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ICAR

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HIGHLIGHTS
अनुसंधान के मुख्य अंश

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MILLETS
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The doodle in the central circle is a metaphorical representation of the emergence of spices as one of the key identities of India across the globe. Spices are depicted as blending seamlessly with important symbols and institutions unique to India.
Doodle Artist: Abhinav

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PREFACE

I am privileged to present the Research Highlights- 2022 of the ICAR-Indian Institute of Spices Research, Kozhikode, Kerala. For the third time, the institute was awarded the "Sardar Patel Outstanding ICAR Institution Award" during 2022. The award, built on painstaking efforts of each and every personnel of the institute, reflects our committed approach to science and our ability to make meaningful change in the lives of key stakeholders.

During the year, we have established a dedicated facility named "Gingerarium" for conserving ginger accessions and related species.The drought tolerant cardamom genotype IC 349537 was recommended for release as a new variety, IISR Manushree. The research on developing recommendations for organic production systems and fertigation requirements for spices has led to customized schedules for crop management. The crop specific micronutrient technologies developed by the institute has found wide acceptance across the spice growing tracts of the country.

This year, the institute received the patent for its micronutrient composition for cardamom (Patent No: 413017). A Decision Support System was developed for Site Specific Nutrient Recommendation (IISR eSOFT) is expected to enhance the adoption of soil test based nutrient application in spice crops. The inking of agreement with Lysterra LLC, a Russian company, for commercialization of the novel microbial encapsulation technology was a watershed moment in the institute's history. A significant development has been the establishment of an Incubation Facility for Microbial Encapsulation (iFAME) to fulfil the long-awaited demand from aspiring start-ups and firms who are willing to license the technology.

During the year, 33 customised training programmes were conducted for the state departments and other organizations. Apart from this, 26 outreach initiatives targeting the SC/ST beneficiaries were also organized. Entrepreneurship incubation services and handholding support offered to commercial business ventures has helped in generating employment and creating confidence in agripreneurship. As a research institute, we remain committed to generation of new knowledge and technologies. The strong foundations built on diligent basic research work and the existing repository of scientific knowledge need to be shepherded to deliver translational research outcomes. This will significantly impact our support to primary production, processing, value addition and in generation of technologies for secondary agriculture.

I am indebted to Dr. Himanshu Pathak, Director General, ICAR and Secretary, DARE for the trust and confidence bestowed on the institute. I am thankful for the guidance received from Dr. T Mohapatra, Former Director General, ICAR in all the endeavours and also immensely thankful to Dr. A K Singh, DDG (Horticultural Science), Dr. Sudhakar Pandey, ADG (Hort I) and Dr. Vikramaditya Pandey, Former ADG (Hort I) for their persistent support and motivation. Special thanks to the editors for bringing out this report.

March 2023
Kozhikode

R Dinesh
Director

CONTENTS

BLACK PEPPER

Genetic resources

Three thousand four hundred and sixty six accessions are being maintained at the black pepper germplasm nursery at ICAR-IISR with new additions from Dibrugarh, Tinsukia and North Lakhimpur districts of Assam (Fig. 1). In the ex-situ gene bank at CHES, Chettalli, Karnataka forty two black pepper germplasm accessions are being maintained.Seedling progenies were established from *Piper barb*eri collected from Anakulam forests (Fig.2). *Piper ornatum* – an exotic ornamental species was collected and added to the Piper germplasm.

Characterization

Forty-two accessions maintained in ex-situ gene bank at CHES, Chettalli were characterized for 10 economically important quantitative traits. Highest coefficient of variation was recorded for fresh yield per vine and lowest for berry size.

Weighted parameter index for identifying promising genotype

Weighted parameters for yield contributing traits to identify promising genotypes were developed with maximum weightage given to yield followed by bulk density, spike length and number of berries. Other traits included in the index are spikes per lateral branch, spike setting, berry size, pericarp thickness, dry recovery, early maturity type, Phytophthora resistance, nematode resistance, drought tolerance, anthracnose resistance, high piperine and high oleoresin.

Participatory plant breeding

Two promising genotypes with long spike and bold berries were identified from Sirsi (Fig.3). Quality analysis of a few farmer's collections revealed highest essential oil (4.3 %) and oleoresin (8.6 %) in Huchmensu and high piperine (5.5 %) in Kuthiravally.

Fig. 1: *Piper lonchites male plant from Assam*

Fig. 2: *Piper barberi with ripened fruits*

Fig.3: a. Promising line with long spike (Master Kere); b. Boldberries (KK king); c. morphological marker on berries (Kudurebala).

Validation of gene specific primers for expression studies related to candidate genes of piperine biosynthesis

BAHD acyltransferase (piperine synthase) is the most important downstream enzyme that catalyzes the terminal formation of piperine from piperidine and piperoyl-CoA. The unigene of BAHD acyltransferase was retrieved from NCBI and Uniprot database and local BLAST with *P. nigrum* chromosome-scale reference genome was done. 63 longest ORF sequences were retrieved and 6 ORF sequences were shortlisted based on the signature motif of BAHD acyltransferase (HXXXD, DFGWG) (Fig. 4).

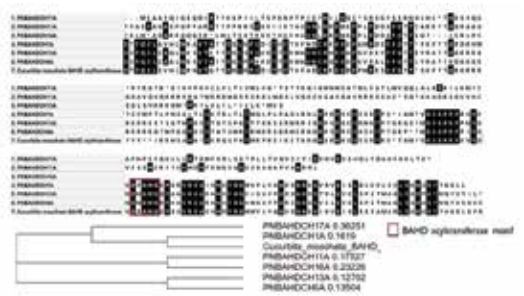

Fig. 4: Multiple sequence alignment & Phylogenetic tree of BAHD isoforms

cDNA library of *P.nigrum* leaf, stem and 4 stages of berry were constructed for the evaluation of candidate genes and for combined co expression analysis involving whole genome and transcriptome data. The result showed that BAHD-AT gene from Chromosome six showed a relative expression (Fig.5) correlating with piperine content.

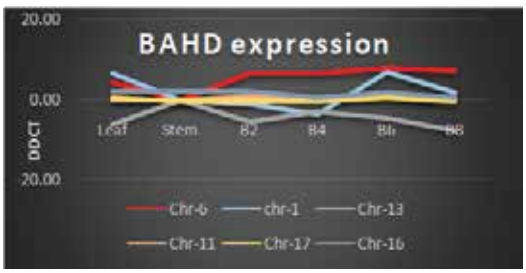

Fig.5: Differential expression profiles of the 6 BAHD isoforms

In-silico analysis of BAHD-AT genes

The six BAHD-AT genes were modelled using SWISS MODEL workspace. The template used to build the model of BAHDch 1, BAHDch 13 and BAHDch 6 were retrieved from Protein data bank with ID: 6ZBS, which has the sequence similarity of 34.96%, 34.83% and 35.10% respectively. Out of the six proteins, only CHR6 BAHD isoform showed negative binding energy (-0.77 kcal/mol) and all other isoforms showed positive binding energy. Figure 6 shows the binding of the ligand molecule piperoyl CoA in the binding site of BAHDCh 6 protein

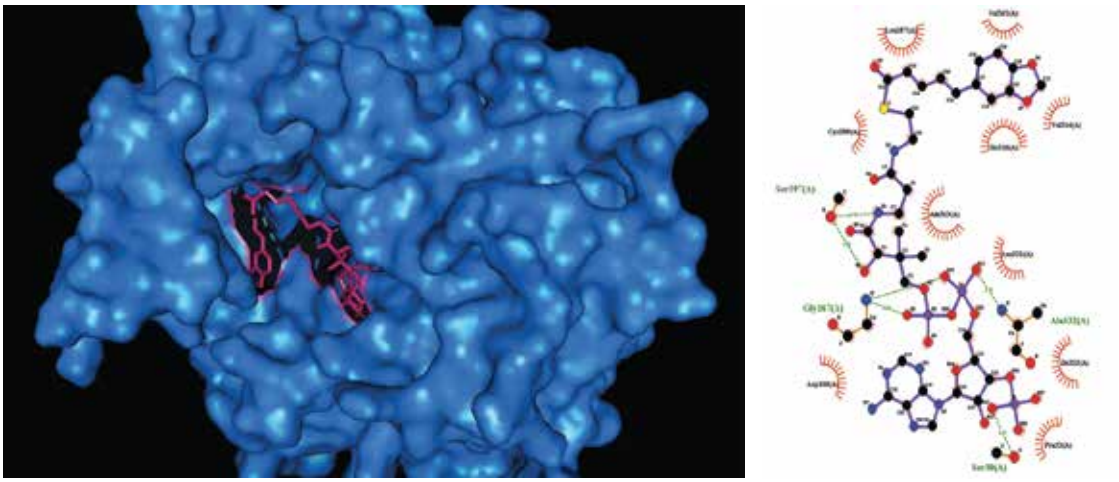

Fig.6: Molecular docking analysis: a) pose view of Ligand molecule Piperoyl CoA (Pink) in the active site of CHR6 BAHD isoform (Blue). b) 2D view of ligand and active site residues, hydrogen bonds are in green dotted lines.

Transcriptome analysis

Rooted cuttings of genotype IC 317179 were grown under normal as well as water stressed (15 days stress by withholding water) conditions. De-novo and reference-based assembly of drought stressed transcriptome was performed using Trinity and STAR respectively. Transcript expression values were estimated using SALMON while the genes were mapped using HTSeq. Differentially expressed genes and transcripts were identified using edgeR. The significantly differentially expressed genes and transcripts (log fold change >1 and p-value) were shortlisted for validation (Fig.7).

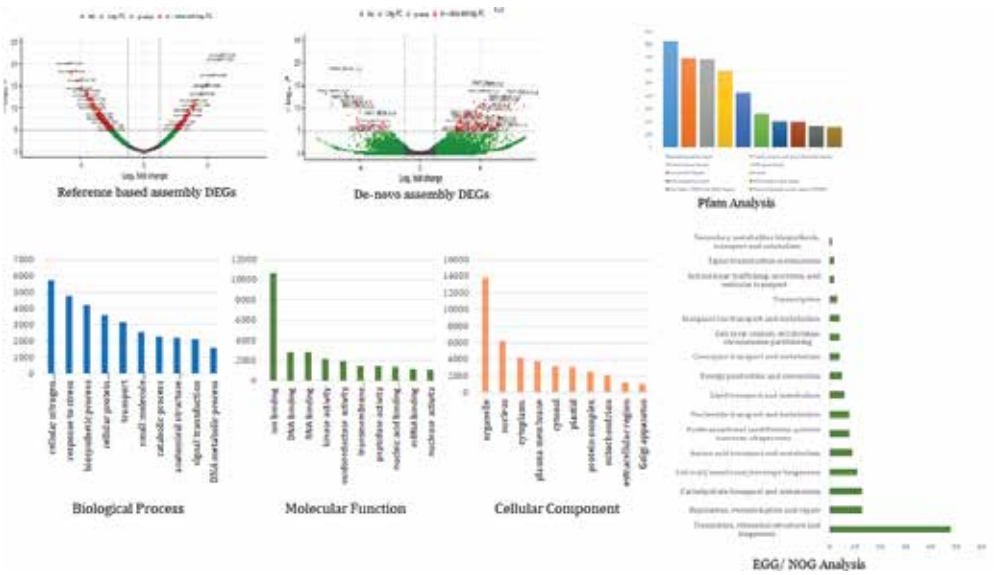

Fig.7 : Results of reference based and *de-novo* analysis of Black pepper transcriptome

Development of fertigation schedule

The varieties IISR Shakthi and Girimunda recorded maximum yield with drip irrigation @ 8 L water per day with 50 % of RDF applied as fertigation in 24 splits, followed by conventional irrigation @ 8 L of water per day with 100% RDF applied in 3 equal splits as basal dose. Whereas, in variety IISR Thevam, maximum yield was observed in recommended dose of fertilizer with conventional irrigation.

Recombinase polymerase amplification (RPA) protocol for the detection of *Phytophthora* spp.

A recombinase polymerase amplification (RPA) protocol to detect *Phytophthora* spp infecting black pepper has been developed. The assay has a sensitivity of detecting upto 1 ng of pathogen DNA in black pepper leaf and root extract. The assay was validated with isolates of *P. capsici* and *P. tropicalis* collected from various locations and also infected leaf samples.

Diversity analysis of *Phytophthora* isolates using REP PCR and RAMS analysis

Forty-eight isolates (24 isolates each of *P. capsici* and *P. tropicalis*) of *Phytophthora* spp. infecting black pepper were analyzed using RAMS, REP, ERIC, and BOX PCR primers. The *P. capsici* and *P. tropicalis* isolates were clearly separated into two major clusters and were further separated into four sub-clusters (I & II- *P. capsici* isolates and III & IV- *P. tropicalis* isolates).

Genome analysis of *Phytophthora* isolates

The genomes of *P. capsici* isolate 05-06 and *P. tropicalis* isolate 98-93 were assembled through hybrid genome assembly of short (Illumina) and long read (PacBio, Nanopore) sequences. The total assembled genome size of *P. capsici* was 68.2 Mb and that of *P. tropicalis* was 63.3 Mb.

Identification and analysis of effector genes

The RxLR 29 gene cloned from *P.tropicalis* isolate 98-93 was sequenced. Blast analysis of the RxLR effector protein (RxLR 29) showed 31.96% similarity with RxLR effector protein PSR2 of *P. sojae*. A three-dimensional (3D) protein structure was constructed using SWISS-MODEL workspace (Fig. 8a). The modeled structure was validated using the Ramachandran plot from the MolProbity program. All amino acid residues of the modeled protein fit within the Ramachandran plot allowed regions (Fig. 8b).

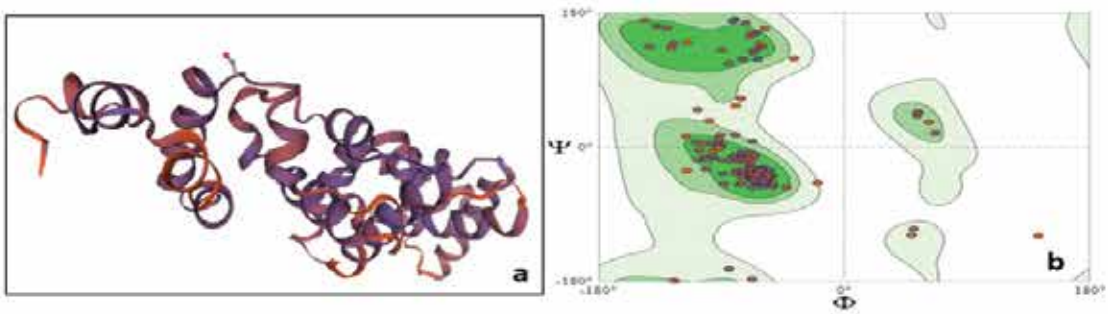

Fig. 8:(a) Predicted structure of the RxLR 29 protein and (b) Ramachandran plot of modelled RxLR29 protein

In vitro evaluation of fungicides against *P. capsici* and *P. tropicalis*

Fifteen fungicides were evaluated *in vitro* at five different concentrations against *P. capsici* and *P. tropicalis* isolates. Among the 15 fungicides tested, Bordeaux mixture, copper oxychloride, fluopicolide-propamocarb, cymoxanil- mancozeb, iprovalicarb-propamocarb, and metalaxyl-mancozeb showed complete mycelial growth inhibition and reduced the sporangial production.

Root colonization studies with temperature-tolerant *Trichoderma* isolates

Endophytic colonization of temperature tolerant isolates of *Trichoderma viz. T. asperellum* (NAIMCC 0049), *T. erinaceum* (APT1), *T. atroviridae* (APT 2), *T. harzianum* (KL3), *T. lixii* (KA15) and *T. asperellum* (TN3) were studied under in vitro conditions. Colonization capacities of the test isolates varied with isolates and the isolate, *T. atroviridae* colonized the most and the least was observed in *T. asperellum.*

Arbuscular mycorrhizal colonization to induce defense responses against *P. capsici* infection

The effect of arbuscular mycorrhizae colonization of black pepper plants to induce defense responses against *P. capsici* infection was studied under greenhouse conditions. Study indicated that AM pre-inoculation up regulated pathogenesis-related genes *viz.,* cAPX, osmotin, and β-1, 3-glucanase, phenylalanine ammonia -lyase and NPR 1 in black pepper leaves and roots upon *P. capsici* inoculation (Fig.9).

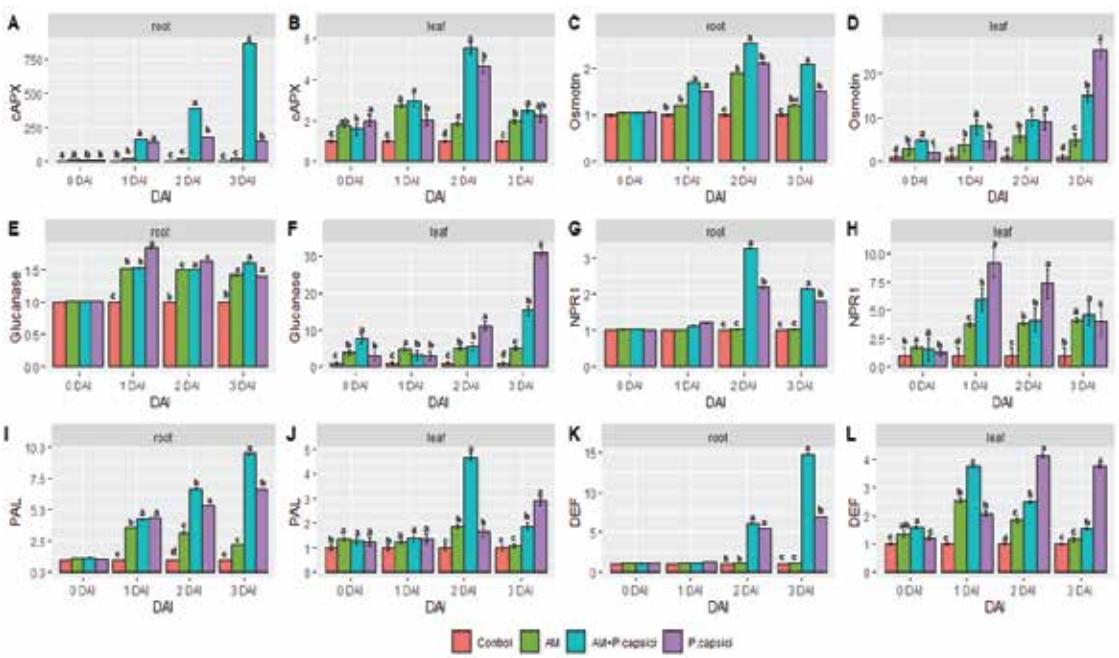

Fig.9: Differential expression of cytosolic ascorbate peroxidase (cAPX), osmotin gene, glucanase gene, NPR1gene, PAL gene, DEF gene in roots and leaves of black pepper

Development and validation of recombinase polymerase amplification -lateral flow assay (RPA-LFA) for the detection of piper yellow mottle virus (PYMoV)

Two rapid assays based on the recombinase polymerase amplification (RPA) coupled with lateral flow assay (LFA) using (i) 6-carboxyfluorescein (FAM) labeled nfo probe and biotin-labeled reverse primer and (ii) FAM labeled forward and the biotin-labeled reverse primer was developed for the detection of PYMoV.The formation of a coloured line at the test line of a lateral flow device was considered positive for PYMoV (Fig. 10). The assays were validated using field samples of black pepper and mealybug vectors. The assay will be useful to identify virus-free black pepper mother plants for the production of virus-free planting material.

Curing of mycoviruses associated with *Fusarium concentricum*

The mycoviruses associated with *F. concentricum* could be cured using ribavirin (concentration >75 µM/ml) amended PDA. Further, *F. concentricum* harbouring viruses showed reduced mycelial growth and conidial production compared to the isolate without mycoviruses.

Screening of low-risk insecticides against mealy bugs

Spiromesifen 22.9 SC, Triflumezopryrim 10 SC (1mLL^{-1}), and Spirotetramat 150 OD (1.5 mL^{-1}) and Clothianidin 50 WDG (1 gL^{-1}) were found to be very effective against the black pepper mealy bug (*Ferrisia virgata*) under *in vitro* conditions.

Efficacy of new nematicides against burrowing nematode

The application of fluopyram twice a year (pre- and post-monsoon) was found to be most effective against burrowing nematode *(Radopholus similis)* under field conditions .

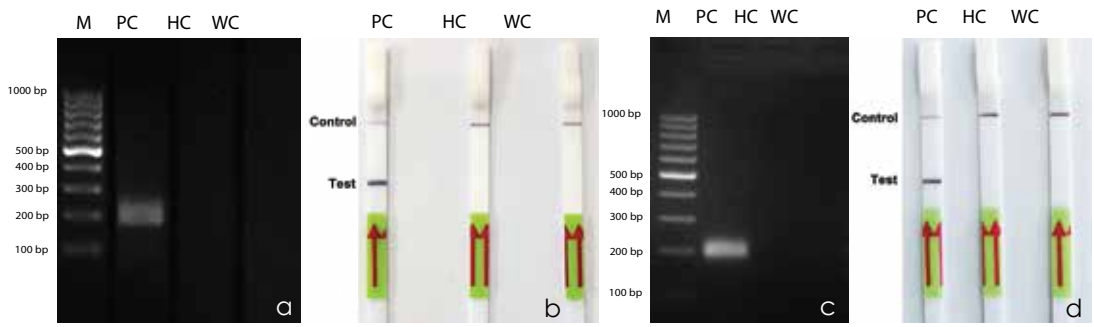

Fig. 10: Detection of piper yellow mottle virus infecting black pepper through recombinase polymerase amplification (RPA) (a, c) and RPA-coupled with lateral flow assay (RPA-LFA) (b, d).

CARDAMOM

Genetic resources

Six hundred twenty five accessions are being maintained in National Active Germplasm Site (NAGS) at ICAR-IISR, Regional Station, Appangala. Characterization of 85 field gene bank accessions was carried out based on different morphological and yield traits.

CVT on hybrids

In CVT trials conducted on hybrids, highest fresh as well as dry yield per plant was recorded in the hybrid PH 13 (6.30 kg/plant and 1.18 kg/plant respectively) this was followed by PH-14 and Bold × IC 547219 hybrid which recorded 0.94 and 0.82 dry yield per plant respectively (Fig.11).

Multi-location trial on leaf blight tolerant lines

Five leaf blight tolerant genotypes *viz.,* IC – 349650, IC – 547222, IC – 547156, IC – 349649, IC – 349648 along with resistant checks: Appangala 1, Njallani Green Gold and susceptible check - IISR Vijetha were tested for tolerance to leaf blight. Disease incidence ranged from 11.11 - 21.11 % and maximum disease incidence was recorded in IISR Vijetha.

Micronutrient formulation gets patent

Institute was awarded with a patent for the micronutrient composition developed specifically for Cardamom (patent No: 413017). The use of micronutrient was found to increase the yield by 10 to 25% and improves the health and vigor of the plants. The formulation is water soluble and compatible with common straight or complex fertilizers.

Identification of genotypes for moisture stress through multi-location evaluation

Eight genotypes, *viz.*, IC 349537, IC 584058, GG×NKE-12, IC 584078, CL 668, HS-1, APG-1 and IC 584090 were evaluated under both irrigation and moisture stress conditions. IC 349537 recorded the minimum Drought Susceptibility Index (DSI) and maximum Drought Tolerance Efficiency (DTE) (%)

Fig.11:Yielding clump, fresh and dry capsules of high yielding hybrid PH13

of 0.89 and 70.71 %, respectively indicating that the genotype performs well under moisture stress condition . The drought tolerant genotype IC 349537 was recommended for release as IISR Manushree (Fig.12).

Multiplex PCR assay for pathogen detection

Multiplex PCR assay was developed to simultaneously detect *Phytophthora* spp., *Pythium vexans*, and *Rhizoctonia solani* pathogens infecting cardamom. The assay could successfully detect the pathogens both singly and in combinations and does not show any cross-reaction with other fungal pathogens of cardamom.

Development of RPA assay for cardamom mosaic virus (CdMV)

A rapid assay based on the reverse transcription recombinase polymerase amplification (RT-RPA) was developed for the detection of cardamom mosaic virus (CdMV). The RT-RPA assay was 10^5 times more sensitive than RT-PCR. The assay was validated using CdMV-infected small cardamom samples from different regions. The developed assay will be useful for the identification of virus-free plants for propagation to produce virus-free suckers for planting.

Dose optimization of low-risk insecticides against shoot and capsule borer

Three low-risk insecticides (spinosad, flubendiamide and chlorantraniliprole) and a combination treatment of spraying chlorantraniliprole and spinosad alternatively along with a standard check (quinalphos) were screened under field conditions for dose optimization (0.3, 0.5 and 1.0 mlL^{-1}) against shoot and capsule borer. The results indicated that all the tested low-risk insecticides were effective in controlling the pest at the highest dose tested ($1 mlL^{-1}$).

Fig.12: Drought tolerant genotype IC 349537

GINGER

Genetic resources

Six hundred and sixty-eight accessions are being maintained in the field gene bank. New additions to the collection were made from Dibrugarh, Tinsukia and North Lakhimpur districts of Assam. Eighteen accessions, which includes *Curcuma inodora*, *C. caulina, C. mutabilis*, *Zingiber neesanum* and *Z. diwakarianum* were collected from Konkan region of Maharashtra in the forest areas in Raigad, Ratnagiri, Sindhudurg (Konkan region) and Kolhapur, Sangli, Satara (Western Maharashtra) (Fig.13).

Gingerarium

"Gingerarium" a dedicated facility for conserving ginger accessions and related species collected from different parts of the country was established at IISR Experimental farm (Fig.14). Besides germplasm accessions, the released varieties and farmer's varieties are also being conserved.

Fig.13: *Zingiber diwakarianum* *Curcuma inodora* *Curcuma caulina*

Colchicine induced polyploidy

Successful induction of tetraploidy (2n=44) was achieved in red ginger, through *in vivo* colchicine treatment. The ploidy level of the selected plants was ascertained using flow cytometric technique. The identified tetraploids were compared with the diploids for morphological and physiological characters. Tetraploids showed a significantly lower stomatal frequency than diploids.

Organic farming

In the evaluation of production systems, integrated management (75% organic + 25% inorganic) recorded maximum yield of 14.9 tha^{-1} which was on par with organic (25%) + inorganic (25%) + seed treatment with Beejamrit (BA), Ganajeevamrit (GJA) and Jeevamrit (JA) (13.3/ha).

Spray schedule optimization of low-risk insecticides against shoot borer

Low-risk insecticides such as chlorantraniliprole (0.01%), flubendiamide (0.02%), and spinosad (0.0225%) and a combination treatment of spraying chlorantraniliprole and spinosad alternatively were evaluated under field conditions for spray schedule optimization at two different spray schedules (i.e.15 and 21 days interval). against shoot borer *(C. punctiferalis)* infesting ginger. The results indicated no significant difference in efficacy among the insecticides at the tested spray intervals.

Fig.14: Gingerarium

Evaluation of mango ginger genotypes

Nine accessions of mango ginger were evaluated for yield performance under field conditions along with check, Amba during 2019-2020, 2020-2021 and 2021-2022. Among the accessions, Acc. 347 recorded the highest yield followed by NVMS 2 (Fig.15).

Fig.15: Rhizome characters of promising mango ginger genotypes

TURMERIC

Phytochemicals, nutraceuticals and bioactivity profiling of varieties

Studies indicated that varieties, NDH 1, Rajendra Sonia, and Kanti could be an alternative source of nutritional supplement based on their proximate (moisture, ash, protein, fat, and carbohydrates) and mineral (Cu, Mn, Fe, Zn, K, Ca, Mg, P) composition.The genotypes recorded 4.0 to 6.4 % essential oil, 5.51 to 15.65 % oleoresin and 0.25 to 6.41 % curcuminoids. The *in vitro* antioxidant activity by inhibiting DPPH (27.34 μgml^{-1} to 53.11μgml^{-1}) and *in vitro* anti-diabetic activity by inhibiting α-glucosidase (32.26μgml^{-1} to 58.72μgml^{-1}) proves the use of turmeric in treating life-style diseases.

Identification of superior genotypes for colour characteristics

CVT on light yellow coloured turmeric for specialty markets indicated that Acc 849 recorded maximum fresh yield followed by Acc. 1545.Maximum fresh yield was recorded in Acc 849 (39.58 t/ha) followed by Acc. 1545 (36.50 t/ha), which were on par. The pooled analysis of variance revealed highly significant variations between genotypes, locations, and genotypes by environment (G × E interaction) for all the colour parameters. The correlation between colour values L*, a*, b*, YIE (yellowness index) and curcuminoids revealed that in all the three production environments, a* value had significant positive correlation (0.74-0.84 %) with curcuminoids. A two-dimensional GGE biplot was generated using the first two principal components (axis 1 and axis 2), which revealed that the genotypes Acc. 379 and Acc. 849 were stable across environments, with low curcuminoids.

Genome wide association analysis (GWAS)for important agronomic traits

GWAS was carried out to obtain SNPs linked to valuable agronomic characters like curcumin content, rhizome girth and rhizome length in turmeric using 51 genotypes with diverse agro-morphological constitution. Structure analysis performed for the elucidation of population structure revealed subpopulation kinship matrix with K =3. Association analysis resulted in identification of nine significant MTAs (marker-trait associations) for the three phenotypic traits. The number of significant MTAs per trait were : curcumin content (1), rhizome length (5), rhizome girth (3).

Organic farming

Twelve varieties (IISR Prathibha, IISR Alleppey Supreme, Varna, Sobha, Sona, Kanthi, Suvarna, Suguna, Sudarsana, IISR Kedaram, IISR Prabha and IISR Pragati) were grown under organic and integrated nutrient management (INM) systems. Organic treatments (100%) recorded maximum soil available organic C, N, P, K, Ca, Mg, Fe, Mn, Zn and Cu. Curcumin (4.6%) and oil (5.6%) content were maximum in organic (75%) + INM (25%), whereas oleoresin content (11.8%) was higher in organic that was on par with INM (50%)+ organic (50%) (11.6%).

Effect of phosphorus solubilising bacteria (PSB) on phosphorus fractions

The microbial solubilization of phosphorus and increase in the amount of plant available phosphorus in soil by two efficient PGPRs were studied with *Bacillus safensis* and *B. cereus* and different levels of sparingly soluble P source on turmeric. At 3 months after planting, Ca – P contributed the highest percentage share in total P followed by Fe – P, saloid P and Al – P. There was a significant positive correlation between Ca – P and saloid P and a significant negative correlation between Ca – P and Al – P revealing that PSB has solubilized a substantial amount of Ca – P into plant available P form.

Land suitability assessment

Land suitability assessment for turmeric in India was analyzed taking in to consideration the 5th Assessment Report Scenarios of IPCC. Climatic parameters such as temperature and rainfall and land characteristics such as soil drainage, texture, pH, depth and slope were considered and land suitability assessment was done for 2020 & 2050 (Fig.16).

Effect of microwave assisted hot air drying on the quality of dried turmeric slices

Microwave assisted hot air drying of turmeric slices (var. Sudarshana) was conducted to determine the quality of dried turmeric slices. Microwave exposure time of 3 min at 620 W showed minimum loss in essential oil and hence duration of 3 min was taken as optimum for microwave cooking followed by hot air assisted drying at 50°C and the time taken for drying slices was 14 h compared to fresh samples where the drying time was 16 h.

Greenhouse evaluation of promising phosphorus solubilizing bacteria (PSB)

Greenhouse evaluation of promising phosphorus solubilizing bacteria (PSB) for soil P solubilization and growth promotion indicated that application of PSB significantly promoted growth as evidenced from the enhanced number of tillers, shoot length, number of leaves, dry weight of shoot and root.

Field evaluation of PSB for soil P solubilization and growth promotion

Combined application of P (75%) and *B. safensis* registered significantly higher rhizome yield of turmeric under field conditions (Fig. 17). With regard to available soil P, the levels were significantly higher in all the treatments involving combined application of *B. safensis* with 50% or 75% or 100% P.

Evaluation of PGPR for Zn solubilization potential

Evaluation of multi-trait PGPR, *Bacillus safensis* for plant growth promotion and Zn solubilization under greenhouse conditions showed that combined application of Zn -5 ppm (source –ZnO) + *B. safensis* was found to be superior with an increase in organic carbon, available nitrogen, etc.

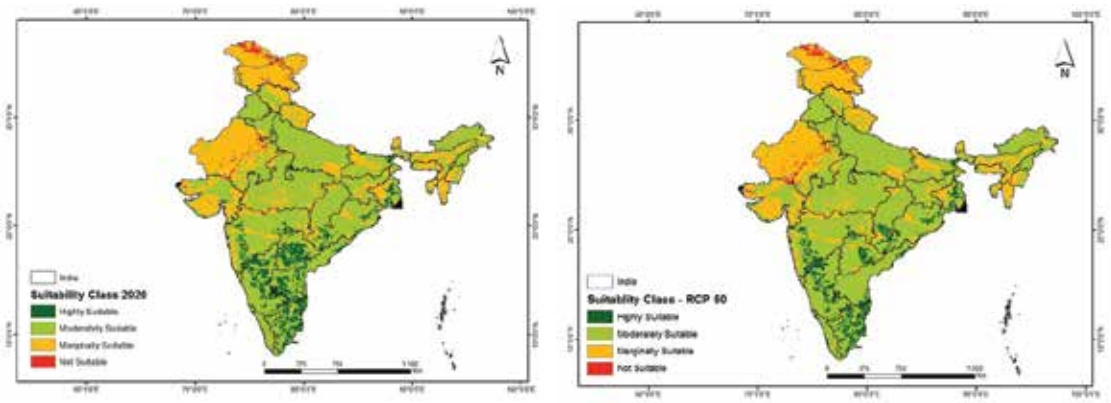

Fig.16: Land suitability assessment in turmeric

and microbial parameters like microbial biomass C, N, and dehydrogenase enzyme activity in soil.

Field evaluation of *Metarhizium pingshaense* against shoot borer

Spray application of *M. pingshaense* at 1 × 10^7 conidia/ml at a spray interval of 21 days was found to be effective in managing shoot borer.

Influence of plant phenology and crop duration on the occurrence of shoot borer

The influence of crop duration and phenology with respect to the seasonal incidence of shoot borer infesting turmeric was studied using two short-duration varieties (IISR-Pragathi and Rajendra Sonia), two long-duration varieties (IISR-Prathibha and Alleppey Supreme), and an extra-long duration variety (ACC. 849). The infestation started in the first week of August irrespective of the variety and peaked during the second fortnight of September.

Population dynamics of lesion nematode, *Pratylenchus spp*

Population dynamics of lesion nematode *Pratylenchus* spp., infecting turmeric was studied and its population was recorded at monthly intervals.The maximum population was recorded during the month of November while the population was minimum during the month of June.

Management of lesion nematodes

Application of fluopyram (0.5 mlL^{-1}) effectively reduced the lesion nematode (*Pratylenchus* spp.) population, enhanced the number of tillers and the yield in turmeric.

Fig. 17. Effect of shortlisted PSB on yield of turmeric clump under field condition

T1- Control
T2- *Bacillus safensis*
T3- B. *safensis* + 50% P
T4- B. *safensis* + 75% P
T5- B. *safensis* + 100% P
T6- 100% P

TREE SPICES

NUTMEG

Conservation

The germplasm repository was expanded with 8 farmer's varieties collected from various districts of Kerala. A high yielding nutmeg accession; IC 645756 was registered with ICAR-NBPGR (INGR 22092) for its monoecious character.

DNA fingerprinting

DNA fingerprinting using ISSR primers was carried out in nutmeg varieties IISR Viswashree, IISR Keralashree, Sindhushree and a monoecious cultivar. Among the 45 ISSR primers used, five primers IS-02, ISSR 05, ISSR-14, ISSR-01 and UBC 834 produced distinguishable markers.

Microwave assisted convective drying system for mace

Drying kinetics, modelling and quality evaluation of nutmeg mace under microwave assisted convective drying system indicated that the drying characteristics data fitted into thin layer drying models and the two-term exponential model was found to be the best fit model under optimized conditions with R^2 = 0.9986, RMSE = 0.01328 and χ^2= 0.000141. The study concluded that microwave at optimal power of 320 W for 1 minute was found to have better retention of essential oil, oleoresin and color of dried mace and that microwave drying can be applied for faster drying of mace with better quality aspects (Fig.18).

Fig. 18. Microwave assisted convective drying

CINNAMON

Five accessions of wild cinnamon comprising of two species each were collected from Dibrugarh, Tinsukia and North Lakhimpur districts of Assam (Fig. 19) and another species of cinnamon was collected from Baba Budan hills of Karnataka.

Fig. 19: *Cinnamomum* sp. collected from Assam

CLOVE

Two accessions, one with bold flower buds and another accession with red pigmented flower buds (Fig. 20) were collected from Tenkasi district of Tamil Nadu. Two accession of bold clove locally called as Madagascar clove and a *Syzygium* species were collected from Nagercoil,TamilNadu and North Lakhimpur district , Assam respectively.

Fig. 20: Variant of clove with red pigmentation

GARCINIA

Twenty six accessions comprising eight species *viz., G. acuminata, G. assamica, G. dulcis, G. lanceifolia, G. pedunculata, G. sibeswarii, G. xanthochymus* and an unidentified species were collected from Dibrugarh, Tinsukia and North lakhimpur districts of Assam (Fig.21). Two accessions of *G. talbotii* were collected from Amboli Ghat, Maharashtra. A new germplasm block has been established at Experimental Farm, Peruvannamuzhi with 96 collections of *Garcinia*.

VANILLA

Conservation

A total of 77 accessions were established in poly house (65 *Vanilla planifolia* and 12 *Vanilla* sp) and in field conservatory. The accession 4766 produced long (22 cm) beans with more fresh weight (38.45 g) followed by the accession 4751 with bean length of 21 cm and fresh weight of 32 g.

Fig.21: *Garcinia pedunculata*

Garcinia sibeswarii

Garcinia dulcis

ALLSPICE

Unique ISSR markers for distinguishing *Pimenta dioica* from *Pimenta racemosa* have been identified. Among the primers tested, five primers ISSR-12, primer IS-10, primer IS-11, IS-02, ISSR-05 produced distinguishable markers.

Fig.22: Production of multiple shoots from seeds *in vitro*

Quantification of major flavour compounds

Protocols for quantification of major flavour compounds *viz.,* vanillin, p-hydroxybenzoic acid,p-hydroxybenzaldehyde and vanillic acid were standardized.

In vitro seed germination

In vitro seed germination was attempted with different media (Fig.22). Seed germination and production of shoot was observed in the media composition MS+BAP (3 mg L^{-1})+NAA (0.5 mg L^{-1}).

GENERAL

Decision Support System for Site Specific Nutrient Recommendation for Spices (IISR-e SOFT)

The software provides fertilizer recommendations for targeted yield based on the factors like initial fertility, nutrient required for per unit yield (NR), contribution of nutrient from soil (CS) and contribution from fertilizer (CF) which were standardized, validated and recommended for major spice crops *viz.,* black pepper, ginger, turmeric and cardamom. It aims to improve fertilizer use efficiency, increase yield and avoid imbalance of nutrients in soil.

Effect of nano ZnO on genesis and regulation of biofilm genes in soil bacterial communities

Nano ZnO (nZnO) and bulk ZnO (bZnO) facilitated profusion of biofilm related genes (BGs) especially at higher Zn levels. In general, nZnO favored enhancement of genes involved in exopolysaccharide biosynthesis and attachment, while bZnO favoured genes related to capsule formation, chemotaxis and biofilm dispersion.

Effect of nano ZnO on bacterial community structure and associated functional pathways

The effect of nZnO on bacterial community structure and associated functional pathways were determined through predictive metagenomic profiling and subsequent validation through Quantitative Realtime PCR. The alpha diversity decreased with increasing ZnO level, with more impact under nZnO, while beta diversity analyses indicated a distinct dose- dependent separation of bacterial communities. The dominant taxa including Proteobacteria, Bacterioidetes, Acidobacteria and Planctomycetes significantly increased in abundance, while Firmicutes, Actinobacteria and Chloroflexi decreased in abundance with elevated nZnO and bZnO levels.

Interrelationships between genera and functional pathways in soils polluted with nano ZnO

The interrelationships between genera and functional pathways under nZnO and bZnO were deduced using co-occurrence network analysis (Fig.23). The network exhibited five modules under nZnO and six modules under bZnO. Topological properties of co-occurrence network indicated contrasting associations under nZnO and bZnO. Thus, the soil bacterial communities under extreme nZnO stress depicted comparatively lesser functional resilience and stability than corresponding level of bZnO.

DNA fingerprinting and barcoding facility

Unique DNA markers have been developed for establishing varietal status in two varieties each of aromatic turmeric, black turmeric, turmeric, fenugreek, nigella and ajwain varieties from various AICRPS centers .

Spice based finger millet products

Steaming as pretreatment to finger millet flour for 10 min and blending finger millet flour to a concentration of up to 40% as replacement to refined wheat flour produced finger millet cookies of acceptable texture. Spice enriched finger millet cookies were prepared by adding different spices like cardamom, black pepper and spice blend to get unique flavour of each spice (Fig.24).

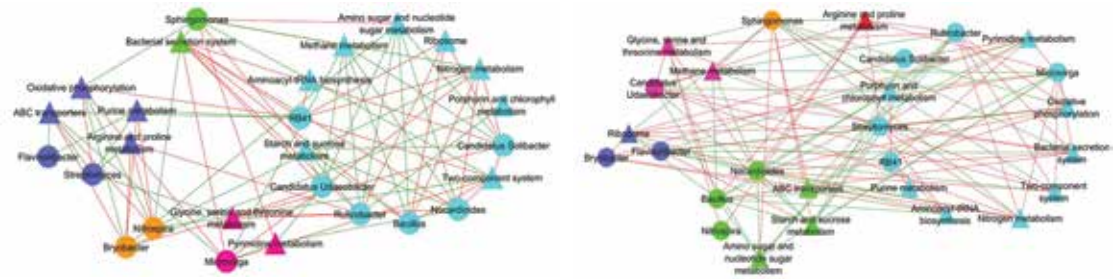

Fig.23: Co-occurrence network indicating the interaction between dominant genera and functional pathway in soils spiked with (a) nZnO and (b) bZnO. Nodes are colored according to modularity classes. Lines connecting two nodes represent a group of significant correlations (P < 0.05). Green lines show positive correlations (R ≥ 0.5); red lines show negative correlations (R ≤ -0.5). Pathways are indicated in triangle and genera in circles.

a. Cardamom b. Black pepper c. Spice blend

Fig.24: Finger millet blended spice flavoured cookies

Economics and policy studies

PESTLE analysis of Indian spice economy A comprehensive analysis of the spice economy was undertaken using a PESTLE framework identifying the key political, economic, social, technological, legal and environmental aspects of the sector which could influence the development trajectory in the coming years. The study identified the critical role played by policy interventions in balancing the interests of primary producers, constituents of spice industry producers and the consumers.

Pesticide regulation policy in spices

Though the pesticide industry in India is highly regulated, there are several lacunae regarding the registration and availability of plant protection products (PPPs) for commercial agriculture. In case of spice crops in India, the limited number of both registered pesticides and the absence of maximum residue limits (MRL) for several pesticides were found to have implications for the domestic and international trade of spices. There is an urgent need to enhance the number of registered pesticides used in spices in India, and to set both national and codex MRLs for pesticides, in order to promote sustainable good agricultural practices (GAP). The approval and registration formalities of biopesticides also need to be eased to address the environmental concerns arising due to over use of chemical pesticides and to ensure food safe spices.

ATIC AND EXTENSION SERVICES

The seed portal of the institute has become fully operational and along with the Visitor management system software, has enhanced the outreach in sale of planting material and other technology inputs. One RAWE programme was organized for students from Kerala Agriculture University and 55 Educational Training Programmes were organised for scholar community spanning graduate, post graduate and other professional courses. The institute participated in exhibitions and were leveraged to showcase the cafeteria of technological advances available in the spice crops and has aided in dissemination of novel technologies across the country. The institute conducted 33 customised training programmes of various duration for the state departments and other organisations on production, crop protection and processing technologies of spices during the year. The institute partnered with AIR Madikeri, Karnataka to develop eight radio talk modules on various aspects of spice farming. The institute conducted 26 focused programmes for beneficiaries belonging to Scheduled Caste and Scheduled Tribe. The handholding services provided to the identified beneficiaries include skill enhancement training programmes, distribution of technology inputs and supply of critical inputs for aiding farming operations. The revenue generation through the sale of planting material of spice crops, bio-inputs and micronutrients and other products from ATIC was 38.8 lakhs during 2022.

Institute Technology Management Unit (ITMU) & Agribusiness Incubation (ABI) Centre

ITMU & ABI commercialized 9 technologies and an amount of Rs. 42.75 Lakhs was earned as revenue through technology commercialization.The technology "A novel method of storing and delivering PGPR/Microbes through biocapsules' was commercialized to Lysterra LLC, Moscow, Russia. Dr T. Mohapatra, former Director General, ICAR, presided over the virtual meet on signing of international MoU. A patent was granted for the technology 'A micronutrient composition for cardamom and a process for its preparation' (Indian Patent No.413017). ITMU organized an awareness programme on Intellectual Property Rights (IPR) under National Intellectual Property Awareness Mission (NIPAM) in collaboration with Indian Patent office, Chennai. IISR entered into MoA with M/s Bayer Crop Science Pvt Ltd, Maharashtra under collaborative research mode for developing best practices for spice cultivation/production, crop protection measures. ITMU & ABI facilitated five consultancy visit to black pepper plantations of various firms in Kerala & Karnataka and earned a revenue of 3.5 lakhs.Three of the startup licensees/firms (SRT Agro Science Pvt Ltd, Chhattisgarh, Codagu Agritech, Karnataka, Hi7 Agri Bio solutions, Karnataka) of IISR were selected to participate and exhibit their products in the Agri Start-up Conclave & Kisan Sammelan-2022. Six startups/ entrepreneurs were enrolled as incubatees during the year 2022 for development of spice based food products, availing spice processing facility at IISR. ABI organized an online Refresher Training Programme (RTP) on Spices Cultivation and Business opportunities in collaboration with MANAGE, Hyderabad. About 55 established agriprenuers from across the country participated in the training program. Mallikaappi, a traditional immunity booster composed of spices was officially launched for commercialization on behalf of incubatee "Suman Research and Rehabilitation Centre", an NGO working for rehabilitating mentally challenged Women at Kozhikode. ABI Unit supported startups/registered farmers/licensees through marketing of quality assured spices, value added products, allied products through the sales outlet SPIISRY and online platform www.spiisry.in. Krishidhan nursery, the incubation facility of ABI could support the production and marketing of quality planting materials of joint liability groups, licensees and registered farmers.

Agricultural Knowledge Management Unit (AKMU)

AKMU facilitates the IT and ICT related activities of the institute, maintains network security aspects,

developing websites, institute & AICRP website updations,maintenance of data center, webservers, databases, technical support to online meetings, and uploading institute activities in the social medias. Apart from this AKMU assists to analyze and interpret geographical data using ArcGIS & DIVA GIS and statistical analysis of scientific data using SAS, JMP and R software.

LIBRARY

Library has a collection of 5683 books and 6010 bound journals. IISR Library is part of the Consortium of electronic Resources in Agriculture (CeRA) and more than 3500 full text journals on agriculture and allied subject are accessible. 25 Indian Journals and Eight Foreign Journals were subscribed during the year in addition to journals accessible under CeRA. The newly added publications were brought in to the Library Automation software 'KOHA' database.

HUMAN RESOURCE DEVELOPMENT

Human resource development (HRD) cell organized training and capacity building programmes on popularization of Hindi language, ICAR service rules, minimum wages act at ICAR-IISR, Kozhikode for scientists, technical and administrative staff. HRD cell also facilitated in organizing virtual training programme on "Data Digitalization and Visualization" and "Life Science meets Programming".

ICAR-ALL INDIA COORDINATED RESEARCH PROJECT ON SPICES (AICRPS)

The XXXIII Annual Group Meeting of ICAR-All India Coordinated Research Project on Spices (AICRPS) was held during 13-15 October 2022 at Acharya Narendra Deva University of Agriculture & Technology, (ANDUAT), Kumarganj, Ayodhya (UP). The workshop was inaugurated by Dr. N.K. Krishnakumar, Former DDG (Horticulture Science), ICAR, New Delhi. Dr. Bijendra Singh, Hon'ble Vice Chancellor, Acharya Narendra Deva University of Agriculture & Technology (ANDUAT), Kumarganj presided over the function. Dr. V. A Parthasarathy, Former Director, ICAR-IISR, Kozhikode was the Guest of Honour during the occasion. During the inaugural session the "Best AICRPS Centre Award 2021-22" was presented to AICRPS centre at Sardarkrushi nagar Dantiwada Agricultural University, Jagudan. Appangala-3, a new small cardamom variety, has been recommended for release by ICAR-AICRPS (Fig.25). The variety was developed by ICAR-IISR Regional station, Appangala, Kodagu, Karnataka. This variety is moisture stress tolerant, with a stable yielding capacity of 550 kg dry capsules ha^{-1} under irrigated conditions and 360 kg dry capsules ha^{-1} under moisture stress conditions. The recommended technologies were: micronutrient management in cumin, standardization of drip irrigation interval and method of micronutrient fertigation in fenugreek and Integrated pest & disease management in cumin

Fig. 25: Released small cardamom variety, IISR Manushree (Appangala-3)

Krishi Vigyan Kendra (KVK)

The KVK, Peruvannamuzhi imparted regular training programmes of various durations in agriculture and allied fields for the farmers, farm women, rural youth and extension functionaries. In total, 61 On-campus, 39 Off-campus capacity building trainings were organized by KVK, which benefitted more than 3545 participants. Paid training programme on Mushroom spawn production and garment making were conducted for rural youth. Sponsored trainings were organized on establishment of nutrition garden and pest and disease management, kitchen waste management and production of organic manures (sponsor- District Kudumbashree mission), mechanized coconut climbing (Sponsor- Coconut Development Board, Cochin), ornamental fish culture and fish health management (Sponsor-NABARD) and Bee keeping (Sponsor -HORTICORP) spices value addition (Sponsor-ATARI Bengaluru and IISR). Six OFTs on" Multiplier onion varieties, strawberry varieties, green gram varieties, assessment of organic methods for the management of aphids in cowpea, efficacy of different drugs for Bovine Papillomatosis were organized.

Eight FLD programmes were demonstrated on high yielding and mosaic resistant variety of Okra, hybrid napier 'Susthira' in coconut-based homesteads, quality ginger seed production, integrated pest and disease management in paddy, integrated management of Tanjore wilt in coconut, culturing of brackish water fishes and integrated fish culture in coconut based farming systems.The Kendra organized seminar on Agri-Nutri garden during Kerala Farmer's Day with expert classes on cultivation, pest and disease management in vegetables. Rashtria Kisan Diwas was celebrated at KVK on 25 November, 2022 and a seminar on "Natural farming and training on pepper planting material production was organized. Soil health awareness programme on 5th December, 2022 was organised with expert classes on soil health management, natural farming and organic pest and disease management.

To commemorate, 75th Year of Independence, KVK, organized trainings, awareness programmes and seminars and participated in live web casting programmes of ICAR viz., Hon'ble PM's address and seminar on "Sustainable Agriculture", nutri-cereals mega convention, Poshan Vatika and tree planting campaign and Natural Farming- Pre Vibrant Gujarat Summit 2021. KVK farmers K.T. Francis, Maruthonkara received Best Coconut farmer award of Coconut Development Board, Kochi and Smt. Bindu Joseph received Pandit Deen Dayal Upadyay Anthyodaya Krishi Puraskar- 2022 of ICAR.

INTELLECTUAL PROPERTY INDIA
PATENTS | DESIGNS | TRADE MARKS
GEOGRAPHICAL INDICATIONS

सत्यमेव जयते

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पेटेंट कार्यालय
THE PATENT OFFICE

पेटेंट प्रमाणपत्र
PATENT CERTIFICATE
(Rule 74 of The Patents Rules)

पेटेंट सं. / Patent No.	:	413017
आवेदन सं. / Application No.	:	1681/CHE/2015
फाइल करने की तारीख / Date of Filing	:	31/03/2015
पेटेंटी / Patentee	:	INDIAN INSTITUTE OF SPICES RESEARCH
आविष्कारक (जहां लागू हो) / Inventor(s)	:	1.SRINIVASAN VEERARAGHAVAN 2.RAGHAVAN DINESH 3.HAMZA SRAMBIKKAL

प्रमाणित किया जाता है कि पेटेंटी को, उपरोक्त आवेदन में यथाप्रकटित A MICRONUTRIENT COMPOSITION FOR CARDAMOM AND A PROCESS FOR ITS PREPARATION नामक आविष्कार के लिए, पेटेंट अधिनियम, 1970 के उपबंधों के अनुसार आज तारीख मार्च 2015 के इकतीसवें दिन से बीस वर्ष की अवधि के लिए पेटेंट अनुदत्त किया गया है।

It is hereby certified that a patent has been granted to the patentee for an invention entitled A MICRONUTRIENT COMPOSITION FOR CARDAMOM AND A PROCESS FOR ITS PREPARATION as disclosed in the above mentioned application for the term of 20 years from the 31st day of March 2015 in accordance with the provisions of the Patents Act,1970.

THE PATENT OFFICE GOVT.OF INDIA

अनुदान की तारीख : 30/11/2022
Date of Grant :

पेटेंट नियंत्रक
Controller of Patent

टिप्पणी - इस पेटेंट के नवीकरण के लिए फीस, यदि इसे बनाए रखा जाना है, मार्च 2017 के इकतीसवें दिन को और उसके पश्चात प्रत्येक वर्ष में उसी दिन देय होगी।

Note. - The fees for renewal of this patent, if it is to be maintained will fall / has fallen due on 31st day of March 2017 and on the same day in every year thereafter.

IMPORTANT DAYS OBSERVED AT ICAR-IISR, KOZHIKODE

Swachatha Hi Seva	11 September to 02 October 202
National Science Day	28 February 2022
International Women's Day	08 March 2022
World Water Day	22 March 2022
World Earth Day	22 April 2022
World Environment Day	05 June 2022
International Yoga Day	21 June 2022
Institute Foundation Day	01 July 2022
World Coconut Day	02 September 2022
Swachatha Hi Seva	11 September to 02 October 2022
Hindi fortnight	14 September to 29 September 2022
Vigilance Awareness Week	28 October to 02 November 2022
World Soil Day	05 December 2022
Swachata Pakhwada	16-31 December 2022

INDIAN COUNCIL OF AGRICULTURAL RESEARCH

ICAR

NATIONAL AWARD OF EXCELLENCE FOR AGRICULTURAL INSTITUTIONS

SARDAR PATEL OUTSTANDING ICAR INSTITUTION AWARD 2021

(For Small Institute Category)

ICAR-Indian Institute of Spices Research, Kozhikode, Kerala

CITATION

ICAR-Indian Institute of Spices Research, Kozhikode, Kerala has been awarded Sardar Patel Outstanding ICAR Institution Award 2021 in the category of Small Institute. The ICAR-Indian Institute of Spices Research (ICAR-IISR), Kozhikode, Kerala is an institute with international repute with primary focus on developing sustainable, cutting-edge technologies for spice cultivation in its mandate crops and product diversification through value addition. As the custodian of the world's largest repository of germplasm in its mandate crops, the institute has been able to harness this priceless asset for furthering its prioritized interventions. The genetic gains embedded in the varieties developed by the institute enhance profitability of spice farming for primary producers. The institute has displayed a steadfast commitment to technology development with focus on prioritized needs of its clientele. The solutions and technology inputs ranging from microbial encapsulation process, designer micronutrients, product processing protocols and comprehensive management packages for diverse ecosystems and farming systems are both critically acclaimed and widely adopted. The institute also house the largest spice research network in the country (AICRPs) that engages in nationwide collaborative and interdisciplinary research in spices. The technology disseminated, and extension services offered by the institute has remained sensitive to emerging needs of its stakeholders and has made visible and distinct impact across the country. The institute has successfully played the role of facilitator in harnessing the commercial potential offered by the spice value chains by handholding commercial ventures and start-up entrepreneurs. A special focus on socially and geographically disadvantaged stakeholder community has resulted in a host of effective intervention programmes targeting improvement in the quality of life for the vulnerable communities. With its persistent pursuit of research excellence, the institute has played a decisive and fundamental role in shaping the spice economy of the country and in making India a global leader in spice sector. An ISO 9001:2015 certified institution, ICAR-IISR stand testimony to the immense societal benefits from engineering to a creative blend of innovative science, quest for excellence and commitment to national

ICAR-Indian Institute of Spices Research

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