituents of *M. fragrans* viz., myristicin, 4-terpineol and α -terpineol were compared by DPPH and phosphomolybdenum methods. Results showed that myristicin had higher antioxidant potential.

Documentation of natural enemies

Surveys for entomopathogens and other natural enemies of spice crop (black pepper, cardamom, ginger, turmeric, garcinia and nutmeg) pests were conducted in Idukki, Wayanad and Kozhikode districts of Kerala, Coimbatore and Nilgiris districts in Tamil Nadu and Dimapur district in Nagaland. The host insects included black pepper scale, *Aspidiotus destructor*, cardamom thrips, *S. cardamomi*, cardamom scale, *Aulacaspis* sp., garcinia hopper, *Busonomimus manjunathi*, and nutmeg shoot borer, *Sinoxylon anale*. The fungus infecting *B. manjunathi* was identified as *Metarhizium* sp. and the fungus infecting *S. cardamomi* as *Isaria* sp. based on morphological and molecular studies. The identity of the four larval and pupal parasitoids of ginger shoot borer collected during the surveys were confirmed as *Eriborus ?ricini*, *Xanthopimpla stemmator*, *Trathala flavoorbitalis* and *Apanteles* sp.



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 Kozhikode, 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. The Cardamom Research Centre, Appangala, was later upgraded as Regional Station of ICAR - IISR in November 2014.

Location

The laboratories and administrative offices of the institute are located at Chelavoor (50 m above MSL), 11 km from Kozhikode (Calicut), Kozhikode District, Kerala, on the Kozhikode - Kollegal road (NH 766), in an area of 14.3 ha. The research farm is located 51 km North East of Kozhikode 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 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* Linn.), cardamom (*Elettaria cardamomum* Maton), ginger (*Zingiber officinale* Rosc.), turmeric (*Curcuma longa* Linn.), cinnamon (*Cinnamomum verum* J. Presl.), cassia (*C. cassia* Nees ex Blume), clove (*Syzygium aromaticum* (L.) Merrill & Perry), nutmeg (*Myristica fragrans* Houtt.), allspice (*Pimenta dioica* (L.) Merrill & Perry), Garcinia (*Garcinia gummi-gutta* (L.) N. Robson and *G. indica* Choisy) and vanilla (*Vanilla planifolia* Jacks. ex Andrews).

Organization

The Director is the administrative head of the institute. The Institute Management Committee, Research Advisory Committee and Institute Research Council 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 Knowledge Management Unit, Bioinformatics Centre and Krishi Vigyan Kendra. The institute also functions as the headquarters for the All India Coordinated Research Project on Spices (AICRPS), and Indian Society for Spices (ISS). The institute has also 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 2215 lakhs during the year, which included 945 lakhs (including



ORP on PhytoFuRA, Network project on HVC and Organic Horticulture) under Plan and 1270 lakhs under Non Plan.

Resource generation

Institute earned a total of 32.0 lakhs through sale of planting materials, biocontrol agents, training, publications and consultancy services.

Staff

The institute has a sanctioned strength of 44 scientific, 33 technical, 24 administrative and 61 supporting staff, of which 31, 14, 16 and 07 of scientific, technical, administrative and supporting staff, respectively are in position. The KVK has a sanctioned strength of 2 administrative, 12 technical and 2 supporting staff.

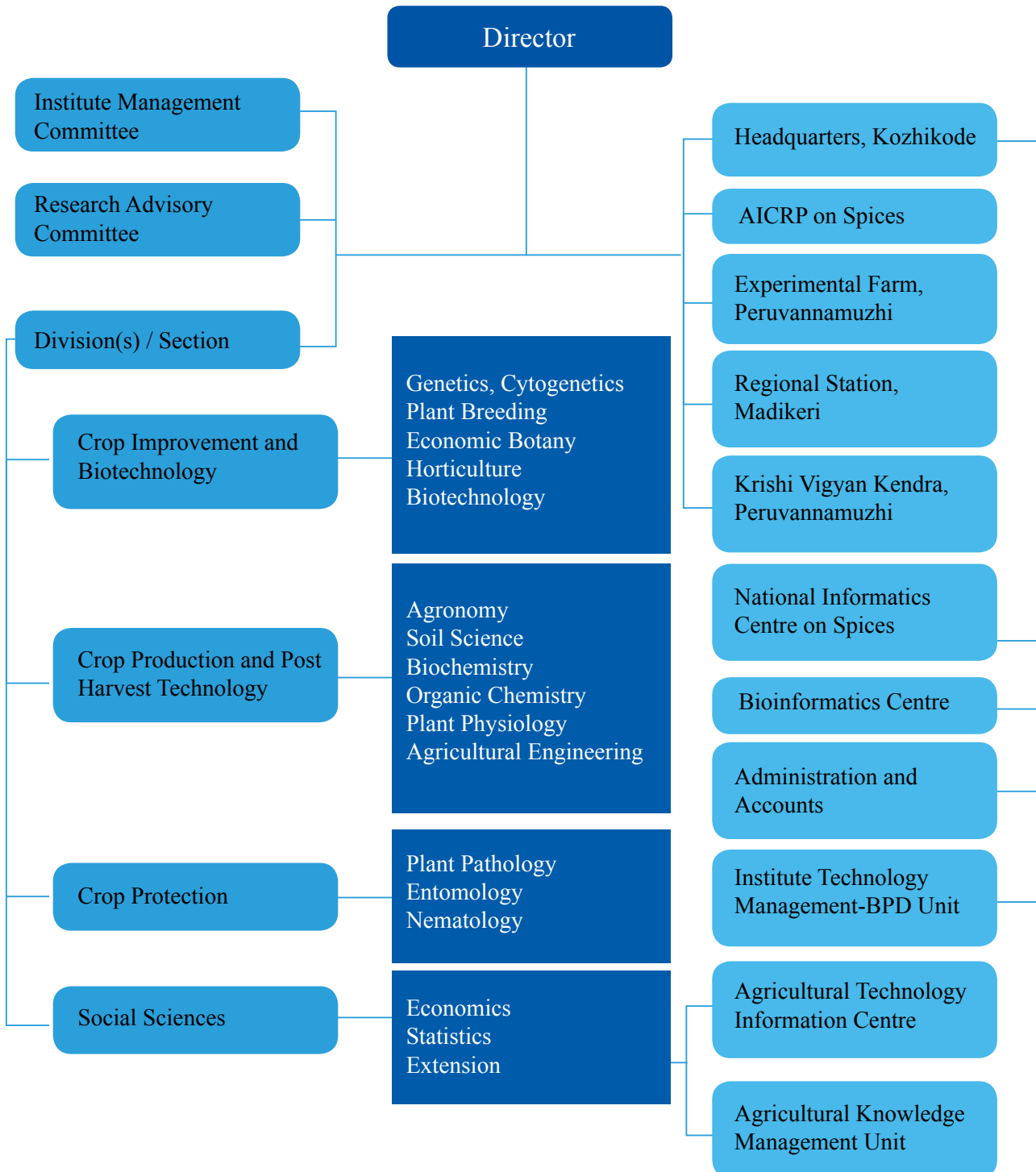
Staff position of the Institute

Category	Sanctioned	Position			Total	Vacant
		Kozhikode	Peruvannamuzhi	Appangala		
Scientific	44	31	-	05	36	08
Technical	33	14	10	04	28	05
Administrative	24	16	-	02	18	06
Supporting	61	07	05	11	23	38
Total	162	68	15	22	105	57

Staff position of KVK

Category	Sanctioned	Position			Total	Vacant
		Kozhikode	Peruvannamuzhi	Appangala		
Scientific	01	-	-	-	-	01
Technical	11	-	10	-	10	01
Administrative	02	-	01	-	01	01
Supporting	02	-	02	-	02	-
Total	16	-	13	-	13	03

Organizational chart of ICAR - Indian Institute of Spices Research, Kozhikode





PAST ACHIEVEMENTS

BLACK PEPPER

Germplasm collections obtained over the years through explorations are being maintained at IISR as well as at other alternate sites viz., Appangala and Chettali of Karnataka for developing improved varieties for yield, quality, abiotic and biotic stresses. The genetic stock has led to release of nine improved varieties such as Sreekara, Subhakara, Panchami, Pournami, PLD-2, IISR Thevam, IISR Girimunda, IISR Malabar Excel and IISR Shakthi. FLD programme was undertaken using the released varieties in the farmers' field. Two accessions, INGR 8099- *P. thomsonii* (IC 398863) - for its unique character for sex change and INGR 8100- *P. nigrum* (IC 563950) – a novel spike variant with proliferating spikes, were registered with NBPGR, New Delhi for their unique characters. Endangered species viz. *P. barberi* and *P. hapnium* were located and collected from Sabari hills. Microsatellites developed for *Piper* species were successfully used to detect polymorphism in black pepper cultivars. Assembly and functional annotation of sequences derived from the transcriptome of *P. colubrinum* and *P. nigrum* helped in the identification of many genes involved in defense and secondary metabolism. Seedlings of *P. colubrinum* on screening with *P. capsici* showed segregation of the resistance character, 21 plants being resistant to *Phytophthora*, two plants susceptible and the rest showing moderate resistance. 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.

The spacing, nutrient and water requirements were standardized for different soil types of pepper growing regions. Irrigating pepper vines once in a fortnight from March to May months at the rate of 50 litres vine⁻¹ enhanced yield substantially. High production technologies and mixed cropping systems were developed for increasing productivity. Among different forms of K, water-soluble K and available K had significant positive correlation

with berry yield, oleoresin and piperine. Organic production technology for black pepper has been standardized. Crops such as ginger, tapioca, coleus, amorphophallus and hybrid napier were found suitable for intercropping in black pepper gardens that are more than 15 years old. Intercropping medicinal plants (*Vetiveria zizanoids* and *Alpinia calcarata*) in juvenile black pepper garden was found to be profitable with a B: C ratio of 2.3. Cost effective method for production of disease-free rooted cuttings was developed. A machine was fabricated in collaboration with CIAE, Coimbatore centre which is capable of mixing, pulverizing, sieving, and filling of potting ingredients in poly bags at desired quantity. Mathematical models for optimum climatic factors for high production of black pepper have been developed. Targeted yield equations for predicting nutrient requirements for fixed yield targets in soils with varying fertility levels were standardized with minimum deviations in black pepper. Major pests, pathogens, viruses and 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) are found to be 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 PYMoV in a single reaction was standardized. SYBR green based real-time PCR was developed for detection of PYMoV and CMV in black pepper. 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 availability of pesticide free produce.

Large scale 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. The presence of β -1, 4 endoglucanase, a major secretory cellulose enzyme in nematodes, was located in *R. similis* through EST analysis. Black pepper accessions, HP-39 and Acc. 1090 were found to be resistant to nematodes besides being rich in caryophyllene. Endophytic bacteria effective against *Phytophthora capsici* and *R. similis* in black pepper have been isolated. Culture filtrates of BRB 13 at 40 μ L mL⁻¹ caused 100% mortality of *R. similis* within 24h. Basal application of *T. harzianum* and aerial spray with 1% Bordeaux mixture was found effective in controlling anthracnose disease.

An integrated pest management schedule for management of root mealy bug has been developed. Metalaxyl-MZ sensitivity of 81 phytophthora isolates was tested and the EC₅₀ and EC₉₀ values ranged from 0.0002 to 14.4 ppm and 1.1-68.5 ppm, respectively. Among the new chemicals tested *in vitro* against *P. capsici*, Acrobat 50 showed 100% inhibition at 50 ppm concentration. Profiling and activity prediction of biochemical compounds using *in silico* tools were completed for *Pseudomonas putida* BP 25 and *Bacillus megaterium* BP 17. PCR based techniques were developed for identification of traded black pepper and to detect adulterants in commercial black pepper powder. The existence of fungicide sensitive or resistant isolates among the field populations of *C. gloeosporioides* infecting black pepper was noticed in Pollibetta and the isolate from this locality was tolerant to recommended doses of Bordeaux mixture and carbendazim. Post harvest technologies for drying, processing, storage and production of value-added product like white pepper were standardized.

Genetic diversity of *Phytophthora* isolates from black pepper was studied by SSR profiling and ITS sequencing with the universal primers ITS 6 and ITS 4. A native isolate of *P. capsici* (Is. No. 98-93) infecting black pepper was completely sequenced using next generation sequencing platform, Illumina - Solexa GA II. ITS region of *R. similis* was amplified with universal primers. A new database, *Phytophthora* Genome Database (<http://220.227.138.212/>)

genomedb/) based on *Phytophthora* whole genome sequencing and annotation was developed. PhytoWeb, a comprehensive portal on *Phytophthora* diseases of horticultural crops in India was developed. Phytolib, an electronic database of research publications on *Phytophthora* and database on *Radopholus* genus RADOBASE were developed and launched.

Impact studies on adoption of IISR varieties of black pepper in farmers' fields indicated that the mean yield for high yielding varieties was 1160 kg ha⁻¹ with the adoption of scientific packages as compared to 620 kg ha⁻¹ for traditional varieties. The estimated cost benefit ratio was 2.48. The level of adoption studies of recommended technologies indicated that the adoption level for aerial spraying of Bordeaux mixture for the control of fungal diseases was 57.14% and for application of biocontrol agents was 64.2%. The adoption level for application of soil fungicides, fertilisers and pesticides were very low at 21.14%, 7.7% and 7.6 % respectively. *Karshika Sankethika Darshanam* and Media Meet were organized to mobilize mass media support for sharing Agro-Information. Video films on Augmenting black pepper production – a success story (Malayalam, English, Hindi) and success story of a 'Prathiba' grower – were produced.

CARDAMOM

Germplasm collections obtained over the years through explorations are being maintained at IISR Regional Station, Appangala, Karnataka and IC numbers have been obtained for all the available germplasm. Meanwhile, germplasm bearing unique characters have been registered with NBPGR, New Delhi. The improved varieties such as Appangala 1, IISR Vijetha, IISR Avinash and Appangala 2 have been developed. Coupled with production technologies, these varieties resulted in increasing productivity of cardamom. About 10 high yielding F1 hybrids were promoted to coordinated varietal trials.

Molecular profiles were developed for 100 accessions of small cardamom germplasm using 25 ISSR markers for studying the genetic diversity and dendrogram of similarity was prepared. Molecular profiling of Indian cardamom revealed the existence of two genetically distinct clusters



such as “Kerala cluster” and “Karnataka cluster” among the germplasm collections. 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. GC-MS study confirmed superiority of Indian cardamom over Guatemalan and Sri Lankan cardamom. High production technology has been standardized. Drip irrigation and sprinkler irrigation once in 12 days significantly improved yield attributing characters. 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.

A procedure for total RNA isolation and detection of CdMV through reverse transcription–polymerase chain reaction (RT-PCR) using primers designed for the conserved region of coat protein was standardized. A protocol for SYBR green based real-time RT-PCR for detection of *Cardamom mosaic virus* (CdMV) and *Banana bract mosaic virus* (BBrMV) in cardamom was developed. Surveys conducted in Karnataka and Kerala, revealed the prevalence of *Banana bract mosaic virus* (BBrMV) infection. A reliable RT-PCR based method was also developed for detection of the virus in plants. The survival of *C. gloeosporioides* infecting cardamom in infected plant part (leaves) was studied under laboratory, greenhouse and field conditions. A new bacterial wilt disease on small cardamom was noticed in Wayanad, Kerala. Phenotypic and genetic characterization revealed that the causative organism is *R. solanacearum* biovar 3 phylotype 1. Multiplex-PCR based phylotyping, 16s rDNA & recN gene sequence based comparison and MLST based comparative genetic analysis further revealed that the strain is 100% similar to the ginger strain of *R. solanacearum*.

GINGER

Germplasm repository of ginger at IISR is the largest collection with several exotic collections and high quality accessions. Six hundred and sixty eight accessions are being maintained in field germplasm conservatory. Three varieties namely, IISR Varada,

IISR Rejatha and IISR Mahima were released for high yield and quality. Cross specific amplification of rice microsatellites was successfully done in ginger. Acc. 195, a tetraploid having $2n=44$, showed mean pollen fertility of 67.73% by glycerol-carmin staining and 60.31% by *in vitro* germination and is suitable for future studies on induction of seed set. Two accessions irradiated with gamma rays showed resistant reaction after three repeated inoculations with *R. solanacearum*. Ginger oil components have been characterized by GC-MS. A relationship between leaf P/Zn ratio and soil P/Zn ratio to rhizome yield has been established. Targeted yield equations for predicting nutrient requirements for fixed yield targets in soils with varying fertility levels were standardized with minimum deviations. Post harvest technologies for processing and technologies for preparation of value added products such as salted ginger were standardized. Comparison of essential oil constituents of fresh and dry rhizomes indicated that fresh rhizomes contained higher level of monoterpenes namely, Z-citral and E-citral whereas the dry rhizomes were predominated by the sesquiterpene hydrocarbons viz., zingiberene, farnesene and sesquiphellandrene. 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*.

Nine actinomycete isolates from ginger soil were found to be antagonistic to *R. solanacearum*. Technique for ginger seed rhizomes treatment (for elimination of bacterial wilt pathogen) and integrated disease management strategy for soft rot and bacterial wilt diseases and shoot borer was developed. *Bacillus amyloliquefaciens* (GRB 35) was effective for disease control and plant growth promotion. PGPR formulation to enhance nutrient mobilization and growth, yield and biocontrol was developed and commercialized.

The life cycle of shoot borer (*Conogethes punctiferalis*) was studied on six resistant and six susceptible accessions. The infectivity of EPNs

strains IISR-EPN 01 to 08 was tested against shoot borer larvae under *in vitro* conditions. One species of EPN belonged to *Oscheius gingeri* was identified as new species on the basis of morphological and molecular characterization. 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 conserved in the field gene bank and were characterized for yield, quality, and resistance to pests, diseases and drought. Seven high curcumin and high yielding varieties, Suvarna, Sudarsana, Suguna, IISR Prabha, IISR Prathiba, IISR Alleppey Supreme and IISR Kedaram were released for commercial cultivation. Open pollinated seedling progenies generated over the years are being evaluated for their yield and quality characters.

Molecular genetic fingerprints of 16 *Curcuma* species using RAPD and ISSR technique revealed high degree of polymorphism among the accessions. A total of 140 microsatellites containing genomic DNA fragments were isolated adopting the selective hybridization method with di and trinucleotide biotinylated probes. Two synonymous *Curcuma* species viz., *C. zedoria* and *C. malabarica* showed identical SSR profiles for 40 microsatellite loci. 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. About 40 seedling progenies with higher curcumin (> 3%) and dry recovery (> 20%) were identified. Three different curcuminoids (curcumin, de methoxy curcumin and bis de methoxy curcumin) could be separated from oleoresin 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.

Targeted yield equations for predicting nutrient requirements for fixed yield targets in soils with varying fertility levels were standardized with minimum deviations. The economic optimum in terms of profitable response for money invested was found to be Rs. 0.65 bed⁻¹ for N, Rs. 0.40 bed⁻¹ for P and Rs. 0.85 bed⁻¹ for K. Increase in curcumin content was recorded when sprayed with micro nutrients like zinc and boron. Processing with or without boiling or different drying methods did not lead to variation in oil, oleoresin and curcumin contents. The optimum spacing, nutrient and water requirement were standardized for different soils and organic farming system was developed for turmeric. Basic data on distribution, bioecology, population dynamics of shoot borer (*Conogethes punctiferalis*) and its natural enemies and crop loss due to shoot borer was generated. Lambda cyhalothrin 0.0125% was more promising in reducing the percentage of shoots infested by the shoot borer. The improved varieties and technologies were disseminated to farmers and other agencies through publications and demonstrations. The adoption of released varieties like IISR Prathiba in Andhra Pradesh, Karnataka and Tamil Nadu were studied. Soil pH based micronutrient mixtures for enhancing growth, yield and quality of turmeric, ginger, black pepper and cardamom were developed.

TREE SPICES

The germplasm holdings of important tree spices like nutmeg, clove, cinnamon including cassia, garcinia and allspice are being conserved. 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 accession. The cassia elite line A1 (IC 370400) has been registered with NBPGR for high cinnamaldehyde content in bark oil (81.5%) and leaf oil (80.5%). Two high quality cinnamon varieties, Navashree and Nithyashree and a nutmeg variety, Viswashree were released. Nutmeg accession, A11/25 was found to be promising for



high yield. Nutmeg accession A9-71 (IC537220), as a source of high sabinene (45.0% sabinene in nutmeg oil and 41.9% sabinene in mace oil) was registered with NBPGR. 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. Green chip budding with orthotropic buds was standardized in nutmeg on *Myristica fragrans* rootstock with 90-100% success.

GC-MS study revealed the presence of two chemotypes in *Cinnamomum verum*. Drying and processing methods for cinnamon, nutmeg and mace have been developed. Antioxidant properties and food color value are being studied in tree spices. GC-MS analysis of the chemical constituents of essential oils in leaves of *C. sulphuratum*, *C. glaucescens*, *C. glanduliferum*, *C. macrocarpum* and *C. perrottetti* revealed that the major chemical constituents in these oils were α -phellandrene, β -phellandrene, camphor, t-caryophyllene and germacrene-D respectively. Vegetative propagation techniques were standardized for nutmeg, cassia and cinnamon. Major pests and diseases on tree spices were documented. The improved varieties and technologies developed on propagation and post harvest processing were disseminated to farming community.

Four species of *Garcinia* viz., *G. kydia* (Kuji Thekera), *G. lancifolia* (Rupohi Thekera), *G. pedunculata* (Bor Thekera) and *G. xanthochymus* (Tepor Tenga) were located in Meghalaya, Assam and Nagaland. Hot water extraction and Solvent extraction (Methanol/chloroform -1:1) of *G. gummigutta* and *G. tinctoria* yielded 50% butter with yellow colour and pleasant aroma.

VANILLA

Vanilla germplasm are being maintained in the repository, which includes a flower colour variant collected from Andaman and Nicobar islands. Comparative anatomical analysis of different vanilla species was carried out. Interspecific hybridization

was made between *Vanilla planifolia* and *V. aphylla*. Reciprocal crosses were conducted between *V. planifolia* and *V. tahitensis* (species reported as resistant to root rot disease) and high percent of fruit set was observed in both the crosses. Fifty interspecific hybrids each of *V. planifolia* x *V. tahitensis*, *V. tahitensis* x *V. planifolia* and selfed progenies of *V. tahitensis* were established *ex vitro*. Chromosome number analysis of two interspecific hybrids between *V. planifolia* and *V. tahitensis* showed $2n=30$ in one (PT-5) and $2n=32$ in the other (PT-17).

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

PAPRIKA

The germplasm collected from various places of cultivation were characterized for various morphological, yield and quality characters such as oleoresin, pungency and colour value. Considerable variability was observed in total extractable colour and capsaicin content (pungency) of selected paprika accessions. The lines ICB-10, Kt-pl-19 and EC-18 were found promising with high colour value and low pungency. PCR based technique was developed to detect adulterants in commercial chilli powder.

RESEARCH ACHIEVEMENTS

BLACK PEPPER

Genetic resources

The germplasm accessions at the NAGS center are maintained at the Experimental farm, Peruvannamuzhi. This year, 52 Kottanadan accessions were augmented in the field genebank. Improved varieties and example varieties were planted and conserved under protected conditions for conservation. Two hundred and seventy five accessions were planted during this year at the alternate center CHES, Chettalli. At present the alternate field gene bank at CHES, Chettalli has 427 accessions. A field gene bank with 223 accessions was established at Kozhikode. For the production of top shoot in large numbers, all the improved varieties were planted in the conservatory under controlled conditions on dead standards with 1.5 m height. Sixty six plants were established in two conservatory sheds. The top shoots were harvested and planting materials were produced. One set of top shoot derived plants of all the released varieties were handed over to farm for establishing a varietal block. Exploration and collection programmes were carried out. The major areas surveyed were Idukki, Thodupuzha, Anakulam, Mankulam and Kannur in Kerala and Kodagu district in Karnataka and 175



Fig 1. Unique accessions collected, 1. Arakulamunda (high yielding); 2. Karimunda (long spikes, 7-8 cm); 3. Karimunda (bold berries); 4. Nedumchola

accessions were collected during the survey. This includes 90 cultivars and 85 accessions of related taxa. Few unique accessions were also collected during the survey (Fig 1). Surveys done in the forest ranges under Mankulam forest divisions (Anakulam, Viripara, Bhagavathi shola, Pettimudi and Kadalar, 590-1780 MSL) yielded 73 related Piper species.

Unique accessions collected

- An Arakulamunda with very good fruit setting and high yield- Kannur
- A Karimunda accession with long spike (8 cm) - Kannur
- Karimunda type with bold berries and good setting- Kannur
- Nedumchola- The black pepper cultivar with smallest leaf and spike. Farmers generally do not prefer this cultivar and it becoming very rare in black pepper gardens

Piper barberi population in Anakulam forests, Western Ghats

A thriving population of *Piper barberi* – an endangered species listed in the Red Data book was located in the evergreen forests of Anakulam forest range for the first time (Fig 2).

Breeding

A replicated yield trial involving 10 improved



Fig 2. Spikes of the back cross progeny [HP1117 x Aimpirian] x Aimpirian

lines/selections along with two controls was laid out at Peruvannamuzhi. Flowering was recorded in the following entries, HP 780-5/30; OPKm-1/30; Thevam-7/30; HP 1411-1/30; Sreekara 6/30 and back cross progeny-3/30 (Fig 2). The fresh yield varied from 0.05 (Acc.820) – 1050g (Thevam). HP 780 recorded highest dry recovery of 37%.

Screening of hybrids for pollu beetle resistance

Twenty pollu beetle resistant hybrids available at Chelavoor farm were screened for leaf damage against pollu beetle, of which, three hybrids exhibited symptoms of pollu beetle damage on the leaves.

Hybridization involving *P. colubrinum* and characterization of the mentor grafted progeny

Double Digest RAD (Restriction site Associated DNA) sequencing was undertaken in mentor grafted progenies in which *P. colubrinum* was used as root stock to confirm nuclear DNA and organellar DNA transport after mentor grafting. The studies indicated the presence of *P. colubrinum* specific sequences in one of the progeny derived from mentor graft (Table 1).

Table 1. List of *Piper colubrinum* specific sequences discovered from mentor progeny of black pepper

Sequence origin	Number of sequences
<i>Piper colubrinum</i> –nuclear specific	302
<i>Piper colubrinum</i> –chloroplast specific	83
<i>Piper colubrinum</i> –mitochondrial specific	49

Comparative anatomical studies also revealed variation in the mentor grafted progeny as compared to the stock and scion (parents) for the presence of trichomes, mucilage canals, arrangement of vascular bundles etc (Fig 3).

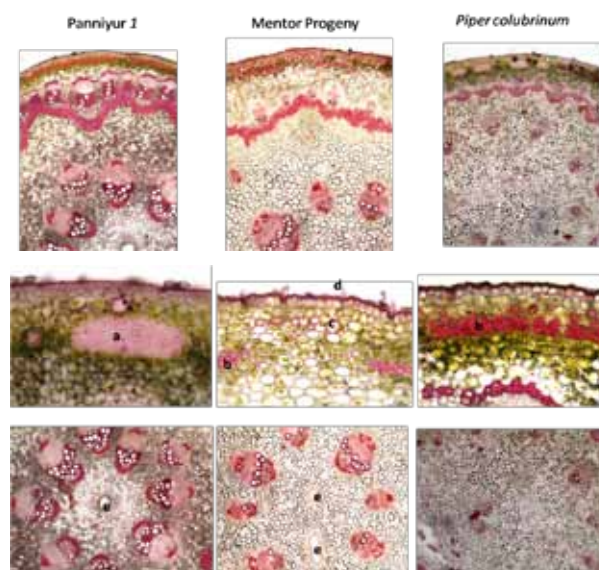


Fig 3. Comparative anatomical features of mentor grafted progeny vs the parents a. Collenchymatous patch; b. Sclerenchymatous patch; c. Glands; d. Trichomes; e. Mucilage ducts

Indexing for drought tolerance using physiological parameters and optimization and selection of molecular markers for screening

Using four polymorphic SSR Markers 20 hybrids were screened and polymorphism detected. Physiological screening of the 20 accessions for linking these polymorphic markers with drought tolerance is in progress.

Expression profiling of genes under water-deficit stress

Ubiquitin gene was found to be the most stable reference gene identified using three different accessions of *Piper nigrum* under water deficit stress using Reffinder software. *GAPD* gene was found to be least stable under the above conditions. Among the genes tested for expression analysis, *Myb* and *NAC protein* genes were found to be expressed 3-fold and above in susceptible Sreekara plants under water deficit when compared to control. The increase in expression of these genes were low in drought tolerant Acc. 4216. *Dehydrin* gene was

again found to be expressed to many folds in Acc. 4216 compared to low expression in Sreekara.

Differential expression proteomics of water deficit stress

Drought was induced in tolerant (Acc. 4226) and susceptible (Subhakara) accessions of black pepper. Subhakara plants started showing wilting symptoms

in 8 days (75 % RWC at 12 % soil moisture) while accession 4226 showed decreased turgidity after 14 days (82 % RWC at 11 % soil moisture) but did not wilt. The total proteins were extracted from control and wilted plants and were analyzed by 2D electrophoresis to elucidate differential profiles of expressed proteins during drought (Fig 4).

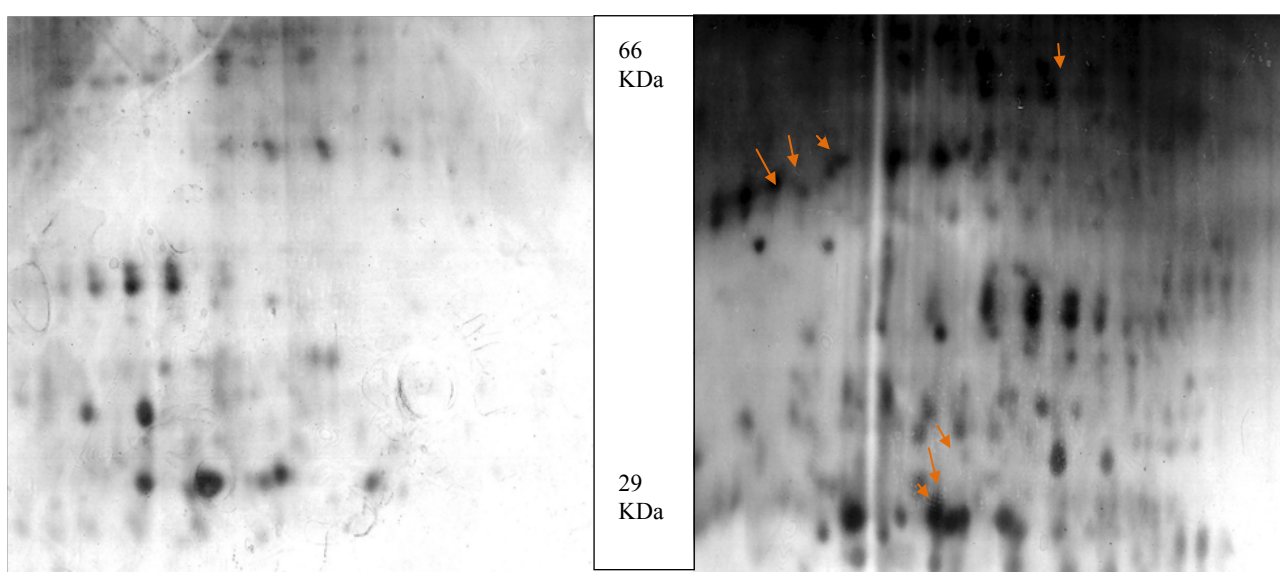


Fig 4. 2D- gel electrophoretic analysis of Subhakara plants showing differentially expressed proteins ranging from 66KDa to 29KDa in drought imposed Vs ABA treated. Arrows shows the unique protein spots present in the particular gel when compared to the other.

ABA response (exogenous ABA) under drought stress

Subhakara plants showing initial wilting symptoms under drought stress (after 7 days) were treated with 250 μ M ABA (both spray and soil drenching). After ABA application, these plants showed complete wilting only after 14 days of stress. Acc. 4226 showed initial wilting symptoms after only 24 days of stress. Wilting of Subhakara could be phenotypically reversed to certain extent with ABA application but not in case of Acc. 4226. Total proteins were extracted from leaves of ABA treated plants and analyzed by 2D electrophoresis to elucidate the differential profile of expressed proteins during drought. Acc. 4226 showed similar profile even

after ABA application. New protein spots along with differentially expressed spots could be identified in Subhakara after ABA treatment which suggests that the exogenous application would have altered the protein pattern to tolerate drought.

Mining for ABA regulatory pathway genes

Important ABA responsive regulatory pathway gene loci, *Aldehyde Oxidase*, *NCED*, *9-cis-epoxy carotenoid deoxygenase* and *Serine Threonine protein Kinase* genes were identified from the *Piper* transcriptome database. Conserved domain search (Interproscan and PROSITE) for the loci yielded important catalytic domains.



Comparison of horizontal and vertical methods in black pepper planting material production

The rooted cuttings of two varieties viz., V1- IISR Thevam and V2- Panchami were allowed to grow both vertically (M1) and horizontally (M2). To support vertical growth, two meters tall hollow vertical column with 0.3 m diameter was made with half an inch thick plastic coated welded wire mesh filled with composted pasteurized cocopeat and vermicompost @ 3:1 ratio fortified with bio-control agent *Trichoderma harzianum*. For horizontal method, a bed having 1.5 m width along the length of poly house to a height of 15 cm with same medium

was made and poly bag cuttings are arranged one side and allowed to creep on the bed. The columns and beds were set up in hi-tech poly house. The result indicated that at the end of 100 days the vines on the column grew to 109.8 cm height with 20.6 nodes and leaves whereas vines on the bed grew to 123.8 cm with 19.5 nodes and leaves (Table 2 and 3). In vertical column method, 48.8% of plants of IISR Thevam and 35.8% of Panchami have produced lateral branches and the vines on the beds have not produced any lateral branch. It was found that three types of cuttings i.e., top shoot (orthotrope), lateral and normal cuttings could be harvested from a vertically grown plant whereas horizontally grown plants on bed could give only normal cuttings.

Table 2. Effect of method of raising shoot on length (cm) of black pepper vine

Treatment	Days after planting						
	15	25	35	50	70	80	100
Method							
Horizontal (M1)	16.14	24.86	37.81	53.36	77.52	98.20	123.83
Vertical (M2)	19.87	29.10	40.80	55.76	75.83	92.61	109.83
SEd ±	1.27	1.11	1.51	2.45	3.11	4.65	4.95
CD(P<0.05)	2.76	2.43	NS	NS	NS	NS	10.79
Variety							
IISR Thevam (V1)	17.65	26.81	39.46	55.12	75.15	92.04	113.00
Panchami (V2)	18.36	27.15	39.15	54.00	78.20	98.77	120.66
SEd ±	1.27	1.11	1.51	2.45	3.11	4.65	4.95
CD(P<0.05)	NS	NS	NS	NS	NS	NS	NS
Interaction							
M1V1	15.48	24.34	37.50	54.58	76.00	95.20	119.66
M1V2	16.80	25.38	38.12	52.14	79.04	101.20	128.00
M2V1	19.82	29.28	41.42	55.66	74.30	88.88	106.34
M2V2	19.92	28.92	40.18	55.86	77.36	96.34	113.32
SEd ±	1.79	1.57	2.14	3.47	4.40	6.57	7.00
CD (P<0.05)	3.90	3.44	NS	NS	NS	NS	15.27
CV (%)	15.72	9.25	8.59	10.05	9.08	10.89	9.45

Table 3. Effect of method of raising shoot on leaf number of black pepper vine

Treatment	Days after planting						
	15	25	35	50	70	80	100
Method							
Horizontal (M1)	4.33	5.71	7.39	9.63	12.76	15.98	19.54
Vertical (M2)	5.03	6.67	8.83	11.41	14.61	17.41	20.65
SEd±	0.30	0.25	0.27	0.40	0.26	0.48	0.66
CD(P<0.05)	0.65	0.54	0.60	0.88	0.57	1.05	1.43
Variety							
IISR Thevam (V1)	4.50	6.02	8.00	10.45	12.96	15.71	18.97
Panchami (V2)	4.86	6.36	8.22	10.59	14.41	17.68	21.22
SEd±	0.30	0.25	0.27	0.40	0.26	0.48	0.66
CD(P<0.05)	NS	NS	NS	NS	0.57	1.05	1.43
Interaction							
M1V1	4.06	5.44	7.22	9.70	11.74	14.70	17.82
M1V2	4.60	5.98	7.56	9.56	13.78	17.26	21.26
M2V1	4.94	6.60	8.78	11.20	14.18	16.72	20.12
M2V2	5.12	6.74	8.88	11.62	15.04	18.10	21.18
SEd±	0.42	0.35	0.38	0.57	0.37	0.68	0.93
CD(P<0.05)	0.92	0.77	0.84	1.24	0.80	1.49	2.02
CV (%)	14.4	9.0	7.6	8.5	4.2	6.5	7.3

Soil carbon pools under cropping systems

The total and particulate organic carbon and nitrogen pools were quantified under different spice based cropping systems and high density multiple cropping system. The particulate organic carbon (POC) and particulate organic nitrogen (PON) pools were higher in coffee + pepper system (56.7 and 16.8 Mg ha⁻¹) with highest total organic C and N (TOC and TON) pools (90.1 and 33.4 Mg ha⁻¹). POC constituted 63% of TOC in this system. The non particulate carbon and nitrogen (NPOC and NPON) pools were higher under cardamom alone and coffee + pepper + cardamom cropping system (67.3 and 58.3 Mg ha⁻¹) constituting 73-78% of total organic carbon pools.

Among different management systems in black pepper, organic management has accumulated higher POC, NPOC and TOC pools as compared to

integrated and conventional management systems. In HDMCS, black pepper basin has accumulated highest TOC, NPOC and POC pools (106.8, 71.6, 35.2 Mg ha⁻¹, respectively) and coconut and nutmeg systems higher NPON and PON (7 and 0.8 Mg ha⁻¹) as compared to other component crops.

Management of virus affected black pepper gardens for yield sustainability

Trials were taken on the management of virus affected black pepper gardens for recouping its health and sustaining the yield at three estates in Madikeri district of Karnataka. Five treatment combinations were tried viz. T1: FYM + Fertilizers as per recommendation, T2: T1 + Micro nutrient spray (twice), T3: T1 + PGPR soil application, T4 : T3+ Micronutrient spray (twice) and T5 : Control. The black pepper vines (predominantly var. Panniyur-1)

were graded for their virus infection status as mild, moderate and severe based on their visual symptoms and canopy coverage. Mild and moderate graded vines were selected for imposing different treatments at three locations viz., Madapura, Chetalli and Pollibetta.

The treatments were applied in two splits, once in June – July and second in August - September. Observations on health of the vine (visual), symptoms on new leaves, spike intensity, leaf nutrient concentrations and yield were recorded. More number of leaves were produced and retained in all the fertilizer/PGPR treatments as compared to control. The spike intensity was also higher in application of nutrients or PGPR. The yellowing/health status and the canopy size of the vines improved with the application of nutrient/PGPR treatments as compared to control in both mild and moderately virus infected vines. Mildly virus affected vines showed better improvement of the health and canopy status than moderately affected vines. The application of nutrients had more influence on the vine health status and the canopy size development than that of PGPR application in Palonjee estate where as PGPR showed a pronounced effect in Mrigarajendra estate. Leaf nutrient contents of mild and moderately virus infected vines were similar.

Development of mechanical unit for production of white pepper from green pepper

Experiments were conducted to determine the effect of replacement of water on the quality of white pepper produced. Freshly harvested green pepper (Panniyur 1) obtained from Experimental Farm, Peruvannamuzhi was used for the production of white pepper. The result obtained from the above experiment indicated that white pepper produced by daily change of water showed maximum whiteness of the produce with a dry recovery of 20.97%. The result of the above experiment was done in a scaled up manner in the fermentation unit installed at the Spice Processing Unit located at IISR, Experimental Farm, Peruvannamuzhi.

In vitro antioxidant activity and cytotoxicity of sequential extracts from selected black pepper varieties and Piper species

Antioxidant activity and cytotoxicity of four medicinally valued *Piper* species viz. *P. nigrum*, *P. chaba*, *P. longum* and *P. colubrinum* were examined. Among all extracts investigated, methanol extracts showed highest antioxidant activity followed by chloroform extracts for all the four assays. Methanol extract of Malabar Excel was found to be highest for all the assays followed by methanol extract of *P. colubrinum*. *In vitro* cytotoxicity was checked on cervical cancer cell line CaSki by MTT assay. Results indicated that chloroform extract of all the samples and hexane extract of *P. colubrinum* showed high cytotoxicity. Cytotoxicity increased with increase in the amount of extract as well as time of exposure of extract with CaSki.

Phytophthora foot rot pathogen diversity

Six new *Phytophthora* isolates from black pepper, colocasia and arecanut were added to the National Repository of *Phytophthora*; 433 isolates of *Phytophthora* are being maintained in the repository.

Multi Locus Sequence Typing (MLST) of *Phytophthora* was done using eight nuclear genes viz., 28S ribosomal DNA, 60S ribosomal protein L10, beta-tubulin, elongation factor 1 α , enolase, heat shock protein 90, TigA gene fusion protein and mitochondrial genome region between gene

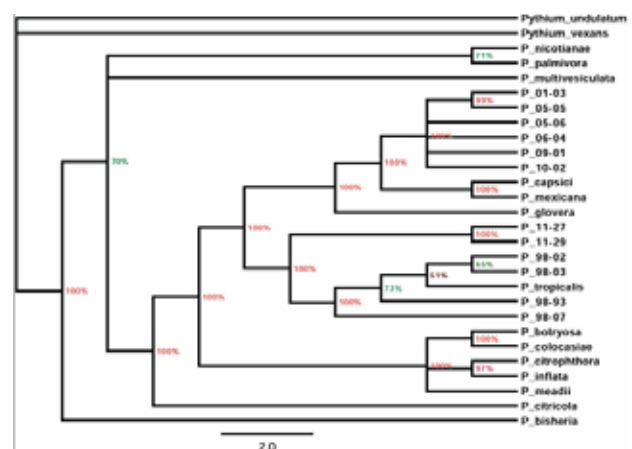


Fig 5. Phylogenetic tree based on Multi Locus Sequence Typing of *Phytophthora* isolates from black pepper.

Cox2 and *Cox1* and *Ras-related protein (Ypt1)* gene. Phylogenetic analysis using Bayesian method showed a separate grouping for *Phytophthora* isolates from black pepper (Fig. 5).

Comparative genomics of *Phytophthora* species

Secretome analyses of *Phytophthora* species were done using different software's like SignalP,

TMHMM and TargetP. Gene encoding proteins from five different *Phytophthora* species namely *P. capsici* (05-06 and 98-93), *P. sojae*, *P. infestans*, *P. ramorum* were taken for this analysis. Proteins with signal peptides were identified by sorting out proteins without transmembrane domains and subcellular localization for further comparative genomics studies (Table 4).

Table 4. *In silico* prediction of proteins with signal peptides from whole genome data of *Phytophthora* spp.

Species	Total number of sequences	No of secretory proteins	% of secretory proteins
<i>P. capsici</i> 05-06	19805	3180	16.056
<i>P. capsici</i> 98-93	9831	2085	21.208
<i>P. sojae</i>	19027	2037	10.705
<i>P. ramorum</i>	15743	1739	11.0461
<i>P. infestans</i>	18140	1848	10.187
<i>P. parasitica</i>	18795	2001	10.646

Screening for *Phytophthora* resistance

Phenotyping of progenies of Panniyur 1 and Subhakara for *Phytophthora* resistance

One hundred and forty progenies of Panniyur 1 x Subhakara were screened for *Phytophthora* resistance by stem and leaf inoculation methods with the virulent isolate 05-06. Among them five progenies of Panniyur 1 x Subhakara tolerated stem infection.

Phenotyping open pollinated progenies of IISR Shakthi and P24-0-4 for *Phytophthora* resistance

One hundred open pollinated progenies of IISR Shakthi and 27 open pollinated progenies of P24-0-4 were screened for *Phytophthora* resistance. Four of the progenies of IISR Sakthi tolerated stem infection. Only one progeny of IISR Shakthi could moderately resist leaf infection. None of the progenies of P24-0-4 tolerated leaf and stem infections.

Molecular profiling to identify markers associated with *Phytophthora* resistance in black pepper

Nine SSR primers were tested on selected progenies of IISR Sakthi. Four genotypes were selected based

on phenotyping data. Two primers namely, PnD10 and PnE3 were polymorphic among the progenies tested.

Reference gene normalization in *Piper* - *P. capsici* pathosystem

As stable reference gene is a prerequisite to study the expression of *Piper* genes during the pathogen infection, six reference genes from the *Piper* transcriptome database were analyzed for its expression stability by geNorm, normFinder, Bestkeeper and Reffinder algorithms and the optimal reference gene combination (PnUbCE and PnGAPDH) was identified. The identified reference genes were validated on the expression of beta glucanase gene between moderately resistant and susceptible genotypes using most stable as well as least stable (PnAct). β -1,3-Glucanase expression was analyzed after challenge inoculating highly virulent *Phytophthora* isolate (05-06) at different time intervals. The relative expression of PnBGlu was found to be up regulated in resistant variety (IISR Shakthi) from 0.5 h with the peak expression at



72 h while susceptible variety (Subhakara) recorded down regulation when analysed using REST 2009 software.

Targeted discovery of R genes

Expression based identification of R genes was attempted using the annotated *Piper* genes from *P. nigrum* transcriptome data. Nine resistance gene loci from transcriptome were taken for targeted gene expression analysis. Cloning of R gene c-DNA was made from pooled RNA samples taken at 0.5, 2, 4, 6, 8, 12, 24, 48 and 72 hpi. Further sequencing of clones and conserved domain search revealed that the R genes involved in *P. capsici* interaction are of coiled coil type of R genes. The phylogenetic analysis grouped the R genes into three different clades. Black pepper R genes showed only 20% similarity to *Arabidopsis* R genes hence pointing out the structural difference of *P. nigrum* R genes.

R gene expression dynamics in *P. capsici* interaction

Two step real time PCR was done to find out the expression pattern of the selected R genes (locus 6113, 81, 21935 and 26441) upon *P. capsici* interaction with extreme black pepper genotypes Subhakara (susceptible) and IISR Shakthi (moderately resistant).

R gene loci 81 and 26441 showed higher expression in susceptible variety in the early hours of infection while resistant genotype recorded its down regulation during the infection till late hours. The R gene loci 6113 and 21935 showed high relative expression in resistant genotype at early hours showing their direct involvement in resistance towards *P. capsici* whereas the expression of these two R genes were down regulated in susceptible. The R gene loci with high relative expression were forming one group in the (R gene) phylogenetic tree when compared to the loci with down regulation which were finding its place as separate group in the phylogeny tree.

Differential protein expression

Label free LC-MS/MS was employed to profile the differential protein expression in moderately

resistant genotype IISR Shakthi. High resolution peptide ion detection method identified spectrum of proteins from protein samples extracted from control and 24 hpi. Relative quantification using non conflicting peptides averaged normalized abundance estimation between treatments yielded the up and down regulated proteins. At 0.05% probability, 52 proteins were identified as differentially expressed. Out of that, 19 proteins were up regulated in infected plants which includes kinases, signal transduction proteins, transport proteins, defense related proteins including repetitive scaffold having role in biotic stress (ATP synthase subunit beta, Protease Do like 1, fructose aldolase, chloroplast rubisco activase, Histone H4, Coffea Canephora genomic scaffold, 20KDa chaperonin family protein, SOD, RNA binding protein, copper transport protein, nucleoside kinase, ATP synthase epsilon chain, Ribosome recycling factor, Enolase, Peptidyl prolyl cis-transferase, TPR repeat containing thio reductase) with the fold change ranging from 1.8 to 20.47 and 30 were down regulated. At 0.03-0.01 probability 37 proteins were identified as differentially expressed. Out of that, 9 showed upregulation in infected plants with fold change ranging from 1.73 to 8.08. The defence response genes and regulation factor proteins suggest their involvement in resistance against *P. capsici*.

Real time quantitative RT-PCR analysis of pathogenicity genes

Quantitative PCR was employed to assess the level of expression of some of the pathogenicity genes of *P. capsici* like *glycoside hydrolase*, *NPP 1*, *RXL*R and *pectate lyase* during *P. capsici*-*P. colubrinum* interaction. *Glycoside hydrolase* and *RXL*R genes showed high levels of expression during early stages of infection (up to 16 hpi), whereas the *NPP1* gene showed maximum expression at later stages of infection (at 72 hpi). *Pectate lyase* gene showed high level of expression at early stages of infection but was then down regulated during the later stages of infection (Fig 6). Phylogenetic analysis of these pathogenicity genes showed maximum similarity to *P. capsici* sequences in the database, except in the case of *glycoside hydrolase* which was grouped along with *P. soj*ae.

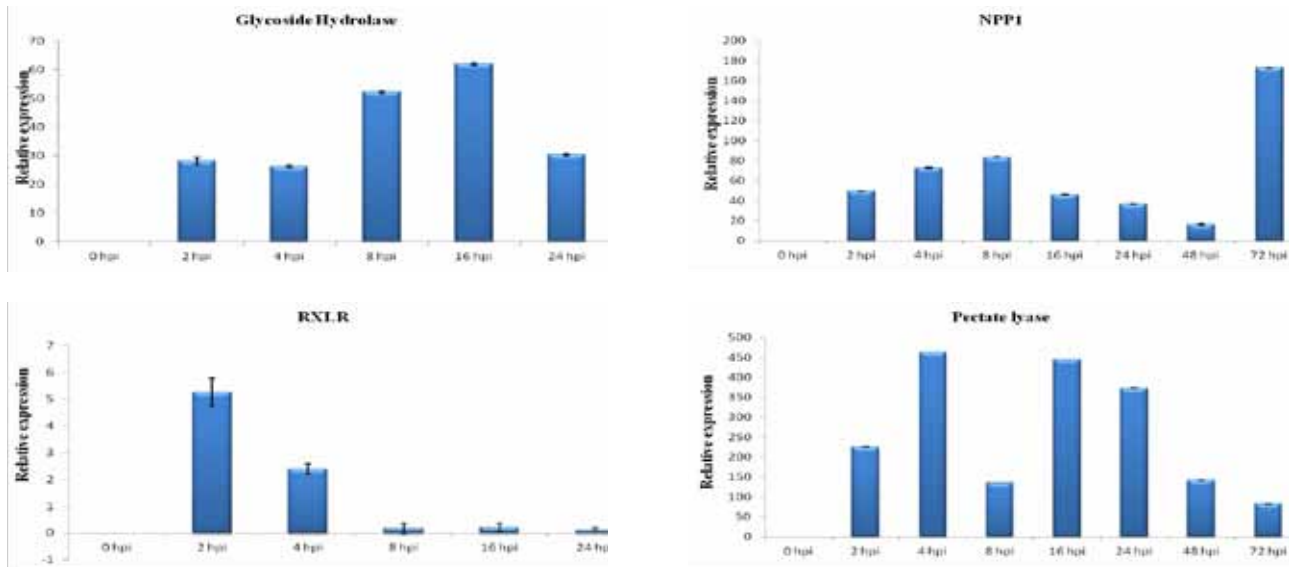


Fig 6. Relative expression of pathogenicity genes expressed during *Phytophthora capsici*–*Piper colubrinum* interaction.

In planta expression and docking studies

Species of the oomycete genus *Phytophthora* employ a matching counter defense system by secreting glucanase inhibitor proteins (GIPs) that specifically bind and inhibit the activity of plant endo-1, 3-glucanases *EGases*. The sequence characterization, *in planta* expression analysis and molecular docking studies of GIP and *P. colubrinum* endo beta-1,3 glucanase genes *pcEGase* based on sequence information derived from the *P. capsici* whole genome sequence data and *P. colubrinum* transcriptome data, respectively, were done. The presence of domains in each functional coding sequences and proteins were confirmed with blastX, PSI-blast and conserved domain database. The *GIP* gene from *P. capsici* have a 1059 bp ORF, encoding a putative peptide of 353 aminoacids and the partial sequence of *pcEGase* gene from *P. colubrinum* have a 936 bp ORF, encoding a putative peptide of 312 amino acids. The expression of these genes was studied *in planta* at different time points by qRT-PCR after challenge inoculation with the pathogen. The *in planta* expression of *GIP* gene from *P. capsici* was at its peak during initial hours of challenge inoculation and the expression of *pcEGase* gene was at its peak at 16 hpi (hours post inoculation). The peak expression of *pcEGase* gene from *P. colubrinum* at

16 hpi and sharp decrease in later periods indicate the successful neutralizing activity of the *pcEGase* gene against the *GIP* gene in this incompatible plant-pathogen interaction.

Three-dimensional model of *GIP* and *pcEGase* gene were constructed and molecular docking studies predicted sites on the surfaces of *pcEGase* gene and *GIP* that may be involved in high affinity binding. Molecular docking studies between *pcEGase* gene

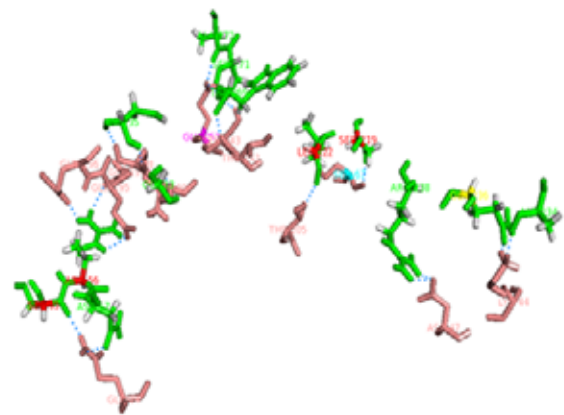


Fig 7. Polar covalent interaction of GIP with *pcEGase* active site residues. The active site residues are shown as sticks HelixSheet Loop and EGase shown in HelixSheetLoop. The interactions are shown in blue dashed lines.

and *GIP* revealed that substrate inhibition is obtained by recognizing arginine and isoleucine residues in the substrate molecule (Fig 7). Docking studies of *EGase* with *GIP* indicates that *GIP* blocks the active site of *EGase*. *GIP* was docked into the binding site of *Endo-β-1, 3-Glucanases* using Cluspro 2.0. The enzyme – substrate complex was analysed using SwissPDB viewer and PyMOL v1.7.2 in order to understand their mode of interactions and key residues involved in binding. The docked complex of *EGase* in complex with *GIP*, revealed 21 polar hydrogen bond interactions.

Expression analysis of defense associated transcription factors

PAMP triggered immunity (PTI) and effector triggered immunity (ETI) leads to biotic stress signalling involving induction of defence related transcription factors. Hence, studies on expression analysis of these transcription factors are very important to understand their importance during host pathogen interaction. Quantitative RT-PCR analysis was done for three transcription factors viz, *MYB*, *MYC* and *WRKY*, to assess the transcriptional activity of these genes during *P. colubrinum* - *P. capsici* interaction. Higher folds of expression of these genes were observed at initial stages of infection when compared to later stages. *MYB* gene showed up to 1.25 folds expression at 4 hpi, *MYC* gene showed 2.6 folds expression at 8 hpi and expression of *WRKY* gene was 1.65 folds at 4 hpi.

Identification and characterization of micro RNAs from *Piper colubrinum*

Dual RNA-seq data of disease-resistant *P. colubrinum* and its pathogen *P. capsici* (05-06) isolate from infected leaves in the early stages of infection (12 and 24 h post-inoculation) were analysed for microRNAs and their corresponding mRNA targets. Primary miRNA predicted from *Piper* transcripts of *P. colubrinum* against the precursor and mature sequences of known plant miRNAs deposited in miRBase version 16 and MIREAP programs. Sequences with an E-value of lower than 0.05 or a score > 32 were processed for further analysis, allowing for a maximum of 1 nt mismatches and 190 precursor miRNAs present in *P. colubrinum* transcriptome were identified.

To understand the corresponding mRNA targets from *P. colubrinum*, which are critical to understanding many pathways and biological systems in which miRNAs are involved, BLASTN searches performed against *P. colubrinum* assembled transcript database to identify putative targets for miRNAs. Similarities with an E-value of less than 0.05 were considered a hit. Around 4542 putative targets were identified from *P. colubrinum* coding transcripts for the predicted miRNAs. Out of these, 881 transcripts were predicted with putative functions and these predicted targets were involved in inhibition and cleavage of various molecular functional genes, such as *cytochrome c biogenesis protein*, *serine/threonine-protein phosphatase*, *NEDD8-like protein* *RUB2*, translation, transcription, etc.

Search for targets in *P. capsici* genomic exons/CDS for the 190 miRNAs from *P. colubrinum* were also done and three of the miRNAs had 13 mRNA targets corresponding targets in *P. capsici* transcripts. These studies will help in understanding miRNAs involvement in *P. colubrinum* that have interactions with *P. capsici*.

DISEASE MANAGEMENT

Mass production of black pepper plants through somatic embryogenesis and testing the presence of mycoendophytes

Embryogenic clumps (25 mg each) from secondary embryogenic cultures were inoculated into five 250 ml flasks containing 250 ml of liquid SH-15 medium and incubated under darkness in shakers for 20 days. After 20 d of culturing, embryos of similar developmental stages were allowed to develop further in SH-30 medium with 12 h light and dark cycles for 10 more days (Fig 8). Twenty ml of medium was withdrawn at 5-days interval and replaced with the same quantity of fresh medium. After 30 days of growth in SH-30, cotyledon stage plantlets were developed and plants with one tap root were transferred to WPM for further growth and maturation. The developed plants were transferred to sterile sand for hardening. Tissues (leaf, stem, root) of somatic embryo derived plants were subjected to endophytic fungal isolation. No fungal growth was observed after 30 days of incubation.

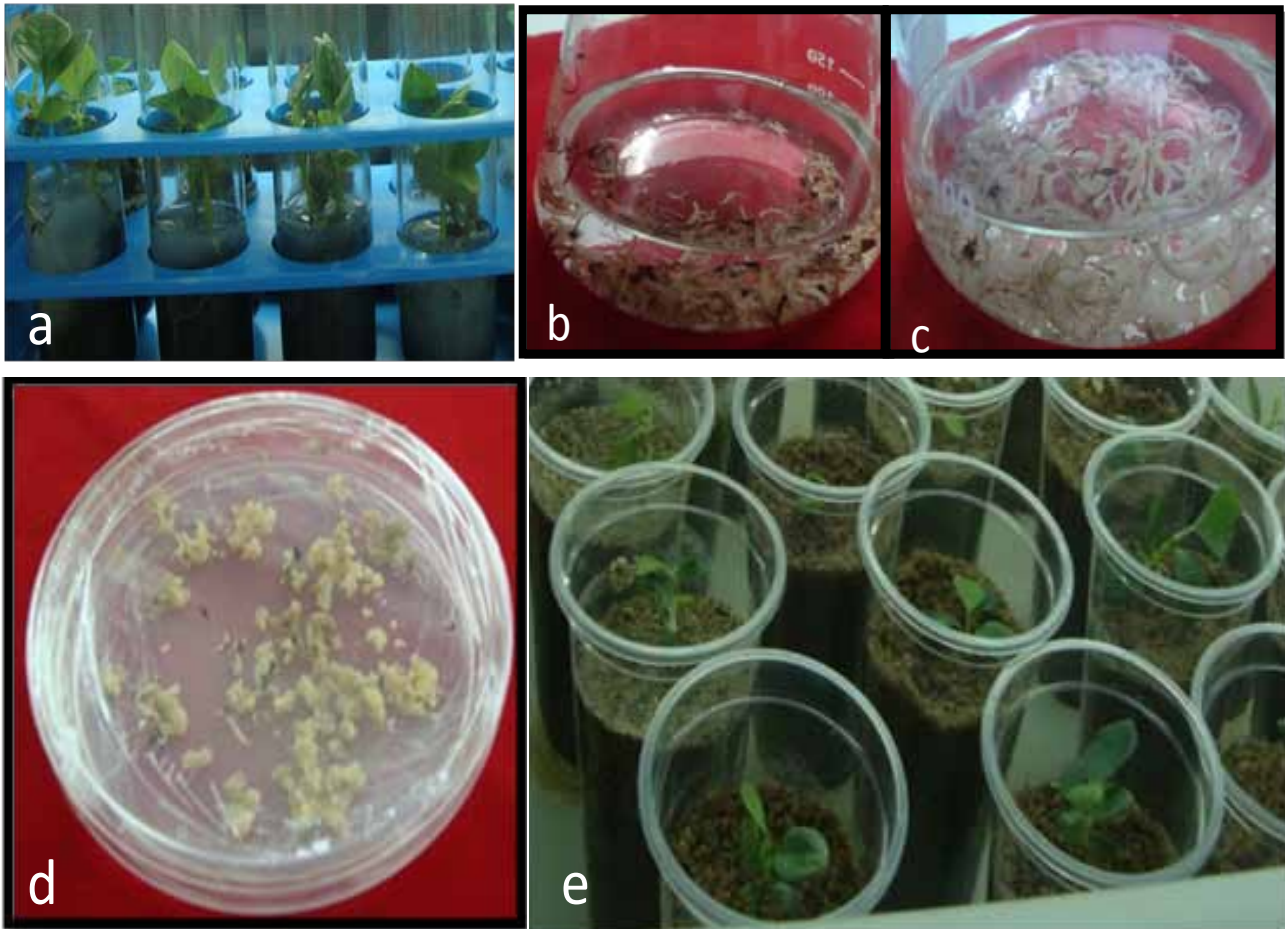


Fig 8. Different stages of plant regeneration (a) embryogenic clumps in SH-15 medium; (b) growth after 10 days of incubation in SH-30 broth; (c) growth after 20 days; (d) maturation of plants in WPM; and (e) hardening of regenerated plants.

Evaluation of promising endophytic bacteria

The trial for IDM in black pepper integrating endophytic bacteria was continued. Among the six treatments, maximum growth with number of nodes and lateral branches and less disease incidence was observed in *Trichoderma* + *Pochonia* integration followed by *Curobacterium leutium* + Metalaxyl Mancozeb and *Pseudomonas putida* + Carbosulfan integration. The establishment of the plants and maximum survival was also observed in these treatments.

Evaluation of consortia of endophytic bacteria

A pot experiment with 15 treatments consisting of different endophytes in combinations and along with recommended chemicals in an integrated mode was initiated. The results showed that consortia containing Bp25 + Bp17, Bp25 + TC10, TC10 + *P. chlamydosporia*, Metalaxyl Mancozeb + TC10 and Metalaxyl Mancozeb + Bp17 were effective in reducing root infection and was on par with the existing recommendation of *Trichoderma* and *P. chlamydosporia* (Table 5). The *Trichoderma* liquid formulation was found to be the most effective when growth parameters are concerned.

Table 5. Evaluation of consortia of endophytic bacteria

Treatment	Details	Root infection (%)	Dry shoot weight (g)	Dry root weight (g)
T1	Bp25 (<i>P. putida</i>)	66.7	60.00	1.66
T2	Bp17 (<i>B. megaterium</i>)	100.0	51.33	0.0
T3	TC10 (<i>C. luteum</i>)	50.0	56.67	3.33
T4	Bp25 + Bp17	0.0	103.00	5.33
T5	Bp25 + TC10	0.0	90.00	4.67
T6	<i>P. chlamydosporia</i>	0.0	56.67	5.0
T7	Bp25 + <i>P. chlamydosporia</i>	100.0	20.00	0.0
T8	TC10 + <i>P. chlamydosporia</i>	0.00	46.67	2.67
T9	Metalaxyl Mancozeb + <i>P. chlamydosporia</i>	66.7	96.67	6.67
T10	Met.mz + TC10	0.00	100.00	5.0
T11	Met-mz + Bp17	0.00	85.00	7.67
T12	Met-Mz + <i>T. harzianum</i> (liquid)	33.3	105.00	2.33
T13	TC 10 + <i>T. harzianum</i> liquid	66.7	55.67	1.67
T14	<i>T. harzianum</i> liquid	0.0	138.33	9.33
T15	Absolute control	66.7	48.33	3.33
	LSD 0.05		94.17	8.09

New target genes in *Radopholus similis*

Potential target genes of *R. similis* involved in parasitism such as FMRF amide-like peptides (nematode FLPs), β -1, 4, endoglucanase, trans-thyretin-like protein-3-precursor, serine-threonine phosphatases and survival such as glutathione-S-transferase(s), acetylcholinesterase, tetratricopeptide TPR-1, superoxide-dismutase and actin were amplified and sequenced. These were submitted to NCBI database (KP027004, KP027005, KM670015 to KM670018).

Diagnostics

In order to develop a real-time PCR based protocol for detection of *R. similis*, a standard graph was prepared by using four different dilutions of DNA isolated from *R. similis* pure culture. DNA sequences from the internal transcribed spacer (ITS) region of this nematode were used to design primers for real-time PCR. SYBR green reliably quantified as

little as 100 pg of *Radopholus* nematode DNA, and could be used to quantify as few as five *Radopholus* nematodes. The *Radopholus* specific primer pair RAD F (GTCCTTTGGTGGGCAGTG) and RAD R (GGTCTGCGCTCATCAAGTC) did not detect other nematodes like *Meloidogyne incognita*.

Nematicidal activity of new generation molecules

Nematicidal activity of carbosulfan and fipronil against *R. similis* was evaluated under field condition at Peruvannamuzhi. Two concentrations i.e. 0.1% and 0.2% for carbosulfan and 15 and 25g plant⁻¹ for fipronil were applied at the base of plant at quarterly (January, April, July and October), twice (pre monsoon: April-May and post monsoon: September-October) and once (September-October).

Results showed that both concentrations of carbosulfan were promising causing 100 per cent mortality of nematodes when treated quarterly and

twice, whereas when the treatment was imposed once in September-October, the nematode mortality was 82 and 93%, respectively. In case of fipronil, higher concentration showed comparatively higher efficacy (57-79% mortality) when compared to lower concentration (35-63% mortality).

Evaluation of actinomycetes against nematodes

Nematicidal activity of actinomycetes was evaluated *in planta* and found that combined application of IISR Act 2 (*Ketosatospora setae*) with IISR Act 5 (*Streptomyces sp.*) and IISR Act 2 with IISR Act 9 (*S. tauricus*) were effective in reducing the nematode population in the soil to an extent of 58-75%. In order to get maximum reduction in nematode population, actinomycetes have to be incorporated into the soil at the time of planting before the buildup of nematode population.

Field demonstration of promising technologies

Demonstration trial was taken at IISR Farm Chelavoor to demonstrate new plant protection technologies with three released varieties *viz.*, IISR Thevam, IISR Shakthi and Pournami. The technologies imposed were metalaxyl-mancozeb 0.125% with carbosulfan 0.1% and *T. harzianum* with *P. chlamydosporia*. The plants are well established.

VIRAL DISEASES

Rapid identification of transgenic black pepper using LAMP and real-time LAMP assays

A loop-mediated isothermal amplification (LAMP) and real-time LAMP based assays were developed for quick and sensitive detection of transgenic black pepper plants. Primers (six each) were designed based on the nucleotide sequence of two target regions [kanamycin and *Cauliflower mosaic virus* (CaMV) 35 S promoter] integrated into the genome of transgenic black pepper. The following conditions: 6 mM of magnesium sulphate, 0.4 M of betaine and

1 h of reaction time proved optimal for amplification of the LAMP assay. Both assays successfully detected the transgenic plants whereas no cross-reaction was recorded with non-transgenic plants. The sensitivity of LAMP was up to 104 times that of conventional PCR while real-time LAMP was up to 103 times that of LAMP. The assays were validated by testing putative transformants of black pepper. The results clearly showed that LAMP and real-time LAMP assays developed in this study can provide a rapid and simple approach for screening transgenic black pepper and other plants transformed by using the above target gene sequences.

Sequencing of RNA2 and RNA3 of Cucumber mosaic virus

Cucumber mosaic virus (CMV) is a tripartite ssRNA virus infecting large number of crops including black pepper. *RNA1* of CMV codes for viral replicase while *RNA2* and *RNA3* each codes for two proteins namely RNA polymerase (2a), silencing suppressor (2b), movement protein (3a) and coat protein (3b). Cloning and sequencing of 2a, 2b, 3a and 3b gene of black pepper isolate of CMV showed that it consists of 2573, 337, 840 and 657 nucleotides respectively potentially encoding proteins with 857, 111, 279 and 218 amino acids respectively. Sequence comparison showed that black pepper isolate of CMV shared 92–95% and 70–71% identity in 2a with CMV subgroup I and II respectively, while it was 82–95% and 65% in 2b; 91–97% and 79% in 3a and 91–99% and 76–77% in 3b. In the phylogeny, all the four genes (2a, 2b, 3a and 3b) showed close clustering with CMV subgroup I strains and distant relationship with subgroup II strains. Among the four genes, 3b showed high level of sequence conservation while 2b showed the least with other members in the subgroup.

Screening against Piper yellow mottle virus

Out of 2554 germplasm accessions screened for resistance against *Piper yellow mottle virus* (PYMoV), four accessions showed resistance in the preliminary test.

Profile of expressed proteins during symptom development in PYMoV infected plants

Previous results have showed that temperature has direct influence on symptom expression in PYMoV infected black pepper plants. Symptomless PYMoV infected plants upon exposure to temperature showed increased virus copy number and symptom upon exposure to high temperature (35°C). Low and high viral copy number plants upon exposure to temperature stress showed similar levels of chlorophyll, proteins, reducing sugars and phenols except for the reducing sugars that were marginally high in low viral copy plants. Total proteins isolated from these plants were analyzed by 2D electrophoresis, and the image was analyzed with Image Master platinum 6.0 software. At the saliency level 1000, the number of spots identified by Image master was 104. Out of that 34 spots were differentially expressed between Low and high copy number plants after symptom expression. Eight and six unique spots were also identified in low and high virus copy plants. Out of 34 differentially expressed proteins 10 major up regulated proteins in low copy number plants were identified as plastocyanin, rubisco large chain, superoxide dismutase, Rubisco activase (AAA Super family protein), membrane kinase, actin family protein, NBD sugar kinase, heat shock protein 60-2, chloroplast rubisco activase and heat shock protein 60 family. The down regulation of these proteins in high copy number suggests their role in virus replication and increase in copy number. Five proteins showed strong up regulation in high copy number plants while they were down regulated in low copy plants. The sequencing of these proteins and other unique proteins would give the insight into the mechanism behind the viral replication and symptom expression

Sequential events in colonization and proliferation of *Colletotrichum gloeosporioides*

Sequential events involved in the infection process of *C. gloeosporioides* in black pepper were studied under laboratory conditions. The leaf samples collected at 4, 6, 8, 12, 16, 20, 24, 48, 72, and 96 h after inoculation were subjected to staining,

destaining and microscopically examined. Conidial germination (Fig 9a) was observed 4 h after inoculation. The germinating conidia were found congregating more towards stomatal region and 75 % of conidia germinated either with one (most cases) or two germ tubes after 10-12h. Higher percentage of germination was noticed, when the conidia were in disaggregated condition, which later produced melanized appressoria (Fig 9b). The infection hyphae originating from appressoria entered through stomata and subsequent intra/intercellular invasion was observed. Invading hyphae in the mesophyll cells and localized tissue death (Fig 9c) were noticed after 48h. Acervulus initials were formed and mature acervuli with prominent setae (Fig 9d) were observed after 48 and 72h, respectively. Several localized necrotic spots manifested on leaf surface after 72h and the invaded epidermal cells turned brown, resulting in rapid collapse and death 72h after inoculation.

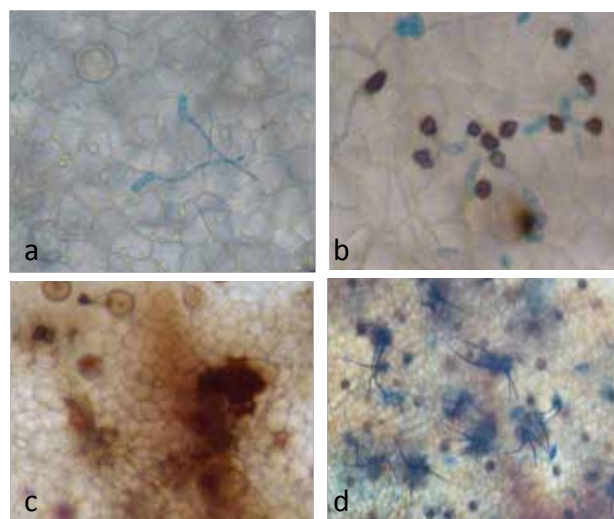


Fig 9. Sequential events in colonization and proliferation of *C. gloeosporioides* a. germinating conidia b. melanized appressoria formation c. localized tissue death d. acervuli with setae

Artificial induction of perfect stage of *C. gloeosporioides* infecting black pepper

The perfect stage (perithecia) was artificially induced under *in vitro* conditions based on mating-test model, in which sterilized toothpicks, dried leaves and twigs of black pepper as well as split, unsplit twigs of silky oak placed between confronting inoculum sources (pathogen culture, infected young and dried leaves of black pepper) served as inert platforms for the induction of perithecia.

Under *in vitro* conditions, production of perithecia was observed in all the combinations. While, formation of ascospores (indication of fertile perithecia) was observed only in the combination of dried black pepper twig + infected young and dried leaves. Exudate (Fig 10) embedded with ascospores produced from fertile perithecia was observed in the combination; black pepper twig + infected young leaf even three months after inoculation indicating, longevity and fertile nature of the perithecia. The twigs with exudate, partly or wholly, when tested for infectivity on black pepper variety, Panniyur-1, under lab and field conditions resulted in the development of characteristic anthracnose symptoms 4-6 days after inoculation.



Fig 10. Exudate embedded with ascospores produced from fertile perithecia

Genome mining of endophytic bacteria for natural products

In vitro assay

Volatile compounds from BP25R and BP17R were checked for antagonistic activity against pathogens such *P. capsici*, *Pythium myriotylum*, *Rhizoctonia solani*, *Gibberella moniliformis*, *Athelia rolfsii*, and *C. gloeosporioides* on tryptone soya agar (TSA) by

sealed plate assay. For this, the fungi were grown on PDA and the mycelial discs were exposed to bacterial volatiles and observed for inhibition of mycelial growth, if any. Significant inhibition was observed for both the bacteria. BP25R was found to be more effective compared to BP17R. Inhibitory activity was also confirmed on a synthetic medium, M9 minimal medium (Fig 11).

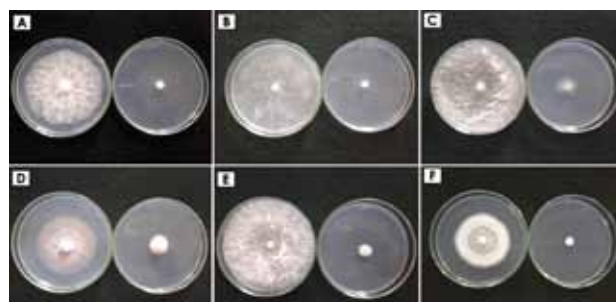
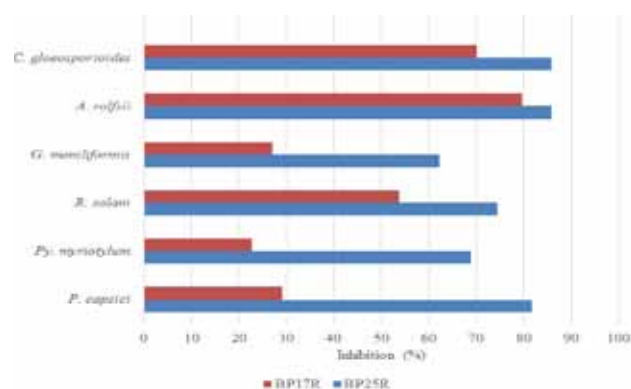


Fig 11. Activity of bacterial volatiles of *Bacillus megaterium* BP17 and *Pseudomonas putida* BP25 against different plant pathogens Top: Percentage inhibition by bacterial volatiles; Bottom: Antagonistic activity of BP25R volatiles on different pathogens A: *P.capsici*, B: *P.myriotylum*, C: *R.solani*, D: *G.moniliformis*, E: *A.rolfsii* and F: *C.gloeosporioides* (Left - exposed; Right - control in each set).

In planta assay

In planta assays were done for BP25R and BP17R volatiles against *P. capsici*, *A. rolfsii* and *P. myriotylum* by detached leaf assay. Detached leaves inoculated with respective mycelial discs were exposed to bacterial volatiles and incubated in a moisture chamber. Total inhibition of *A. rolfsii* on black pepper leaves and *P. myriotylum* on ginger and turmeric leaves was observed (Fig 12). Percentage of inhibition was found to be 54 and 29 against *P. capsici* for BP25R and BP17R, respectively.



Fig 12a. Activity screening of bacterial volatiles against *Athelia rolfsii* on black pepper leaves (From L to R) BP25 R, BP17R, pathogen alone and uninoculated leaf.



Fig 12b. Activity screening of bacterial volatiles against *Pythium myriotylum* on turmeric leaves (From L to R) BP25 R, BP17R, pathogen alone and uninoculated leaf.

Identification of bacterial volatiles using GC-MS

BP 25 R and BP 17 R volatiles from tryptone soya broth were extracted using a specially fabricated extraction apparatus at CPCRI. Identification of the bacterial volatiles was done using GC-MS (Agilent technologies 7890 A) which revealed the presence

of many potential inhibitory compounds such as 2,5-dimethyl pyrazine; Isoamyl alcohol; 2-ethyl pyrazine; 2-methyl pyrazine; 2-ethyl-3-methyl pyrazine; 1,8-Nonadien-3-ol; dimethyl disulphide, dimethyl trisulfide etc. (Table 6).

Table 6. GC-MS identification of microbial volatile organic compounds (MVOCs) from *P. putida* BP 25R and *B. megaterium* BP 17R

Name of the compound	BP 25 R	BP 17 R
Pyrazine, 2,5-dimethyl-	39.526*	16.932*
Isoamyl alcohol	20.984	12.382
Pyrazine, ethyl-	-	9.206
Pyrazine, methyl-	10.506	8.069
1-Undecene	7.411	-
Disulphide,dimethyl	6.395	-
Pyrazine, 2-ethyl-3-methyl-	-	4.927
Pyrazine, 2-ethyl-5-methyl-	3.049	-
Dimethyl trisulfide	1.449	-
Heptamethyl-2-nonene	1.199	2.545
β -Naphthol	1.164	1.922
Octadecyl vinyl ether	1.071	-
Tetradecane, 2,6,10-trimethyl	0.937	1.093
Cyclobutene, 2-propenylidene-	0.794	-
Heptamethyl-1-nonene	0.771	4.394
Dodecane	-	4.192
Tridecane	-	3.500
Tetradecane	-	2.641
Pentadec-7-ene, 7-bromomethyl-	-	1.953
n-Nonadecanol-1	-	1.220
1,8-Nonadien-3-ol	0.411	-
Octadecanal,2-bromo	0.385	-
17.2-Undecanethiol, 2-methyl-	-	1.140
Ethylhexanol	-	1.237
2-Ethyl-3,6-dimethyl pyrazine	0.231	-
Sulfurous acid, butyl cyclohexylmethyl ester	-	5.076

* Peak area %

Activity screening of synthetic volatile compounds

In vitro assay

Volatile compounds such as 2,5-dimethyl pyrazine, 2-ethyl-3-methyl pyrazine, 2-methyl pyrazine, 2-ethyl pyrazine, 2-ethyl-5-methyl pyrazine, 2-ethyl 3,6-dimethyl pyrazine, and dimethyl trisulfide were screened for activity against *P. capsici*, *P. myriotylum*, *R. solani*, *G. moniliformis*, *A. rolfsii*, *C. gloeosporioides* and *R. similis*. For fungal bioassay, PDA was inoculated with mycelial disc

and sealed against a lidless plate containing filter paper impregnated with volatile compound. The amount taken was adjusted so as to get different concentrations ranging from 21 to 679 $\mu\text{g cm}^{-3}$. After incubation, mycelial diameter was measured and percentage of inhibition was calculated. For *R. similis*, water agar was used instead of PDA and mortality was checked by counting the number of live/dead nematodes at various intervals. Significant inhibition of all the fungal pathogens was observed with all the tested compounds (Table 7). The study has shown the multifaceted interaction of MVOCs

Table 7. Broad spectrum antimicrobial and nematocidal activities of microbial volatile organic compounds produced by *Pseudomonas putida*.

Name of Compound	<i>P. capsici</i>	<i>A. rolfsii</i>	<i>C. gleosporioides</i>	<i>G. moniliformis</i>	<i>P. myriophyllum</i>	<i>R. solani</i>	<i>R. similis</i>
2, 5-dimethyl pyrazine	60.53*	74.00	60.00	52.60	93.30	85.00	26.50**
2-ethyl 3-methyl pyrazine	100.00	80.50	82.86	81.57	95.50	95.80	31.00
2-methyl pyrazine	37.50	53.47	60.00	52.60	41.85	80.00	05.30
2-ethyl pyrazine	43.75	100.00	57.13	68.42	96.25	100.00	09.00
2-ethyl 5-methyl pyrazine	53.13	100.00	85.29	84.20	100.00	100.00	29.00
2-ethyl 3,6-dimethyl pyrazine	76.32	100.00	100.00	100.00	100.00	100.00	39.00
Dimethyl trisulfide	100.00	100.00	100.00	100.00	100.00	100.00	100.00
2-ethyl hexanol	100.00	100.00	100.00	100.00	100.00	100.00	NT

* % inhibition at 169 $\mu\text{g cm}^{-3}$ ** % mortality at 339 $\mu\text{g cm}^{-3}$

with various pathogens and among the compounds, the highest nematocidal activity was found with dimethyl trisulfide.

In planta assay

Black pepper cut shoots inoculated with *P. capsici* mycelial discs were exposed to volatile compounds in a sealed box and incubated under room temperature for three days. Cut shoots exposed to compounds alone were also kept to check their phytotoxicity.

Cut shoots inoculated with *P. capsici* alone served as control. After three days, the cut shoots were checked for lesion suppression and phytotoxicity. Among the tested compounds, 2-methyl pyrazine and 2-ethyl 3-methyl pyrazine @ 42 $\mu\text{g cm}^{-3}$ showed lesion suppression with no visible signs of phytotoxicity. The minimum inhibitory concentration (MIC) of these compounds is being worked out using live plant materials.



CARDAMOM

Genetic resources

A total of 618 cardamom accessions have been maintained at National Active Germplasm Site (NAGS) which consist of 442 accessions from Appangala; 73 accessions from Pampadumpara; 47 accessions from Mudigere and 56 from Sakleshpur. Fifty seven cardamom accessions were characterized in NAGS for morphological, yield parameters; leaf blight and rhizome rot resistance under field conditions. FGB 82 recorded maximum yield and more number of capsules per plant.

The accessions were grouped based on the per cent disease index for leaf blight and rhizome rot. The accessions *viz.*, FGB 67, FGB 87 and FGB 113 were classified as resistant to leaf blight disease whereas the accession FGB 118 was classified as highly resistant to rhizome rot. Physiological parameters *viz.*, specific leaf weight, relative water content and per cent of leaf folding were recorded in 57 accessions. Significant variation was recorded for relative water content and specific leaf weight. Two cardamom lines (Mysore type) from Mankulam forest range in Kerala were collected and added to the germplasm.

Breeding

The accession, IC 547167 (Appangala 1 x NKE 19) with potential yield of 1393.12 kg ha⁻¹ (first crop on three years after planting) and mean yield of 456.79 kg ha⁻¹ over locations, with mosaic resistance and good quality characters has been recommended for release in the state of Karnataka as new cardamom variety under the name Appangala 2 by XXV AICRPS meeting held at UBKV, Pundibari on September 2014 (Fig 13).



Fig 13. New cardamom hybrid Appangala-2

PET III was laid out with 21 inter-varietal F1 hybrids for its evaluation for yield. The hybrid GG x IISR Vijetha has recorded highest plant height and number of leaves while maximum tiller production was observed in the cross Mudigere 2 x Appangala 1. PET IV was laid out with 24 selfed and seedling progenies of TTL lines and is evaluated for thrips tolerance. TTL 21 has recorded highest plant height and number of leaves while maximum tiller production was observed in TTL 16.

Standardizing the parameters for target yield

Based on the previous year's crop yield under different treatments and the nutrient uptake data, the nutrient removal for producing 100 kg of capsule was worked out and fertilizer recommendations were made for fixed target yield levels based on the soil test values for Appangala-1 and Green gold varieties. In both the varieties, the recorded yield parameters were higher in target specific applications as compared to the blanket recommendations.

In green gold, recorded yield levels per plant basis was 0.7, 0.9 and 0.9 kg plant⁻¹ for the targets 0.4, 0.6 and 0.8 kg plant⁻¹ with a positive mean deviation of 72, 55 and 15%, respectively. Similarly, in Appangala-1 yield per plant has shown a positive mean deviation of 83, 76 and 14% for the fixed target levels. The mean bias error and root mean square deviation for the prediction model and the recorded (projected) yield were also minimum, indicating better fitness of the target yield equation

Evaluation of cardamom elite lines for yield and quality under moisture stress

Twelve short listed and three checks of cardamom genotypes were planted in replicated trial with control and stress treatments. Moisture stress was imposed by withholding irrigation for two months during fourth year. Growth and yield, physiological parameters (relative water content and specific leaf weight) were recorded in control and stress treatments. Soil moisture content was recorded by

gravimetric method. Soil moisture content ranged from 12-14% under stress treatment.

Growth and yield parameters

Growth parameters like plant height (cm), total number of tillers per clump, total number of bearing tillers per clump, total number of non bearing tillers per clump and yield parameters like number of panicle per clump, panicle length (cm), number of capsules per panicles and fresh yield of capsule per clump were recorded. Growth and yield parameters were generally reduced under stress. Total number of tillers per clump ranged from 23.66 (IC 584059) to 35.66 (Appangala 1) with a mean of 29.58 in control and in stress it ranged from 17.93 (IC 584071) to 28.4 (APG 224) with a mean of 22.49 (Fig 14).

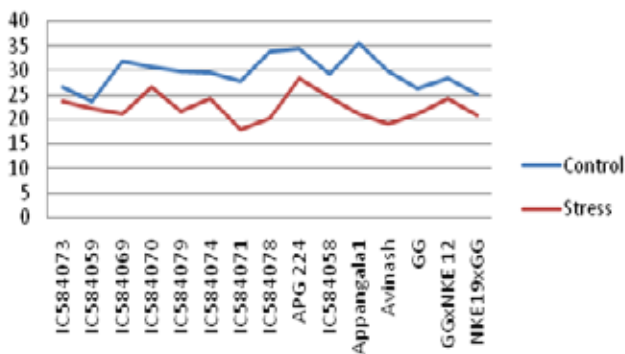


Fig 14. Tillers per clump as influenced by moisture stress

Total number of panicles ranged from 15 (Green Gold) to 28.93 (Appangala 1) with a mean of 20.14 in control and from 12 (IC 584059) to 18.26 (GG x NKE12) in stress. The panicle length (cm) ranged from 39.32 (IC 584071) to 60.24 (Appangala 1) with a mean of 49.83 in control and in stress it ranged from 23.16 (IC 584071) to 45.92 (IC 584070) with a mean of 36.28.

Dry cardamom yield (kg ha⁻¹) ranged from 98.05 (IC 584073) to 419.5 (IC 584058) with a mean of 201.26 in control and in stress it ranged from 48.91 (IC 584071) to 115.91 (IC 584070) with a mean of 82.72.

Chemoprofiling of cardamom

The cardamom collected from Myladumpara (Idukki) showed 5.8-7.4% oil content on capsule weight basis whereas those from Appangala had 4.5-6.0% oil. Pannikulangara-2 recorded the highest

essential oil content, which was followed by ICRI-2 with 7%. The essential oil constituents of cardamom from both locations had low 1,8-cineole and high α -terpinyl acetate. GC-MS analysis indicated 22-32% 1,8-cineole and 36-46% α -terpinyl acetate in Myladumpara varieties whereas 22-30% 1,8-cineole and 37-51% α -terpinyl acetate in Malabar, Mysore and Vazhukka varieties from Appangala. Pannikulangara-2 contained 29% 1,8-cineole and 44% α -terpinyl acetate.

Monitoring cardamom quality by HEN

Using the modified Hand – held electronic nose (HEN) developed in collaboration with C-DAC, Kolkata, cardamom samples could be analyzed for essential oil content and graded into low (< 4.0%), medium (4.0-6.0%) and high (>6%) based on oil yield. The results indicated good correlation with that of chemical analysis. The instrument will be further validated at traders' level.

Differential reaction of *C. gloeosporioides* isolates on cardamom varieties

Differential reaction of 20 *C. gloeosporioides* isolates from cardamom was studied on cardamom varieties viz., Appangala 1 (Fig 15), IISR Avinash and IISR Vijetha by employing prominence of yellow halo and streak as well as lesion area as criteria for recording observation. The isolates exhibited differential reaction as indicated by prominence and non-prominence of yellow halo and streak. The area of lesions developed on young leaves varied between 4.91 – 40.82, 7.85 – 60.45 and 11.78 – 38.47 mm² in IISR Avinash, IISR Vijetha and Appangala 1, respectively.

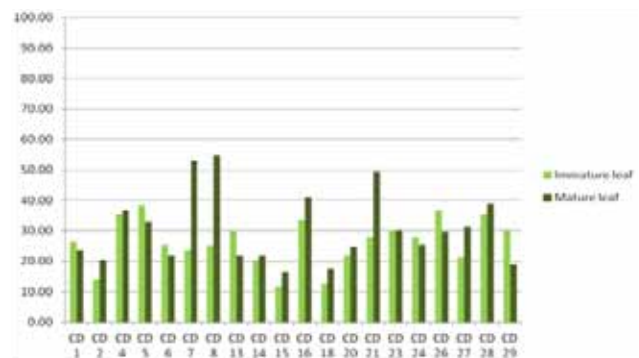


Fig 15. Lesion area – Appangala 1 x *C. gloeosporioides* (cardamom isolates)

Occurrence of perfect stage of *C. gloeosporioides* infecting cardamom

Surveys carried out in cardamom plantations revealed manifestation of different types of foliar symptoms viz., spot, blight and shredding. The cultures isolated from these symptomatic samples exhibited variations in colony morphology and

was found to be the most effective isolate against *Pythium vexans* and *Fusarium oxysporum* whereas KA-20 was effective against *Rhizoctonia solani*. The plants inoculated with *P. vexans*, *R. solani* and *F. oxysporum* showed 94.0, 81.5 and 85.6% increase in biomass respectively in treatments with most efficient *Trichoderma* isolates KA-3 and KA-20.

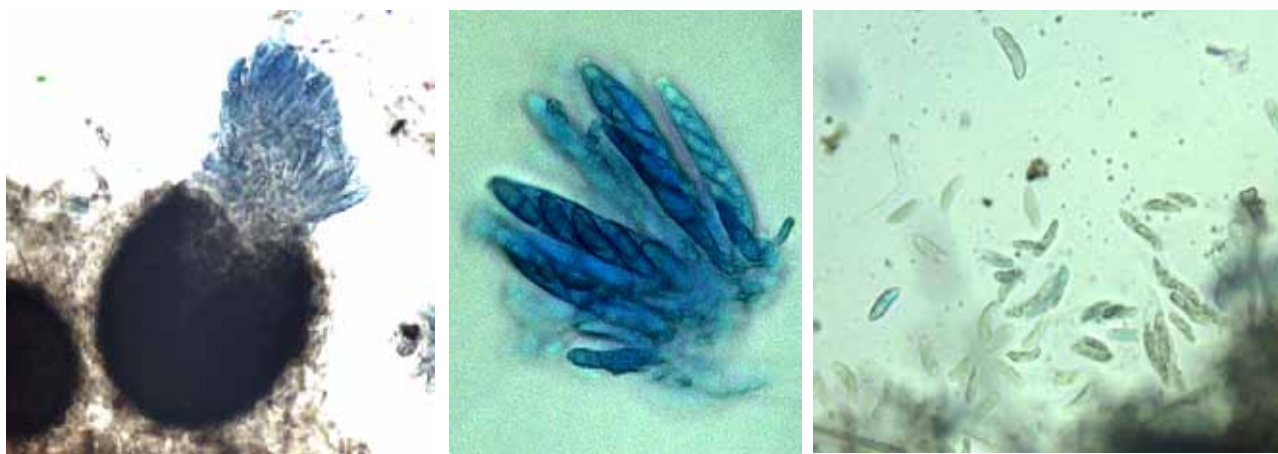


Fig 16. Perfect stage of *C. gloeosporioides* infecting cardamom a. globose perithecia b. Unitunicate asci c. ascospores

colour. Among the cultures, greyish white culture appeared puffy with a faster growth rate (14 mm/day) and produced dark brown-black, globose perithecia, 4 weeks after incubation. Microscopic examination of perithecia (Fig 16a) revealed the presence of narrow, cylindrical, unitunicate asci (Fig 16b) with hyaline, aseptate, cylindrical ascospores (Fig 16c).

Molecular characterization of pathogens and antagonists

The shortlisted efficacious *Trichoderma* isolates KA-1, KA-3, KA-20 (Karnataka), KL-3, KL-10, KL-13, KL-17, KL-19 (Kerala) and TN-3 (Tamil Nadu) were characterized based on ITS r-DNA sequencing.

Evaluation of potential antagonists

The shortlisted nine efficacious *Trichoderma* isolates viz., TN-3, KA-1, KA-20, KA-3, KL-3, KL-10, KL-19 and KL-17 were evaluated against *Pythium vexans*, *Rhizoctonia solani* and *Fusarium oxysporum* under green house conditions. The isolate KA-3

Evaluation of chemicals

The suckers of cardamom (Appangala 1) were established in pots to evaluate shortlisted fungicides, fenamidone + mancozeb (0.2%), captan + hexaconazole (0.2%) and tebuconazole (0.05%) against *P. vexans*, *R. solani* and *F. oxysporum*. Among the three fungicides tested, tebuconazole (0.05%) was effective against *R. solani* and *F. oxysporum*, whereas the tested fungicides were not effective against *P. vexans*. The average disease incidence in *R. solani* inoculated pots treated with tebuconazole was 11.6% whereas in pots inoculated with pathogen alone showed 94.0%. The average disease incidence in *F. oxysporum* inoculated pots treated with tebuconazole was 10.5%.

Natural incidence of rhizome rot

The natural incidence of rhizome rot disease was recorded in 57 accessions maintained at field gene bank (FGB). The accessions were grouped into various categories based on the reaction towards rhizome rot (Table 8).

Table 8. Reaction of field gene bank accessions against rhizome rot

Disease Index (%)	Category	Accessions
0.0 – 5.0	Highly resistant (HR)	FGB 118
5.1 – 10.0	Resistant (R)	FGB 107, FGB 117, FGB 119
10.1 – 25.0	Moderately susceptible (MS)	FGB 70, FGB 75, FGB 76, FGB 78, FGB 81, FGB 82, FGB 83, FGB 84, FGB 87, FGB 97, FGB 105, FGB 106, FGB 108, FGB 110, FGB 112, FGB 113, FGB 114, FGB 115, FGB 116
25.1 – 50.0	Susceptible (S)	FGB 61, FGB 62, FGB 63, FGB 64, FGB 65, FGB 66, FGB 67, FGB 68, FGB 69, FGB 71, FGB 72, FGB 73, FGB 77, FGB 79, FGB 80, FGB 85, FGB 86, FGB 88, FGB 89, FGB 90, FGB 91, FGB 92, FGB 93, FGB 94, FGB 95, FGB 98, FGB 99, FGB 100, FGB 101, FGB 102, FGB 104, FGB 109, FGB 111, FGB 120
> 50 %	Highly susceptible (HS)	Nil

Evaluation of microbes against rhizome and root rot pathogens

The endophytic fungal isolates from cardamom varieties viz., Appangala 1, IISR Avinash and IISR Vijetha were evaluated for antagonistic efficacy against *F. oxysporum*, *R. solani* and *Pythium vexans* under *in vitro* conditions. Among the isolates tested, Va 4-2 (IISR Vijetha), Cb 4-1, Cb 6-2 (Appangala I) and Aa 1-1 (IISR Avinash) were found promising against *F. oxysporum*. While, Cb 4-1, Cb 6-2 (Appangala 1) and Ab 6 (IISR Avinash) were effective against *P. vexans* and Cb 2 (Appangala 1) was inhibitory to *R. solani*.

Evaluation against leaf blight and rhizome - root rot pathogens

Endophytic fungi and bacterial isolates from *Alpinia mutica*, *Alpinia galanga* and *Amomum microstephanum* were evaluated for antagonistic efficacy against *R. solani*, *F. oxysporum* and *C. gloeosporioides* under *in vitro* conditions. Among the isolates, AmL 1C (*A. mutica*), AgR 5A, AgR 5D (*A. galanga*) and AmiPs 4C (*A. microstephanum*) were found promising against *R. solani*. AmL 1B (*A. mutica*), AgR 5D (*A. galanga*) and AmiPs 4A (*A. microstephanum*) were effective against *F. oxysporum*. Whereas, AmL 1B (*A. mutica*) and AmiPs 4A (*A. microstephanum*) were found inhibitory to *C. gloeosporioides*.

Standardization of spray schedule for *Sciothrips cardamomi*

Four promising insecticides (fipronil, imidacloprid, quinalphos and spinosad) were evaluated in the field at Appangala for standardization of spray schedule for the control of cardamom thrips. The spray schedule included three sprays during March, May and August along with standard spray schedule (five sprays during March, April, May, August and September). The percentage of capsules damaged by thrips was recorded in each harvest. The trial indicated that the percentage of capsules damaged by thrips in the standard spray (five sprays) schedule was lowest in spinosad (4.3%) that was on par with all other treatments except control. The percentage of capsules damaged by thrips in the discontinuous spray (three sprays) schedule was lowest in fipronil (5.4%) that was on par with all other treatments except control.

Documentation of natural enemies

Surveys were conducted in Idukki District for documentation of natural enemies of cardamom thrips. Cadavers of dead thrips, on laboratory culturing, showed the presence of a fungus, identified as *Isaria* sp. (IISR-EPF-05). Molecular confirmation of the identity of the fungus is in progress.

Field evaluation of entomopathogenic fungus

Field trials with the promising entomopathogenic fungus *Lecanicillium psalliotae* for the control of cardamom thrips was conducted at Kodagu, Wayanad and Idukki. The trial in Wayanad was undertaken in a partnership mode with M/s A V Thomas & Company, Meppadi. The trial indicated that combined application of *L. psalliotae* as spray and basal application gave better control than other treatments at Wayanad.

Studies on *Wolbachia* endosymbiont

The effect of tetracycline treatment in removal of *Wolbachia* endosymbiont from cardamom thrips was studied by treating the insects with tetracycline treated cardamom leaf bits and also by feeding with a mixture of tetracycline and sucrose. DNA was extracted from adult thrips of first generation and screened for *Wolbachia* using *wsp* primers. The primers failed to amplify *wsp* gene indicating that *Wolbachia* was removed from the test insects; the control insects showed the presence of the bacterium. The above method was found to be suitable for removal of *Wolbachia* from thrips

Shoot Borer Molecular characterization

PCR conditions for molecular characterization of *Conogethes punctiferalis* populations from cardamom using mitochondrial COI gene region primers (Lep F1 & Lep R1) were standardized.

Evaluation of EPNs

The infectivity of four promising EPNs, *Heterorhabditids* sp. (IISR-EPN 01), *Steinernema* sp. (IISR-EPN 02), *O. gingeri* (IISR-EPN 07) and *Oscheius* sp. (IISR-EPN 08) is being tested against shoot borer infesting under field conditions at Appangala

Infectivity of EPNs against root grub

The infectivity of EPNs against cardamom root grub, *Basilepta fulvicorne* was tested *in vitro*. Among the test EPNs, *Heterorhabditis* sp. (IISR-EPN 01) and *O. gingeri* (IISR-EPN 07) were more pathogenic to root grub as they brought about 100 % mortality to the insect within 72 h post exposure, followed by *Steinernema* sp. (IISR-EPN 03), *S. carpocapsae* (IISR-EPN 06) and *Oscheius* sp. (IISR-EPN 08). *Steinernema* sp. (IISR-EPN 02) and *Oscheius* sp. (IISR-EPN 04 and 05) took 120 h, respectively, to kill the test insect.



GINGER

Genetic resources

Six hundred and sixty eight ginger accessions have been maintained in the field gene bank. Germplasm conservatory was enriched with 17 ginger accessions (extra bold local ginger from West Bengal, black and pink ginger from Meghalaya and bold ginger from Kerala, Fig 17).



Fig 17. a. Bold ginger collection from Kerala; b. Bold ginger collection from West Bengal; c. Pink ginger from Meghalaya; d. Bold ginger from Meghalaya; e. *Kaempferia* sp. (black ginger?); f. black ginger, *Zingiber* sp.

Yield evaluation of promising extra bold accessions

To evaluate the yield performance of extra bold germplasm selections, an experiment was laid out

in a randomized complete block design with three replications. Among the 13 accessions studied, highest yield was recorded in Acc. 247 followed by Acc. 713 and Acc. 723 (Table 9).

Table 9. Evaluation of extra bold ginger accessions for yield

Genotypes	Yield bed ⁻¹ (3m ²)		Mean	Projected yield (t ha ⁻¹)
	2013/14	2014/15		
727	4.35	5.15	4.75	11.88
714	4.81	8.65	6.73	16.83
287	4.75	7.47	6.11	15.28
397	5.00	6.01	5.51	13.76
249	3.91	5.65	4.78	11.95
689	3.88	9.04	6.46	16.15
713	9.98	9.69	9.84	24.59
247	10.09	10.92	10.51	26.26
723	11.12	8.03	9.58	23.94
726	6.72	7.00	6.86	17.15
702	6.52	7.79	7.16	17.89
821	3.21	6.12	4.67	11.66
701	7.28	8.22	7.75	19.38
IISR Varada	9.11	8.89	9.00	22.50
Mean	6.48	7.76		16.20
CV (%)	11.86	14.22		
CD (0.01)	1.69	1.88		

Low fibre

The promising low fibre accessions of ginger were evaluated during 2014/15 for yield and other growth characters. The experiments were laid out in randomized complete block design (RBD) with three replications. Among the seven low fibre ginger accessions along with check IISR Varada studied, mean yield per bed (kg) ranged from 4.15 to 8.17. Maximum yield was recorded in Acc. 278 followed by Acc. 82, which was statistically on par.

Mutation induction

Four genotypes (IISR Varada, IISR Mahima, Acc. 182 and Acc. 247) were subjected to gamma irradiation (900 buds each) at different doses of 0.80, 1.00 and 1.20 kR at Mangalore University, Mangaluru, Karnataka. Differential response of the varieties was observed for germination. The M1V1 mutants were established in the green house for screening against *Pythium* sp.

Three putative mutants each against *Ralstonia solanacearum* (HP 0.5/2, HP 0.5/15 and M 0.5/1) and *Pythium* sp. (V 0.5/2, R 0.8/1 and R 1.25/4) were multiplied for further screening. *In vitro* cultures were initiated using buds of released varieties namely Mahima, Rejatha, Varada, Suprabha and natural polyploid Acc. 821. Callus was successfully induced from *in vitro* established buds of Varada and Acc. 821. These are being multiplied further *in vitro*.

Results on partitioning percentage at 180 days after planting revealed that all treatments had similar partitioning percentage. However, maximum partitioning to rhizomes (55 %) was noticed in 100 % solid fertilizers at monthly interval. Per plant yield varied from 160 to 240 g at harvest. The 100 % solid fertilizer treatment showed highest yield (240 g plant⁻¹) followed by 100 RDF through fertigation (220 g plant⁻¹). Fertigation once in two or four days showed similar results (Table 10).

Influence of coloured shade nets on ginger

The objective of this experiment was to study the influence of coloured shade nets on growth, partitioning percentage, yield and quality in ginger and turmeric.

Coloured shade nets viz. red, green, black and white were used for the study with open as control. Light intensity under different shade nets was around 60 %

Table 10. Partitioning percentage of fertigated ginger at 180 days after planting

Treatment	Partitioning Per cent				Fresh weight (g)				
	Stem	Rhizome	Leaf	Root	Stem	Rhizome	Leaf	Root	Total
75% RDF*	33bc	53ab	10a	4a	95ab	150ab	27a	12b	284b
100% RDF	31a	55b	10a	4a	100b	170b	30a	14c	314c
125% RDF	32ab	52a	9a	6b	85a	140a	25a	16d	266a
100% RDF Solid fertilizer, monthly	33bc	52a	11ab	4a	125c	203c	44b	15cd	387d
75% RDF + PGPR	34c	51a	12b	3a	105b	160 b	30a	10a	305c
Mean	32.6	52.6	10.4	4.2	102	165	31.2	13.4	311.2

* Recommended dose of fertilizers

Values followed by different alphabets indicate significant difference

Standardization of fertigation schedule

The objective was to standardize fertigation schedule under soil-less production of ginger. The medium used was coir pith + FYM (1:1). Five dosages of NPK fertilizers were included as treatments. Other nutrients (applied uniformly for all treatments once in a week) were CaNO₃ (5g L⁻¹), micronutrient mixture Ginger Plus (5g L⁻¹) and CuSO₄ (5g L⁻¹). Two fertigation intervals (once in two days and once in four days) were tried for all the above treatments.

of light under open condition. Results showed that at 140 DAP, fresh weight and partitioning to rhizomes was more in red shade net (52 %) in ginger. Black, red and white recorded similar rhizome fresh weight at harvest & least in open. Not much variation in photosynthetic and transpiration rate was noticed among treatments. However, red and white showed slightly higher photosynthetic and lower transpiration rate (Table 11). Quality parameters viz. oil and oleoresin were slightly higher under red and black shade nets compared to other treatments.

Table 11. Partitioning percentage under different coloured shade nets at 140 DAP in ginger

Treatment	Rhizome	Stem	Leaf	Root	Rhizome FW at harvest (g plant ⁻¹)
Red	52c (300)*	27a	17a	3a	451c
Black	45a (240)*	31b	20b	4a	414b
Green	48b (255)*	27a	18a	7b	406b
White	45a (235)*	30b	22c	3a	449c
Open	45a (270)*	33c	20b	2a	358a
Mean	47	29.6	19.4	3.8	415

*Fresh weight (fw) of rhizomes

Biodiversity of bacterial wilt pathogen

Five new isolates of *Ralstonia solanacearum* were collected from bacterial wilt infected fields of ginger and tomato and tested for cross infectivity of the isolates to ginger and *vice versa*. None of the isolates from tomato were found infecting ginger. But ginger isolates infected tomato and brinjal.

Development of LAMP for detection of *R. solanacearum* biovar 3

A set of six primers were designed using the software LAMP Designer 1.12 from <http://www.premierbiosoft.com>. This was validated with ginger *R. solanacearum* isolates as well as *R. solanacearum* isolates from solanaceous crops like tomato, brinjal and potato (Fig 18, Table 12). Specific amplification was obtained for only ginger *R. solanacearum* showing the high specificity of the LAMP primer. Further validation was done with soil DNA extracted from *R. solanacearum* sick soil and also with genomic DNA from the same. The same set of primers was used for detection of *R. solanacearum* of ginger using Real Time LAMP. In RT LAMP also, the primers detected only *Ralstonia* strain of ginger. Analyzing the annealing curve and Ta value (92°C) confirmed the amplification of correct product. The sensitivity of the method was found to be 1pg of pathogen DNA. To standardize the sensitivity, different quantities of genomic DNA were amplified and the tests revealed that in RT LAMP even 50pg of the DNA can be amplified. This can be used as a diagnostic kit for testing ginger *Ralstonia* in the field before planting as well as for seed testing.

Standardization of Real Time LAMP

Real Time LAMP was standardized and validated with ginger *Ralstonia* isolates along with *Ralstonia* isolates from solanaceous crops (Table 13). Further validation was done with genomic DNA and soil DNA from artificially inoculated soil and also with different quantities of genomic DNA to find out the specificity.

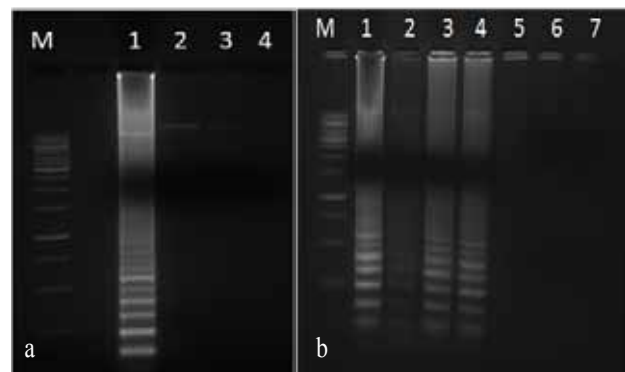


Fig 18. a. Amplification of DNA specific to *Ralstonia solanacearum* using LAMP primers. M. 1 Kb ladder, 1- GRs Sik, 2-TRs Klm, 3-PRs Pun, 4-Water control, b. Specificity of LAMP primers to amplify ginger *R. solanacearum*. M-1kb ladder, 1-GRs Sik, 2-GRS Mnt2, 3-GRs Vyt, 4-GRs Ktm, 5-ORB-3 BrinjalRs, 6-ORP-1 PotatoRs, 7-water control

Table 12. Validation of Real Time LAMP with different quantities of genomic DNA of *R. solanacearum*

Sample	Amp.time (mm:ss)	Ann.tem (°C)
GRs Sik	36:31	91.97
GRs Vyt	35:31	92.07
GRs Mnt 2	38:01	91.97
ORB-3	-	-
ORP-1	-	-
Water control	-	-

Table 13. Validation of Real Time LAMP with different quantities of genomic DNA of *R. solanacearum*

Sample	Amp.time (mm:ss)	Ann.tem (°C)
5×10^1	22:31	92.02
5×10^0	25:15	92.07
5×10^{-1}	29:15	92.07
5×10^{-2}	32:30	92.07
5×10^{-3}	34:31	92.03
5×10^{-4}		
5×10^{-5}		
5×10^{-6}		

Genetic diversity of *Curcuma amada* in response to *R. solanacearum*

Different accessions of *C. amada* were screened against *R. solanacearum* using pseudostem and soil inoculation methods. Only two accessions were found resistant under the two methods of screening.

Pathway analysis of transcriptomes of ginger and mango ginger and tissue specific expression analysis of shortlisted genes/ESTs

NBS-LRR, ABC transporters, 4-coumarate: coenzyme A ligase (4-CL), WRKY transcription factor 8 and callose synthase were studied for their expression level in ginger and mango ginger at different time intervals (0, 1, 4, 8, 16, 24, 48, 72, 96 and 120 hpi) in leaves and pseudostem. The expression of 4-CL was higher in *C. amada* than all the transcripts studied and its expression peaked up 48 hpi. Increase in the expression of PR genes such as 4CL might help in forming physical barriers (lignin) to prevent bacterial movement and proliferation.

Light and fluorescent microscopic studies

Light microscopic studies indicated no difference between the *R. solanacearum* inoculated and

uninoculated sections of the root of *C. amada*. In ginger, there appeared to be the presence of gaps in cortical regions which can lead to the assumption that they could be bacterial pockets. Under fluorescent microscope, the stele of unstained sections of *C. amada* showed thick casparian thickenings compared to *Z. officinale*. Bright field microscopic observation of the sections of ginger showed large intercellular pockets (bacterial pockets) in the root inner cortex. The cortical cells next to them showed the symptoms of degeneration. Bacteria advance from the cortex to vascular cylinder through vascular parenchyma crossing the endodermis.

Evaluation of apoplastic microbes against bacterial wilt

Around 150 bacteria were isolated from ginger apoplastic fluid. Based on *in vitro* and *in planta* evaluation against *R. solanacearum*, six isolates *viz.*, GAB5, 24, 43, 48, 107 and 148 were short-listed. The biocontrol and growth promoting traits of the short-listed apoplastic bacteria were tested *in vitro*. All the isolates showed siderophore and ammonia production. Two isolates *viz.*, GAB48 and GAB107 showed amylase, protease and cellulose activities (Table 14). Based on enzyme production and growth promoting as well as biocontrol traits, four apoplastic bacterial isolates were short-listed and evaluated under greenhouse and field conditions (Table 15). Under greenhouse condition, almost 50% reduction in disease incidence was noticed with GAB 43 in the initial stages of infection when compared to positive control and chemical treatment. In the field no bacterial wilt or soft rot incidence was observed during the period. However, dry rot incidence and scale infestation were observed during harvest. Comparison of different treatments indicated that dry rot due to *Macrophomina phaseolina* and scale infestation were comparatively lesser in GAB 48 and GAB 107 applied plots.



Table 14. Growth promoting traits of apoplactic bacteria isolated from ginger

Isolate name	Siderophore production	HCN	NH ₃ production	Phosphate solubilization	IAA
IISR GAB 5	+	-	+	+	-
IISR GAB 24	+	-	+	+	+
IISR GAB 43	+	-	+	-	+
IISR GAB 48	+	-	+	-	+
IISR GAB 107	+	-	+	+	-
IISR GAB 146	+	-	+	+	-

Table 15. Enzyme activities of apoplactic bacteria isolated from ginger

Isolate name	Amylase	Protease	Cellulase	Lipase
IISR GAB 5	+	+	+	-
IISR GAB 24	+	-	+	-
IISR GAB 43	-	-	-	-
IISR GAB 48	+	+	+	-
IISR GAB 107	+	+	+	-
IISR GAB 146	+	-	+	-

Whole genome sequencing of *R. solanacearum*

Two strains of *R. solanacearum* (GRs-SIK and GRs-MEP) were Illumina sequenced and the raw data has been assembled using A5-miseq. Both the strains have been annotated using Prokka (a software tool for the rapid annotation of prokaryotic genomes). In GRs-MEP there are 5120 CDS, 80 tRNA, and 1 tmRNA while GRs-SIK possesses 5080 CDS, 63 tRNA and 1 tmRNA. To better classify the predicted proteins from Prokka, a refined annotation has been done using Blast2GO with $1.0E^{-3}$ as e-value cut off and 33 as HSP cut off length. The genomes were mined for various effector proteins and other virulence factors. Gene ontology (GO) distribution from the predicted CDS of GRs-SIK and GRs-MEP were classified into three major components viz. biological process (BP), molecular components (MF) and cellular components (CC).

Sequence assembly and annotation

Sequence assembly of two strains of *R. solanacearum* (GRs-MEP and GRs-SIK) was performed using

A5 pipeline and yielded 286 and 213 scaffolds respectively. For both, GRs-MEP and GRs-SIK genes were predicted using Prokka and revealed 5,201 and 5114 genes with 5120 genes were predicted as CDS in GRs-MEP and 5080 in GRs-SIK. The annotation using BLAST2GO for GRs-MEP resulted in 5039 annotated sequences and 81 were without any hits, whereas in GRs-SIK, 4891 CDS could be annotated and 73 CDS were without any blast hits against NR. 4CL might help in forming physical barriers (lignin) to prevent bacterial movement and proliferation.

Proteome comparison

Proteomes of both strains were compared against 10 other available strains of *R. solanacearum* using OrthoMCL. In total, 6510 orthologues protein clusters have been identified in the 12 strains of *R. solanacearum*. Singletons were also predicted for each strain, GRs-SIK had 20 singletons, whereas GRs-MEP had 59 singletons.

Prediction of pathogenic proteins

Collection of already characterized virulent genes

from different strains of *R. solanacearum* was done and 41 virulence genes were found. Sequence similarity between these genes and CDS from GRs-MEP and GRs-SIK were analyzed. The predicted CDS of both the strains were further scanned for the presence of pathogenic protein using MP3 and 316 CDS each were predicted in both (Fig 19).

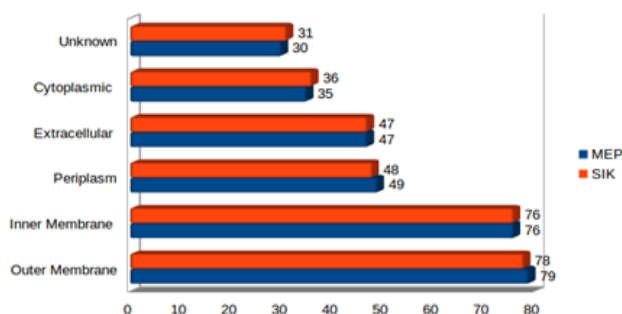


Fig 19. Localization of predicted pathogenic proteins in two strains of *R. solanacearum* strains, GRs- MEP and GRs-SIK.

Reference based alignment and SNP calling

Reference based alignment with GMI1000 using BWA has shown that 83% of reads properly paired with the reference genome. SNP calling using GATK and Samtools indicated 4368 SNPs in GRs-MEP and in GRs-SIK 4648 SNPs were reported.

Type 3 effector prediction

All the Type 3 effectors (T3E) have been predicted in both the strains using T3E prediction tool. The effectors present in our strain was compared to those of 11 other strains of *R. solanacearum*.

Classical secretory protein prediction

The classical secretory protein prediction for both GRs-MEP and GRs-SIK was done using the stand alone tool SignalP4.1. GRs-MEP had 522 secretory proteins while GRs-SIK had a total of 517 secretory proteins. The localization of these classical secretory proteins was predicted using SOSUI Gram N to estimate the target regions for the secretory proteins (Fig 20).

Evaluation of EPNs against shoot borer

The infectivity of four promising EPNs, *Heterorhabditis* sp. (IISR-EPN 01), *Steinernema* sp. (IISR-EPN 02), *O. gingeri* (IISR-EPN 07) and *Oscheius* sp. (IISR-EPN 08) was tested against shoot borer infesting ginger under field conditions at Peruvannamuzhi. Among the test EPNs, *O. gingeri*

(IISR-EPN 07) treated plants showed minimum shoot damage (19.4%) in comparison to control (36.9%) which was on par with Malathion (18.4%).

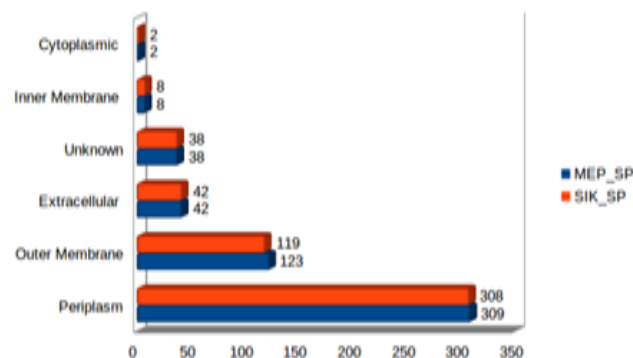


Fig 20. Localization of predicted signal peptides of *R. solanacearum* strains, GRs-MEP and GRs-SIK

Symbiotic bacteria of EPNs

The symbiotic bacterium associated with the EPN, *Heterorhabditis* sp. (IISR-EPN 01) was identified as *Photorhabdus luminescens* (IISR-EPN BC 09) on the basis of morphological, biochemical and molecular characterization.

Documentation of natural enemies of insect pests

Surveys for entomopathogens and other natural enemies of spice crop (black pepper, ginger, turmeric, allspice) pests were conducted in 16 locations in Idukki, Wayanad and Kozhikode districts of Kerala. In Tamil Nadu, six locations were surveyed in Coimbatore district and four locations in Nilgiris district for the occurrence of entomopathogens and other natural enemies. Survey was also conducted in Dimapur district of Nagaland for the occurrence of entomopathogens and other natural enemies. Five entomopathogens including an NPV (IISR-NPV-03) were isolated from spice crop pests (*Aspidiotus destructor*, *Busonomimus manjunathi*, *Aulacaspis* sp., *Zeuzera* sp. and *Pericallia ricini*) during the surveys. The fungus infecting *B. manjunathi* was identified as *Metarhizium* sp. (IISR-EPF-03) and the fungus isolated from *Sinoxylon* sp. was identified as *Beauveria* sp. (IISR-EPF-04). based on morphological and molecular studies. These isolates are maintained in the entomopathogen repository of the institute.

TURMERIC

Genetic resources

1404 *Curcuma* accessions have been maintained in the field gene bank. Germplasm conservatory was enriched with nine *Curcuma* accessions. As part of National Active Germplasm Site (NAGS) on spices and germplasm exchange programme, 25 accessions were supplied to other research centres and 21 accessions were received.

Two hundred and forty seven first generation seedlings besides 476 second generation seedlings, 43 first generation inbreds and three inter-varietal hybrids are also being maintained.

Yield evaluation - germplasm selections

A multilocational trial with three promising turmeric accessions (Acc. 48, Acc. 79 and Acc. 849) along with IISR Prathiba and local check was laid out in Kerala (Peruvannamuzhi), Andhra Pradesh (Vijayawada), Tamil Nadu (Erode) and Karnataka (Chamrajanagar and Chettali). The Acc. 849 (long duration type) recorded maximum yield across locations followed by the short duration genotypes Acc. 48 (Fig 21).

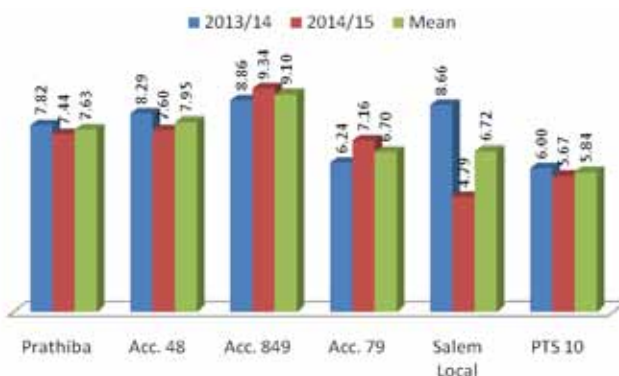


Fig 21. Mean yield data of turmeric genotypes tested in four locations

Evaluation of second generation seedling progenies and inter varietal hybrids

Morphological characters namely plant height, number of tillers, number of leaves in the main tiller, leaf lamina length, leaf lamina breadth, petiole length, collar girth and flowering status have been recorded from 61 second generation seedlings (pot culture).

Rhizomes were harvested in the month of February 2015 and mean yield per pot was calculated. Three seedlings showed multiplication above ten times.

Morphological characters namely plant height, leaf lamina length, leaf lamina breadth, petiole length, collar girth and number of tillers and colour of emerging secondary shoots were also recorded from the three F1 hybrids. Emerging shoots of Hybrid-1 and Hybrid-2 had red colour like the female parent 389/1 while Hybrid-3 had whitish green colour like the male parent Surajana. Hybrid-1 produced 242.0 g rhizomes while Hybrid-2 and Hybrid-3 produced 128.0 g and 74.0 g, respectively. Chromosome number analysis of Hybrid-2 showed $2n=80$ while the female parent 389/1 had $2n=78$ and male parent Surajana had $2n=63$

Chromosome number analysis was completed in 20 second generation seedlings and 25 inbreds. All the second generation seedling progenies showed $2n=84$. Among the first generation inbreds of 138/11/1 analyzed (21), there were variants showing $2n=86$ (2) $2n=88$ (1) and $2n=90$ (1) while the majority showed $2n=84$ (18). All the first generation inbreds of 138/7/1 analyzed (4) showed $2n=84$ (Table 16).

Morphological characters namely plant height, leaf lamina length, leaf lamina breadth, petiole length, collar girth and number of tillers were recorded from the 43 first generation inbreds and their mother plants after 6 months of planting. Variation in inflorescence morphology and flower structure was also recorded from those flowered. Variation in style length, separation of style from staminal column and variation in quantity of pollen were observed among inbreds. On harvest, the weight of rhizomes and morphology were recorded. The weight of rhizomes ranged from 2.0 g to 240.0 g among the inbreds of 138/11/1 and 4.0 g to 86.0 g among the inbreds of 138/7/1.

Table 16. Chromosome number analysis in inbreds

Identity	Chromosome number (2n)
138/7/1(Mother)	84
138/7/1/I1 -1	84
138/7/1/I1 -2	84
138/7/1/I1 -3	84
138/7/1/I1 -5	84
138/11/1(Mother)	84
138/11/1/I1-1	84
138/11/1/I1-2	84
138/11/1/I1-3	84
138/11/1/I1-4	86
138/11/1/I1-5	84
138/11/1/I1-6	84
138/11/1/I1-7	84
138/11/1/I1-8	86
138/11/1/I1-9	84
138/11/1/I1-10	86
138/11/1/I1-11	84
138/11/1/I1-12	84
138/11/1/I1-13	84
138/11/1/I1-15	84
138/11/1/I1-16	84
138/11/1/I1-17	84
138/11/1/I1-18	84
138/11/1/I1-19	90
138/11/1/I1-20	88
138/11/1/I1-21	84
138/11/1/I1-22	84

Pollination and seed setting studies

Self pollination was attempted in cultivars Sudarsana, Narendra Haldi; high curcumin line 389/1; 12 First generation inbreds and two inter-varietal hybrids involving 389/1 x Surajana. Fruit set was observed in two first generation inbreds and the inter-varietal hybrids. The seeds collected from the fruits were sown for germination. Seed germination started in

Hybrid-1 and Hybrid-2 of inter-varietal hybrids to produce F2 generation and inbred 138/11/1/I1-8 to produce second generation inbreds. Fifty two F2 hybrids and three second generation inbreds have been established so far (Table 17).

Self pollination in 24 second generation seedling progenies resulted in fruit set in only 65/9/22. Of the 21 flowers pollinated, only two produced fruits and resulted in 25 seeds. Ten seeds germinated to produce first generation of inbreds.

Table 17. Self pollination, fruit/seed set and germination in turmeric

Identity of selfed genotype	No. of Flowers pollinated	No. of Flowers set fruit	No. of seeds recovered	No. of seeds germinated
Sudarsana	89	-	-	
Narendra Haldi	77	-	-	
389/1	107	-	-	
138/7/1/I1 -2	24	1	5	
138/7/1/I1 -4	20	-	-	
138/11/1/I1-1	39	-	-	
138/11/1/I1-2	24	-	-	
138/11/1/I1-5	101	-	-	
138/11/1/I1-8	31	2(1perished)	4	3
138/11/1/I1-18	31	-	-	
138/11/1/I1-19	38	-	-	
138/11/1/I1-30	25	-	-	
138/11/1/I1-31	38	-	-	
138/11/1/I1-32	38	-	-	
138/11/1/I1-33	45	-	-	
138/11/1(Mother)	19	1	6	
389/1xSurajana (Hybrid 1)	93	8(1Perished)	36	7
389/1xSurajana (Hybrid 2)	49	18	128	45

Cross pollination was performed in three first generation inbreds with pollen from 389/1. Fruit set was observed in one (138/11/1/I1-30 x 389/1). Seven seeds obtained from two fruits were sown and are being observed for germination. Only five seeds germinated out of 28 open pollinated seeds collected from high curcumin line 389/1. Also, open pollinated seeds were collected from 19 second generation OP seedling progenies and sown so as to



raise the third generation. Germination started in 13 of them till date.

Pollen fertility was assessed through carmine staining in seven first generation inbreds and one hybrid. Variation in pollen fertility among inbreds ranged from 0.0% to 67.63%. Pollen fertility analysis of 16 second generation seedlings showed variation ranging from 28.28% to 98.37% (Table 18).

Table 18. Pollen fertility by staining in second generation seedlings

Identity	Pollen fertility (%)
18/22/8	39.75
18/22/20	35.29
69/5/20	98.37
69/5/17	25.68
69/5/15	90.91
69/5/4	77.89
69/5/19	74.10
138/17/7	77.39
138/17/7	83.94
138/12/20	32.46
138/4/2	28.28
138/12/20	80.78
20/7/18	45.95
18/7/7	61.94
18/22/16	37.37
18/22/12	60.15

Identification and validation of microRNAs from turmeric

About 33 conserved miRNA families and 94 novel turmeric specific miRNAs were identified using high throughput Illumina sequencing of small RNAs. Out of these, 14 conserved miRNAs viz., miR156, miR157, miR159, miR160, miR161, miR166, miR167, miR167g, miR169, miR171, miR172, miR319, miR396, miR398 were validated by stem loop RT-PCR. Ten novel miRNAs specific to turmeric were also validated by the same method. Four miRNAs viz., miR156, miR167, miR172 and miR396 were also cloned from turmeric. Two of the

cloned miRNAs (viz., miR156 and miR167) were validated by northern blotting. The target mRNAs of the identified miRNAs were predicted using psRNA target. Some of the targets identified were transcription factors like squamosa promoter binding like protein genes (SPLs), growth regulating factors (GRFs), NAC domain containing proteins, F-box family proteins, floral homeotic protein APETALA 2 like isoform X1, homeobox leucine zipper proteins, TCP transcription factors and auxin response factors were targeted by miR156, miR396, miR164, miR394, miR319, miR172 and miR160 respectively, while targets of novel miRNAs included transcription factors and metabolic enzymes.

Protocol optimization for amplification of full length cDNA

A protocol was optimized for amplifying full length genes for cloning key enzymes of the curcumin biosynthetic pathway. First strand cDNA was synthesized from 1µg of RNA from the pooled tissues of Megha turmeric using SMARTer PCR cDNA synthesis kit and double stranded cDNA was prepared using PrimeSTAR HS DNA polymerase. The cDNA was circularized using T4 DNA ligase and used as a PCR template for the amplification with gene specific (curcumin synthase3-curs3) inverse primers. The PCR product of 1.1kb was purified, ligated in pGEM-T vector and transformed in *Escherichia coli* DH5α cells. The positive recombinants were sequenced bi-directionally using M13 primers. Sequence analysis revealed that 1.1 kb amplicon contained both 5' (137bp) and 3' (299bp) regions of the curs3 cDNA.

SSR genotyping

Ten highly polymorphic SSR primers (CLM 2, CLM 33, CLM 34, CLM 45, CLM 61, CumiSat 8, CumiSat 18, CumiSat 20, CumiSat 28 and CumiSat 22) were selected for genotyping in 96 turmeric accessions using MultiNa, microchip based electrophoresis system. The varieties Suvarna, Suguna and Sudarshana could be separated from other released varieties with primers CLM 2, CLM 34 and CLM 61. Turmeric released variety Prabha showed a distinguishable pattern from other released varieties with the SSR primer CLM 25 and CumiSat 8 (Fig 22).

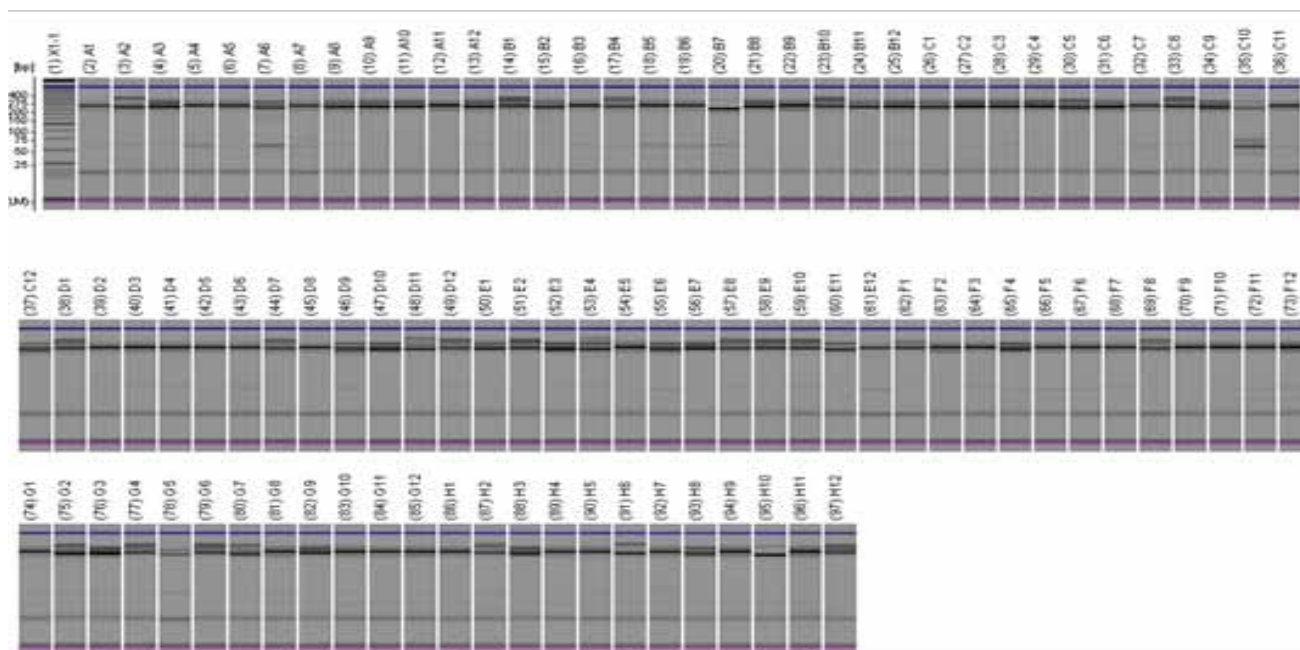


Fig 22. Profiling of SSR primer CLM 25 in 96 *Curcuma* accessions

Influence of coloured shade nets

In turmeric, not much variation in partitioning to rhizomes (28-31%) was observed among treatments at 140 DAP. The least fresh weight was recorded under open conditions. Photosynthetic and transpiration rate among treatments was on par. Maximum rhizome yield was noted in white shade net followed by black and red shade nets at harvest. Leaf oil was on par though black and red recorded slightly higher leaf oil percentage.

Organic farming

Under Network Project on Organic Farming field experiments were conducted to evaluate the impact of organic, conventional and integrated management practices on crop productivity and soil health. Eleven varieties of turmeric were tested under five treatments viz., organic 100%, organic 75%, INM (75% org + 25% chemical), INM (50% org + 50% chem) and 100% chemical. Yield parameters were found to be higher under INM with innovative technology (50% org + 50% chem. + micro nutrient). Under organic treatment, Aleppy supreme has recorded maximum (20 t ha⁻¹) yield whereas Sudarsana (31.3 t ha⁻¹)

and Suvarna (18.45 t ha⁻¹) varieties have recorded maximum yield under INM and chemical treatments, respectively. Soil analysis after 120 DAP showed higher micronutrients availability in INM (50% org + 50% chemical + micronutrient). Population of *Azospirillum*, Phosphate solubilising bacteria, and *Pseudomonas* sp. was maximum under organic management system.

Farming system model plot was established with spices, fodder and vegetables at Chelavoor farm. Black pepper, turmeric, ginger, fodder grasses (hybrid Napier grass, CO-3, CO-4, Congo signal grass), yams, tapioca, banana, pineapple and cowpea were planted and established in the model plot. Fodder grasses 656 kg, Tapioca 50 kg and vegetable cowpea 8 kg were harvested.

Cytotoxicity of turmeric extracts on cancer cell lines

Cytotoxicity of 80 % ethanol extracts of turmeric on A375 cancer cell lines was tested at concentrations ranging from 5-100 µg mL⁻¹. Inhibition of cell growth of 64-72% was observed at these concentrations after 24h incubation and 72-76% inhibition after 48 h incubation.

Evaluation of EPNs against shoot borer

The infectivity of four promising EPNs, *Heterorhabditis* sp. (IISR-EPN 01), *Steinernema* sp. (IISR-EPN 02), *O. gingeri* (IISR-EPN 07) and *Oscheius* sp. (IISR-EPN 08) was tested against shoot borer infesting turmeric under field conditions at Peruvannamuzhi. Among the tested EPNs, *O. gingeri* (IISR-EPN 07) treated plants showed minimum shoot damage (28.4 %) which was on par with Malathion (24.6%) in comparison to control (51.9%).

Characterization of NPV isolated from *Spilarctia obliqua*

The NPV isolated from *S. obliqua* (IISR-NPV-02) (Fig 23) infesting turmeric was characterized as Group I NPV based on the sequence information of *polyhedrin* and *lef-9* genes and restriction enzyme analysis. The genome size was estimated as 99kb. The biological activity (LD_{50} & ST_{50}) of the SpobNPV

was tested under laboratory conditions against larvae of *S. obliqua*. The results indicated that the isolated virus is highly virulent to the host insect.

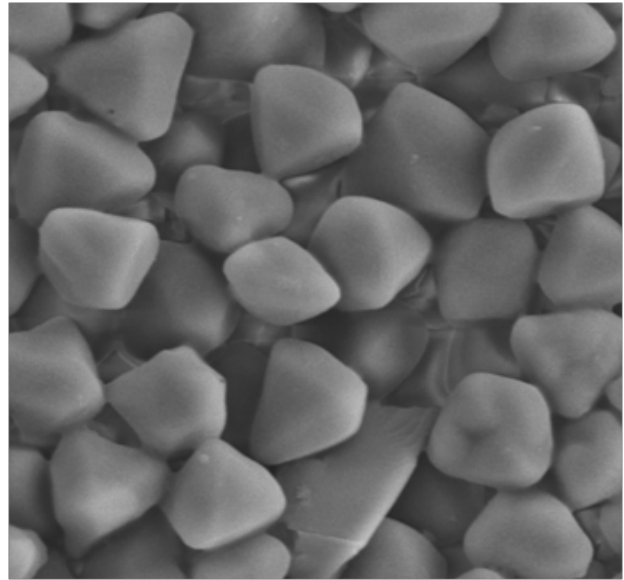


Fig 23. Scanning electron microscopy of NPV infecting *S. obliqua*



VANILLA

Genetic resources

Ninety three germplasm collections, 100 seedlings and 75 inter-specific hybrids of different combinations are being maintained.

Interspecific hybrids

Ten interspecific hybrids involving *Vanilla* sp. (A & N) x *V. aphylla* were characterized based on morphological characters (Fig 24a) and chromosome number (Fig 24b). in addition to those characterized earlier. Characters namely leaf length, leaf breadth,

internode length and stem girth were recorded. Variation was observed particularly with respect to leaf length and breadth. Chromosome analysis showed $2n=56$ in all the ten interspecific hybrids analyzed.

Seed germination was observed on *in vitro* seed cultures of H-1 x *V. aphylla* and H 1 x *Vanilla* sp. (A&N) supporting the earlier observation that the sterility in hybrids of *Vanilla* sp. (A & N) x *V. aphylla* is restricted to the level of pollen and ovules are fertile.



Fig 24a. Morphology and flower of *Vanilla* sp. (A&N), *V. aphylla* and their interspecific hybrids. A. Plant habit of *Vanilla* sp. (A&N) (female parent). B. Plant habit of *V. aphylla* (male parent). C. Plant habit of Interspecific hybrids. D. Flower of *Vanilla* sp. (A&N) E. Flower of *V. aphylla* F. Flower of interspecific hybrid

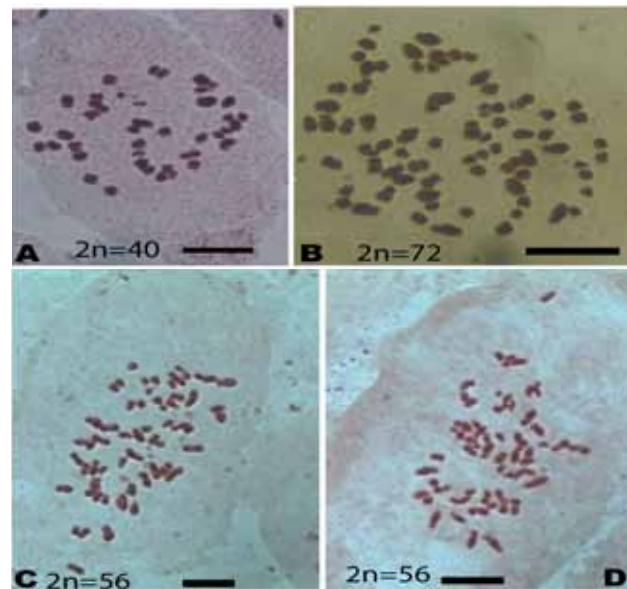


Fig 24b. Chromosome number of *Vanilla* sp. (A&N), *V. aphylla* and their interspecific hybrids. A. Mitotic metaphase of *Vanilla* sp. (A&N) (female parent) showing $2n=40$. B. Mitotic metaphase of *V. aphylla* (male parent) showing $2n=72$. C&D. Mitotic metaphase of interspecific hybrids showing $2n=56$. Bar represents $5\mu\text{m}$

NUTMEG

Genetic resources

A farmer participatory germplasm collection for nutmeg germplasm was made in Idukki, Kottayam, Kozhikode and Malappuram districts of Kerala. Thirty one nutmeg germplasm including few farmers varieties were collected and conserved. The unique germplasm collected include a nutmeg with rudimentary sterile seed (Fig 25), nutmeg with bold nut, thick and entire mace type, high yielding monoecious nutmeg and Punnathanam jathi, a farmer's variety which had very bold nut and thick mace.

Registration of nutmeg germplasm

A nutmeg germplasm (IC 053718), with bold nut, high sabinene and myrcene content has been registered with NBPGR, New Delhi with the registration number INGR-14039.



Fig 25. Fruit, mace and seeds of sterile a. and fertile b. nutmeg

Conservation and evaluation of seedling progenies of monoecious nutmeg trees

Seedlings of nutmeg collected from a monoecious tree at Ankola, Karnataka and two grafts collected from Kottayam, Kerala were planted for field evaluation. All the plants have established and the growth of the seedlings is good.

Evaluation of grafts of elite lines

Grafts of four short listed elite lines namely, A9/4-3 (IC 537153); A9/4-11 (IC 537153); A4/17 (IC 537043); A9/20 (IC 537169) having high myristicin and elemicin in nutmeg and mace oils were planted for field evaluation at Chelavoor. 90% of the plants have established and the growth is good.

High density planting

An experiment was initiated at Chelavoor campus this year for studying the suitability of the nutmeg for high density planting with plagiotropic grafts of nutmeg variety IISR Viswashree. The spacing adopted was 4m x 2m, 4m x 3m, 4m x 4m, 4m x 5m, 4m x 5m (control, no pruning). A study to determine the effect of pruning nutmeg in 25 year old trees at IISR-Regional Station, Appangala by detopping nutmeg at 20 feet above ground level and pruning the side branches at 1 and 2 m distances from the main trunk was also initiated.

Ethrel treatment in nutmeg induces synchronous fruit splitting

A simple technique of hormone treatment was developed to split open fruits without exposure to soil. The methodology involves harvesting physiologically mature fruits when the colour of the rind change from green to pale yellow/yellow and dipping them in 500 ppm ethrel (2-Chloroethylphosphonic acid) solution for 10 minutes and then storing them in shade. By this method, 90-100 % of fruits will be split in 18-20 h. The quality in terms of nut and mace dry weights and essential oil content were comparable with that of naturally split fruits. Width of the split was also equal to that of naturally split fruits. Dipping in 100 ppm naphthalene acetic acid (NAA) induced 70-80 % and water dipping induced 40-50 % fruit split in 18-20 h. Width of the split was lesser in NAA and water dip

treatments compared to ethrel treated or naturally split fruits. The method is very simple and can be easily practiced by farmers which save time, labour and money for harvesting and processing of nutmeg. The cost of ethrel treatment works out to be less than Rs 1000 t⁻¹ of fruits.

Characterization of *Phytophthora* spp. causing leaf and nut fall

During the monsoon period of 2011, occurrence of a serious leaf fall and nut fall was observed in major nutmeg growing areas of Thrissur, Ernakulam, Idukki, Kottayam and Kozhikode districts of Kerala. The disease was characterized by severe defoliation and nut fall. Leaf and stem infections resulted in extensive defoliation. A detailed study on morphology, temperature requirement and molecular characterization using ITS marker was undertaken. The sporangial morphology of the isolates (13-01 to 13-06) was characterized by papillate/semi papillate sporangia, ovoid to obovoid, with intermediate pedicel lengths. The oogonia was amphigynous and formed on pairing with *P. meadii* from cocoa which is of A2 mating type except for 13-06 which formed oogonia in presence of 05-06 (A1) and designated as A2. All the isolates grew between 15-30°C with an optimum temperature of 25°C and no growth was observed at 35°C. In ITS sequencing the nutmeg *Phytophthora* isolates formed a separate clade with *P. colocassiae* and showed close similarity to *P. meadii*. In ITS sequence, 100% similarity was shown to *P. meadii* by isolates 13-02, 13-04 and 13-06 in Q-bank fungal identification database.

Chemoprofiling of *Myristica prainii*

Nuts collected from *M. prainii*, a wild *Myristica* species were subjected to phytochemical analysis. The nuts contained 38-40% butter. The fatty acid profile of the butter indicated 78-83% myristic acid, 7.7-8.3% myristoleic acid, 5.8-7.4% palmitic acid, 3.0-3.5% lauric acid, 2.5-3.9% elaidic acid as chief constituents. Butter of *M. fragrans* nuts contained 80-82% myristic acid, 5.0-6.0% palmitic acid, 3.0-3.5% lauric acid and 3-3.6% oleic acid.

Antioxidant potential of *M. prainii*

Antioxidant potential of methanolic extract of *M. prainii* nut and mace were compared by DPPH and Phosphomolybdenum methods. The antioxidant activity was found to be concentration dependant between 1 to 10 mg mL⁻¹. The peroxide scavenging activity of the seed extract ranged from 16-76% and that of mace from 12-59%. By phosphomolybdenum method, the seed and mace showed antioxidant potential of 335 and 239 µg ascorbic acid equivalents per ml.

Garcinia bark exudates

The biochemical composition of the exudates from the barks of *G. gummi-gutta*, *G. indica* and *G. xanthochymus* were studied. Total phenol content of the resins varied from 53-67% and xanthenes ranged from 20-35%. IC₅₀ of the antioxidant activity measured by DPPH method showed 18-22 mg ml⁻¹, which were of the range of aqueous extracts of fruit rinds. The biochemical content of the exudates is depicted in Table 19 and composition of resin extract in Table 20.

Table 19. Biochemical composition of exudates

Exudate Parts	<i>G. gummi-gutta</i> (%)	<i>G. indica</i> (%)	<i>G. xanthochymus</i> (%)
Resin	68.3	60.4	40.0
Total sugars	14.2	20.3	35.1
Insoluble part	17.5	19.3	24.9

Table 20. Biochemical composition of resin part of the exudate

Biochemical content	<i>G. gummi-gutta</i> (%)	<i>G. indica</i> (%)	<i>G. xanthochymus</i> (%)
Total phenol (g 100g ⁻¹)	56.37	53.43	67.13
Total flavonoids (g 100g ⁻¹)	16.64	18.80	37.61
Xanthone content (g 100g ⁻¹)	35.57	32.42	20.12
Antioxidant activity (IC ₅₀ µg mL ⁻¹)	20.4	18.2	21.7

DUS TESTING OF SPICES

On-site DUS testing of spices were undertaken for the 12 farmer's varieties and six varieties of common knowledge (IISR varieties). Three black pepper (Pepper Thekkan, Kumbakkal Selection and Agali pepper) and six small cardamom varieties (Ela-Elarajan, Ela-White flowered, Thiruthali, Wonder Cardamom, Panikulangara bold-1 and Panikulangara

bold-2) from farmers were recommended for registration by Protection Plant Varieties and Farmers Right Authority (PPV&FRA), New Delhi based on the unique characteristics of the candidate varieties. Four farmer's varieties in turmeric, three in ginger and four in turmeric from ICAR-IISR are also being tested under DUS.



Fig 26. Field visit of DUS expert committee in Kerala



BIOINFORMATICS

Identification of antimicrobial peptides through genome mining

Around 17422 antimicrobial peptides (AMPs) were identified from *Z. officinale* (68946 sequences) using AMPA tool. On analyzing further with CAMP database, the number reduced to 7661 AMPs. Similarly, 19688 AMPs were predicted from 86617 sequences of *C. amada* by using AMPA tool which was refined to 8282 AMPs by using CAMP tool. On checking reported and non-reported AMPs using CD-search, it was found that ginger contains 557 reported and 7104 non reported AMPs while *C. amada* contains 505 reported and 7777 non reported AMPs. Protein blast was used in this study to classify the antimicrobial peptides as antibacterial, antifungal, anticancer, antiHIV, insecticidal, antiprotist, antiparasite and antiviral peptides. The property of each peptide was identified by using APD server. AMPs were also predicted from *Bacillus megaterium* DCM 319 strain and *Pseudomonas putida* NBRC 14164 using AMPA and CAMP tools.

Bioinformatics data analysis

Technical help and support was extended to the following works of various scientists of the Institute:

- Genome assembly and annotation of two strains of *Ralstonia* from Sikkim and Meppadi (Rs-SIK and Rs-MEP) (Fig 27)
- Comparative genomics of five species of *Phytophthora* and their secretome analysis
- Transcriptome analysis of *P. colubrinum*-*P. capsici* dual RNA-seq to discover miRNA targets and their Blast2GO annotation
- Analysis of data for comparison of the transcriptomes of ginger (*Z. officinale*) and mango ginger (*C. amada*) in response to the bacterial wilt infection
- EST analysis and identifying resistance gene in *C. amada* and *Z. officinale*
- Secretome and EST analysis of *R. similis*
- 18s RNA structural alignment and comparison, development of comprehensive SSR & SNP markers for studying the genetic diversity and association analysis in *Curcuma* species
- QRT primer designing, phylogenetic analysis and domain studies for real time quantitative RT-PCR analysis of some pathogenicity genes expressed during *P. capsici* - *P. colubrinum* interaction.
- QRT primer designing, phylogenetic analysis and domain studies for targeted discovery of R genes in black pepper - *P. capsici* interaction.
- KEGG annotation for *Curcuma* species.
- Protein-protein docking study for NB-LRR and AVR gene from *Piper nigrum*
- MLST and phylogenetic analysis of various isolates of *P. capsici* and *R. solanacearum*
- Molecular modeling, biological activity prediction, docking and validation studies of inhibitory activity of secondary metabolites of spices against various pathogens
- *In silico* biological activity prediction on molecular level druggability studies on phytochemicals in spices has been done to understand and validate therapeutic use of spice compounds.
- Docking studies with telomerase and cancer associated drug targets with spice phytochemicals
- Docking studies of a glucanase inhibitor from *P. capsici* and beta 1-3 glucanase from *P. colubrinum*.
- Docking studies with telomerase and cancer associated drug targets for the project evaluation of spice extracts for anticancer effect in relation to telomerase activity
- Antioxidant activity prediction for components in black pepper that have activity against cervical cancer
- Virtual screening and *in vitro* assay to explore novel inhibitors from black pepper against potential targets of *R. similis*
- Docking and phylogenetic analysis for comparative study of pathogenesis related protein-5 of different Zingiberaceae species.

Collaboration and support

Technical support was extended to the following institutions in data analysis and bioinformatics training.

- NIT, Kozhikode - Primer designing and validation for human myeloperoxidase gene & bioinformatics training
- Calicut University-Phylogenetic analysis for *Bacillus thuringiensis* BPU5, a novel isolate from Malabari goat that efficiently combats *Tetranychus macfarlanei*, a spider mite
- ICAR-Sugarcane Breeding Institute, Coimbatore- Transcriptome data analysis for *Colletotrichum falcatum* and functional annotation for protein sequences of 2.94 lakhs sugarcane ESTs

- ICAR-Indian Institute of Horticultural Research, Bengaluru- Analysis of *R. solanacearum* whole genome
- ICAR-Central Institute of Fisheries Technology, Kochi- Transcriptome analysis of Gammaproteobacteria and Mangrovibacter

Bioinformatics training

Supported the following DBT sponsored training programs

- Genomics and proteomics in plant and microbes towards translational research, 21 January to 10 February 2015, IISR, Kozhikode.
- Workshop on Bioinformatics tools and applications, 18-19 February 2015, BIF-Calicut University

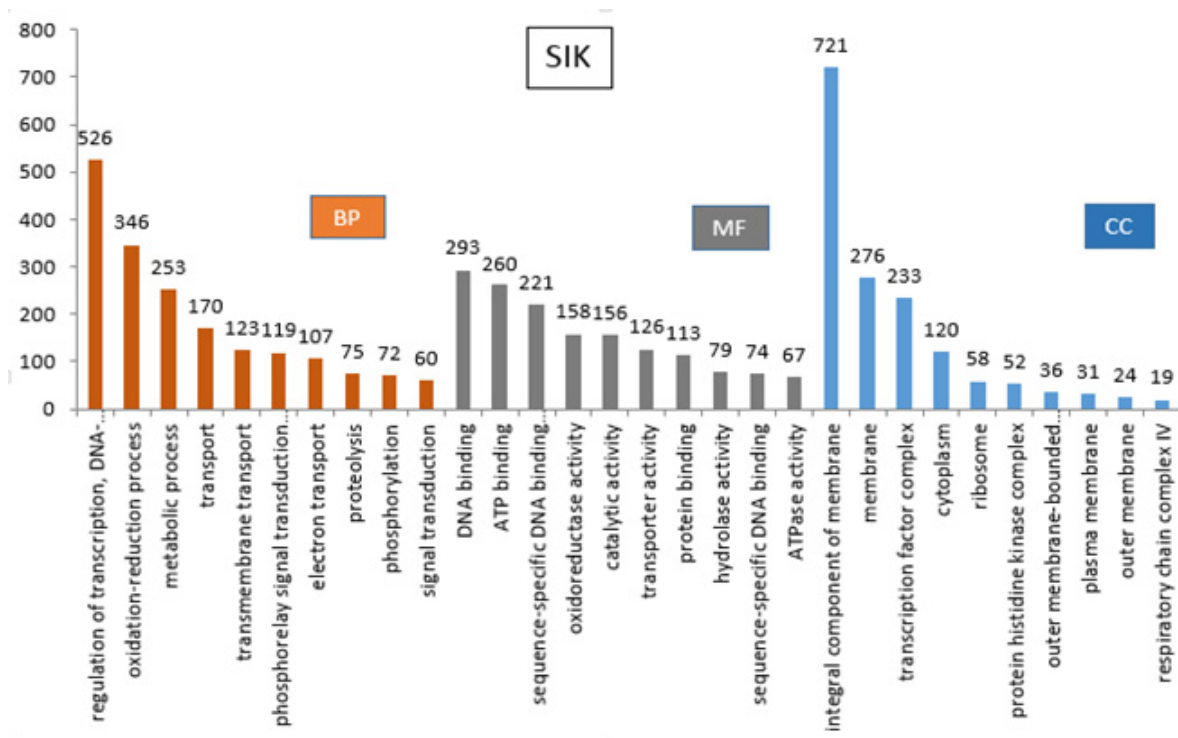


Fig 27. Gene ontology (GO) distribution – from the predicted CDS of SIK, classified into three major components (molecular components, biological process and cellular components).

NATIONAL INFORMATICS CENTRE FOR SPICES

Subscription of library resources

IISR Library continued its subscription to EBSCO Discovery service to facilitate the access of various library resources/databases through a single window. The subscription of CAB Direct, 14 foreign and 54 Indian Journals were continued. Thirty six reference books, 23 e-books, 19 technical reports, eight theses, nine project reports and 15 *gratis* books were added to the stock during the period.

E-services

The library website, Spice Bibliography, OPAC, Spice-books, DSpice etc. were updated with newly added information resources. The metadata of 1500 newly bound journals added to OPAC database. Thirty eight new books were scanned and converted to searchable e-books. Twelve issues of the Agrititbits, the agricultural news service, were brought out. Online alert service called 'Latest

in spices research' was continued for the benefit of spices workers for the latest research articles published globally on 12 mandate crops of IISR. The institutional digital repository DSpice was updated with 61 new documents.

Training on library use

Hands on sessions of effective use of library resources using EBSCO Discovery tool was held in the library on 14 November 2014.

Library services

Library continued to be a part of CeRA, the e-journal consortium of ICAR and catered to the requests from various CeRA members. During the reporting period, 2050 users availed the e-library facilities while 1200 internal and 500 external users visited the library.





EXTENSION AND IMPACT ASSESSMENT

Capacity building and front line intervention programmes for spice sector development in tribal settlement areas of Kerala and NE states

The project is implemented under Tribal Sub Plan (TSP) XII Plan aiming at livelihood improvement of tribal communities. The selection of the target areas/ institutions and feasible areas/ themes of technology interventions were carried out through collection and analysis of secondary data and participatory rural need assessment and appraisal. Based on the assessment of the secondary data on agricultural production plan for tribal hamlets, Anakkal hamlet and Attappady Cooperative Farming Society (ACFS) were selected as target institutions. Anakkal tribal hamlet has got 148 tribal farm families cultivating a total land of 40 ha. The location receives a rain fall of 1500-2000 mm and has got a functional tribal development “samithy”. A need assessment meeting was organized with the tribal community leaders of the hamlet, officials of ACFS and Department of Agriculture, Government of Kerala. The technology interventions selected for demonstration were:

- Improved varieties of black pepper for area expansion in Anakkal tribal hamlet
- Provision of naturally ventilated poly house for establishment of a black pepper nursery as a community asset to be managed by farming

cooperative society.

- Creation of community owned capital assets like prayers in the target area.

Through a similar exercise, nine Krishi Vigyan Kendras in Nagaland state were identified as participating institutions for programme in NE states. The two lead institutions identified were ICAR Research Complex for North Eastern Region, Nagaland center, Dimapur and the Central Institute of Horticulture, Dimapur. The theme areas selected for interventions are:

- Single sprout transplanting method of ginger and turmeric.
- Production of nucleus plating material of improved varieties of ginger and turmeric.
- Providing small scale turmeric steam boiling units as community facilities.

Four awareness programmes, three in Wayanad and one in Palakkad were organized for tribal farmers of Kerala (Fig 28, 29). In North East India, one workshop on need assessment and action plan preparation involving ICAR stations at Guwahati and one workshop on recent advances in production management in spices involving nine KVKs in Dimapur, Nagaland were organized (Fig 30). The details of awareness programmes and workshops held are provided in Table 21.

Table 21. Awareness programmes/seminars organized under TSP

Programme	Participants	Location
Production management in black pepper for tribal farmers	80	ICAR-IISR, Kozhikode, Kerala
Training programme on production management in black pepper	48	Pudur, Palakkad, Kerala
Training programme on production management in black pepper	70	Meenangadi, Wayanad, Kerala
Community need assessment workshop for Tribal farmer leaders	30	Kalpetta, Wayanad, Kerala
Workshop on action plan preparation for spices development in NE states	25	Guwahati, Assam
Workshop on recent advances in production management of spices	30	ICAR research complex for NEH Region, Dimapur, Nagaland
Training on improved production technology in black pepper	65	Sulthan Battery, Wayanad, Kerala
Training on improved production technology in black pepper	62	Mathamangalam, Wayanad, Kerala
Training on ginger production and management	70	Kanencherry Kuruma colony, Kaniambetta, Wayanad, Kerala



Fig 28. Black pepper cuttings distribution to tribal women during an awareness workshop in Wayanad district, Kerala



Fig 30. Method demonstration of pro-tray method of ginger planting at workshop in Dimapur for KVK staff

Trend in area, production and productivity of black pepper in Kerala

The time series analysis of area, production and yield of black pepper in Kerala during the period 1980-2011 was carried out to identify the trends in these variables. Secondary data from published official documents of Directorate of economics and statistics, Government of Kerala was used for the study. Comparison of compound growth rate was done for two distinct time periods; 1980-1995 and 1995-2011. The cost of cultivation trends during the recent past were analyzed with respect to the “Paid out cost” component of black pepper cultivation during the period 2000-01 to 2012-13.

The study revealed that the growth rates in all the key parameters which determine the domestic availability of black pepper declined for Kerala, which accounts for a significant share of black pepper production in country. This has adversely



Fig 29. Power sprayer being handed over to Puthenkunnu Adivasi Karshaka Sangham, Wayanad

affected the total production of black pepper in India and simultaneously affected the exportable quantity of black pepper of domestic origin. The growth rates of both area and production of black pepper has become negative during the second period whereas growth rate declined significantly in case of yield (Table 22).

Table 22. Growth rate in area, production and yield of black pepper in Kerala

Period	Area	Production	Yield
1979-80 to 1994-95	4.84	6.31	1.38
1995-96 to 2011-12	-1.94	-2.24	0.02

Analysis of cost of cultivation indicted a sharp rise in the cost of cultivation. The slow growth rate of productivity with sharply rising cost of cultivation has resulted in an increase in cost of production of black pepper. This has made the produce from Kerala to be non competitive in the international market. The cost of hired human labour, which alone accounts for more than 50% of the total paid out cost of cultivation in black pepper increased from Rs.8185 per hectare during 2000-01 to Rs. 30147 cost per hectare in 2012-13, which followed a steadily increasing average daily wages in the state during the period.

Potential of technology interventions in black pepper

The district wise data on district wise area production and productivity of black pepper in Kerala and Karnataka for the period corresponding to the XI five year plan was used to estimate the yield gap and the



potential for yield enhancement through technology adoption in these states. The technology backed yield potential was calculated for individual states based on the yield of pepper obtained in the varietal trials conducted in the state under All India Coordinated Research Project on Spices (AICRPS) during the period 2007-08-2011-12. The average yield gap of black pepper in Kerala and Karnataka was estimated to be 309 kg ha⁻¹ and 634 kg ha⁻¹ respectively (Table 23). The total production gap in quantity terms due to non-adoption of technology was estimated to be about 50,000 tonnes at the national level. The

cultivators would result in an additional production of 12343 tonnes of dry pepper valued at 414 crore rupees.

Trends in export of spice oils and oleoresins from India

The quantity of spice oils and oleoresins exported from India grew at a compound annual growth rate of 8.0% during the period 1998-99 to 2012-13. Though the nominal value of the spice oils and oleoresins exported have shown a growth rate of 11.62% per annum, the growth rate drops down to

Table 23. Production gap in black pepper

Particulars	Value
Kerala	
Area under pepper (ha)	151679
Technology backed yield (kg ha ⁻¹)	532
Average yield (kg ha ⁻¹)	282
Average yield gap (kg ha ⁻¹)	309
Production gap (tonnes)	37971
Production gap in value terms (crore Rs)	1212
Karnataka (based on Adjusted yield)	
Area under pepper (ha)	17483
Technology backed yield (kg ha ⁻¹)	1321
Average yield (kg ha ⁻¹)	682 #
Average yield gap (kg ha ⁻¹)	634 #
Production gap (tonnes)	11403
Production gap in value terms (crore Rs)	364
All India	
Total Production gap in quantity terms (tonnes)	49374
Total Production gap in value terms (crore Rs)	1655*

Calculated using the adjusted yield of pepper in Karnataka based on Spices Board estimates

* This figure includes an addition of 5 per cent over the total value of production gap in Kerala and Karnataka to account for other pepper producing areas.

technology generated by the public funded research institutions offer tremendous scope for enhancing the productivity levels of pepper in the country.

The approximate value of additional returns to the pepper farmers from complete spread of technology adoption would be of the order of 1655 crores at 2011-12 prices. Even an additional adoption of complete technology package by 25% of pepper

6.0% per annum in real terms (Base: 2004-05 = 100). Based on a conservative estimate based on the linear trend analysis, the exports from the country can be expected to grow at a rate of approximately 400 tonnes an year in the short run. Based on the study, it is estimated that the quantity of Spice oils and oleoresins exported from India would be nearly 11,000 tonnes during 2014-15 (Fig 31).

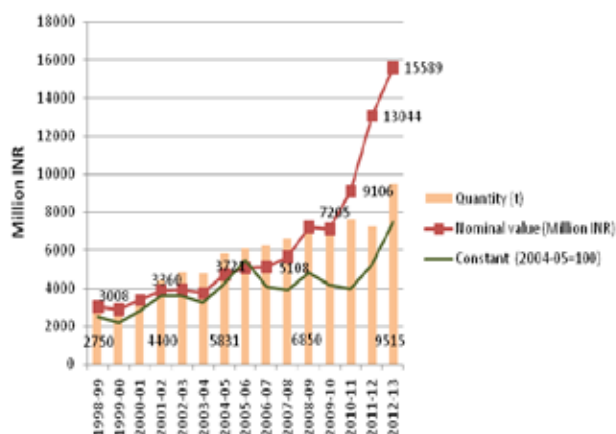


Fig 28. Trend in Export of Spice oils and oleoresins

Cost of production of turmeric

The cost of production of turmeric in three southern states (Kerala, Tamil Nadu and Andhra Pradesh) was estimated based on available secondary data, key informant survey and expert estimates. The input use level was marked up for Andhra Pradesh by 30%, based on expert opinion in comparison to states like Kerala. Though the cost of cultivation is higher in Andhra Pradesh, the higher productivity of the crop in the state brings down the cost of production and is the lowest among the three states studied. Table 24 presents the estimates for the cost of production of dry turmeric in the three states.

Table 24. Comparative cost of production of turmeric

State	Average yield (t ha ⁻¹)	Cost of Cultivation (Rs ha ⁻¹)	Cost of Production (Rs quintal ⁻¹)
Andhra Pradesh	6.52	103582	1589
Kerala	2.43	87276	3592
Tamil Nadu	5.20	90000	1731

Notes

1. Cost estimate for Andhra Pradesh based on cost of cultivation for turmeric in Kerala as the benchmark and increasing the variable cost elements by 30% to account for the intensive cultivation practice followed.
2. Cost estimate of Tamil Nadu based on key informant survey in Erode district
3. The interest on the value of the land is not included in the calculation which will further raise the cost of production

Spicepedia – A Knowledge base for spices

In tune with the NAIP initiative, Agropedia, an interactive portal for grey literature on spices, spicepedia was developed in collaboration with IIT, Kanpur. It is now maintained by IASRI, New Delhi. The main objective was content development and management of audio, video and text messages pertaining to major spices from different sources. Totally, content was developed for 320 collected documents including popular articles from various publications, radio talks and knowledge capsules pertaining to spice crops and agriculture. During the current year 25 videos capsules on major innovative technologies of ICAR-IISR were developed. The major themes covered for developing knowledge capsules were propagation in major spice crops, technologies developed for integrated nutrient management in spices and post harvest technology with emphasis on state of the art mechanized processing and value addition spices.

Varietal spread

Turmeric

Feedback from farmers’ plot revealed very good acceptance of the released turmeric variety Prathibha (Fig 32). Mr. Ramaprasad Reddy of Hyderabad, an young IT professional turned turmeric grower, who grew Prathibha in 3 acre plot got an average yield of 38 t ha⁻¹ with 23% dry recovery. He also got a better price for Prathibha owing to its better quality.



Fig 32. Mr. Ramaprasad Reddy’s Prathibha plot

Ginger

Mr. K.C. Joseph Kavukattu, Venappara, Kozhikode, a small farmer gets a multiplication rate of 1:25 for Varada from his 40 cents of land. Mr. Joseph attests that a multiplication rate of 1:40 is achievable with Varada under better management (Fig 33).



Fig 33. Mr. Joseph with the harvested Varada ginger

Area-wide integrated management of wilt diseases

The project on area-wide integrated pest management (AWIPM) for wilt disease in black pepper was launched in three panchayats of Wayanad, Kozhikode and Idukki districts. A total of 71 farmers in Korangad and Thekkumthottam in Kozhikode, Rajakkad in Idukki and Pulpally in Wayanad were selected. Demonstration plots were taken in nine farmer's plots. Three nurseries each were also established in these areas. Trainings and awareness programmes were conducted in all the areas. The status of foot rot and slow decline diseases along with other black pepper diseases and insect pest incidence in Thamarassery, Koduvally, Anakkampoyil, Koodaranjii, Omassery, Madavoor, Kakkur, Chelannoor, Chathamangalam, Mavoor, Koorachund, Chakkittapara, Koothali, Kayanna and Nochad panchayats in Kozhikode District were recorded using a standard proforma. A similar survey was conducted at Rajakkad and Pulpally panchayats. Soil samples were collected from these areas and analyzed for nutrient status as well as biological properties. In Kozhikode, a contiguous area of about 5 ha was selected for implementation of the project in Thekkumthottam and Korangad in Thamarassery Panchayat. In Rajakkad many of

the plantations are inter cropped with cardamom. A few plots with pepper as the main crop was also included. In Pulpally, a traditional pepper growing area, Adikolly was selected for the implementation of the project. Baseline data was collected from selected farmers' fields regarding the variety being cultivated, standards used, fertilizer and organic manure application, irrigation schedule if any and plant protection measures being taken up.

Integrated black pepper research and development in North Kerala districts

The project is being implemented in four Panchayats of Kozhikode district (Koorachundu, Chakkittapara, Thamarasery and Olavanna). About 180 samples were collected from farmer's plots for analyses of nutrients and pathogen load. The soils were acidic with medium organic carbon, potassium, calcium and high phosphorus content.

Six trainings were conducted for farmers and Agriculture Officers on pepper production technology and soil management. Twenty four FLDs on improved technologies and 23 participatory nurseries were initiated at farmer's plots in four Panchayats of Kozhikode district. Relevant nursery and field inputs were supplied in time and are being monitored continuously. The establishment of plants in the field ranged from 84-98% and in nurseries 150-800 cuttings were produced by the farmers. Three visits were made by the scientists to the FLD plots for giving advisories.

Rehabilitation package for Wayanad

Soil samples collected from hot spot areas in four panchayats (180 nos) were analysed for the pathogen load of which 10 were found to be *Phytophthora* positive and advisories were given for control. All the soil samples showed the presence of nematode population irrespective of the health of the plant (whether yellowing or healthy), warranting immediate *ad hoc* recommendation for their control. The soil and leaf samples were also analyzed for major, secondary and micro nutrients and results with crop specific recommendations passed on to the farmers. Healthiness of the vines showed good correlation with the leaf N, K and Mg concentration.

Imbalance of leaf N & K content was seen in yellowing vines.

Five visits were made by team of scientists to hot spot areas to educate on soil health and disease problems including three farmers' seminars on pepper nursery/cultivation. Seventy five FLD plots, spread across Poothadi, Mullankolli, Pulpally, Thirunelli and Meppadi panchayats are being maintained to demonstrate combating yellowing

of black pepper through supply of inputs like neem cake, vermicompost, bio control agents and micronutrient mixtures. The plots with moderate to high yellowing have become healthy by the adoption of site specific technology package involving soil acidity correction, biocontrol application and micronutrient management. Those plots treated with gypsum + lime or lime for subsoil acidity has shown reduction in yellowing as compared to control.





KRISHI VIGYAN KENDRA

Training programmes

During 2014/15, KVK has conducted 131 training programmes for practising farmers and farm women, rural youth and extension functionaries in the disciplines of agronomy, horticulture, animal sciences, home science, fisheries, plant protection and allied fields. A total of 4215 trainees benefited from the programmes.

FLD programmes

Ten Front Line Demonstrations (FLD) programmes were undertaken during the period as detailed below.

- Demonstration on integrated management of Thanjavur wilt of coconut.
- Demonstration of high yielding foot rot tolerant variety of Black pepper *viz.*, IISR Thevam
- Demonstration of use of PGPR encapsulated bio-capsules for management of soft rot in ginger.
- Introduction of a high yielding variety of amaranthus, Renusree
- Demonstration on feeding Anionic mixture to prevent milk fever in cows
- Demonstration on complete feed mixture in dairy cattle
- Seed production of pearlspot fish in fresh water area
- Production and marketing of value added products from fruits, vegetables and spices
- Demonstration of a short duration, semi tall upland rice variety, Vaisakh
- Integrated Farming System

OFT programmes

The major On Farm Trials (OFT) programmes carried out during the period are listed below:

- Management of *Phytophthora* foot rot of black pepper
- Assessment of transplanting technique for ginger using pro-trays

- Efficacy of termite soil for udder oedema in dairy cattle
- Culture of Asian Seabass (*Lates calcarifer*) in brackish water ponds
- Performance evaluation of grafted black pepper

Revolving fund programme

KVK has a strong 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. Income was also generated by way of sale of layer chicks, goats, heifers and bulls and consultation and doorstep services through the clinic. During the period, an amount of Rs.7.56 lakh has been realized through sale of planting materials, bioproducts, bioagents and the activities of Plant and Animal Health Centre.

Plant and animal health centre

The Kendra operates a plant and animal clinic offering various services to the farmers. An artificial insemination facility is also available at the centre to upgrade the genetic stock of livestock. The centre offers consultation, treatment and doorstep services with a nominal fee. In addition to the various treatments, the centre also provides vaccination facility and organises animal health camps in association with the state animal husbandry department. The various activities taken up by the Clinic during the period are furnished below:

Consultancy/advisory/home service carried out	1236
Artificial insemination carried out	118
No. of Animal health campaigns/infertility camps	4
Vaccination of poultry birds and animals	11750
Block <i>ksheeroltsavam</i>	2

Other extension activities

Nature of Extension Programme	No. of Programmes
Field day	10
Exhibition	11
Film show	63
Farmers' seminar	9
Workshop	4
Group meetings	5
Newspaper coverage	27
Radio talks	3
Popular articles	4
Extension literature distributed	100
Advisory services/ helpline	2464
Scientific visit to farmers field	16
Field visits	224
Exposure visits	7
Consultancy services	667
Farmers visit to KVK	4087
E-mails	306
Meetings attended	17
Diagnostic visits	25
Trainings attended	3
Animal health camps	4
Kisan mela/ technology week	1

Demonstration units

The following demonstration units are maintained by the KVK.

- Medicinal plant unit
- Model homestead garden
- Model arecanut seed garden
- Guava demonstration unit
- Sapota demonstration unit
- Vermiculture unit
- Nutmeg scion bank
- Dairy unit
- Goatary unit
- Layer unit
- Broiler unit
- Hatchery unit
- Ornamental fish culture unit

- Anthurium unit
- Pot culture of vegetables
- Coconut nursery

Kisan mobile SMS service

KVK started short message Service (SMS) to all registered farmers on latest updates in agriculture and allied fields over their mobile phones. The SMS are being sent to farmers regarding new interventions, latest technologies, market price of agriculture produce, weather forecast, disease management measures, planting material availability, forthcoming trainings etc. KVK has so far sent 18 SMS, 5 voice message benefitting 743 farmers and 141 Extension functionaries.

Technology week

Technology week (*Eruthum Kathirum*) of the Kendra was conducted from 20 - 24 March 2015 in which about 450 persons including farmers, extension functionaries attended (Fig 34). Experience sharing of farmers, seminars, exhibition, sale of techno inputs and method demonstrations were also organized as part of the celebration.



Fig 34. Dr. M. Anandaraj, Director, ICAR-IISR inaugurating technology week.

Establishment of hatchery unit

A hatchery unit with 30,000 eggs capacity per month was established at KVK with the financial support of NABARD.

Gardeners training programme

One gardeners' training programme of six months

duration was organized under the sponsorship of State Horticulture Mission empowering 25 rural youth.

Out of these, 12 trainees started self-employment units in various nursery activities.



Fig 35 Glimpses of KVK activities

ALL INDIA COORDINATED RESEARCH PROJECT ON SPICES (AICRPS)

All India Coordinated Research Project on Spices is a coordinating unit with 38 centres (19 regular, 11 co-opting and 8 voluntary centres) supplemented by three more in project mode funding, spreading over various agro climatic zones in 23 states of the country. Black Pepper, Large Cardamom, Small Cardamom, Ginger, Turmeric, Cinnamon, Nutmeg, Clove, Coriander, Cumin, Fennel and Fenugreek are the mandate crops. Annual budget for the year 2014/15 was Rs. 462 lakhs as ICAR share.

Genetic resources

A total of twenty new collections were made from black pepper, large cardamom, ginger and turmeric. Dwarf clove, king clove and extra bold Madagascar clove (for the first time) from Simpson and Rajan estates of Nagercoil were collected. 10 Bold types of ginger and 3 *Curcuma* species from Nagaland and Arunachal Pradesh and one unique black ginger was collected from Nagaland. Jamaican ginger, Jamaican turmeric and Singapore ginger were collected from local farmer's fields of Kerala and Tamil Nadu respectively.

One hundred and eighty three ginger collections were evaluated for rhizome yield and other horticultural traits at Solan. The yield range varied from 100.63 q ha⁻¹ (SG-865) to 141.20 q ha⁻¹ (SG-857). Yield of three lines viz., SG-1134 (142.48 q ha⁻¹), SG-857 (141.20 q ha⁻¹) and SG-12-4 (134.45 q ha⁻¹) excelled the check Himagiri which yielded 124.03 q ha⁻¹. The rhizome rot disease incidence varied from 11.47-25.47% with 11.47% and 15.60 % in SG-12-4 and Himagiri, respectively.

In a nutmeg germplasm evaluation trial at Dapoli, maximum dry nut yield (1505.0 g) and dry mace yield (315.0 g) was recorded in the genotype DBSKKVMF 29 (2006 to 2014). The genotype DBSKKVMF 29 is found promising considering its fruit weight, nut weight and mace weight.

Crop improvement

Five high yielding varieties of spices were recommended for release in XXVth AICRP on Spices workshop held at UBKV, Pundibari. Two cardamom varieties Appangala 2 (first hybrid resistant to *Katte* virus) from ICAR-IISR Regional Station, Appangala and PV-3 (Moderately resistant to drought) from Cardamom Research Station, Pampadumpara, 2 coriander varieties RCr 475 (bushy and erect plant type) from SKN college of Agriculture (RAU), Jobner and Narendra Dhania 2 (dual purpose variety) from NDU&T, Kumarganj and a high yielding variety of fenugreek LFC-103 suitable for both irrigated and rainfed conditions from Horticulture Research Station, Dr. YSRHU, Guntur are the varieties recommended for release. Pepper hybrids PRS160 and PRS 161 developed through inter-varietal hybridization from Panniyur were found to be promising with maximum green berry yield of 4.2 kg vine⁻¹ and 4 kg vine⁻¹ respectively. Number of spikes vine⁻¹ was 560 and 472 in PRS 160 and PRS 161 respectively. Spike length was maximum in PRS 161 (20.1 cm). GCP 49 a ginger genotype showed highest yield 23.31 t ha⁻¹ followed by Karthika with a yield of 17.84 t ha⁻¹ in a CVT trial at Pundibari. NDH-98 of turmeric gave maximum rhizome yield (36.41 t ha⁻¹) followed by NDH-79 (35.74 t ha⁻¹), which were significantly superior over National Check Pratibha (21.45 t ha⁻¹) and local check Megha Turmeric-1 (22.81 t ha⁻¹) in a turmeric CVT trial at Pasighat. In a MLT of coriander at Coimbatore among 70 genotypes seed yield of genotypes varied from 325 to 656 kg ha⁻¹ and the genotype LCC-168 registered maximum seed yield (656 kg ha⁻¹) which was on par with DH 246, LCC 144, CS 66, ND 80 and ND 82. In a CVT of fenugreek at Navsari, FGK-74 (1358.02 kg ha⁻¹), FGK-67 (1345.68 kg ha⁻¹), FGK-68 (1246.91 kg ha⁻¹) and FGK-69 (1234.57 kg ha⁻¹) recorded higher seed yield. Number of pods per plant and number of seeds per pod were also high in all these entries. At the Coimbatore centre, FGK-43 recorded highest seed yield of 431.70 kg ha⁻¹ as compared to



Hisar Sonali and Rmt-362 (national check) (302.50 kg ha⁻¹ and 312.60 kg ha⁻¹ respectively).

Crop production

At Panniyur, in a trial to standardize drip fertigation in black pepper, maximum green berry yield (4.89 kg vine⁻¹) was recorded by application of 50% RDF with 8 l of water day⁻¹ vine⁻¹ through drip. In a fertigation trial in small cardamom at Mudigere, application of irrigation at 9 l clump⁻¹ day⁻¹ with 100% RDF through drip recorded the highest capsule yield (207.41 kg ha⁻¹) and this is on par with irrigation at 9 L clump⁻¹ day⁻¹ with 75% RDF (201.23 kg ha⁻¹). In source sink relationship trial in ginger, variety Mahima produced the highest fresh yield of 4.68 kg plot⁻¹ (9.43 t ha⁻¹) and highest dry yield of 1.21 kg plot⁻¹ at Pundibari. In turmeric, drip once in a day at 80% PE recorded highest rhizome yield (38.32 kg plot⁻¹) followed by drip once in 2 days at 80% PE treatment (37.21 kg plot⁻¹) at Kammarpally whereas in Pundibari, surface irrigation 5 cm at 0.90 IW/CPE recorded highest rhizome yield (11.40 kg plot⁻¹).

Crop protection

In a trial for biological management of slow decline in black pepper, least disease incidence (17.5 %) was observed by soil application of *Trichoderma harzianum* + Neem cake @ 2 kg vine⁻¹ and this was on par with the soil application of *Trichoderma harzianum* followed by soil drenching with *P. fluorescens* @ 2% (18.8% disease incidence). Adoption of phytosanitation and application of bioagents in large cardamom has resulted in controlling the incidence pests (shoot fly and leaf caterpillar) and diseases (blight, chirke and foorkey) in farmers field at Singhik, North Sikkim. In a ginger trial at Pundibari to test the efficiency of different fungicides including new molecules against leaf spot disease, it was found that foliar spray with Hexaconazole (0.1%) first at disease appearance and

then 2 times at 20 days interval registered lowest leaf spot disease severity (PDI 17.48) and highest yield of 4.13 kg plot⁻¹ (8.33 t ha⁻¹). A trial to test the efficacy of new generation fungicides for the management of coriander powdery mildew was laid out at Coimbatore. In this trial the incidence of powdery mildew was less (5.14 PDI) in propiconazole sprayed plants and these plants also recorded higher grain yield of 663.33 kg ha⁻¹ and was followed by Tebuconazole, Difenconazole (11.67 PDI), while in control the disease incidence was 91.55 PDI with grain yield of 556.11 kg ha⁻¹.

Production and distribution of quality planting material

- Produced and supplied about 20 t of pure seed material of high yielding high curcumin turmeric variety Roma in tribal areas of Andhra Pradesh and 100 t of Megha turmeric in Meghalaya for establishing areas of high quality turmeric for industrial use.
- 10 q each of cumin, coriander, fennel and fenugreek seed material was produced and distributed.

Success stories

Black pepper grafted on resistant root stock - *P. colubrinum*, an eco friendly way to manage *Phytophthora* foot rot, reducing excessive use of fungicides. This grafted pepper cultivation is already spread to about 80 ha in Uttara Kannada district in Karnataka.

Highly efficient single node portray technology in turmeric, was successfully demonstrated in over 20 acres in many farmers field and over 20 awareness and training programmes were conducted in Tamil Nadu, Andhra Pradesh, Maharashtra and Odisha.

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EDUCATION AND TRAINING

Trainings attended

Name of the official	Training programme	Date	Organization
Ms. S. Aarthi Ms. H.J. Akshitha Dr. R. Praveena Dr. Sharon Aravind	XXI days short term training course on Genomics and proteomics of plants and microbes towards translational research	21 Jan. to 10 Feb. 2015	ICAR-IISR, Kozhikode
Dr. Awadhesh Kumar	FOCARS training (Part I)	22 Nov. to 31 Dec. 2014	ICAR-NRCG, Pune
	FOCARS training (Part II)	1 Jan. to 18 Feb. 2015	ICAR-DMAPR, Anand
Dr. C.N. Biju	International training programme on biosecurity and incursion management	08-28 April 2014	NIPHM, Rajendranagar, Hyderabad
Dr. E. Jayashree Dr. R. Praveena	Training programme on ISO 22000:2005 - Food safety management systems	5 June 2014	IISR, Kozhikode
Dr. T.K. Jacob	Training of Dr. KS Krishnan school of chemical ecology	16-27 Feb. 2015	NCBS, Bengaluru
Mr. K. Jayarajan	Management development programme on cyber security	16-20 Feb. 2015	NIFM, Faridabad
Dr. Prativa Lakhotia	Professional attachment training	21 Nov. 2014 to 21 Feb. 2015	ICAR-IIHR, Bengaluru
Mr. M. Radhakrishnan	Public financial management & accountability	26-30 May 2014	ICISA, Noida
	Management development programme on analysis of financial statements	14-18 July 2014	NIFM, Faridabad
Dr. C. M. Senthil Kumar	Two weeks refresher course on Agricultural research management for directly recruited senior/principal scientists	14-26 July 2014	NAARM, Hyderabad
Mr. R.N. Subramanian	Effective office management and administrative, general financial rules and CCS(CCA) rules	23-27 Feb. 2015	NPC, Goa
Mr. V.C. Sunil	Special training programme for the employees of ICAR	24 Nov. to 05 Dec. 2014	ISTM, New Delhi
Ms. P. Umadevi	Training on Next generation sequencing (NGS)-bioinformatics and data analysis	15-19 July 2014	MIT, Anna University, Chennai

Training course on genomics and proteomics

Conducted DBT sponsored short term training course (STTC) on Genomics and proteomics in plants and microbes towards translational research during 21 January – 10 February, 2015. Eighteen trainees from ICAR institutions, SAUs and other Universities participated in the training programme (Fig 36). The course content was structured with basics to advanced genomics and proteomics techniques in

three modules. The training was implemented with five core guest faculties, five in-house experts, four faculties from corporate/ private companies and a visit to a next generation sequencing service facility at Cochin. Dr M Anandaraj, Director, IISR was the course Director, Dr D Prasath and Ms. P Umadevi were the course co-ordinators.



Fig 36. Participants and faculty of DBT sponsored 21 days training programme.

INSTITUTE TECHNOLOGY MANAGEMENT UNIT

During the year 2014-15, the ITM-BPD Unit issued four licenses to Rainbow Agri Life, Kadapa, Andhra Pradesh through NRDC for the commercialization of micronutrient mixtures for black pepper, ginger, turmeric and cardamom. M/S. Shrey Agritech, Hubli, Karnataka availed license for commercialization of micronutrient mixture for black pepper. A non exclusive license for commercializing *Trichoderma harzianum* was given to District Agricultural Farm, Thaliparamba. The license for turmeric varieties, IISR Pratibha and Alleppey Supreme were renewed.

The office and incubation facilities were leased to M/S. Natura Nursery and Agro Products for the production of micronutrient mixtures for ginger and turmeric. A contract research project was signed with Novozymes Ltd., Bangalore to test their chemical “Actinovate” against the nematodes of spice crops with the funding of Rs. 2.08 lakhs. During 2014-15 four consultancy visits were carried out to various plantations by the scientists to deliver technical advices regarding different aspects of crop production.

ITM-BPD unit has generated Rs. 3.36 lakhs through consultancy, licensing of bio agents, contract research and field visits. An amount of Rs. 9.5 lakhs was obtained through licensing of micronutrient technologies. Testing of manures or fertilizers or bioagents for quality assessment contributed Rs. 8.36 lakhs to the institute revenue. Hence, the total income generated through various activities is Rs. 21.30 lakhs.

The highlight of the BPD unit at IISR is the spice processing facility established at IISR Farm, Peruvannamuzhi. This unit is envisaged to promote entrepreneurship development and improvement of the competitiveness of the spice industry through scientific training, capacity building and implementation of ISO standards to spice processing. The centre is equipped with facilities for cleaning and grading of black pepper white pepper and curry powder production units. Successful trial run of the unit was conducted during 29 - 30 July 2014.

Three entrepreneurs were registered for utilizing the facility. The unit has got manufacturing license from Food Safety and Standards Authority of India (FSSAI). IISR has entered into a Memorandum of Understanding with Kerala Industrial and Technical Consultancy Organization Ltd. (KITCO) on 31 March, 2015 in an effort to jointly promote entrepreneurship development (Fig 37). IISR is planning to provide pre-incubation training, market potential studies etc. to the aspiring entrepreneurs through KITCO.

As a part of BPD activities, scientists from the institute participated and delivered lectures regarding the processing of spices and business incubation facility of IISR during the workshops organized by the District Industries Centre, Wayanad, Kerala State Small Industries Association etc. A class on “Value addition in spices at Business Planning and Development Unit in IISR” was given to 300 participants during National Agri Fiesta 2015 organized by Regional Agricultural Research Station (KAU), Ambalavayal. One news paper article on biocapsule and four articles on signing of MoU with KITCO were published in various news papers.

In an attempt to promote commercialization of technologies, ICAR-IISR has participated and exhibited the technologies in the Fourth Horticulture Institute-Industry Interface Meet held on 10 February 2015, organized by ICAR-Indian Institute of Horticultural Research, Bengaluru, Karnataka.



Fig 37. Signing of MoU between ICAR-IISR and KITCO

HINDI CELL ACTIVITIES

OLIC meeting

The Official Language Implementation Committee (OLIC) meets once in every quarter; first on 14 July 2014; second on 29 September 2014; third on 30 December 2014 and fourth on 30 March 2015 under the chairmanship of Dr. M. Anandaraj, Director and reviewed the official language implementation activities of the institute.

Workshops conducted

Hindi workshops were organized at ICAR-IISR, Kozhikode to popularize official language. First on noting and drafting in OL on 18 June 2014. Second on rules of OL and its implementation on 26 September 2014, third on Hindi translation and words pronunciation on 17 December 2014 and fourth on how to popularize official language in the office on 20 March 2015.

Hindi day and Hindi fortnight celebration

Hindi Day was celebrated on 15 September 2014 and Hindi fortnight from 15 - 29 September 2014. Hindi fortnight inauguration was held on 15 September 2014 under the presidentship of Dr. M. Anandaraj, Director (Fig 38). During this period various competitions viz., extempore speech, song, debate, noting and drafting, memory test, caption writing and anthakshari were conducted for the staff members and prizes were distributed to the winners in the valedictory function on 29 September 2014. Dr. Suneeta Yadav, Deputy Director, Regional Office for the official language implementation, Cochin was the chief guest. Institute official language magazine, *Masaloon ki Mehak* was released on this occasion.

TOLIC activity

- Dr. M. Anandaraj, Director; Dr. Rashid Pervez, Senior Scientist & Hindi Officer and Ms. N. Prasannakumari, Hindi translator attended the half yearly TOLIC meeting at Malabar Palace Kozhikode on 18 November 2014.
- Dr. Rashid Pervez, attended the editorial board

meeting of the TOLIC magazine *Malabar Jyoti* at SBT on 5 September and 3 November 2014.

- Dr. Rashid Pervez attended Hindi workshop conducted by TOLIC, Kozhikode on 29 April 2014.
- Dr. Rashid Pervez and Ms. N. Prasannakumari attended the half yearly and subcommittee TOLIC meeting on 13 May 2014; 27 May 2014; 11 July; 31 October and 29 December 2014.

OL implementation Inspection

Dr. Suneeta Yadav, Deputy Director (OL), Deputy Director, Regional Office for the official language implementation, Cochin inspected official language implementation activities in the institute on 29 September 2014.

Publications

- Annual report (2013-14), Page 100 + iv
- Anusandhan ke mukhya ansh (2013-14), Page 28 + iv
- Executive summary of annual report of the institute and AICRPS (Spices)
- Masala Samachar (4 issue)
- Masaloon ki Mehak (OL magazine), Page 79 + iv
- Bulletins (Kalimirch), Page 24 + iv
- 12 scientific popular hindi articles in various bulletins/journals.

OL Reports

Quarterly and annual reports on official language activities of the institute was prepared and sent to ICAR, New Delhi, TOLIC, Calicut and Regional Implementation Office, Cochin. The half yearly report on Official Language implementation have been prepared and submitted to Regional Implementation Office, Cochin.

Other activities

Translated various items viz., office orders, circulars, documentaries, rubber stamps, name boards, envelopes and web site into Hindi. Displayed daily a word/phrase in Hindi and its meaning in English.

Rajbhasha Award

The Institute was awarded Rajbhasha Shield Award 2014 among the 70 central government organizations of Kozhikode in the annual meeting of the TOLIC

at Malabar Palace Kozhikode on 18 November 2014. Dr. M. Anandaraj, Director received the Rajbhasha Shield Award 2014 from the chief guest of the function (Fig 39).



Fig 38. Release of Hindi magazine *Masoolon ki Mehak* and Hindi fortnight valedictory function



Fig 39. Dr. M Anandaraj, Director, receiving Rajbhasha Shield Award 2014

RECOGNITIONS

ISO 9001:2008 Certification

ICAR-Indian Institute of Spices Research has been certified for ISO 9001:2008 from 10 March 2015.



ICAR-Indian Institute of Spices Research won the second prize in the science and technology exhibition titled 'Swasraya Bharat-2014' held during 14-19 October 2014. The exhibition was organized by the Swadeshi Science Movement at College of Agriculture, Padannakkad, Kasaragod.

ICAR-IISR, Regional Station, Appangala, Karnataka won second best exhibition stall award under Government stall category, in Krishimela 2015 at ICAR-CPCRI, RS, Vittal, Karnataka on 10 January 2015.

PLACROSYM XXI Best Logo Award

Dr. Santhosh J. Eapen bagged the best logo award instituted by PLACROSYM XXI for the logo design.

Best Woman Scientist Award

Dr. Utpala Parthasarathy, Chief Technical officer awarded NABS- Best Woman Scientist for the year 2013.

National Innovation Foundation of India – Award

The National Innovation Foundation of India, an autonomous body under the Department of Science and Technology, Govt. of India has honoured ICAR-Indian Institute of Spices Research, Kozhikode with a Partnership Award for helping the foundation to achieve their objectives. Some of the spice farmers, Mr. P.G. George, Puliamakkal House, Zion, for pepper and Mr. Varkey Thomman, Punnathanam House for nutmeg and others, whose claims have been validated by ICAR-IISR were among national award winners. The award was given in the award function organized at Rashtrapathi Bhavan cultural centre on 7th March 2015 at 1030 hrs. Shri Pranab Mukherjee, the President of India inaugurated the award function and also the festival of innovation.



Distinguished Scientist Awards

Dr. M. Anandaraj, Director, ICAR-IISR and Dr. K. Nirmal Babu, Project Coordinator, AICRP on Spices have bagged Distinguished Scientist Awards instituted by Dr C.S. Venkataraman Memorial Trust for their outstanding contribution in the field of Plantation Crops Research and Development. The award was conferred during the inaugural function of International Symposium on Plantation Crops (PLACROSYM XXI) held at Kozhikode on 10 December 2014.

MAJOR EVENTS

ICAR-IISR hosts International Symposium on Plantation Crops

The XXI International Symposium on Plantation Crops (PLACROSYM XXI) was hosted by ICAR-Indian Institute of Spices Research, Kozhikode. The event, organized by 12 research and development organizations dealing with plantation crops in the country, was held during 10-12 December 2015 at Kozhikode. The symposium was inaugurated by Prof. M.S. Swaminathan, Emeritus Chairman, MSS Research Foundation, Chennai, and Dr. P. Rajendran, Vice Chancellor, Kerala Agricultural University presided over the function. Dr. N.K. Krishna Kumar, Deputy Director General (Hort. Science), ICAR, New Delhi delivered the keynote address. Dr. M. Anandaraj, Director, ICAR-Indian Institute of Spices Research, Kozhikode and President ISPC & General Chairman, PLACROSYM XXI welcomed the gathering while Dr. S. Devasahayam, General Convenor, proposed the vote of thanks.



Fig 40. Prof. M.S. Swaminathan, Emeritus Chairman, MSS Research Foundation, Chennai inaugurating the XXI International Symposium on Plantation Crops

Several publications viz., special issues of Indian Horticulture, Spice India and Journal of Arecanut, Spices and Medicinal Plants and a book on '*Phytophthora* Diseases of Plantation Crops' apart from Abstract of papers and Souvenir of PLACROSYM XXI were released to mark the occasion. Awards were presented to distinguished

scientists and farmers which included the following. Eleven lead talks, 26 oral papers and 220 poster papers were presented in six technical sessions. The symposium was attended by over 350 delegates from India and abroad.

67th Annual meeting of Indian Phytopathological Society and National symposium on Understanding host-pathogen interaction through science of omics

The 67th Annual meeting of Indian Phytopathological Society, New Delhi and National symposium on Understanding host-pathogen interaction through science of omics was held at ICAR-Indian Institute of Spices Research, Kozhikode during 16-17 March 2015. The event was inaugurated by renowned plant pathologist, Dr. Y.R. Sarma, FAO Consultant and former Director of ICAR-IISR. Dr. M. Anandaraj, Director, ICAR-IISR, and President of the Indian Phytopathological Society presided over the inaugural meeting. Dr. S. Devasahayam, Head Division of Crop Protection, Dr. Pratibha Sharma, Secretary, Indian Phytopathological Society and Dr. A.I. Bhat, organizing secretary also spoke on the occasion.



Fig 41. Dr. Y.R. Sarma, FAO Consultant, inaugurating the National symposium on Understanding host-pathogen interaction through science of omics

RESEARCH ADVISORY COMMITTEE

Name and address	Position
Dr. K.V. Peter Ex-Vice Chancellor, KAU, Thrissur & Director, World Noni Research Foundation, Chennai-600096	Chairman
Dr. M.N. Venugopal Door No.11, Block -3 Rangarao colony, Vasu Layout Ramakrishna Nagar, Mysore-22	Member
Dr. M. R. Sudharshan Ex-Director (Research) Spices Board, 222, 9 th Main Road Srinagara, Bengaluru- 560050	Member
Dr. K. K. Sharma National Coordinator, AINP on Pesticide Residues, IARI, LBS Building, New Delhi-110 012	Member
Sh. Philip Kuruvilla Chairman, World Spices Organisation, 8/1386, Palliyarkavu Road, Mattancherry, Kochi 682 002	Member
Dr. R. Viswanathan Professor & Head, Department of Post – Harvest Technology, TNAU, Coimbatore	Member
Dr. T. Janakiram, ADG (Hort-II), KAB-II ICAR, New Delhi-110012	<i>Ex-officio</i> Member
Dr. M. Anandaraj Director, ICAR-Indian Institute of Spices Research, Marikunnu PO, Kozhikode-673 012	<i>Ex-officio</i> Member
Dr. R. Dinesh Principal Scientist (Soil Science) ICAR-Indian Institute of Spices Research Kozhikode-673 012	Member Secretary

Recommendations of RAC

New germplasm collections may be for specific agronomic traits such as higher quality parameters, synchronous flowering in cardamom etc. The germplasm characterized so far may be published immediately.

Food safety and supply chain management of important spices may be taken up along with post harvest technology to enable development of code of practices for production of contaminant free spices.

The Regional station at Appangala may be strengthened to cater to the needs of the farming community in Karnataka region where cultivation of black pepper, ginger, turmeric and nutmeg are taken up in large scale.

While undertaking management trials for thrips in small cardamom, effect of the treatments on other insect pests and pollinators may be recorded. Data on efficacy and residues for newer molecules may be generated and shared with CIB for fixing MRL levels.

For optimizing the use of water, quantity of water required for producing a unit volume of spices may be worked out.

A technical bulletin on aflatoxin management in nutmeg and other spices may be published for disseminating the knowledge to the stake holders.



Fig 42. RAC meeting at IISR, Kozhikode

INSTITUTE MANAGEMENT COMMITTEE

Name and address		Position
Dr. M. Anandaraj	Director, ICAR-Indian Institute of Spices Research Marikunnu P.O, Kozhikode – 673 012	Chairman
Dr. R. Viswanathan	Head, Plant Protection ICAR-Sugarcane Breeding Institute Coimbatore – 641 007, Tamil Nadu	Member
Dr. V. Niral	Principal Scientist ICAR- Central Plantation Crops Research Institute Kudlu P.O, Kasaragod – 671 124	Member
Dr. K. Kandiannan	Principal Scientist ICAR-Indian Institute of Spices Research, Marikunnu P.O, Kozhikode – 673 012	Member
Dr. P.K. Asokan	Principal Scientist & Scientist – In-Charge Calicut Research centre of CMFRI West Hill P.O, Calicut - 673 005	Member
Assistant Director General (H)-II	Indian Council of Agricultural Research Krishi Anusandhan Bhavan -II Pusa, New Delhi – 110 001	Member
Mr. M. Radhakrishnan	Finance and Accounts Officer ICAR-IISR, Kozhikode	Member
Mr. K.V. Pillai	Administrative Officer ICAR-Indian Institute of Spices Research, Marikunnu P.O, Kozhikode – 673 012	Member Secretary



LIST OF PROJECTS

Mega Project I: Conservation, characterization and sustainable utilization of genetic resources of spices [Project leader: Dr. K.V. Saji]

1. Gen. XXVIII 813: Conservation and characterization of *Piper* germplasm (2008-2020) [Dr. K.V. Saji, Dr. B. Sasikumar and Ms. P. Umadevi]
2. Gen. XIX (813): Conservation, characterisation, evaluation and improvement of *Zingiber* and *Curcuma* sp. (2007-2015) [Dr. D. Prasath, Dr. B. Sasikumar and Dr. K.V. Saji]
3. Gen. XXXIII (813): Identification of core collection, characterization and maintenance of cardamom germplasm (2012-2017) [Dr. Sharon Aravind, Dr. S.J. Ankegowda and Dr. C.N. Biju]
4. DBT-CIB-5: Development of comprehensive SSR and SNP markers for the study of genetic diversity and association analysis in *Curcuma* (2012-2015) [Dr. T.E. Sheeja, Dr. D. Prasath and Dr. B. Sasikumar]

Mega Project II: Development of trait specific and improved varieties of spices through conventional breeding and biotechnological approaches (Project Leader: Dr. B. Sasikumar)

1. Gen. XXXI (813): Breeding black pepper for high yield, quality and resistance to stresses (2012-2017) [Dr. B. Sasikumar, Dr. Johnson K. George, Dr. K. V. Saji, Dr. T.E. Sheeja, Dr. T. John Zachariah, Dr. R. Suseela Bhai, Dr. K.S. Krishnamurthy, Dr. S. Devasahayam and Ms. S. Aarthi]
2. Gen. X (813): Breeding cardamom for high yield and disease resistance (2007-2015) [Dr. Sharon

Aravind, Dr. R. Praveena and Dr. C. M. Senthil Kumar]

3. Gen. XXVI (813): Evolving high yielding and high quality nutmeg clones by selection (2007-2016) [Dr. J. Rema, Dr. K.V. Saji and Dr. B. Sasikumar]
4. Gen. XXXIV (813): Induction of variability in ginger through induced mutation for yield and disease resistance (2012-2017) [Dr. D. Prasath, Dr. R. Ramakrishnan Nair and Dr. R. Suseela Bhai]
5. Gen. XXXII (813): Expression profiling and allele mining of genes induced under water-deficit stress in black pepper (*Piper nigrum* L.) (2012-2015) [Dr. Johnson K. George, Dr. K.S. Krishnamurthy and Ms. P. Umadevi]
6. Gen. XXXV (813): Genetic improvement in turmeric through seedling selection and hybridization (2013-2020) [Dr. R. Ramakrishnan Nair and Ms. S. Aarthi]
7. Biotech. XII (813): Mining of DNA markers and genes from expressed sequence tags of *Curcuma longa* (2012-2015) [Dr. T.E. Sheeja and Dr. B. Sasikumar]
8. Gen. XXX (813): Evaluation of genetic variability in vanilla with emphasis to disease tolerance (2010-2015) [Dr. R. Ramakrishnan Nair]

Mega Project III: Development of resource conservation and management technologies for improving productivity of spices (Project leader: Dr. K. Kandiannan)

1. Phy. X (813): Evaluation of black pepper and cardamom elite lines for yield and quality under moisture stress (2010-2015) [Dr. S.J.

Ankegowda, Dr. K.S. Krishnamurthy and Ms. H.J. Akshitha]

2. Phy. XI (813): Source sink relationship, endogenous hormone levels and their relationship with rhizome development in ginger and turmeric (2011-2016) [Dr. K.S. Krishnamurthy, Dr. K. Kandiannan and Dr. V. Srinivasan]
3. SSC VI (813): Nutrient cycling and soil C sequestering potential of spice crops under different management systems (2011-2015) [Dr. V. Srinivasan, Dr. R. Dinesh, Dr. S.J. Ankegowda and Dr. S. Hamza]
4. ICAR Mega Seed Project: Production of nucleus planting materials of improved varieties of spice crops (2006-2017) [Dr. K. Kandiannan, Dr. S.J. Ankegowda, Dr. J. Rema, Dr. K.V. Saji, Dr. D. Prasath and Dr. P. Rajeev]
5. DBT Twinning Programme for the NE: Seed system development in major spice crops (ginger, turmeric and Naga Chilli) of NER through *in vitro* techniques (2012-2015) [Dr. K. Nirmal Babu and Dr. K. Kandiannan]
6. ICAR-CPPHT-4: Micronutrient management in horticultural crops for enhancing yield and quality (2014-17) (Dr. R. Dinesh, Dr. V. Srinivasan and Dr. S. Hamza)

Mega Project IV: Development, refinement and demonstration of integrated cropping system for improved total factor productivity in spices (Project Leader: Dr. V. Srinivasan)

1. Kerala State – CPPHT-3: Integrated pepper research and development Project for North Kerala districts (2013-2016) [Dr. V. Srinivasan, Dr. P.S. Manoj, Dr. K.M. Prakash, Dr. K.K. Aiswariya, Dr. P. Rajeev, Dr. S. Hamza, Dr. R. Suseela Bhai, Dr. T.K. Jacob, Dr. A. Ishwara Bhat, Dr. Santhosh J. Eapen, Dr. Rashid Pervez, Dr. R. Dinesh, Dr. C.K. Thankamani,

Dr. K. Kandiannan, Dr. K.S. Krishnamurthy and Dr. K.V. Saji]

2. Hort. VII (813): Evaluation of nutmeg for its suitability for high density planting (2011-2016) [Dr. J. Rema and Dr. Sharon Aravind]

Mega Project V: Development, refinement and demonstration of organic production technology of spices for improved productivity, quality and soil health (Project leader: Dr. C.K. Thankamani)

1. ICAR-CPPHT-1: Network project on organic farming (2007-2017) [Dr. C.K. Thankamani, Dr. V. Srinivasan, Dr. T. John Zachariah and Dr. R. Praveena]
2. ICAR-CPPHT-2: Network on organic farming in horticulture crops (2014-17) (Dr. J. Rema, Dr. V. Srinivasan, Dr. K. Kandiannan, Dr. R. Dinesh, Dr. S.J. Ankegowda, Dr. C.N. Biju, Dr. C.M. Senthil Kumar and Mr. Narendra Chaudhary)

Mega Project VI: Development and refinement of post harvest handling, processing and value addition technologies for minimization of post harvest losses and diversified use of spices (Project leader: Dr. N.K. Leela)

1. PHT VII (813): Developing energy efficient processing technologies for spices (2013-2017) [Dr. E. Jayashree, Dr. N.K. Leela and Dr. Ankur Nagori (CIFT, Cochin)]
2. Org. Chem. IV (813): Chemoprofiling of *Myristica* species for nutraceutical and medicinal properties (2013-2018) [Dr. N.K. Leela and Dr. T. John Zachariah]
3. Biochem. VIII (813): Evaluation of spice extracts for anticancer effect in relation to telomerase activity (2012-2016) [Dr. N.K. Leela, Dr. T. John Zachariah and Dr. K. Sujathan (RCC, Thiruvananthapuram)]



4. DST-CPPHT-1: Development of mechanical unit for production of white pepper from green pepper (2012-2015) [Dr. E. Jayashree, Dr. R. Suseela Bhai, Dr. T. John Zachariah and Dr. Ravindra Naik (CIAE, Coimbatore)]
5. DoE-CPPHT-1: Developing electronic nose for monitoring cardamom aroma (2012-2015) [Dr. N.K. Leela and Dr. Nabarun Bhattacharya (C-DAC, Kolkata)]
6. ICAR-CPPHT-3: Network project on high value compounds and phyto-chemicals (2014-17) (Dr. T. John Zachariah, Dr. N.K. Leela, Dr. Santhosh J. Eapen and Dr. Awadhesh Kumar)

Mega Project VII: Bio-Intensive management of pests in spices (Project Leader: Dr. T.K. Jacob)

1. Ent. XIV (813): Survey and documentation of naturally occurring entomopathogens in spice cropping systems (2012-2015) [Dr. C.M. Senthil Kumar, Dr. T.K. Jacob and Dr. S. Devasahayam]
2. Nema. VI (813): Mass production and field evaluation of promising entomopathogenic nematodes against insect pests infesting major spices (2012-2016) [Dr. Rashid Pervez, Dr. Santhosh J. Eapen and Dr. S. Devasahayam]
3. Outreach Programme on Management of sucking pests in Horticultural Crops: (2009-2017) [Dr. T.K. Jacob, Dr. S. Devasahayam and Dr. C.M. Senthil Kumar]
4. ICAR-Consortium Research Platform (CRP) on borers in network mode.

Mega Project VIII: Integrated management of fungal and bacterial diseases of spices (Project leader: Dr. R. Suseela Bhai)

1. Crop. Prot. 1.5 (813): Integrated management of *Phytophthora* foot rot and slow decline diseases

- of black pepper (2008-2016) [Dr. R. Suseela Bhai, Dr. Santhosh J. Eapen and Dr. Rashid Pervez]
2. Path. XXI (813): Diversity of rhizome – root rot pathogens and their antagonists in cardamom (2010-2015) [Dr. R. Praveena and Dr. C.N. Biju]
3. Path XXII (813): Investigations on the endophytic and rhizospheric microflora associated with cardamom and allied genera (2012-2015) [Dr. C.N. Biju and Dr. R. Praveena]
4. Outreach Programme on *Phytophthora*, *Fusarium* & *Ralstonia* Diseases of Horticultural and Field Crops (2008-2017) [Dr. M. Anandaraj, Dr. R. Suseela Bhai, Dr. Santhosh J. Eapen, Dr. K. Nirmal Babu, Dr. Johnson K. George, Dr. D. Prasath, Dr. R. Praveena and Ms. P. Umadevi]
5. DBT-CP6: Genome mining of spice associated endophytic bacteria for natural products (2011-2015) [Dr. Santhosh J. Eapen and Dr. R. Suseela Bhai]
6. Outreach programme on diagnosis and management of leaf spot diseases in field and horticultural crops (2009-2017) [Dr. C.N. Biju and Dr. R. Praveena]
7. Kerala State –CP-1. Area wide integrated pest management for wilt diseases in black pepper (2014-2017) [Dr. R. Suseela Bhai, Dr. Santhosh J. Eapen, Dr. Rashid Pervez and Dr. K.K. Aiswariya]

Mega Project IX: Development of diagnostic kits and integrated management viral diseases of spices (Project Leader: Dr. A. Ishwara Bhat)

1. Path XX (813): Screening of *Piper* germplasm accessions against *Piper Yellow Mottle Virus* (PYMoV) (2008-2015) [Dr. A. Ishwara Bhat, Dr. T.K. Jacob, Dr. K.V. Saji, Dr. K.S. Krishnamurthy and Ms. P. Umadevi]

2. DBT-CP5: Testing transgenic black pepper for resistance to viruses (2011-2014) [Dr. A. Ishwara Bhat and Dr. D. Prasath]

J. Eapen, Dr. S.J. Ankegowda, Dr. Rashid Pervez, Dr. K.S. Krishnamurthy, Dr. P. Rajeev, Dr. C.N. Biju and Dr. S. Hamza]

Mega Project X: Improving knowledge and skill of stakeholders for increasing production of spices (Project Leader: Dr. P. Rajeev)

1. Ext. VI (813): Spicepedia – A knowledge base for spices (2013-2015) (Dr. P. Rajeev and Mr. K. Jayarajan)

2. Kerala State – CPPHT-2: Pepper Rehabilitation Package – Technology Mission on Black pepper for Wayanad – SUGANDHI (2010-2015) [Dr. V. Srinivasan, Dr. T.K. Jacob, Dr. R. Suseela Bhai, Dr. R. Dinesh, Dr. C.K. Thankamani, Dr. K. Kandiannan, Dr. A. Ishwara Bhat, Dr. Santhosh

3. DBT-SS1: Distributed Information Sub-Centre (2000-2017) [Dr. Santhosh J. Eapen]

4. Capacity building and front-line intervention programmes for spice sector development in NE states and tribal empowerment (2014-17) (Dr. P. Rajeev and Dr. Lijo Thomas)

5. Economic analysis technology, market dynamics and policy scenario in major spice crops (2014-19) (Dr. Lijo Thomas and Dr. P. Rajeev)

6. Network project on Economic Impact studies on crop diversification and technology adoption in Horticulture (2014-17) (Dr. P. Rajeev and Dr. Lijo Thomas)

NEW NETWORK PROJECTS

ICAR has approved two new network projects *viz.*, High Value Compounds and Organic Horticulture in XII plan with ICAR-IISR as the Nodal Institute. The projects started functioning by November 2014.

The total budget of High Value Compounds and Phytochemicals is Rs. 2560 lakhs for the XII plan period with nine ICAR partner institutes. Prediction and validation of nutraceuticals and functional properties of phytochemicals and high value compounds identified from selected crop plants, developing knowledge base, *in silico*, *in vitro* and

in vivo validations, and developing formulations are the major objectives of the project.

The broad objectives identified for Network Project on Organic Farming in Horticulture Crops are evaluation of suitable organic amendments for meeting the nutrient requirement and pest and disease management, developing an organic package for different horticulture crops. This network includes nine ICAR research institutes with a budget allocation of Rs. 300 lakhs for the XII plan period.



RESULTS - FRAMEWORK DOCUMENT (RFD) (2013-2014)

Section 1: Vision, Mission, Objectives and Functions

VISION

Enhancing productivity of spices for meeting growing domestic demand and to be the global leader in spices export

MISSION

Utilize the scientific, technological and traditional strengths for sustainable spice production

OBJECTIVES

1. Production management, value addition and transfer of technology in spices
2. Conservation of genetic resources for sustainable use

FUNCTIONS

To attend to the research and development of high yielding and quality varieties and sustainable production, protection and post harvest technologies, training and dissemination of developed technologies to the stakeholders for increasing the production and productivity of spices.

Section 2: Inter se priorities among key objectives, success indicators and targets

S. No.	Objective (s)	Weight	Action(s)	Success Indicator (s)	Unit	Weight	Target / Criteria Value				
							Excellent 100%	Very Good 90%	Good 80%	Fair 70%	Poor 60%
1.	Production management, value addition and transfer of technology in spices	59	Optimization of horticultural/ INM/ IPM technology management and development of value added products of spices	Technologies developed/evaluated on INM/IPM/IDM	Number	15.0	6	5	4	3	2
				Diagnosics/value added products developed/identified	Number	10.0	4	3	2	1	-
				Nucleus planting materials produced	Number ('000s)	10.0	120	110	100	90	80
			Dissemination/commercialization of technologies	Nucleus seed rhizomes produced	('000 kg)	5.0	7	6	5	4	3
				Trainings conducted (farmers/ agr. officers and others)	Number	9.0	17	15	12	10	8
							13	12	10	8	6
Partnership development including licensing of technologies	Number	5.0	6	5	4	3	2				

2.	Conservation of genetic resources for sustainable use	30	Collection, conservation and cataloguing of spices germplasm and characterization for useful agronomic traits	Germplasm accessions collected, conserved and catalogued	Number	20.0	160	150	130	110	90
				Accessions evaluated for specific agronomic traits	Number	10.0	120	110	100	90	80
	Efficient functioning of the RFD system	3	Timely submission of draft RFD (2013-14) for approval Timely submission of results for RFD (2012-13)	On-time submission	Date	2.0	15/05/2013	16/05/2013	17/05/2013	20/05/2013	21/05/2013
On-time submission				Date	1.0	01/05/2013	02/05/2013	05/05/2013	06/05/2013	07/05/2013	
	Administrative reforms	4	Implement ISO 9001 as per the approved action plan Prepare an action plan for Innovation	% Implementation	%	2.0	100	95	90	85	80
				On-time submission	Date	2.0	30/07/2013	10/08/2013	20/08/2013	30/08/2013	10/09/2013
	Improving internal efficiency /responsiveness / service delivery of Ministry / Department	4	Implementation of Sevottam	Independent audit implementation of Citizen's Charter	%	2.0	100	95	90	85	80
				Independent audit implementation of public grievance redressal system	%	2.0	100	95	90	85	80

Section 3: Trend values of the success indicators

S. No.	Objectives	Actions	Success indicators	Unit	Actual value for FY 11/12	Actual value for FY 12/13	Target value for FY 13/14	Projected value for FY 14/15	Projected value for FY 15/16
1.	Production management, value addition and transfer of technology in spices	Optimization of horticultural/ INM/ IPM technology management and development of value added products of spices	Technologies developed/ evaluated on INM/IPM/IDM	Number	5	5	5	6	6
			Diagnostics/ value added products developed/ identified	Number	2	3	3	4	5
		Production of breeder seed/ planting materials	Nucleus planting materials produced	Number ('000s)	128	80	110	120	140
			Nucleus seed rhizomes produced	('000 kg)	7	5	6	7	8
2.	Conservation of genetic resources for sustainable use	Dissemination/ commercialization of technologies	Demonstrations / exhibitions conducted	Number	18	15	15	18	20
			Trainings conducted (farmers/ agrl. officers and others)	Number	12	15	12	15	18
			Partnership development including licensing of technologies	Number	3	3	5	5	6
		Collection, conservation and cataloguing of spices germplasm and characterization for useful agronomic traits	Germplasm accessions collected, conserved and catalogued	Number	236	157	150	165	180
			Accessions evaluated for specific agronomic traits	Number	100	100	110	115	120
			One-time submission	Date	-	-	16/05/2013	-	-
Efficient Functioning of the RFD System	Administrative reforms	One-time submission	Date	-	-	02/05/2013	-	-	
		% Implementation	%	-	-	95	-	-	
		On-time submission	Date	-	-	10/08/2013	-	-	
		Independent audit of implementation of Citizen's Charter	%	-	-	95	-	-	
Improving internal efficiency /responsiveness / service delivery of Ministry / Department	Implementation of Sevottam	Independent audit of implementation of public grievance redressal system	%	-	-	95	-	-	

Acronyms

S. No.	Acronym	Description
1.	INM	Integrated Nutrient Management
2.	IPM	Integrated Pest Management
3.	IDM	Integrated Disease Management
4.	NEH	North Eastern Hill Region
5.	NGOs	Non Governmental Organizations
6.	IISR	Indian Institute of Spices Research

Section 4:

Description and definition of success indicators and proposed measurement methodology

S. No.	Success indicator	Description	Definition	Measurement	General Comments
1.	Technologies developed / evaluated on INM/IPM/IDM	Integrated nutrient/ pest/ disease management is practiced encompassing conjunctive use of both chemical and organic nutrient/ bioagent/ botanical sources for improving environmental health & sustaining higher productivity	Integrated nutrient/ pest/ disease management refers to the maintenance of soil / plant/ ecosystem health at an optimum level and control the pest/disease incidence for sustaining the desired productivity through optimization of the benefits from all possible sources of organic, inorganic and biological components in an integrated manner	Developing integrated nutrient, pest and disease management technologies for different spice crops and cropping systems	To ensure balance fertilization, control of biotic stresses and sound soil/ plant/ environmental health
2.	Diagnostics/ value added products developed/ identified	The development of diagnostic kits would involve delineation of process (processes) for detection of specific pest/ diseases. Value addition involves identification/ development of new products from the raw agro-produce. These would involve specific number for field testing / validation through various institutes, State Departments, NGOs, private production houses/ industry	To develop sensitive tests for detection of causative agents for specific pest/ diseases of spices and identification of different products from agro-produce	Number	Development of new diagnostics will be needed for disease surveillance that are likely to cause high economic loss. The value added products will diversify the use there by increasing the profit.
3.	Nucleus planting materials produced	Production of nucleus planting material of black pepper and nutmeg, vegetatively propagated for producing quality materials for distribution to extension agencies/ farmers	It is a process of vegetative means by which new individuals arise without production of seeds or spores	Numbers produced (in thousands)	In a wider sense, planting material arise from vegetative propagation include cutting, budding, grafting and tissue culture

4.	Nucleus seed rhizomes produced	Production of nucleus seed rhizome material of ginger and turmeric, vegetatively propagated for producing quality materials for distribution to extension agencies/ farmers	It is a process of vegetative means by which new individuals arise without production of seeds	Quantity produced (in tonnes)	In a wider sense, planting material arise from vegetative propagation include cutting, budding, grafting and tissue culture
5.	Demonstrations / exhibitions conducted	Trials and demonstrations conducted for technology testing and proving the technology potential and the knowledge and skills of primary and secondary stakeholders shall be enhanced by organizing exposure visits to on-farm trials/ demonstrations/ exhibitions	On-farm trials aims at testing new technologies under farmers condition and management, by using farmers own practice as control. Frontline demonstration is the field demonstration conducted on farmers field under the close supervision of scientists	Number	
6.	Trainings conducted (farmers/ agrl. officers and others)	Capacity building activities related to knowledge and skill improvement/ development programmes conducted for farmers, rural youth and extension personnel	Training is a process of acquisition of new skills, attitude and knowledge in the context of preparing for entry into a vocation or improving productivity in an organization or enterprise	Number	
7.	Partnership development including licensing of technologies	With respect to commercialization of technologies and services for promoting partnerships with both public and private sector agencies, it is envisaged to bring commercial ethos in agricultural research system. The increasing numbers of partnerships over the years points towards emphasis on transfer of knowledge, skills and technologies, thereby contributing to improved socioeconomic impact from contribution of IISR	Partnership development, includes licensing of IISR's technologies and/or services	Number	
8.	Germplasm accessions collected, conserved and catalogued	Diverse germplasm is the basic requirement to bred new improved varieties	Basic genetic resource for crop improvement	Number of germplasm accessions	Cataloguing is done for morphological and yield attributes
9.	Accessions evaluated for specific agronomic traits	Promising source material for the improved varieties to be evaluated	Material generated from the basic germplasm	Number of promising/ breeding lines evaluated	Evaluation is done for potential agronomic (yield attributes), quality or stress (biotic/ abiotic) tolerance

Section 5: Specific performance requirements from other departments

Location Type	State	Organisation Type	Organisation Name	Relevant Success Indicator	What is your requirement from this organisation	Justification for this requirement	Please quantify your requirement from this organisation	What happens if your requirement is not met.
State Governments	Kerala, NEH	Departments	Forest Department	Germplasm accessions collected, conserved and catalogued	Permission to survey/ collection	Without permission it is illegal to enter the reserved forest for collection	Number of permission letters issued	Less or more numbers of germplasm of spices will be collected

Section 6: Outcome / impact of activities of the organization

S. No	Outcome / Impact of organisation	Jointly responsible for influencing this outcome / impact with the following organisation (s) / departments/ministry(ies)	Success Indicator (s)	Unit	2011-2012	2012-2013	2013-2014	2014-15	2015-16
1.	Production of quality seed and planting materials of improved varieties and processing technologies of spices crops	Ministry of Agriculture, Ministry of Commerce, Ministry of Environment & Forests, Ministry of Rural Development and State Governments, NGOs and Private partners	Increase in spice crops productivity Enhancing the quality of turmeric (curcumin content)	% %	4.0 3.0	4.1 3.25	4.15 3.5	4.20 4.0	4.25 4.25
2.	Commercialization of technologies	State Departments, NGOs and Private partners/ entrepreneurs	Research converted in to commercialized technologies	Number	3	3	4	5	6

Annual (April 1, 2013 to March 31, 2014) Performance Evaluation Report in respect of RFD 2013-2014 of RSCs i.e. Institutes

Name of the Division: Horticultural Science
Name of the Institution: ICAR-Indian Institute of Spices Research, Kozhikode, Kerala
Name of RFD Nodal Officer: Dr. V. Srinivasan

S. No.	Objective (s)	Weight	Action(s)	Success Indicator (s)	Unit	Weight	Target / Criteria Value					Achievements	Performance	
							Excellent 100%	Very Good 90%	Good 80%	Fair 70%	Poor 60%		Raw Score	Weighted Score
1.	Production management, value addition and transfer of technology in spices	59	Optimization of horticultural/ INM/ IPM technology management and development of value added products of spices	Technologies developed/evaluated on INM/IPM/ IDM	Number	15.0	6	5	4	3	2	7	100	15.0
				Diagnostics/value added products developed/ identified	Number	10.0	4	3	2	1	-	4	100	10
			Production of breeder seed/ planting materials	Nucleus planting materials produced	Number ('000s)	10.0	120	110	100	90	80	118	98	9.8
				Nucleus seed rhizomes produced	('000 kg)	5.0	7	6	5	4	3	6	90	4.5
			Dissemination/ commercialization of technologies	Demonstrations/ exhibitions conducted	Number	9.0	17	15	12	10	8	26	100	9.0
				Trainings conducted (farmers/ agrl. officers and others)	Number	5.0	13	12	10	8	6	17	100	5.0
				Partnership development including licensing of technologies	Number	5.0	6	5	4	3	2	6	100	5.0

2.	Conservation of genetic resources for sustainable use	30	Collection, conservation and cataloguing of spices germplasm and characterization for useful agronomic traits	Germplasm accessions collected, conserved and catalogued	Number	20.0	160	150	130	110	90	157	97	19.4
				Accessions evaluated for specific agronomic traits	Number	10.0	120	110	100	90	80	137	100	10.0
	Efficient functioning of the RFD system	3	Timely submission of draft RFD (2013-14) for approval	On-time submission	Date	2.0	15/05/2013	16/05/2013	17/05/2013	20/05/2013	21/05/2013	14/05/2013	100	2.0
	Administrative reforms	4	Timely submission of results for RFD (2012-13)	On-time submission	Date	1.0	01/05/2013	02/05/2013	05/05/2013	06/05/2013	07/05/2013	01/05/2013	100	1.0
	Improving internal efficiency /responsiveness / service delivery of Ministry / Department	4	Implement ISO 9001 as per the approved action plan	% Implementation	%	2.0	100	95	90	85	80	-	0.0	-
			Prepare an action plan for Innovation	On-time submission	Date	2.0	30/07/2013	10/08/2013	20/08/2013	30/08/2013	10/09/2013	30/07/2013	100	2.0
			Implementation of Sevottam	Independent audit implementation of Citizen's Charter	%	2.0	100	95	90	85	80	100	100	2.0
				Independent audit implementation of grievance redressal system	%	2.0	100	95	90	85	80	100	100	2.0

Total Composite Score: 96.70

Rating: Excellent

Procedure for computing the Weighted and Composite Score

1. Weighted Score of a Success Indicator = Weight of the corresponding Success Indicator x Raw Score / 100
2. Total Composite Score = Sum of Weighted Scores of all the Success Indicators

PERSONNEL

HEADQUARTERS

Scientific

Name	Designation
Dr. M. Anandaraj	Director
Dr. K. Nirmal Babu	Project coordinator (Spices)
Dr. S. Devasahayam	Head, Crop Protection Division
Dr. T. John Zachariah	Head, Crop Production & PHT Division
Dr. B. Sasikumar	Head CI & BT Division w.e.f. 04.11.2014
Dr. T.K. Jacob	Principal Scientist (Entomology)
Dr. J. Rema	Principal Scientist (Horticulture)
Dr. Johnson K. George	Principal Scientist (Gen. & Cytogenetics)
Dr. C.K. Thankamani	Principal Scientist (Agronomy)
Dr. R. Dinesh	Principal Scientist (Soil Science)
Dr. R. Suseela Bhai	Principal Scientist (Plant Pathology)
Dr. A. Ishwara Bhat	Principal Scientist (Plant Pathology)
Dr. R. Ramakrishnan Nair	Principal Scientist (Gen. & Cytogenetics)
Dr. K.S. Krishnamurthy	Principal Scientist (Plant Physiology)
Dr. K. Kandiannan	Principal Scientist (Agronomy)
Dr. N.K. Leela	Principal Scientist (Org. Chemistry)
Dr. Santhosh J. Eapen	Principal Scientist (Nematology)
Dr. K.V. Saji	Principal Scientist (Economic Botany)
Dr. P. Rajeev	Principal Scientist (Agril. Extension)
Dr. V. Srinivasan	Principal Scientist (Soil Science)
Dr. T.E. Sheeja	Senior Scientist (Biotechnology)
Dr. Rashid Pervez	Senior Scientist (Nematology)
Dr. D. Prasath	Senior Scientist (Horticulture)
Dr. E. Jayashree	Senior Scientist (AS & PE)
Dr. C.M. Senthilkumar	Senior Scientist (Entomology)
Ms. P. Uma Devi	Scientist (Biotechnology)
Dr. Lijo Thomas	Scientist (Agri. Economics) w.e.f. 04.06.2014
Dr. R. Praveena	Scientist (Plant Pathology)
Ms. Aarthi S	Scientist (Spices Plantation Medicinal & Aromatic Plants) w.e.f. 08.04.2014



Ms. Akshitha H J	Scientist (Spices Plantation Medicinal & Aromatic Plants) w.e.f. 08.04.2014
Dr. Awadesh Kumar	Scientist (Plant Biochemistry) w.e.f. 20.10.2014
Dr. Prativa Lakhotia	Scientist (Spices Plantation Medicinal & Aromatic Plants) w.e.f. 20.10.2014

Technical Officers

Dr. Hamza Srambikkal	Chief Technical Officer (Lab) (T9)
Dr. Utpala Parthasarathy	Chief Technical Officer (T9)
Mr. K. Jayarajan	Sr. Technical Officer (Stat.) (T6)
Dr. C.K. Sushama Devi	Sr. Technical Officer (T6) (Lib.)
Ms. N. Prasannakumari	Sr. Technical Officer (T6) (Hindi Translator)
Mr. K.T. Muhammed	Technical Officer (T5) (Farm)
Mr. A. Sudhakaran	Technical Officer (T5) (Artist-cum-Photographer)
Mr. N.A. Madhavan	Technical Officer (T5)
Mr. K. Krishnadas	Technical Officer (T5)
Mrs. P.K. Chandravalli	Technical Officer (T5)

Administrative

Mr. K V Pillai	Administrative Officer
Mr. M Radhakrishnan	Finance & Accounts Officer
Ms. P.V. Sali	Private Secretary
Mr. K.G. Jegadeesan	Astt. Finance & Accounts Officer
Mr. R.N. Subramanian	Astt. Administrative Officer
Mr. P Sundaran	Astt. Administrative Officer w.e.f. 22.12.2014

IISR EXPERIMENTAL FARM, PERUVANNAMUZH

Technical Officers

Mr. V.K. Aboobacker Koya	Chief Technical Officer (T9)
Mrs. E. Radha	Asst. Chief Technical Officer (T 7-8)
Mr. E S Sujeesh	Sr. Technical Officer (T6) w.e.f. 05.06.2014
Mr. K. Kumaran	Technical Officer (T5)

KRISHI VIGYAN KENDRA**Subject Matter Specialist**

Dr. P.S. Manoj	Subject Matter Specialist (T9) (Horticulture)
Dr. S. Shanmugavel	Subject Matter Specialist (T9) (Veterinary Science)
Mr. K.M. Prakash	Subject Matter Specialist (T9) (Agronomy)
Dr. B. Pradeep	Subject Matter Specialist T6 (Fisheries)
Ms. A. Deepthi	Subject Matter Specialist T6 (Home Science)
Mrs. K K Aiswariya	Subject Matter Specialist T6 (Plant Protection)

IISR REGIONAL STATION, APPANGALA, KARNATAKA**Scientific**

Dr. S.J. Ankegowda	Principal Scientist (Plant Physiology)
Dr. C.N. Biju	Scientist (Plant Pathology)
Dr. Sharon Aravind	Scientist (Spices Plantation & Aromatic Plants) w.e.f. 22.04.2014
Mr. Narendra Chaudhary	Scientist (Spices Plantation & Aromatic Plants) w.e.f. 13.10.2014
Ms. Rajna S	Scientist (Entomology) w.e.f. 26.12.2014

Technical Officer

Mr. K. Ananda	Technical Officer (T5)
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Administrative

Mr. P. Muraleedharan	Asst. Administrative Officer
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WEATHER DATA

Month	IISR Main Campus, Kozhikode, Kerala			
	Rainfall (mm)	Rainy days	T max (°C)	T min (°C)
January	15.2	1	32.2	22.0
February	0.0	0	32.7	22.9
March	0.0	0	33.9	24.2
April	27.8	2	34.4	25.5
May	201.6	7	33.3	25.1
June	486.2	19	31.4	24.2
July	856.0	26	29.1	23.3
August	669.1	24	29.4	23.3
September	304.3	14	30.8	23.7
October	222.2	17	31.4	23.8
November	101.8	10	32.0	23.2
December	4.1	0	31.8	22.9
Total/mean	2888.3	120	31.87	23.68

Month	Experimental Farm, Peruvannamuzhi, Kozhikode, Kerala			
	Rainfall (mm)	Rainy days	T max (°C)	T min (°C)
January	0.0	0	34.17	21.12
February	16.2	2	34.23	21.98
March	9.0	1	35.61	22.80
April	259.8	8	35.10	24.50
May	377.0	14	33.03	24.50
June	845.6	18	30.96	24.28
July	1495.9	29	27.90	19.18
August	996.6	27	28.96	21.46
September	571.0	23	31.03	21.48
October	727.4	20	31.46	22.85
November	109.4	7	33.01	22.58
December	70.2	6	33.22	22.48
Total/mean	5478.1	155	32.39	22.43

Month	IISR Regional Station, Appangala, Karnataka			
	Rainfall (mm)	Rainy days	T max (°C)	T min (°C)
January	0.0	0	31.19	13.34
February	0.0	0	31.80	14.80
March	11.2	1	33.06	16.24
April	113.4	8	33.13	19.10
May	184.3	10	31.00	19.10
June	240.3	19	27.37	19.00
July	1074.9	31	24.10	18.60
August	682.8	29	24.60	18.30
September	355.9	25	25.60	18.20
October	90.6	14	28.80	18.70
November	9.1	2	28.40	15.80
December	15.5	5	27.90	15.90
Total/mean	2778	144	28.91	17.26

INTENSIFYING BLACK PEPPER PRODUCTION

Traditionally pepper is trailed on live support trees in India, whereas, in few other pepper producing countries non-living support, mainly wooden poles, are also used. A new and novel idea of providing support with rooting media was conceptualized and six months old rooted top shoots with one or two lateral branches were planted during April 2014. The vertical column (3 m height, 50 cm width) was made with a plastic coated welded wire mesh filled with composted pasteurised cocopeat and powdered dry cow dung @ 3:1 ratio. The column was irrigated regularly with drip system; nutrient was applied in liquid form through media and foliar application. As and when vines put-forth new node, it was firmly fixed along the rooting medium filled in the vertical column and this facilitated in converting the clinging root to absorbing root, which in turn accelerated the growth of pepper. In this way the pepper vine covered the entire column within 10 months time and started producing spike in the same year.





हर कदम, हर दमर
किसानों का हमसफर
भारतीय कृषि अनुसंधान परिषद

Agri search with a human touch



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