



A SCIENCE AND TECHNOLOGY NEWSLETTER

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## PROMISING TECHNOLOGIES

### Rapid- and- efficient RNA isolation protocol for recalcitrant tissues of mango

Mango (*Mangifera indica*) is an economically important fruit-crop of India. Biochemical changes occurring in the fruit during its ripening impart it softening, carotenoid accumulation and flavour-production qualities. And all the biochemical events are regulated at the gene level, and thus understanding them is of paramount importance in improving fruit quality and its storage potential. Thus, there is a need to isolate good quality RNA from fruits at different stages of development and ripening.

Ribonucleic acid (RNA) isolation is a critical step for molecular experiments involving reverse transcription polymerase chain reaction (RT-PCR), rapid amplification of cDNA ends (RACE), Northern hybridization, and microarray analysis and transcriptome analysis for deciphering mechanisms of gene expression, gene regulation, signal transduction and in post-translational studies.

Mango is one of the most complex crops from which RNA isolation was found very difficult due to significant differences in chemical composition of its tissues at varied stages of development, such as sudden shift in pH, changes in fatty acid, lipid and protein

#### Special Advantages of the Protocol

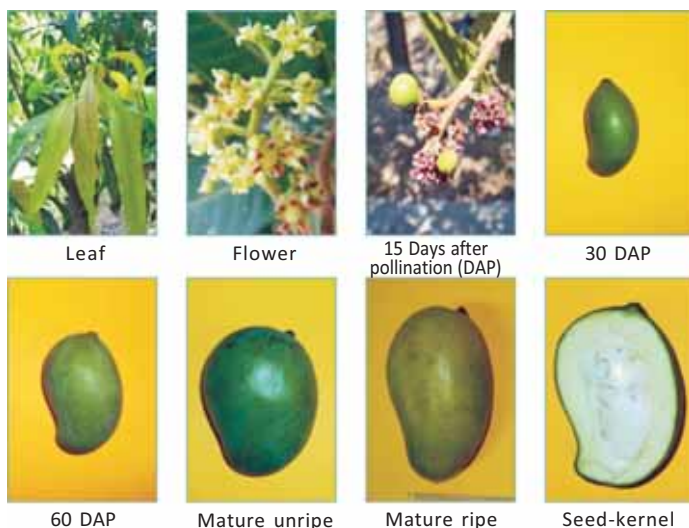
- This method is quite efficient for isolation of good quality (i.e., high purity and integrity) and quantity of RNA from problematic tissues of mango.
- It has been developed to reduce chemical usage and to lower toxicity (CTAB-free, guanidine-free and LiCl-free), compared to conventional protocols.
- Through reduction in number of steps, it takes lesser time of 1 - 2 h for RNA isolation.
- This method can be used for high-throughput sampling (10-12 samples in a day).
- The RNA isolated using this protocol has been found suitable and highly competent for molecular downstream applications such as construction of a cDNA library and for RT-PCR.

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# PROMISING TECHNOLOGIES



Different tissues of mango used for RNA isolation

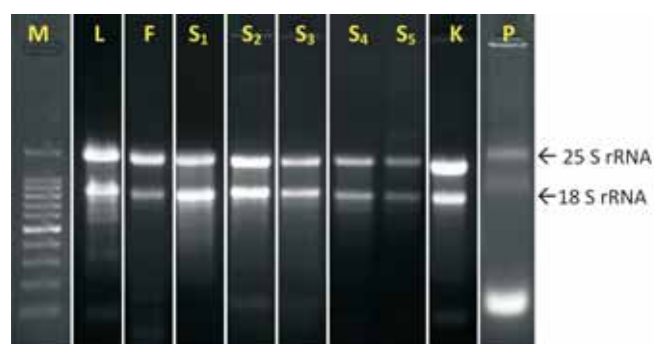
concentrations, and conversion of starch into sugars and protopectins into pectin. Many protocols were tried for isolation of good quality RNA from the tissues rich in polysaccharides and secondary metabolites, but most of them failed.

This newly developed protocol has worked well for extracting RNA from tissues of leaf, flower, fruit, fruit-peel and seed- kernel of mango. The quality ( $A_{260}/A_{280}$ : 1.6-2.05 and  $A_{260}/A_{230}$ : 1.6-2.2) as well as the quantity (16-80  $\mu\text{g}/\text{g}$  tissue) of the RNA were better with this in



## Quality and yield of RNA isolated from mango tissues with the protocol

Mango sample	$A_{260}/A_{280}$	$A_{260}/A_{230}$	Concentration ( $\mu\text{g}/\text{g}$ tissue)
Young leaf	1.57	1.39	84.83
Flower	1.59	1.41	40.89
Fruit stage-S <sub>1</sub> (30 DAP)	2.00	2.20	41.68
Fruit stage-S <sub>2</sub> (60 DAP)	2.01	2.13	28.66
Fruit stage-S <sub>3</sub> (90 DAP)	2.03	2.14	21.48
Fruit stage-S <sub>4</sub> (mature unripe)	2.05	1.98	18.37
Fruit stage-S <sub>5</sub> (mature ripe)	2.03	1.68	16.57
Fruit-peel	1.47	1.48	19.44
Seed-kernel	1.78	1.61	52.94



Agarose gel 1.2% (w/v) electrophoresis of RNA isolated from different tissues of mango. RNA was stained with 0.1  $\mu\text{l}/\text{ml}$  ethidium bromide and observed under UV light. (Lane M, 100 bp DNA ladder; lane L, leaf; lane F, flower; lanes S<sub>1</sub>-S<sub>5</sub> fruit stages: lane S<sub>1</sub>, 30 DAP; lane S<sub>2</sub>, 60 DAP; lane S<sub>3</sub>, 90 DAP; lane S<sub>4</sub>, mature unripe; lane S<sub>5</sub>, mature ripe; lane K, seed-kernel; lane P, fruit-peel)



RT-PCR amplification of transcripts of the *actin* gene isolated from various mango tissues using the protocol. (Lane M, 100 bp molecular markers; lane L, leaf; lane F, flower; lanes S<sub>1</sub>-S<sub>5</sub> fruit stages: S<sub>1</sub>, 30 DAP; S<sub>2</sub>, 60 DAP; S<sub>3</sub>, 90 DAP; S<sub>4</sub>, mature unripe; S<sub>5</sub>, mature ripe; lane K, seed-kernel; lane P-fruit-peel)

comparison to other methods. In addition, the shorter period of the protocol allows simultaneous processing of 10-12 samples in a single working day.

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## Magnetic field application catalyzes growth of sunflower-crop

High-quality sowing material is an important yield-enhancing factor in cultivated plants. Most often seed-dressing and priming are done with different chemical substances as pre-sowing seed treatments. Such methods have been considered effective but are not found environment-neutral. Magnetic field treatment is one of the physical pre-sowing methods which impacts different processes in the seed, and stimulates development of plants.

Sunflower-seeds were exposed to static magnetic field for different durations and their germination attributes were measured in terms of seedling growth, root characteristics and vigour. Magnetic field application enhanced significantly seed performance in terms of germination, speed of germination, seedling length and seedling dry weight compared to the control. Among the various combinations, 200mT for 2h exposure was the best for sunflower-crop. Exposure prior to germination enhanced seed performance; showed improvement over untreated control by 5-11% for germination, 9-15 % in speed of germination,

6-41% for shoot length, 16-80% for root length, 12-57% for total seedling length and 5-13% for seedling dry weight. Calculated vigour indices I and II also increased by 18-74% and 10-25%. In exposed seeds, there was significant reduction (6-14%) in electrical conductivity of seed leachates. All seedling parameters such as shoot height, root length, shoot and root dry weight of the one-month-old

seedlings of treated seeds exhibited significant increase over the control—shoot height by 7 to 10% and root length by 20 to 42%; shoot dry weight by about 83 to 94% and root dry weight by 69 to 107%; total root length by 57 to 73%; root-surface area by 55 to 81%; projected area by 55 to 82%; average root diameter by 17 to 35%; and even the root volume doubled in plants raised from treated seeds as compared to the control.

Sunflower-seeds exposed to 200 mT static magnetic field for 2h were sown in the field along with the untreated control. The exposed seeds showed increased field emergence index, leaf area index, shoot length, number

of leaves, chlorophyll content, biomass, 1,000 seed weight and seed yield. With magnetic treatment significantly higher values were observed for total root length, root surface area and root volume as compared to untreated control. The value of the total root length at different fractional groups was 78 to 86% of the total root length; 36 to 55 % of the total surface area was found up to 1- mm fractional range; and 86 to 90 % of the total root volume was found above 2- mm range. Also with magnetic treatment higher values for water-use efficiency, radiation-use efficiency and percentage of

intercepted photosynthetically active radiation were noticed as compared to control. The treatment yielded 3-8 % more seeds.



Effect on the growth and yield of sunflower with seeds exposed to static magnetic field before sowing



Pre-sown seed exposure to static magnetic field effects growth and yield of sunflower raised at farmer's field

Water uptake as well as enzyme activity were found higher during imbibing in magnetically treated seeds. The water uptake and spin-spin relaxation were greater in lag phase II and rapid hydration phase III in the treated seeds. Nuclear

magnetic resonance (NMR) relaxation time of seed water and its analysis indicated early appearance of structural/hydration water and greater amount of cytoplasmic bulk water and hydration water in the magnetically exposed seeds. Also in these treated seeds, molecular mobility of cytoplasmic bulk water and hydration water of macromolecules was higher. This might be responsible for early germination and higher seedling vigour of the treated seeds. A highly significant correlation between the relaxation time of cytoplasmic bulk water and the activities of the germination-related enzymes in general emphasizes the fact that this fraction of water is vital for metabolic activities taking place during germination.

# PROMISING TECHNOLOGIES

The study indicated that magnetic field application enhanced seed performance, and magnetic exposure gave best performance for rooting characteristics, growth characteristics and yield. Improvement in root parameters suggests that this technique can be used for growing crops under lesser water where enhanced root

growth will be useful in extracting moisture from the deeper soil layers.

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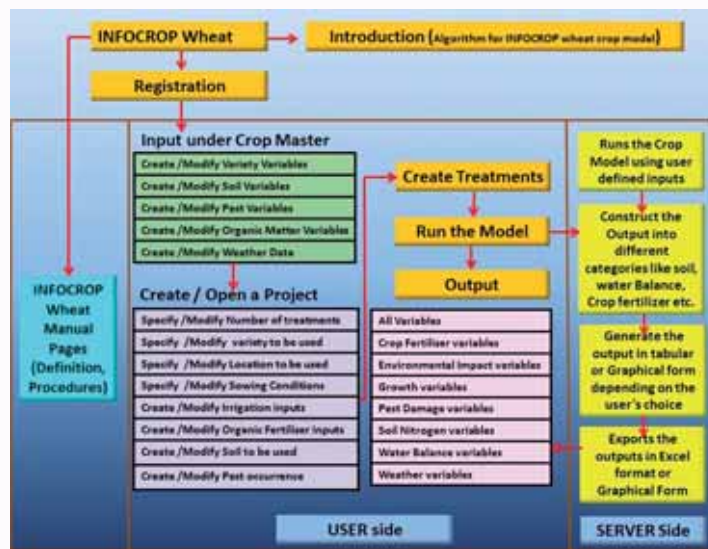
## Web-based crop model – INFOCROP Wheat

INFOCROP Wheat simulates wheat-crop growth on a daily basis, based on the weather, soil and variety and management practices, and facilitates simulation of growth and yield of the crop. It has been developed using Visual Studio Express, SQL Server, NET framework 4.0, and has been hosted at <http://InfoCrop.iari.res.in>.

The system has been designed using modular approach and has separate modules for input variables, management conditions

change on wheat growth and yield.

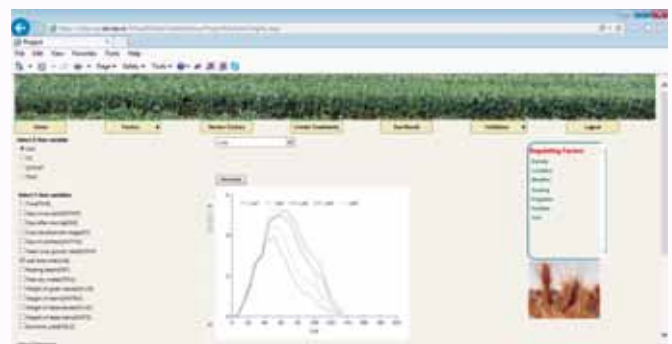
To upsurge the convenient use of the model and to improve its practical use, a tremendously easy menu-determined form has been established in the model. The clients are not required to have any knowledge in software design or development, and are given assistance to enter/choose necessary data. The interface of the present web-based model is written in Microsoft.Net and the back-end has C# models with databases in MS-SQL. In



Workflow and web interface of the INFOCROP Wheat



Results can be viewed in a tabular form by clicking on the respective treatment from the list appearing on the left panel of the screen



After running simulation, results can be viewed in a graphical form

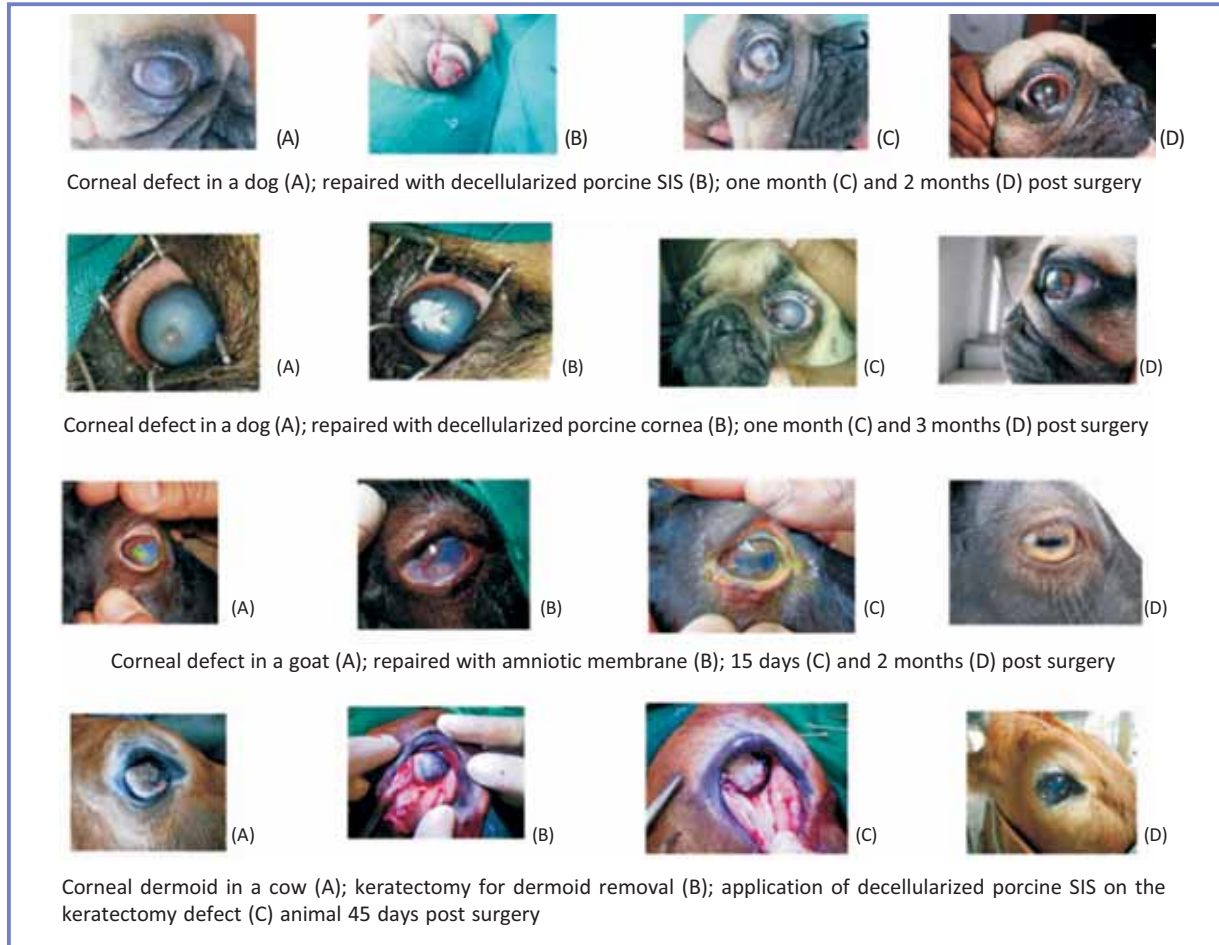
and output-results. Registered users can insert, edit/update and delete data within their private domain. System manager has only the administrative right to add data to public domain. And general users have the flexibility to run and simulate growth of wheat-crop and can have information on environmental impact, crop growth and yield, pest damage, soil-nitrogen dynamics, water balance and weather at a single-day interval or as defined by them. The system can be operated on mobile device to simulate wheat-crop yield and effects of climate

addition, a huge database of varieties, soils, pests and weather have been incorporated into the database master. INFOCROP Wheat demonstrates dissemination of crop simulation model system over universally available web-based application; providing tailored crop simulation-based decision support to end-users. Many of the pertinent and the existing crop growth processes observed under the field, which are pragmatic under real-world conditions, have been implemented in the crop simulation processes of this web-based INFOCROP Wheat.

## Biomaterials for corneal grafting in animals

Cornea is a transparent dome-shaped structure that helps transmit light onto retina hence is important for vision in animals.

intestine sub-mucosa and cornea and caprine amniotic membrane have been processed to make them acellular and less immunogenic. After proper sterilization and



Application of biomaterials for corneal defect repair in animals

Corneal surface defects owing to trauma or other causes are common in domestic and companion animals like cows, buffaloes, horses and dogs. Some conditions like corneal dermoid leaves a defect in the cornea even after surgery.

Protocols for use of biomaterials in surgical reconstruction of corneal defects have been developed. Porcine small

experimental trials, these biomaterials have been used successfully to repair corneal defects in many clinical cases.

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INFOCROP Wheat would aid as an architecture for upcoming web-based decision support systems using crop-simulation models; proposing a common user interface, common input data and opportunity to relate same data in different applications. The framework would guarantee quick dissemination of crop simulation to end-users.

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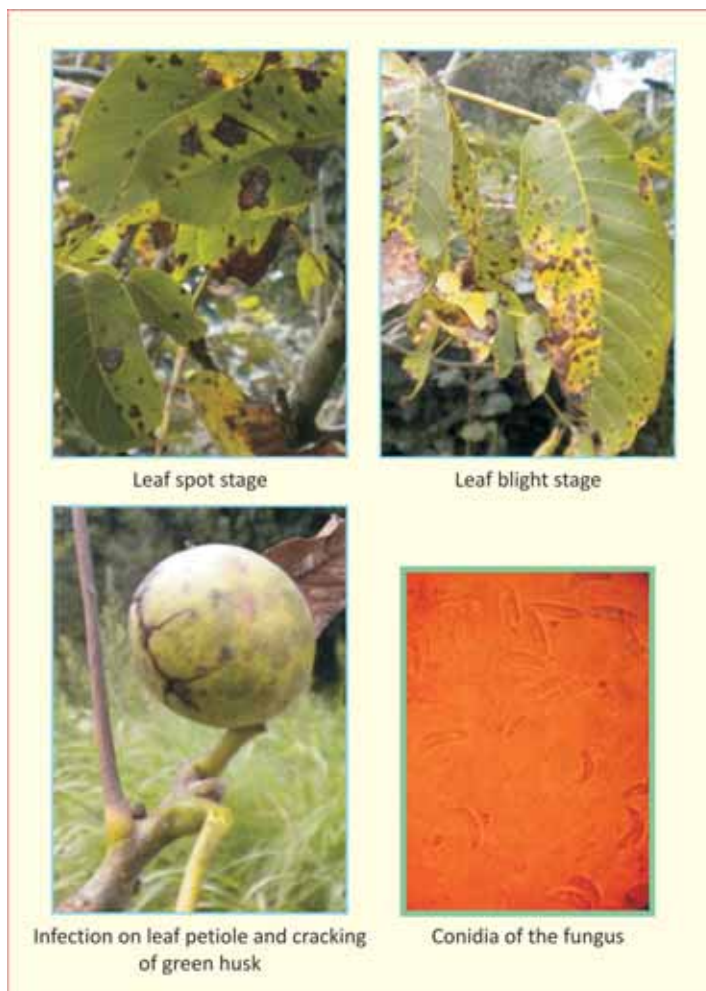
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### Leaf blotch on walnut in Uttarakhand

Nature has bestowed Uttarakhand with congenial weather and favourable soil for cultivation of walnut. Many projects have been launched there for area expansion under walnut. Most of the planting material is usually procured from Jammu and Kashmir, and many of the newly established orchards have come up to fruit-bearing stage.

Since 2014, leaf blotch of walnut has appeared in a severe form in walnut-growing areas, including Mukteshwar in Nainital district of Uttarakhand; and this is threatening walnut cultivation by causing premature leaf-fall in July-August at the fruit-development stage. Prolong wet weather during the period seems to be the most favourable factor for this disease. Many infected fruits either drop prematurely or remain intact on the trees but with shrivelled half developed forms and with very poor quality kernels. Consequently, the disease is causing huge yield losses to the crop.

In India, this disease was first observed in almost all walnut-growing localities of Kashmir. Appearance of leaf blotch, caused by *Marssonina juglandis* (Lib) Magn. [Tel.: *Gnomonia leptostyla* (Fr.) Ces. and De Not.], is a new



threat to walnut in the region. Asexual stage of fungus (acervuli) appears as the small pin-head-like structures in August on the abaxial surface of the diseased leaves. Fungal conidia are variously shaped — comma, straight, falcate or with one end rounded and the other pointed. They are hyaline and 1-septate with prominent oil globules and measure 10 to 22µm × 2-4µm.

This disease affects leaves, young fruits and nuts. On leaves, it initially produces round reddish brown spots with greyish brown centre; which later develop into a large blotch, covering major part of the lamina. Infection gradually spreads to whole leaf and extends towards leaf petiole, which

gives blighted appearance to trees. On young shoots of the current growth, lesions are oval, sunken, greyish brown, surrounded by reddish brown margins. Blotch also causes depressed circular to irregularly circular dead spots on the husk of the nuts, and early infection on the nuts results in premature fruit drop. Green husk cracking can also be seen on the susceptible cultivars at the later stage.

The leaf blotch of the walnut can be managed successfully by spraying Captan (0.25%) or Mancozeb (0.30%) or copper oxychloride (0.30%) at (i) leafing stage i.e. when leaf starts unfolding, (ii) at full-leaf stage, and (iii) two weeks after the second spray. None of the walnut cultivars have exhibited complete resistance against the disease.

ICAR NEWS wishes all readers and contributors

*A Very  
Happy New Year*

2016

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## Cashewnut – a valuable nutritional package

Cashew (*Anacardium occidentale*, family Anacardiaceae) was introduced into India by the Portuguese in the beginning of 16th century. Among dry fruits, cashew is the only nut commercially grown in the tropical region for its delicious and nutritious kernels. Cashew is grown mainly in Kerala, Karnataka, Goa and Maharashtra along the West Coast, and in Tamil Nadu, Andhra Pradesh, Odisha, and West Bengal along the East Coast, and is also spreading in the non-traditional areas of Chhattisgarh, Jharkhand, Gujarat and North-Eastern Region. GIS- based delineation of cashew cultivation has indicated that it can be grown successfully away from coastal belt and in areas with 600 to 1,500 mm rainfall per annum. Its productivity is higher in frost-free regions where minimum temperature ranges from 10 to 22 oC. Globally, India not only occupies largest area under cashew cultivation but is also its largest processor, exporter, importer and consumer. India lags in productivity of raw cashewnut (706 kg/ha) compared to the Philippines (5,103 kg/ha), Peru (4,987 kg/ha), Vietnam (3,692 kg/ha), Mexico (3,013 kg/ha), Nigeria (2,500 kg/ha) etc.

### Improving cashew productivity

#### Quality planting material:

Instead of growing seedling plants of cashew, softwood-grafting technique has been standardized and found ideal for commercial multiplication of cashew varieties. Application of microbial inoculants, 5 g each of *Azospirillum*, *Pseudomonas*, phosphate solubilizing bacteria and 2.5 g of arbuscular mycorrhizal fungi, per poly-bag containing 2 kg of potting mixture for cashew rootstocks resulted in production of healthy grafts in the nursery, and later their better establishment in the field.

**High-yielding varieties:** Commercially cultivated varieties are: Bhaskara, Selection 2, Vengurala 4, Vengurala 7, VRI 3, BPP 8, Ullal 3, Priyanka, Dhana, Kanaka, Chintamani 1, Chintamani 2, Jhargram 1, Jhargram 2, Jagannath and Balabhadra.

**High- density planting:** Planting at a spacing of 4m × 4m with 625 plants/ha has been found better than normal spacing of 8m × 8m with 156 plants/ha; resulting in an increased yield by 2.5 times over the wider spacing during initial ten years. Optimal fertilizer requirement for high-density planting system is 50 % of the recommended dose of fertilizers (500 g N and 125 g each of P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O per tree/year). Further, for improving productivity, plants should be irrigated during fruit-set to fruit maturity; from December to January 7 L/day and 9 L/day from February to March, respectively, which is equivalent to 20 % of the evaporative demand.



#### Main factors for low productivity

- Large area under cashew plantations is of seedling origin
- Most of the cashew plantations are on the degraded lands and under rainfed conditions
- Poor adoption of latest cashew production technologies

#### Soil -and- water conservation techniques:

Cashew has generally been grown on wastelands having poor fertility, prone to soil erosion and problem of moisture deficit during reproductive phase. Thus, using soil-and-water conservation (SWC) techniques, like crescent bund and staggered trenches with coconut husk burial, conserved soil moisture, lowered annual

runoff / soil loss and increased cashewnut yield by 30-40 %. Terrace with crescent bund for individual cashew-tree base is the best SWC measure for cashew grown on slopes.

#### Nutritional content

Cashew-kernels are rich in proteins, carbohydrates and fat; along with some minerals and vitamins. Cashew-kernel gives more energy (611 K cal/ 100 g) as compared to average animal food (147-272 K cal/ 100 g) and fish (234 K cal/ 100 g). Kernels contain high percentage of fat, but 82% of its fat is unsaturated fatty acid. Major fatty acids in the kernel are oleic acid (73.4%), followed by linoleic acid (11.9%). The principal polyunsaturated fatty acid found in cashew is Omega-6, important for health.

The protein in the kernel comprises all the essential amino acids such as Glutamic Acid (28.0%), Leucine (11.93%), Aspartic Acid (10.78%), Arginine (10.30%), Serine (5.76%), Glycine (5.33%), Valine (4.53%), Phenylalanine (4.35%), Iso Leucine (3.86%), Proline (3.72%), Lysine (3.32%), Tyrosine (3.20%), Alanine (3.18%), Threonine (2.78%),

## Myths related to the nut

- **Increases body cholesterol:** Cashewnuts are rich in good fats and have **zero cholesterol**.
- **Increases heart problem:** Eating cashewnuts help **lower low density lipoprotein (LDL), the bad cholesterol and triglyceride**, making one's heart healthy.
- **Accelerates high blood pressure:** Cashewnuts are **low in sodium and high in potassium** and thus keep blood pressure under check.
- **Enhances stone formation:** Studies suggested that daily intake of cashewnuts can **reduce risk of developing gallstones** up to 25%. While owing to over-eating, oxalates in cashew become concentrated in the body fluid, crystallize and cause health problems in people with pre-existing kidney or gall bladder problems.
- **Prone to diabetes:** Monounsaturated fats found in cashews can **reduce triglyceride levels in diabetics**; protecting from further complications.

content, from 1 to 3% and non-reducing sugars from 2.4 to 8.7%. Starch content ranges from 4.6 to 11.2% and oil content also shows a wide variability from 34.5 to 46.8%. The nut is also good in many essential vitamins, pantothenic acid (vitamin B 5), pyridoxine (vitamin B 6), riboflavin and thiamin (vitamin B 1), and a good proportion of vitamin E. In addition, the kernels also contain a small amount of zeaxanthin, an important

Histidine (1.81%), Tryptophan (1.37%), Methionine (1.30%) and Cystine (1.02%). Cashew-kernels are comparable with casein in terms of protein efficiency ratio (PER). The PER of cashew-kernel protein is 3.2 which is comparable with milk-casein.

flavonoid antioxidant, which is selectively absorbed into the retinal macula lutea of eyes.

Analysis of cashew-kernels from different regions of India have revealed that there are variations in reducing sugar

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## Milkman's screen touch to deliver feed formula

Android Touch Screen is featuring door-step delivery of the Feed Chart for crossbred cows and buffaloes for the South Asian dairy farmers in pursuit to be the cantilever between feed mangers and milk bowls. This operating system nurtures conventional bovine-feed ingredients – from dry roughages, green fodder, and concentrates of the sub-continent – commensurating rural milk production from cow (body weight: 400 kg) and buffalo (body weight: 450 kg), yielding five to twenty litres, spaced at an increment of 2.5 litres. The distributive proportion of the classified ingredients has been made in accordance with the relative availability of high, moderate and scarce vegetation. The supportive literature delineates user guide, compositional details confirming to real-time barnyard stocks and feed-back portal for advice and applicability, adding efficiency to conventions.

The device has been applied to reach aspirations of the tech-savvy generation-next small and middle holder dairy



farmers, who report to co-operative milk societies or private entrepreneurs. This conduit of information delivery is likely to step-in as model accelerator to retain dairy farming as a profitable venture. Link: <https://play.google.com/store/apps/details?id=nianp.exempl.feedchart&rdid=nianp.exempl.feedchart>

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## New chemotypes of sweet basil – Rich source of methyl chavicol

*Ocimum basilicum* is a herb of family Lamiaceae, used for culinary and ornamental purposes. Originated in tropical Asia, sweet basil is a classic culinary herb of Mediterranean and South-east Asian cuisines. In India, basil is considered sacred and is cherished in almost every Hindu house. For centuries, basil leaves have been a very popular spice. Widely cultivated commercially and in kitchen-gardens, sweet basil may occasionally escape, though it is likely ephemeral near cultivation sites. Its habitats are anthropogenic meadows and fields as it is an essential ingredient in many cooking traditions.

Many lines of sweet Basil were evaluated for fragrance, flavour, leaf colour, leaf size, herbage yield and oil yield. Oil content in dry herbage varied from 0.07% to 1.92%. Considering, constant human demand for new flavours, new lines are being bred. On account of high degree of variability in economic characters, several accessions were evaluated from the existing populations.

Easily grown tender leafy herb is cultivated for its aromatic leaves containing essential aromatic oils. Its essential oils are used extensively since many years in food products, cosmetics, perfumery, dental and oral products, and hygiene and cleaning products.

Basil essential oils and their principal constituents are also found to exhibit antimicrobial activity against a wide range of Gram-negative and Gram-positive bacteria, yeast and mold; basil is also used as a local anaesthetic and antiseptic. Besides, its strongest medicinal use as an antimicrobial, it is very high in vitamins and minerals. Basil can be used as an ointment for insect bites, is effective as a mosquito-repellent and its oil is applied directly on the skin to treat acne.

Following international codex and market preferences, a comparative study was undertaken to assess variability of phenotypic and chemotypic characters present among the genotypes. Phenotypes are found variable in terms of leaf (colour, shape and type), panicle (colour and type) and anther (colour). Lower leaf colour is green with purple spots in all the lines, except DOB 1, DOB 5, DOB 6 and DOB 10, where it is green. Upper leaves are observed greenish except DOB 2, DOB 3, DOB 7 and DOB 8, which are purplish. Panicle colour is purple in all genotypes, except DOB 4 and DOB 6, which have greenish-purple panicle. The panicles are either bunched (DOB 2, 3, 5, 10 and PG 2), individual (DOB 1, 4, 6, and 7) or intermediate type (DOB 8). Branches are smooth, except of DOB 2 and DOB 7, where they have sparse hairs. Leaf is serrate in DOB 6, DOB 7 and DOB 8 while in the rest it is smooth. Anther colour is orange in DOB 7 and in the rest it is whitish.

A wide variability in chemotypic characters among



Morphological variations in leaves and vein colour



Morphological variations in inflorescences, leaves and branches

# NATURAL RESOURCE MANAGEMENT

Major volatile compounds in fresh basil-leaf oil expressed as a relative percentage

Lines	Methyl chavicol	Linalyl formate	Methyl eugenol	Naphth-alene	Eugenol
Anand Basil 1	22.34	13.93	1.11	7.6	35.37
DOB 1	62.28	13.58	1.02	4.09	4.15
DOB 4	7.38	6.96	62.57	2.28	-
DOB 5	63.44	12.23	1.72	3.83	3.51
DOB 6	1.07	1.61	3.22	20.88	31.62
DOB 7	53.83	-	24.41	1.44	4.53
DOB 8	64.25	1.92	19.96	3.97	-
DOB10	86.66	-	2.25	2.04	-
DOB 3	87.13	-	2.26	2.17	-
DOB 2	86.52	1.15	2.39	2.27	-

these accessions offers opportunity for production of basil to meet specific market requirements for particular compounds (linalool, eugenol, methyl chavicol, methyl

cinnamate, or methyl eugenol). DOB 8 and DOB 7 are observed rich in both methyl chavicol and methyl eugenol, while DOB 6 is found rich in eugenol and naphthalene. DOB 3, DOB 10 and DOB 2 are rich in methyl chavicol.

Among the seedling selections from the population, DOB 2 has highest content of total chlorophyll and carotenoids (2.023mg/litre and 16.20 mg/litre, respectively), followed by DOB 6 (1.89 mg/litre and 14.89 mg/litre, respectively). The maximum leaf area has been observed in natural crosses: DOB 8 (189.47cm<sup>2</sup>), followed by DOB 7 (141.92 cm<sup>2</sup>). Overall these diverse selections have been found superior from the rest of germplasm in leaf yield and oil quality.

**Parmeshwar Lal Saran, Vandana Tripathy, Ram Swaroop Jat and Deepak Sharma**

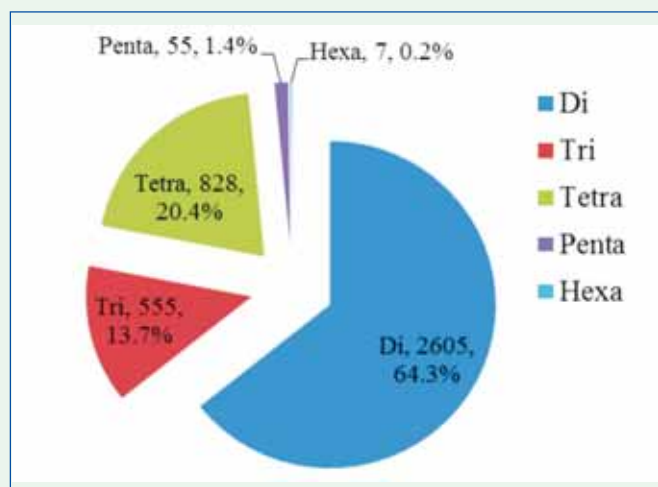
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## SSR markers identified in *Chitala chitala* through third generation sequencer

SSR markers are being increasingly used in molecular genetic studies in the recent years. With the advent of next-generation sequencing technologies, marker identification in genomes of non-model organisms has revolutionized the field of genomic research in humans, plants, animals and microorganisms. Pacific Biosciences Single Molecule Real-Time sequencer, PacBio RSII, was worked on to generate SSRs for fish *Chitala chitala*. Two SMRT cells were used for sequencing a 1.5 Kb SMRT bell library of genomic DNA, which generated 101.9 Mb polymerase read data. Primary analyses and removal of adapter sequences have generated 37.8 Mb read of insert data, which contain 27,562 Circular Consensus Sequence (CCS) reads with >99% accuracy. Assembly of these CCS reads

has generated a total of 9,746 high quality sequences having 776 contigs and 8,970 singletons, which when analysed further for SSR repeats have resulted in the identification of 4,050 different SSRs motifs (Di- 2605; Tri- 555; Tetra- 828; Penta- 55 and Hexa- 7) in 2,129 sequences. A total of 780 sequences were found to have more than 1 SSR, while 612 sequences having 1,941 SSRs were found present in compound form and/or in imperfect repeat units; 2,109 perfect repeats present in 1,517 sequences consist of 1,353 di, 412 tri, 300 tetra, 43 penta and 1 hexa repeat motifs.

Additionally, the assembled sequences were further annotated for prediction of protein coding genes using homology based search against the NCBI non-redundant database. Analyses resulted in the annotation of 3,097 (31.7%) protein sequences, which were further categorized into respective gene ontology (GO) terms based on the annotation. The search of 52 randomly selected gene annotations in the genome database of *Danio rerio* resulted in the identification of their location on 23 chromosomes of *D. rerio* genome database, which strengthens random sharing of *C. chitala* genomic DNA.



SSR repeat types distribution in *Chitala chitala*

**Vindhya Mohindra, Rajeev K. Singh and J.K. Jena**

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## PROFILE

# ICAR-Indian Institute of Soil Science, Bhopal



ICAR-Indian Institute of Soil Science (ICAR-IISS) (an ISO 9001:2008 certified research institute on soil science) came into existence as a follow-up of the recommendations of a Working Group of the ICAR and a Task-force Committee, which felt that fundamental research in many aspects of soils was not being taken up by any of the then ICAR institutes.



The Institute formally started functioning on 16 April 1988 from another ICAR Institute (CIAE, Bhopal), located at G.T.B. Complex, T.T. Nagar, Bhopal; later shifted to a rented building at M.P. Nagar, Bhopal, and then in 1995 shifted to its present campus at Nabibagh, Berasia Road. Some of the major areas in which the institute conducts research are: improving soil quality, input-use efficiency, carbon sequestration, GIS-based fertility mapping, conservation agriculture, organic farming, microbial diversity and biofertilizers, soil genomics, bio-fortification, waste management, climate change mitigation strategies and nano fertilizers.

The ICAR also shifted the Project Coordinating Units of six AICRPs concerned with soil and nutrient management to the IISS, Bhopal.

### SPECIFIC OBJECTIVES

- To carry out basic and strategic research on soils; especially physical, chemical and biological processes

related to management of nutrients, water and energy.

- To develop advanced technologies for sustainable systems of input management, which are most efficient and least environment polluting.
- To develop database repository of information on soils in relation to quality and productivity.
- To develop expertise and backstop other organizations engaged in research on agriculture, forestry, fishery and on various environmental concerns.
- To exchange information with scientists engaged in similar pursuits through group discussions, symposia, conferences and publications.
- To collaborate with the State Agricultural Universities, National, International and other Research Organizations to fulfill above objectives.

### INFRASTRUCTURE

The institute has 50 hectares of research farm with the required facilities for conducting field experiments. It has Agricultural Knowledge Management Unit (AKMU) equipped with more than 120 network computers and also has dedicated high speed connectivity of 100mbps through natural knowledge network (NKN). The AKMU also maintains institute's website. The IISS has a well



Information centre

established library with more than 3,500 books and 80 international and national journals.

## SALIENT ACHIEVEMENTS

### ENHANCING SOIL FERTILITY THROUGH INPUT/NUTRIENT MANAGEMENT

**IPNS system for soybean-wheat cropping:** Realizing the low soil fertility status of Malwa and Vindhyan plateau regions of Madhya Pradesh where soybean-wheat cropping system is being practised, a specific Integrated Plant Nutrient Supply (IPNS) recommendation has been developed to make the system more profitable. The recommendation is 50% recommended rate of NPKS (12.5 kg N, 30.0 kg P<sub>2</sub>O<sub>5</sub>, 10 kg K<sub>2</sub>O, and gypsum 55 kg/ha) + 5 tonnes FYM/ha + *Rhizobium* (750 g/ha) to soybean and 75% of the recommended NPKS (90.0 kg N, 45.0 kg P<sub>2</sub>O<sub>5</sub>, 25 kg K<sub>2</sub>O and gypsum 83 kg/ha) + phosphate solubilizing bacteria (PSB) (3.5 kg/ha) to wheat. The technology is viable since many of the farmers can produce 5 tonnes FYM per annum. But, for the farmers who cannot have FYM, 100% NPKS<sub>Zn</sub> to soybean and 100% NPKS to wheat



RT-PCR



DNA isolation bench

can have higher productivity from the system.

**Mechanical harvest-borne residue management:** Field burning of crop residues is undoubtedly a wasteful practice. Soil incorporation of wheat residue plus N supplementation through FYM at 28 kg N/ha (approx. 4 tonnes FYM/ha) along with 25 kg P/ha for rainfed

### Micro- and Secondary- Nutrients and Pollutant Elements in Soils and Plants

- Collected 127,812 GPS -based surface soil samples from 217 districts, covering Assam, Bihar, Gujarat, Punjab, Haryana, Jharkhand, Madhya Pradesh, Tamil Nadu, Odisha, Maharashtra, Uttarakhand, Uttar Pradesh and West Bengal, and analyzed them for available Zn, Cu, Fe, Mn, B, Mo and S. About 43.4, 14.4, 6.1, 7.9 and 20.5% soil samples were deficient in Zn, Fe, Cu, Mn and B, respectively.
- Physiological as well as biochemical changes including metabolism of different crops as influenced by low and excess supply of micronutrient and pollutant elements were studied.
- The deficiency symptoms of various micro- and secondary-nutrients were identified, and critical limits for nutrients for different crops have been established.
- Several amelioration techniques have been standardized for mitigation of micro- and secondary-nutrient deficiencies.
- Geo-referenced profile soil samples were collected from fields where sewage effluent was used for irrigating vegetable crops, cereals, forage etc. and also from the fields irrigated with well water. Micronutrients (Zn, Fe, Mn, Cu) and heavy metals (Co, Cd, Cr and Pb) were higher on the surface and concentration declined with soil depth.

The Institute Celebrated International Year of Soils – 2015



2015  
International  
Year of Soils

The 68th UN General Assembly recognized the year 2015 as the International Year of Soils. The institute has celebrated with fervor this very important year. Noteworthy in this is the institute's active association with the

Government's Soil Health Card programme.

The institute has also come out with a mini laboratory "Mridaparikshak", which can aid in the preparation of soil-health cards. It has also prepared soil-fertility maps, depicting availability of organic carbon, available nitrogen, phosphorus, potassium, zinc, iron, manganese and copper, and pH and electrical conductivity (EC), of 173 districts of India. Institute has developed enriched compost and generated efficient biofertilizers, balanced fertilization and Integrated Plant Nutrition Systems for different crops, and has also generated and tested efficiency of tillage systems.

An important event in this series was the celebration of World Soil Day on 5 December 2015, wherein several soil-health cards were distributed to farmers by the Hon'ble Chief Minister of Madhya Pradesh, Shri Shivaraj Singh Chauhan.



soybean and 68 kg N + 30 kg P/ha for irrigated wheat (3 irrigations including presowing irrigation) was more effective and profitable. Wheat residue incorporation resulted in 20-22% higher yield of soybean and 15-25% higher yield of wheat as compared to residue burning.

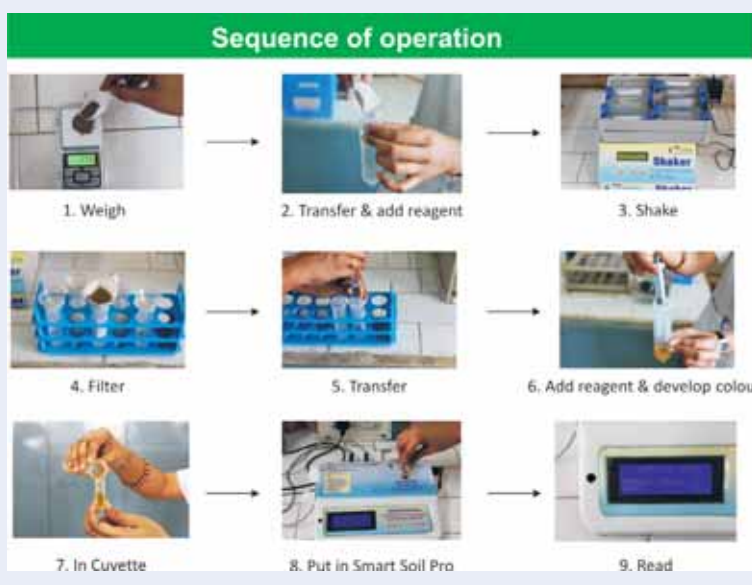
**P management in Vertisols:** P is sorbed as an outer-sphere surface complex by weak bonds in smectite dominant clayey Vertisols. It is available to soybean and wheat crops due to rhizosphere effect. It is recommended that fertilizer P may initially be applied in amount equivalent to about 1.5

times of the P removal by the crop, and in subsequent years, P may be applied in amount equal to that removed

Mini Soil Lab – Mridaparikshak

It is a digital mobile quantitative mini lab to provide soil-testing service at farmer's doorstep. It comes with 'Smart Soil Pro', an instrument for determining soil parameters and displaying fertilizer nutrient recommendations.

Mridaparikshak gives crop and soil specific (alluvial soils, red soils and black soils) fertilizer recommendations directly to farmer's mobile through SMS.



by crop harvest products. By doing so Olsen-P values would remain almost at the same level or at slightly higher levels.

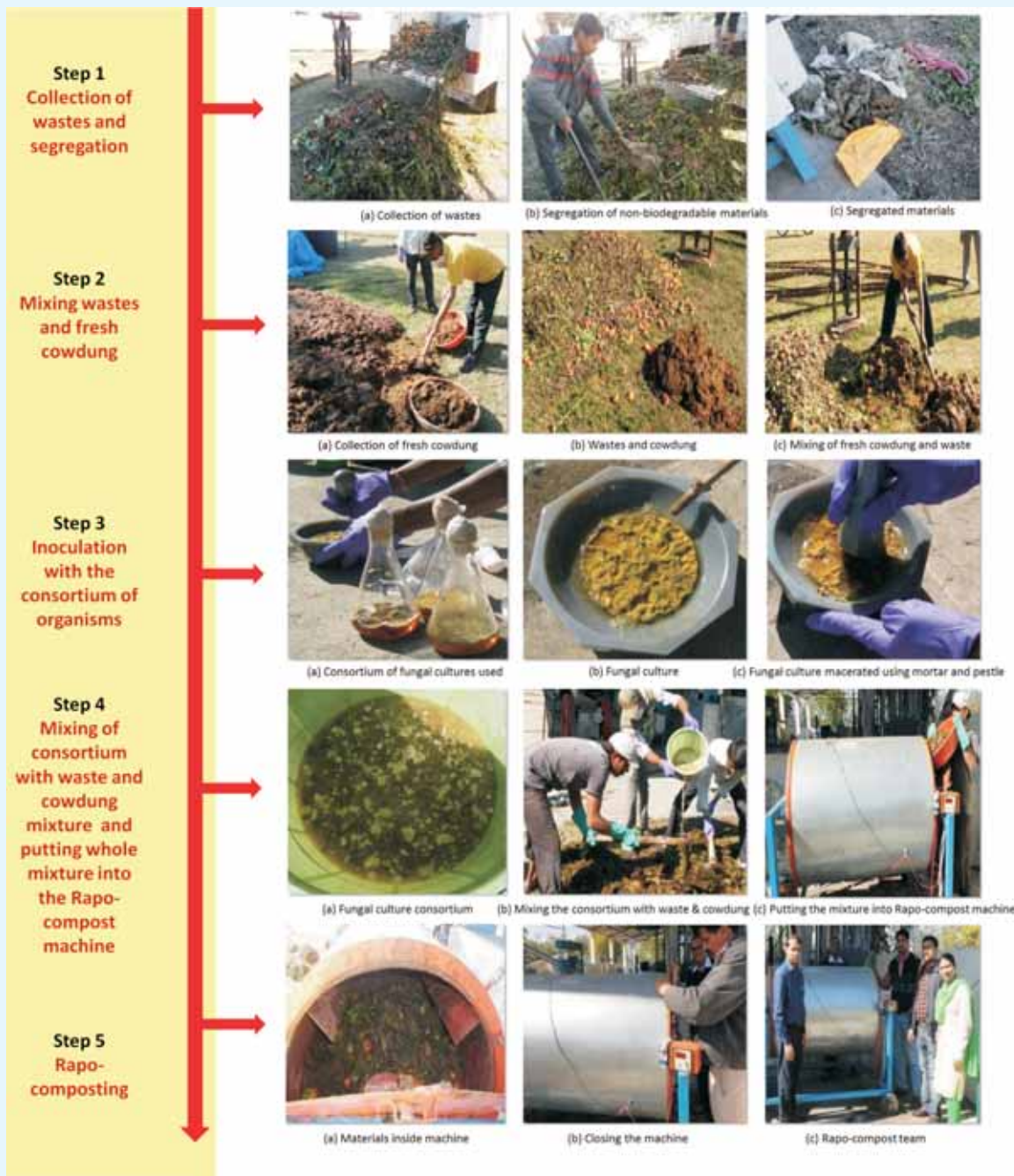
**Enriched compost production:** Enriched composting is a process where ordinary compost is fortified with necessary plant nutrients. Different types of enriched composts have been developed.

**Phospho compost:** It is developed using phosphate solubilizing microorganisms (*Aspergillus awamori*, *Pseudomonas straita* and *Bacillus megaterium*), phosphate rock, pyrite

and bio-solids to enhance manurial value. The average P content of this varies from 2-3.50 % and cost incurred

## Rapo-Composting Technique for Recycling Farm and City Wastes

- This technology can prepare compost within 1-1.5 months from domestic and vegetable wastes.
- Compost manurial value improves, such as total nitrogen (1.5 to 2.3 %) and total phosphorus (0.5 %  $P_2O_5$ ).
- Large quantities of waste materials can be recycled back to the field after converting them to quality manure.
- Rapid composting kills all plant-disease producing organisms.
- Insects do not survive at high temperature during composting process.
- Most weeds and weed-seeds are also killed.



to obtain one kg  $P_2O_5$  through this is around ₹9.00 as compared to ₹16.0-17.0 supplied through single superphosphate (SSP) or diammonium phosphate (DAP).

**Phospho-sulpho-nitro compost:** In this, urea at 0.5-1% (w/w), rock phosphate at 12.5% (w/w) and pyrite at 10% (w/w)

are added into the composting mixture. The average nutrient value of this is 1.5-2.3% N and 3.2-4.2% P. This compost at 5 tonnes/ha can replace 25% of the recommended fertilizer dose in soybean-wheat system.

**Spent wash amended compost:** This is prepared by heap

method but water requirement is met by spent wash (major waste material from distillery industry). The nutrient value of amended compost is 1.37% N, 1.30% P and 1.82% K.

**Enriched organo - mineral compost:** In this, crop residues are mixed with cow- dung, low-grade rock phosphate, waste mica and mineral gypsum. The nutrient value of the compost is 1% N, 1.0% P, 2.1% K, 1.7% S and addition of 1 tonne of this compost can supply 10 kg N, 10 kg P, 21 kg K, and 17 kg S to the crop.

**Microbial enriched municipal solid waste (MSW) compost:** To make use of the untapped nutrient value of the municipal solid waste as well as to manage environmental pollution issues, this microbial enriched compost technology has been developed. For making

1,000 kg microbial enriched compost, 1,600 kg waste material, 320 kg fresh cow-dung and 21kg urea are required. The nutrient value of this compost is 0.73% N, 0.79% K with 11.3% total organic-carbon content.

**Biofertilizers:** Biofertilizers are preparations of living organisms that are useful for promoting plant growth. Some selected technologies on biofertilizers developed are as follows.

**Mixed consortium biofertilizers:** Mixed biofertilizers (BIOMIX) containing a consortium of N fixers, P solubilizers and plant growth promoting rhizobacteria (PGPR) to promote crop growth of cereals, legumes and oilseeds has been developed. Use of BIOMIX could save 25% of N and P fertilizers.

### Soil-Test Crop Response

Considerable agronomic and economic benefits were accrued when farmers applied fertilizer nutrient doses based on the soil test. Studies on soil biological parameters in guar-wheat cropping system under arid condition revealed that soil microbial biomass, dehydrogenase activity and organic carbon were higher in STCR (soil-test crop response)-based nutrient application as compared to general fertilizer recommendations. Ready reckoners in the form of fertilizer prescription equations have been developed for profitable use of fertilizers based on soil-test values, and the same has been demonstrated through various multi-location / verification follow-up trials as well as front-line demonstrations. STCR prescription equations are also being used for development of customized fertilizers.

#### Economic analysis of fertilizer doses associated with different yield targets

An appraisal of the effect of nutrients (NPK) applied on crop yield and benefit: cost ratio (BCR) with NPK alone and under IPNS showed that out of 66 crop ×target combinations, the BCR was between 1 and 2 in 35 % cases and between 2.1 and 3.0 in 62% cases; in 3% cases, BCR was above 3. Irrespective of the crops, higher yields were recorded at higher yield targets over lower targets coupled with higher net returns and BCR.

#### Fertilizer recommendations for fixed cost of investment and allocation under resource constraints

A new dimension to the value of utility of soil testing has been added by the concept of fertilizer application for targeted yield demonstration in farmers' fields by choosing yield target at such a level so that the cost of fertilizer requirement becomes more or less same as what is being practised by the farmers.

#### Expert system developed by AICRP (STCR)

An expert system has been developed, which calculates nutrients required for specific yield targets of crops based on the fertility on farmers' soil. It is accessible on Internet (<http://www.stcr.gov.in/>). Also developed a Decision Support System for Integrated Fertilizer Recommendation (DSSIFER 2010) for Tamil Nadu state, encompassing soil-test and target-based fertilizer recommendations through Integrated Plant Nutrition System. If site specific soil-test values are not available, database included in the software on village fertility indices of all the districts of Tamil Nadu will generate soil-test based fertilizer recommendation. Besides, farmers' resource-based fertilizer prescriptions can also be computed.



**Enhancing biofertilizer efficacy:** Bioinoculants (*Azospirillum*, *Azotobacter*, PSB) mixed with well decomposed FYM/ vermicompost in 1:25 ratio and incubated at 30% moisture for a week improved microbial population 2-15 fold. Demonstrations revealed a yield increase of 8-12% in above ground and 25-30% in below ground vegetation with this formulation.

**Liquid biofertilizer formulations:** Liquid biofertilizer formulations are found ideal to increase shelf-life of biofertilizers. In an evaluation period of 360 days, Liquid *Rhizobium* medium (LM3) maintained  $2.710 \times 10^8$  cfu/ml, Liquid PSB medium (LM3) maintained  $1.612 \times 10^8$  cfu/ml, and *Azospirillum* liquid medium (LM2) maintained  $4.395 \times 10^8$  cfu/ml even after 360 days.

**Oleoresin-coated urea fortified with nano-particles:** To manage micronutrient requirement of the crop, a protocol has been developed for fortifying urea with a consortium of nano-particles of zinc, copper, iron, and silicon by using oleoresin. This product contains 0.438g N, 2.2 mg Zn, 1.10 mg Fe, 0.66 mg Cu and 1.06 mg Si per gram of coated urea product. Application of this fortified urea product at 200 kg/ha would supply 87.68 kg N, 440g Zn, 220g Fe, 132g Cu, and 212g Si to the crops.

**Nano-rock phosphate:** Development of this is an effort towards commercial utilization of low-grade rock phosphate; available in India as a direct phosphatic fertilizer. Experiments conducted in four soils (Vertisols of Bhopal,

Alfisols of Betul, Inceptisols of Ludhiana, and Aridisols of Jodhpur) revealed that crop utilization of P from nano-rock phosphate is on par with that of normal sized SSP in Vertisol and Inceptisol; and biomass growth of maize could be enhanced with nano-rock phosphate. Two types of rock-phosphate materials SRP (Sagar Rock Phosphate) and HGRP (High Grade Rock Phosphate, Udaipur) were tested. Nano-rock phosphate (size:110.1 nm) prepared from SRP showed yield advantage of 20% in Vertisols, 61% in Alfisols, 31% in Inceptisols, and 14% in Aridisols over normal sized rock phosphate (size:13.4  $\mu$ m) from SRP. Further, nano-rock phosphate (size:70.89 nm) prepared from HGRP showed an yield advantage of 31% in Vertisols, 88% in Alfisols, 27% in Inceptisols, and 15% in Aridisols over normal sized rock phosphate (size:12.9  $\mu$ m) from HGRP.

**Nano-zinc oxide:** A protocol for zinc delivery to plants through seed coating with nano-Zn oxide particles has been developed, which can be successfully used for production of customized seed for zinc- deficient areas. Nano ZnO can be used as a direct source of Zn for crops. Nano Zn particles at relatively lower level (0.28ppm) enhanced growth of maize compared to normal  $ZnSO_4$  (0.5ppm); further, seed treatment with nano-ZnO was found promising to meet Zn requirement of the crop and there was no toxic effect observed on seed germination and further plant growth.

## ASSESSING AND SUSTAINING SOIL HEALTH

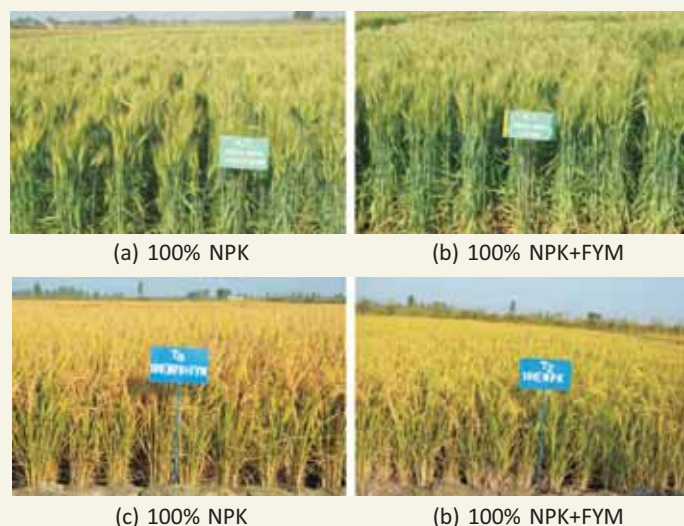
**Soil fertility maps based on nutrient index (NI):** Nutrientindex based soil fertility maps of the country has

### Long-term Fertilizer Evaluation

Long-term experiments have generated large and valuable information, which is being used for developing crop and location-specific strategies for sustainability in intensive agriculture.

Analysis of data generated on physical, chemical and biological condition of soils across India revealed that there is no adverse effect of balanced fertilizer application on soil properties; rather, improvement was noticed with balanced and integrated nutrient management practices.

To evaluate soil quality, different soil indicators have been chosen in different soils. In Inceptisols, available nutrient status is found prime indicator; in Vertisols, soil physical properties like bulk density, soil aggregate and infiltration rate appear important; and in Alfisols, soil pH, Ca, Mg and K status are important indicators. However, in all the types of soils, one or other forms of soil-organic carbon have been as one of the soil indicators.



Impact of balance (a, c) and integrated nutrient management (b,d) on crop growth (Wheat at Jabalpur and rice at Pantnagar)



### Soil Biodiversity – Biofertilizers

- Major Programme on “Genetic Diversity of Rhizobia of Indian Soils”, involving >1,200 rhizobial isolates of 20 major legumes for 16S rRNA diversity, identified effective strains for different soils, including arid and hyper-arid zones. Proteomics of rhizobia in acid soils showed unique features conferring adaptation to acid soils.
- Major Programme on “Soil Metagenomics for Soil Health Assessment” indicated that organically farmed soils showed higher eubacterial species richness and diversity and key functions. Only extremely high inputs of fertilizers (5× recommended) and pesticides (2×) adversely affected diversity and function. Actinobacteria are key phyla involved and are the key indicators of improved soil health.
- Effective biofertilizer strains for legumes, cereals, millets, oilseeds, fibre and vegetable crops have been isolated, characterized and deployed all-over India; biofertilizer packages demonstrated particularly in disadvantaged and tribal areas, eastern India and NEH region showed saving of 25% in chemical fertilizers, and improved NPK-use efficiencies, produced quality and improved livelihood.
- Novel microbial inoculants developed based on Actinomycetes and *Arthrobacter*.
- Apple white root-rot was controlled by plant-growth-promoting rhizobacteria *Bacillus licheniformis* formulation and yield increases were demonstrated in Himachal Pradesh.
- Liquid biofertilizer formulations developed are giving excellent response in pulses, cereals and oilseeds. There is increased demand for liquid inoculants from farmers in South and West India.
- The research efforts contributed to significant awareness and increase in biofertilizer production in the country; which grew at an annual growth rate of ~14% in the last ten years.

estimated that about 57% of the soils are low in available nitrogen, 36% are in medium category and 7% are in high category for available nitrogen. For available phosphorus 51% of the soils are low in available phosphorus, 40% are in medium category and 9% are in high category and for potassium 9% of the soils are low in available potassium, 42% are in medium category and 49% are in high category.

**Conservation tillage for soybean-wheat cropping system:** An improvement in soil water retention, soil organic carbon concentration, soil aggregation and stability was observed under conservation tillage treatments and yield advantage was observed in soybean grown under reduced tillage system. Conservation agricultural (CA) practices have been found to add 2.30 tonnes/ha wheat residues (0.90 tonnes C/ha) in Vertisols compared to 0.70 tonnes/ha (0.30 tonnes C/ha) in farmers’ practices under soybean-wheat system indicating more C in soil through CA.

**Broad bed furrow (BBF):** It consists of semi-permanent broad beds, approximately 100-cm wide, separated by about 50 cm wide and 15-cm deep furrows with a rolling slope of 0.4-0.7% for safe drainage of excess water; crops can be grown on beds in 2-4 rows. The system is a good option for cultivating crops in waterlogged areas; beneficial for high productivity, improved drainage, and also for *in-situ* moisture conservation. An yield increase of 11-18% in BBF system was noticed compared to that of flat bed system among the following cropping systems

— soybean-chickpea, maize-chickpea, soybean/maize-chickpea, soybean/pigeonpea, and maize/pigeonpea.

**Organic farming practices:** Specific package of practices for organic farming for crops like soybean, wheat, isabgol, chickpea, pomegranate, mustard, and pigeonpea have been developed.

### Phytoremediation of Chromium through Ornamental Plants

The leather industry is the major cause for high influx of Cr to the biosphere. There are about 3,000 tanneries in India, mostly spread over Tamil Nadu, Karnataka, Maharashtra, Uttar Pradesh, Rajasthan and Punjab. There was a reduction in net sown area by 10 – 41% and reduction in crop yield by 25 – 57% owing to contamination by tannery effluents.



The xerophytic plants i.e., *Furcraea*, *Agave* and *Euphorbia* can be suitable for phytostabilization of soils contaminated with Cr.

## Maintaining soil organic-carbon equilibrium (SOC):

Fundamental information developed on SOC equilibrium with continuous application of 100% recommended rates of NPK plus FYM established a new equilibrium of SOC much earlier ( $t_{1/2}$ , 2.4 years in Vertisol, 7.7 years in Inceptisol and 2.1 years in Alfisol) than imbalanced use of either N or NP fertilizer ( $t_{1/2}$ , 8.1 - 25.7 years in Vertisol, 14.9 - 50.3 years in Inceptisol and 2.1 - 2.8 years in Alfisol). This information is very useful assessing management for impacts to sustain yield, assess soil quality and restore degraded soils.

**Bioremediation of heavy metal contaminated sites:** In phytoremediation green plants are used *in-situ* for cleaning contaminated sites. Some floricultural plants like marigold, chrysanthemum, gladiolus, tuberose and bio-agents like *Trichoderma viride* have been found useful in managing heavy metal contaminated areas.

**Limits of heavy metals loading in soils established:** The maximal permissible limits of heavy metals in soil have been determined by 'phytotoxicity', 'food contamination,' and 'soil microbial activity diminution' approach. Considering lowest values by all three approaches as protective for all target organisms, the limits are: 392 mg/ kgZn, 179 mg/ kgCu, 0.34 mg/ kgCd, 81 mg/ kgPb, 30.7 mg/ kgNi, and 31 mg/kgCr. In terms of dilute calcium chloride extractable concentrations, these are: 0.003 mg/kg Cd, 0.052 mg/ kg Cr, 0.637 mg/ kg Cu, 0.022 mg/ kg Ni, 0.008 mg/kgPb, and 3.800 mg/kg Zn.

**Characterization of municipal solid waste (MSW)-** Municipal solid waste composts from 29 cities across India showed that heavy metal contents in composts

from bigger cities (>1 million population) were higher as compared to those from smaller cities (<1 million population). Majority of MSW composts did not conform to the quality control guideline of 'the Fertilizer Control Order 1985' in respect of total organic C, total P, total K as well as heavy metals Cu, Pb and Cr. Composts prepared from source separated biodegradable wastes contained on an average, higher organic matter (by 57%), total N (by 77%) and total P (by 78%), but lower concentrations of heavy metals Zn (by 63%), Cu (by 78%), Cd (by 64%), Pb (by 84%), Ni (by 50%), and Cr (by 63%) as compared to those prepared from mixed wastes. A protocol for compost quality evaluation and utilization for different quality of composts in different agro-ecosystems has been developed.

## DATABASE, MAPS AND SOFTWARES FOR SOIL-HEALTH MANAGEMENT

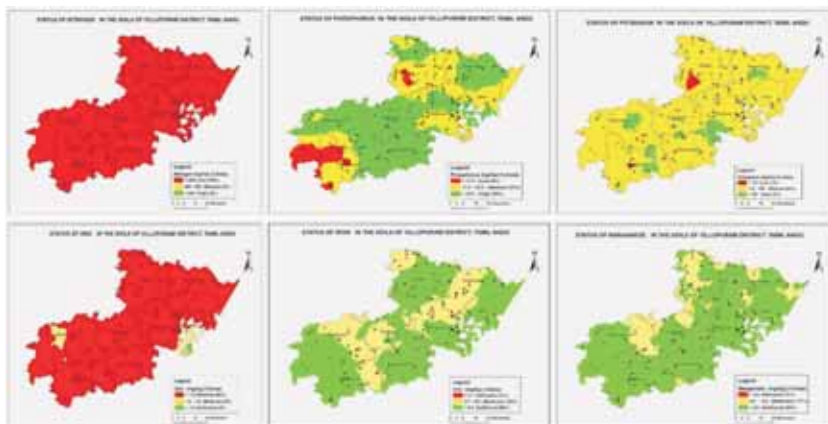
**Online fertilization recommendation system:** This software has been developed to recommend fertilizer doses for the targeted yields of various crops. It has the facility to input actual soil test values at the farmers' fields to obtain optimum fertilizer doses for nitrogen, phosphorus, and potassium. The software can be accessed at <http://www.iiss.nic.in>. The software is compatible with Internet Explorer.

**Database of different sources of plant nutrients:** This database has been generated in MS access. To access the data one has to open the file 'Nutrientdatabase.mdb' and then click on various queries.

## Bioremediation of Heavy Metals from Municipal Solid Waste

In India, every year about 64.8 million tonnes of municipal solid waste, 12.7 million litres of wastewater and 8.58 lakh million litres of industrial wastewater are generated which carry heavy metals and other pollutants. These if enter in the food chain or drinking water supply degrade soil health, groundwater quality, affect plant growth, livestock and human health.

- Six mesophilic fungi as biofilters have been identified namely *Trichoderma viride*; *Aspergillus heteromorphus*; *Rhizomucor pusillus*; *Aspergillus flavus*; *Aspergillus terreus* and *Aspergillus awamori*.
- Biofilter was developed in a perforated stationary phase (34cm×32cm×5cm); individual fungus was grown with standard medium at 30°C.
- A series of biofilters were inserted in the compost at a distance of 6 inches.
- Biofilter was harvested at 15 days interval and quantity of total heavy metals removed was quantified.
  - By mesophilic fungi, removal of Pb was maximum (18-51.5%), followed by Ni (15.5-35.3%) and Zn (4.6-32.8%) when biofilter was used during humification or curing phase.
  - Among six isolated fungi *Trichoderma viride* was found promising for removal of Pb, Ni, Zn and Cd; followed by *Aspergillus flavus*.
  - Mesophilic fungi would be a good biofilter to remove heavy metals from MSW compost and would improve compost quality.



GIS-based soil fertility map of Villupuram District of Tamil Nadu

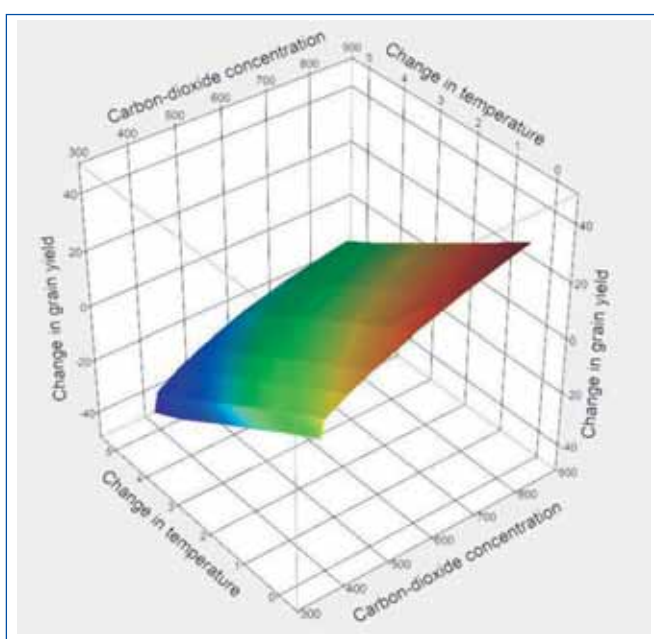
**GIS-based soil-fertility maps of different states:** The soil fertility data on N, P, and K index values at district level for Andhra Pradesh, Maharashtra, Chhattisgarh, West Bengal, Haryana, Orissa, HP, Karnataka, Punjab, Tamil Nadu and Bihar of India have been developed in MS-Access. From the attribute database, the different thematic layers have been reclassified to generate various thematic maps on N, P and K index values (IVs).

**Soil carbon and nitrogen turnover model:** A new soil carbon and nitrogen turnover model has been developed by using soil and crop dataset of long-term fertilizer experiments of India. The model works on the principle of soil carbon saturation theory, which suggests that soil carbon sequestration rate decreases as the soil carbon content increases and vice-versa. The model computes total organic carbon, Walkley & Black C content, carbon in resistant (passive) and mineralizable (active+slow) pools, carbon stocks, total and available N. Soil carbon

and nitrogen prediction model uses a yearly time step and the users have to define only initial soil carbon content. The model itself determines relative allocation of carbon in different pools. The model automatically computes carbon and nitrogen turnover based upon these parameters, and output is displayed in excel sheet.

**Software for evaluating municipal solid waste (MSW) compost:** A new method has been developed that enables grading of MSW compost based on its quality. With this software, grading can be done for marketable class on a four point scale or for Restricted Use class on a three point scale based on the *Fertilizing Index* and *Clean Index* of the MSW compost.

**Climate change effect on wheat yield:** The results of the modeling study showed that an increase in temperature and elevated CO<sub>2</sub> concentration have had an interactive effect on wheat yield in a sub-humid part of central India. It revealed that temperature played a negative role while CO<sub>2</sub> played a positive role on grain yield of wheat under future climate change scenarios. It was predicted that 1°C increase in temperature from the base would decrease wheat (Sujata) grain yield by 8.4%. Increase in CO<sub>2</sub> concentration from the base (350 ppm) to 850 ppm, would increase the wheat grain yield. With increasing the CO<sub>2</sub> level from 400 ppm to 700 ppm and corresponding increase in temperature to 5°C, negative effect of temperature on wheat yield diminished.



Combined effects of change in temperature and CO<sub>2</sub> concentration on wheat yield

**Early detection of nitrogen stress in maize and wheat crop through hyper-spectral reflectance based vegetation indices:** Nitrogen stress in maize and wheat crops could be predicted with two hyper-spectral vegetations indices – Red Edge Position (REP) and Slope of the REP data. However, the predictability improved with the use of two new indices – Combined Index and Double-peak Canopy Nitrogen Index (DCNI). Both these indices utilized rapidly changing spectral reflectance pattern in the red to infra-red regions and the soil-base line developed for soil. These indexes could predict more than 60% variability of the nitrogen content in maize leaf. DCNI predicted 71% variability of nitrogen content in maize leaf under our controlled experimental condition. It is concluded that DCNI can be used for prediction of nitrogen stress in maize and wheat. The information generated will be useful for development of models for yield prediction in large area and also in precision farming for nitrogen management.

## THRUST AREAS

### Soil health management and input-use efficiency

- Increasing inputs-use efficiency
- Precision agriculture
- Nano-technology
- Integrated nutrient management: Indigenous mineral and by-product sources
- Bio-fortification
- Fertilizer fortification
- Organic farming and quality characterization of produce
- Improving soil quality through organic matter addition and correcting nutrient imbalance
- Soil quality assessment, imbibing influence of different physical, chemical and biological soil attributes
- Understanding resilience of degraded soils and restoration of their productivity.

### Conservation agriculture and carbon sequestration

- Carbon sequestration research in context of sustainable management of land and soil resource and conserving deteriorating environment
- Conservation agriculture and carbon sequestration, especially in semi-arid and sub-humid regions
- Tillage and nutrient interaction in soil

- Crop simulation modeling and remote sensing in climate change research
- Crop adaptation to climate change-rhizosphere study.

### Soil microbial diversity and genomics

- Efficient composting techniques and integrated plant nutrient supply systems
- Characterization and prospecting of large soil bio-diversity
- Characterization of functional communities of soil organisms
- Testing mixed biofertilizer formulations.

### Soil pollution and remediation

- Bio-remediation/ phyto-remediation of contaminated soils
- Quality compost production and establishing quality standards
- Solid wastes and waste waters-quality assessment and recycling.

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## NATIONAL ACADEMY OF AGRICULTURAL SCIENCES

invites Nominations for different Awards for the Biennium 2015-2016

### I. MEMORIAL AWARDS (6 Nos)

The nominee should be a distinguished scientist above 55 years in age. The period of assessing the contributions would be life-time up to the year of nomination. Each award consists of a citation and a silver plaque.

### II. ENDOWMENT AWARDS (2 Nos)

The awards will be given to outstanding scientists for their contributions towards ensuring (i) food and nutritional security and (ii) overall contribution to agriculture. The nominees can be from any branch of science relevant to agriculture. The award comprises a citation and a silver plaque.

### III. RECOGNITION AWARDS (6 Nos)

The awards would be given to distinguished scientists in the age group of 35-55 years, who are Fellows of the National Academy of Agricultural Sciences. Each award consists of a citation and a silver plaque.

### IV. YOUNG SCIENTISTS AWARDS (6 Nos)

Scientists below the age of 35 years are eligible for this award. Each award consists of a citation and a gold plated silver medal.

For details, please visit Academy website at [www.naasindia.org](http://www.naasindia.org) or write to:

The Executive Secretary  
National Academy of Agricultural Sciences, NASC, DPS Marg, New Delhi 110 012  
*Tel.:* (011) 25846051, *Fax:* (011) 25846054, *Email:* naas@vsnl.com

Last date for receipt of the nominations in the Academy is March 31, 2016

*Note: Self Nominations are not acceptable.*

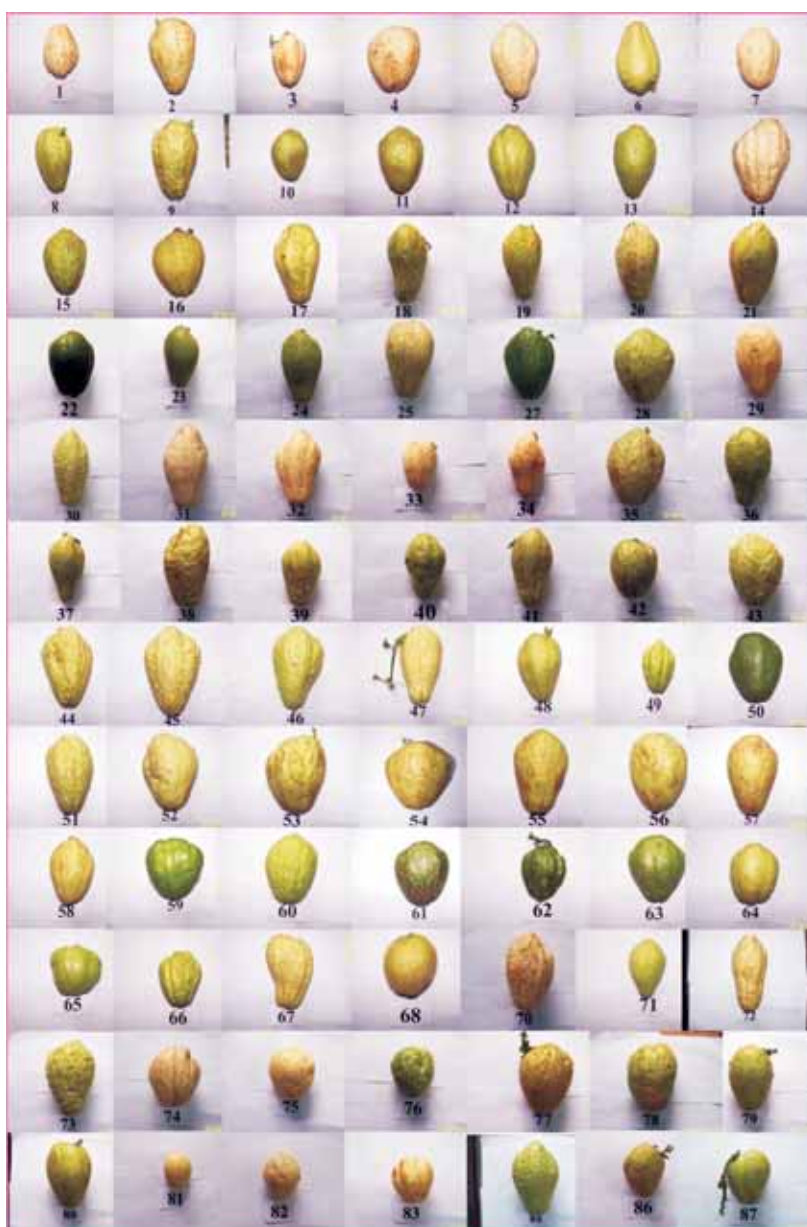
## Genetic diversity of chayote (*Sechium edule* (Jacq.) Sw.) in Sikkim Hills

Chayote, often called poor man's vegetable, is very popular in North Eastern hilly region and grows abundantly without much care and attention at mid to high hills of Sikkim. Its edible parts include fruits, flowers, seeds, tendrils, young leaves, shoots and roots. The young and tender leaves are eaten and cooked like other leafy vegetables; seeds are fried, roasted for consumption, and tubers can be removed after two years for vegetable purpose. The fruit is, however, the most popular chayote product in the market place; the fruit can also be curried, fried, creamed or pickled. All plant parts are good food for livestock also.

In Sikkim, varying morphological characteristics of the chayote-fruit are the most obvious sign for its high genetic diversity. Eighty-six accessions were collected to document its existing genetic diversity in the region by classifying shape, colour, texture, and other characteristics of the fruit. Morphologically different types and colours of the fruits observed are: round, oblong, spiny, very spiny, without spine, and creamy white to green, dark green fruits. High range of fruit phenotypic variations observed among the accessions for several parameters such as fresh fruit weight, dry fruit weight, fruit length, fruit

Chayote is a good source of fibre and vitamin C content; and potassium, calcium, iron are high in fruits. Sikkim has several native cultivars of green, creamish and white types, including spiny and non-spiny, green types.

width, spine density per unit area and spine length in spiny types, peel thickness, seed weight, seed length, seed width and total soluble solid (TSS) contents are as follows: fresh fruit weight ranges from 93 g to 1,250 g; dry weight from 7.09 g to 16.47 g/100 g fresh fruit; fruit length from 37.5 mm to 178 mm; fruit width from 47.2 mm to 102.7 mm; spine density from 3.20 to 62.56 per square inch and spine length 0.5 mm to 7.17 mm in spiny types; peel thickness from 0.14 to 0.51 mm; seed weight from 10 g to 26 g; seed length from 26.31 to 91.46 mm; seed width from 9.52 to 47.31 mm and TSS from 2° Brix to 7.1° Brix. The yield of chayote accessions varied from 0.5 to 62 kg / plant.



Genetic diversity of chayote

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## Technology for remote and degraded coastal lands of Andaman Islands

Decreasing rainy days, increasing length of dry spell along with soil salinity during dry season makes agriculture highly vulnerable in the coastal lowlands of Andaman. After

the furrows, which was good enough for culturing Indian Major Carps. Subsequently, farmers restored backyard poultry farming of Vanaraja birds; they also used regular



Waterlogged-saline land (before intervention)  
(11° 41' 36.363 N ; 92° 36' 24.213 E)



After tsunami (Waterlogged-saline)



Raised bed and furrow systems made in the affected areas with vegetables/fruits+fish

2004 tsunami, seawater is more often gushing into land inundating paddy fields with saline water. Collinpur village of South Andaman is one among the several villages affected by this problem.

Realising that agriculture and livelihood are affected by saline-water intrusion and climate change, raised bed and furrow (RBF) system was constructed at the farmer's field. RBF system comprising three raised beds of 4-m width and three furrows of 6-m width to a convenient length (~ 36 m) in an area of about 0.3 acre of coastal lowland was formed in March 2013. Salts and other toxic substances from raised beds leached out with rainwater in subsequent rainy season. Rainwater was harvested in the furrows from 2014 onwards after draining off water harvested in the first year. By the end of July 2014, nearly 465 m<sup>3</sup> of freshwater could be harvested and stored in

agro-advisories received from the agromet unit of the institute.

Cultivation of bananas in the beds of RBF system was started organically by using compost from farm-wastes and poultry manure and Panchagavya. Water from furrows was used to irrigate banana-crop during dry season and short-duration vegetables (amaranthus and radish) were harvested as intercrops. Thus, farmers could earn livelihood even in the backdrop of drought-like situations

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## Knocking down *myostatin* gene in chicken for enhancing body weight

Of the several genes, *myostatin*, sometimes called GDF8, is a major gene having negative effect on the growth of chicken by inhibiting cellular differentiation during early embryonic stage.

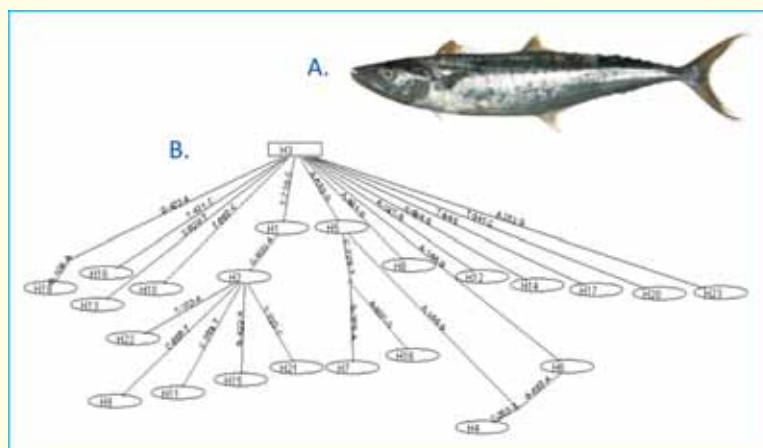
The protein acts through its receptors, ACVR2A and ACVR2B; of which ACVR2B plays a major role, while ACVR2A has only a minor role in deciphering its activities. Of several approaches, blocking myostatin protein

synthesis by gene silencing has been found an important tool to inhibit myostatin activities in the chicken-body.

A total of 5 shRNA molecules were designed on coding sequence of mRNA of chicken *myostatin* gene. The molecules were cloned in BLOCK-iT U6 RNAi entry vector and pBLOCK-iT DEST6 vector (Invitrogen). The cloned molecules were first tested in primary chicken-embryo myoblast culture for activities. Molecules

## Population stock structure of narrow-barred Spanish mackerel in Indian waters

Narrow-barred Spanish mackerel, *Scomberomorus commerson*, of family Scombridae (Lacepède, 1800) is one of the economically important marine fishes in Indian waters. There is regular fishery of this species from Indian waters, and 31,412 tonnes of *S. commerson* were caught in 2014 from different areas. However, there were no population structure studies available for this species. The population genetic structure of this commercially important fish species using variable mitochondrial ATPase 6/8 gene has been carried out for conservation and sustainable management. A total of 87 tissue samples were collected from five different geographical locations along the Indian coast (Mangaluru, Kochi and Veraval in the west coast; Chennai and Vishakhapatnam in the east coast). Amplification of ATPase 6/8 gene yielded 842 bp with 23 haplotypes across the sampling sites. Pair-wise  $F_{ST}$  values did not show any significant differentiation ( $p > 0.05$ ) among populations of *S. commerson*, which means this species can be considered as a unit stock in Indian waters. As this species



A. Image of *S. commerson*; B. Haplotype network tree of *S. commerson*

is having high commercial importance, the findings of the present study would be of immense value in managing the species.

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showed significant silencing of *myostatin* gene in myoblast cells. All the shRNA molecules then were transferred through embryonic manipulation on day 4 to 7, and through spermatozoa with lipofection and electroporation. The efficiency of the technique for developing knock-down birds was up to 32%. The effect of shRNA molecules on post-hatch body weight was analyzed and found significantly ( $P < 0.05$ ) higher at 4 and 6 weeks of age. The molecule 5 showed the highest effect, followed by molecule 2, molecule 4, molecule 3 and molecule 1. The molecule 5 showed 18.8, 37.8, 25 and 33% higher body weight than control group at 2<sup>nd</sup>, 4<sup>th</sup>, 5<sup>th</sup> and 6<sup>th</sup> weeks of age. Biochemical parameters such as total serum cholesterol and total protein were compared between knock-down and control birds, and found that there were no significant differences between these groups. Gene silencing technique has been perfected in chicken to be used further in improving body weight of birds.

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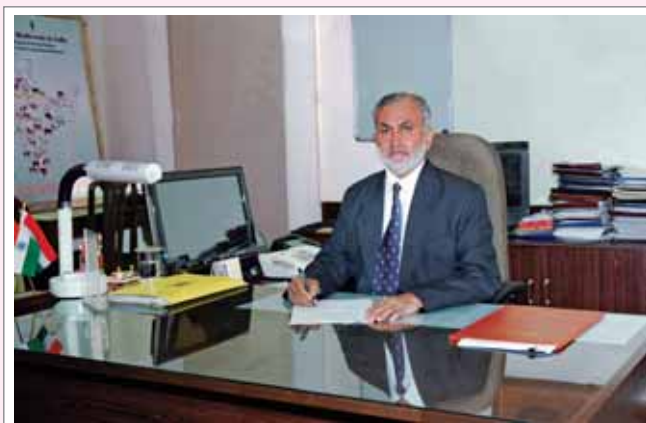
## WAY FORWARD

**S**OIL is the most precious nature's gift to mankind and forms the basic medium to sustain life on the Earth, as 95% of our food is produced on our soils. Sustaining of food and nutritional security of our teeming millions in the country, representing 17.5% of the global human population and 11% of the world's livestock population, depends only on 2.3% of the global land resource. In 2035, per capita land availability is likely to decline further to 0.08 hectare from 0.13. In India, nearly 104 Mha of the 141 Mha net arable land is affected with different types of land degradations, comprising 73.27 and 12.40 Mha under water and wind erosion, respectively. About 11 Mha of arable land suffers from acute soil acidity ( $pH < 5.5$ ) with very low productivity ( $< 1$  tonne/ha) and around 6.74 Mha is salt-affected and 1.07 Mha is under physical degradation, mostly waterlogging. In addition, polluted surface water and groundwater add many harmful chemicals into the soil body. More than 35 billion litres of urban wastewater and 25 billion litres of industrial wastewater are released every day, and a significant part of this enters into agricultural land.

Deterioration of soil health and quality due to over dependence on chemical fertilizers is another important reason threatening food and environmental security. By soil health, we generally mean physical and chemical health of the soil (oblivion of the biological health). Soil and its living organisms are also integral part of the agricultural ecosystem, and play a critical role in maintaining soil health. The consequences of neglecting or abusing soil life would weaken soil functions and contribute to greater losses of fertile land.

A real challenge for the future is to provide viable farming practices, scientific and judicious land-use planning, and appropriate bio-engineering measures for soil- and water-conservation, organic inputs, soil amendments and resource-conservation technologies. These would definitely influence soil biological population and processes and would bring in transformation from unsustainable agricultural practices to sustainable ones.

Realizing the role of soils in Global Food Security, the 68<sup>th</sup> UN General Assembly declared 2015 as the International Year of Soils (IYS) with the slogan 'Healthy Soils for a Healthy Life'. The Food and Agriculture Organization of the United Nations has been nominated to implement the IYS 2015 within the framework of the Global Soil Partnership and in collaboration with the Governments and the Secretariat of the United Nations.



Dr S. Ayyappan, Secretary (DARE) and Director General (ICAR)

Keeping our soil healthy and productive is of paramount importance. The six key messages identified are: (i) soil health as the basis for healthy food production, (ii) soils are the foundation for vegetation, which is cultivated or managed for feed, fibre, fuel and medicinal products, (iii) soils support our planet's biodiversity and they host a quarter of the total, (iv) soils help combat and adapt to climate change by playing a key role in carbon cycle, (v) soils store and filter water, improving our resilience to floods and droughts and (vi) soil is a non-renewable resource; its preservation is essential for food security and for our sustainable future.

Our Hon'ble Prime Minister has laid greater emphasis on Soil Health, and on many occasions, he expressed his concern over soil management. The Government of India launched a *National Mission on Soil Health Card* in year 2015, as being the International Year of Soils. The Mission provides soil-test-based recommendations to farmers to manage their soil resources. Incidentally, World Soil Day was also celebrated nation-wide on 5 December 2015. As part of the same, the National Agricultural Research and Education System through ICAR Institutes, KVKs and SAUs distributed about 2.5 lakh Soil Health Cards to farmers. With concerted efforts and technological backstopping, the country can achieve sustainability in agriculture by a greater understanding of the agroecosystem ecology, input-use efficiency and natural resource management. This is vital, as the country is looking forward for a second green revolution in the near future.



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