### January-March 2011



Agresearch with a Buman touch



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A SCIENCE AND TECHNOLOGY NEWSLETTER

**RESEARCH UPDATE** 

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- Speciality soybeans developed
- Treatment of chickpea-seeds for enhancing crop growth
- Agricultural knowledge sharing

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PROMISING TECHNOLOGIES

# Top-working in sweet-gourd for conservation and for increasing productivity

The woody stems of the unwanted male-plants of sweet-gourd Momordica cochinchinensis are cut-back at a height of 1.5 m above the ground level. Tender scion-sticks collected from the growingtips of the high-yielding female-vine can be grafted on the beheaded plants that have sprouted after two weeks. Greencoloured scion-stick 10-20-cm long with 3-5 nodes matching with the girth of rootstock sprout is ideal. Tip of the rootstock sprout



is cut-back and a vertical split of 2.5-3.5 cm is made in the middle, when it is still green but hardened. Slanting cut of 2.5-3.5 cm is made at both the sides at the base of the scion in such a way that it fits tightly into the split made on the rootstock.

The tapered base of the scion is inserted into the split, ensuring cambial union through contact of cut-surface skin of the rootstock and the scion. The scion is fixed by tying gently with a polythene grafting strip. The scion, including graft juncture, is covered with

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### **PROMISING TECHNOLOGIES**



Grafted plant in flowering (female); Harvested ripe fruits (inset)

a transparent polythene to prevent wilting through transpiration. A few drops of water are sprinkled inside the polybag before tying to maintain high humidity and mist effect. Temporary shade is provided to protect from scorching sun. The graft union normally takes place within a week. Grafting can be done on multiple sprouts. The polythene strap should be removed after two weeks when the graft juncture starts thickening. Success rate of about 98% has been normally observed when sufficient moisture is in the surroundings. The grafted-plant starts flowering within 6 weeks.

On an average, 25 fruits per grafted-plant (350-700 g/ fruit) were obtained within 3 months of grafting compared to 16 fruits from seedling-plants and 8 from rooted-cuttings that too after a gestation period of 12-18 months. Further, grafted-plants are compact,

Momordica cochinchinensis (Lour.) Spreng. is a wild or less cultivated, nutritionally rich vegetable occurring in Andaman Islands, northeastern states and eastern India. Germplasm collection is hampered by non-availability of sufficient seeds; and one of the major problems for its cultivation is the segregation of seedling progenies into male and female in 50:50. Normally, surplus male-plants are uprooted; one or two healthy plants are retained as pollen source. To save resources and to increase seed productivity, a grafting technique has been standardized to convert unproductive males to productive females and to aid in establishment of the collected germplasm.

require less space (4-5 m<sup>2</sup>) compared to uncontrolled vine growth of seedling- plants and rooted-cuttings. Multiple accessions and male and female plants can be grafted on different sprouts of the same rootstock, enabling judicious space utilization. Softwood grafting has been found useful in collection, establishment and multiplication of germplasm of this rare species of economic importance.

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### Speciality soybeans developed

Oil and protein are the major economic attributes of soybean-seeds. However, soybean genotypes with special quality traits, also known as speciality soybeans, are also being sought after for specific end-uses.

Kunitz-trypsin inhibitor- free soybeans: Soybean has been endowed with the 'functional food of the century' as certain nutraceutical components present in it reduce risks of hormonedependent cancer, cardiovascular diseases, diabetes and osteoporosis.

In India, wheat-flour fortified with soybean (9:1) has often been recommended. This fortification requires boiling of soybean-beans for the best possible inactivation of kunitz-trypsin inhibitor (KTI), an antinutritional factor, before grinding with wheat, and requires an extra effort at the household level; and at the industrial level incurs considerable energy cost for processing different soy-products. Moreover, heat treatment insolubilizes much-valued soy proteins, and causes losses of essential amino acids.

**NRC 101** (INGR 10054) and **NRC 102** (INGR 10055), two new soybean genotypes free from KTI polypeptide, have been developed using exotic germplasm accession PI 542044 as the donor for the null KTI allele and early-duration popular cultivar Samrat as the recipient parent. Both these newly developed genotypes mature early.

**High oleic acid soybeans:** Fatty acid composition of the oil from the conventional soybean cultivars shows 11% palmitic acid (C16:0), 3% stearic acid (C18:0), 23% oleic acid (C18:1), 53% linoleic acid

### **PROMISING TECHNOLOGIES**

(C18:2) and 8% linolenic acid (C18:3). High unsaturated fatty acids are responsible for the poor oxidative stability of the oil. Oleic acid is the least susceptible unsaturated fatty acid compared to linoleic acid and linolenic acid.

In the vegetable-oil industry, oxidative stability is often improved by partial hydrogenation; this process is cost-ineffective, but results in the formation of trans fats, which are atherogenic and diabetogenic.

Soybean genotypes with high oleic acid content are desired that can deliver naturally oxidatively stable

oil; not requiring even partial hydrogenation. Out of the 2,100 soybean germplasm lines screened, an indigenous germplasm line **IC 210** (INGR 10053) has been identified for high oleic acid, and **NRC 106** (INGR 10052) has been developed with high oleic acid. Across three consecutive years, average value for this fatty acid has been found 42.7 and 41.9% in IC 210 and NRC 106, respectively.

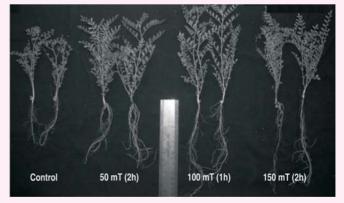
> Vineet Kumar, Anita Rani, S.M.Husain and S.K.Srivastava Directorate of Soybean Research, Khandwa Road Indore (Madhya Pradesh) 452 001

### Treatment of chickpea-seeds for enhancing crop growth

Pre-sowing exposure of seeds of different crops to static magnetic field (SMF) has been reported to stimulate plant development. Among the various combinations of the static magnetic strengths and durations, exposure to 100 mT for 1hr was found best for maximum enhancement of germination and root-shoot growth in *desi* and kabuli chickpeaseedlings.

Water imbibing characteristics of seeds showed temporal change in water absorption; and activity in chickpea through the SMF increased by 8-20%. Seed-water diffusivity also significantly increased (30-70% over control). In the SMF-treated plants, consistent and significant increase (20 - 150%) in the root parameters (length, weight, volume and surface area) was recorded under two soil-water regimes (12, 20%, v/v). Study in the nethouse with two soil-water potentials (-0.1 and -0.2 MPa) also exhibited significant improvement in the various physiological and biophysical traits (photosynthesis, transpiration, water relations) and in the root and growth parameters in the plants grown from the SMF-treated seeds; thereby improving radiation and water-use efficiencies. With the optimum soil-water regime, plants from the treated seeds produced more biomass, number of pods, and more grain weight than untreated controls.

In the fields, root-water uptake was steadily higher from the surface layer (0-30 cm), and it was 68-75 mm more in the treated crop. Improved root growth substantially increased water uptake, thereby



Growth behaviour of chickpea under different strengths and durations of static magnetic field

increasing biomass in the treated chickpea. The treated chickpea also developed significantly higher number of pods and seeds, leading to higher seed yield. Owing to better yield (15-20%) and biomass (20-25%), water-use efficiencies (10-15%) were higher in the treated crop.

Improved root characteristics suggest that this technique may be used for growing chickpea in rainfed areas, where enhanced root growth will be able to tap moisture from deeper layers of the soil.

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### National Agricultural Innovation Project

### Agricultural knowledge sharing

Knowledge sharing programme at the Directorate of Knowledge Management in Agriculture (DKMA) is involved in sharing Indian agricultural research knowledge at the global level, and also for providing improved communication links among knowledge generators – public research organizations – and their users – researchers, farmers' groups, *Panchayati Raj* Institutions, private sectors and other stakeholders, for poverty alleviation and income generation. This new initiative has harnessed benefits of the Internet and ICTs.

Since its inception, the ICAR has been sharing knowledge through traditional channels of knowledge-delivery. The ICAR publishes two flagship research journals – *The Indian Journal of Agricultural Sciences* and *The Indian Journal of Animal Sciences*.

Under the knowledge-sharing, *ICT infrastructure* has been established for providing gateway to all user-level services (search and browse) for these agricultural research journals and other value-added functionalities (post authoring tools, online review, monitoring tools through analysis of document and usage statistics).

*ICAR e-Newsletters* are also being e-published in an interactive format. The e-publishing platform has now been made available to agricultural research societies for hosting their research journals online free of cost—Journal of Medicinal and Aromatic Plants Association of India and Indian Phytopathology have been hosted online.



Screenshot of ICAR's online journals' portal and e-Newsletters

#### Important Web URLs

e-Publishing portal: <u>http://epubs.icar.org.in/ejournal</u> ICAR News: <u>http://news.icar.org.in</u> ICAR Reporter: <u>http://reporter.icar.org.in</u>

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Online access of ICAR Journals

# Key features of e-publishing and knowledge dissemination

- Online submission of articles and online review
- Automation of key publishing activities
- Efficient and reliable record keeping through database
- Paperless processing of research articles
- Full-text searchable repository of back issues of journals

#### Access Statistics of e-publishing portal for Nov. 2010–March 2011

Visits	Pageviews	Unique visitors	Avg. time on site
37,472	275,070	19,404	6.33 minutes

350-450 visits daily on the online system in week days and 75-150 visits during weekends *Source:* Google Analytics

#### **Benefits of e-Publishing**

- 1. More effective and timely dissemination of Indian Agricultural Research Knowledge to researchers, students, extension-workers and farmers.
- 2. Enhanced visibility (Access from 142 countries worldwide and 82 cities nationally).
- 3. Increased readership: more than 5,500 registered readers (and increasing).
- 4. Increase in research article submission nationally as well as internationally.

Owing to online discoverability, requests are being received from the international universities for collaboration in research with the authors of the articles that are published in the ICAR's journals. Requests for purchasing copyrights of articles are also being received. E-publishing and open access may result in increased citation index of articles and may improve impact factor of the journals in the forthcoming years.

#### Himanshu

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### **NEW INITIATIVES**

### Molecular approaches for variability studies in Jatropha curcas, a biofuel crop

Bio-diesel can be an efficient and 100% clean natural energy, an alternative to petroleum fuel, which may provide desired environment-friendly solution. *Jatropha curcas*, known as *Ratanjyat* or Physic-nut, can be easily grown in the arid and semi-arid regions of



 A. Red circles on the map indicate collection sites.
B. Seedlings of *J. curcas*, raised from 3-yearcryobanked seeds



India. Accessions of *Jatropha* were collected from different states; and a set was selected, representing *Jatropha* diversity, for DNA profiling. Selections were based on the available data on the passport information with details of the collection site and oil

content.

So far, there have been only a few studies on the analysis of the genetic variations within *J. curcus* using molecular markers — RAPD, AFLP, SSRs and SRAP. In this study, wide variability of *Jatropha* obtained from 12 agro-climatic regions, covering 19 states and 90 districts, was analyzed.

Molecular characterization of 260 Jatropha curcas accessions was conducted, and a set of 50 RAPD, 20 SSR and 25 AFLP primers have been identified that are suitable for genetic diversity analyses. Using random amplified polymorphic DNA (RAPD) marker, 96.8% polymorphism was studied across the accessions on the basis of the banding patterns. Twentyseven primers, out of 30, generated 100% polymorphic patterns. Jaccard's coefficient of similarity varied from 0.31 to 0.98 among different genotypes, indicating high level of genetic variation. Overall grouping pattern of clustering

corresponded well with the principal component analysis, confirming patterns of genetic diversity observed among accessions. Similar patterns of variability were observed using AFLP and SSR markers also. The accessions from Madhya Pradesh, Maharashtra, Uttar Pradesh and Jammu and Kashmir are found most diverse, and can be used for basebroadening of *Jatropha* cultivars; and these regions can be targeted for more collections.

In the National Cryogenebank, 767 *Jatropha curcas* germplasm have been cryostored successfully since the past 4 years.

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# NEW INITIATIVES

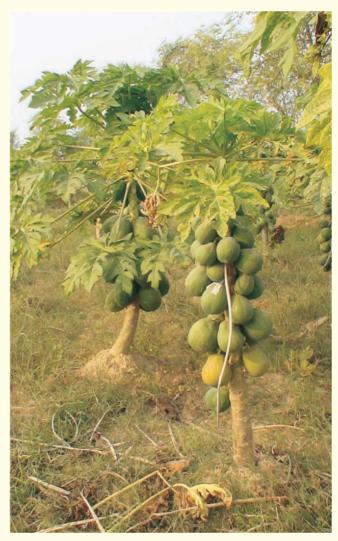
### Cost-effective sex identification in dioecious papaya

Papaya (*Carica papaya*) is an important cash-crop, valued for its nutritional, industrial, pharmaceutical and medicinal properties. It is conventionally propagated by seeds but the dioecious papaya varieties do not ensure right plant type. As the result, 50-60 % of the seeds sown turn out to be males, and are to be uprooted after 6-7 months of planting. Identification of sex type in papaya at a young stage would result in higher fruits production, and would consequently increase profitability.

Shortage of the right planting material of papaya from elite varieties in the country has impeded its cultivation. To overcome this, and to make papaya cultivation profitable, PCR-based sex diagnostics for the crop are available. But they are expensive isolation of plant DNA alone is expensive and labourintensive. Recently, a cost-effective method of sex identification has been devised by circumventing DNA isolation phase; using directly crude leafextract.

Young leaf tissue (10g) of cv. Pusa Nanha was taken in 1.5ml centrifuge tubes; to this 40µl of 0.25N NaOH was added, and tubes were placed in the boiling water-bath for 1minute. After that 40µl of 0.25N HCl was added, followed by addition of 20µl 0.5M Tris HCI (pH8.0) and 25µl of 1.25% PEG (Poly ethylene glycol) solution. These all together were boiled for 2 minutes in water-bath. After boiling, tissue was disintegrated using plastic mortar or pipette tips. The suspension was mixed well and kept at 4°C for 10-15minutes, followed by pulse centrifugation at room temperature. The supernatant (crude extract) was directly used for PCR reaction. To standardize PCR reactions by using crude extract, different PCR mixes were made using different amounts of the extract (2µl to 16µl), primer concentration, Taq polymerase, dNTP mix and MgCl<sub>2</sub>.

Crude extract (10  $\mu$ l) derived from fresh, young leaves was treated with Tris-Cl and poly ethylene glycol for identification of papaya-plant sex. Optimum PCR condition was worked out, which included 1 $\mu$ l (25pmol) primer, 1 $\mu$ l (3U/ $\mu$ l) dNTP mix and 2.5  $\mu$ l MgCl<sub>2</sub>. Male specific primer (Forward 5'-GAGGATCCCTATTAGTGTAAG-3'; Reverse 5'-



Field evaluation of sex-identified Pusa Nanha plantlets

GAGGATCCCTTTTGCACTCTG-3') showed amplification in 1% agarose gel. Presence of band suggests male-plant and its absence female-plant; as the primers used are male-specific.

Normal DNA isolation costs around Rs15.50 per sample (DNA Isolation Kit-Sigma-Aldrich, USA), and this method reduces cost to only Rs 2.25 per sample; thus bringing down cost of DNA isolation to approximately 70%.

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## NATURAL RESOURCES MANAGEMENT

# Soybean rhizobia and mycorrhiza-helper bacterium isolated from Malwa Region

Two rhizobia—*Bradyrhizobium liaoningense* and *B. japonicum*—have been isolated from the root nodules of soybean cultivar JS 95-60 and JS 93-05, respectively, cultivated in Malwa Region of Central India. These rhizobia are found positive to nitrate reductase and IAA production, and have potential to enhance



nodulation, nitrogen fixation and growth of soybean. *B. liaoningense* was found extra-slow growing, and could grow at 36°C, when tested under *in-vitro* liquid.

One bacterium identified as *Burkholderia arboris* was also isolated from soybean-root nodules of JS-93-05, and was found fast growing, plant-growth promoting and synergistic to AM fungi, and this was able to enhance AM fungi production when tested in microcosm conditions.

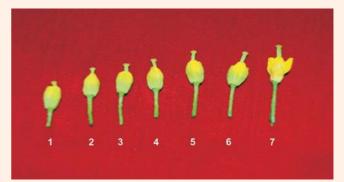
These bacteria were identified through full sequencing of 16SrRNA gene, and are registered as MTCC-10753, MTCC-10751 and MTCC-10752 respectively with the MTCC, IMTECH, Chandigarh.

> Mahaveer P. Sharma, Sushil K. Sharma and V.S. Bhatia Directorate of Soybean Research Khandwa Road Indore( 452 001)Madhya Pradesh *e-mail*: mahaveer620@gmail.com

# Protogyny in Indian mustard—a tool for easy hybrid development

Rapeseed-mustard is an important oilseed crop, contributing 26.1 and 29.1% respectively, to total oilseeds area and production in India. Yield improvement in Indian mustard (*Brassica juncea* (L.) Czern. & Coss.) can be achieved by changing breeding strategies and through development of commercial hybrids.

Protogyny (Pg) system (protogyny is the condition where stigmas are extruded from buds prior to anthesis) can offer an option for easy hybrid development, besides cytoplasmic-genetic male sterility (CGMS) and self-incompatibility (SI). In Indian mustard, protogynous plants were observed in  $F_3$ generation in a cross between Agra Local and Varuna. These lines were studied for protogynous interval, stigma receptivity, pollen viability, fertility index and seed set. Results indicated protogynous interval extended up to 9-11 days and stigma receptivity up to 3 days from its protusion. Protogynous plants have normal viable pollens. Pollination methods applied



Progress in flowering [1-6: stigma exserted in bud stage; 7: opened flower]

showed very high fertility index; indicating selfincompatible nature of protogynous plants. These Pg lines, hence, can be used in hybrid-development programme of Indian mustard.

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### NATURAL RESOURCES MANAGEMENT

# Tissue-culture protocol for developing resistant sunflower

Sunflower cultivation has been limited owing to susceptibility of released varieties and hybrids to a wide array of biotic and abiotic stresses at all phenological stages of crop growth.

A regeneration protocol for sunflower has been developed with a combination of two cytokinins for shoot induction (thidiazuron TDZ and 2-isopentenyl adenine) and also for shoot elongation (benzyl aminopurine, BA and 2-isopentenyl adenine, 2-iP). Combination of a less potent cytokinin 2-iP and a more potent one This protocol has been tested on a wide range of cultivars, and has been found **genotypeindependent**.

Whole plantlets could be established in the soil within 8 weeks of culture initiation via adventive organogenesis pathway. Cytokinin 2-iP helped in overcoming premature flowering.

The protocol is being currently used in the laboratory for the development of sunflower transgenics resistant to sunflower necrosis



TDZ facilitated quality shoot production without vitrification and precocious flowering, besides triggering high frequency of shoot differentiation. Cytokinins 2-iP+BA in the shoot-elongation medium promoted shoot development with distinct nodes and lesser vitrification. Shoot-regeneration frequency was >90%; with up to 100-120 shoots per regenerating explant. disease through deployment of TSV coat protein (*TSV-cp*) gene in sense and antisense directions.

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### Varieties Developed

**Pusa Mustard 26 (NPJ 113):** It is an Indian mustard variety, identified during the 17<sup>th</sup> AICRP Rapeseed-Mustard Annual Group meeting held at the Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior, for the late (November)-sown, irrigated, mustard- growing areas of Zone II, comprising Jammu and Kashmir (Plains), Punjab, Haryana, Rajasthan, Delhi and western Uttar Pradesh. It possesses terminal heat tolerance and tolerance to high temperature also to some extent at the seedling stage. Average maturity period of the variety is 126 days.



It is a small-seeded variety with an average oil content of 37.6%, and it is suitable for multiple cropping particularly in rice and cotton belt where fields generally vacate in November and also for areas where long-duration *guar* varieties are grown and fields vacate in November.

**Pusa Mustard 27 (EJ 17):** This is an early-maturing Indian mustard variety identified at Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior, for early-sown, irrigated areas of Zone III, comprising Uttar Pradesh., Madhya Pradesh, Uttarakhand and Kota region of Rajasthan. It possesses terminal heat tolerance and also tolerates high temperatures at the seedling stage to some extent. Average maturity duration of the variety is of 118 days, and its average oil content in seeds is 41.7%. This variety is suitable for multiple cropping systems of Zone III, and fits well to have an additional crop between September and December or in the cropping systems, where sugarcane or vegetables are grown in January.

Division of Genetics, Indian Agricultural Research Institute, New Delhi 110 012 *e-mail:* dkygenet@gmail.com **Brinjal DBL-02:** A long-fruited brinjal variety DBL02 has been identified for release, and is recommended for cultivation in Zone I (Humid Western Himalayan

Region i.e. Jammu and Kashmir, Himachal Pradesh and Kumaon and Garwal in Uttarakhand), Zone IV (Subhumid Sutlej-Ganga Alluvial Plains i.e. Punjab,



Delhi, Uttar Pradesh and Bihar) and Zone VI (Arid Western Plains i.e. Haryana, Rajasthan, Gujarat, Dadra and Nagar Haveli, and Daman and Diu). Its plants are non-spiny with erect branches, having light purple pigmentation partially on the younger leaves, mid-rib and veins. Its fruits are long, violet-purple with round distal end, and each fruit weighs 80-90 g. Its maturity period (days to first fruit harvest) is about 55 days from transplanting.

> Ravinder Kumar Division of Vegetable Science Indian Agricultural Research Institute New Delhi 110 012 *e-mail*: ravinderkuagarwal@rediffmail.com

Arka Flame (*Dianthus caryophyllus*): This first Indian Carnation variety, developed by the IIHR, has been identified for release. It produces red flowers having smooth-edged petals. And yields 300-360 flowers/m<sup>2</sup>/yr with 65-cm long flower-stalk and 5- cm long flower-bud. Its flower-stalks are thick and straight,



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hence transportation of these cut-flowers can be done conveniently. Compared to other commercial varieties, it has shown tolerance to nematodes and *Fusarium* wilt.

The Karnataka State Seed Sub-Committee has approved and recommended Arka Flame for

Notification by the Central Variety Release Committee for commercial cultivation in Karnataka.

Indian Institute of Horticultural Research

Hessaraghatta Lake Post Bengaluru (Karnataka) 560 089 *e-mail:* director@iihr.ernet.in

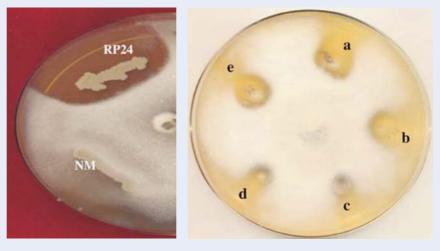
### Deciphering antifungal traits of Bacillus subtilis

Bacillus subtilis RP 24, a promising plant-growth-promoting rhizobacterium and a potent biocontrol agent, was mutagenized with ethyl methane-sulphonate to study possible mechanism/s involved in the potential antagonistic properties of the strain.

Production of hydrogen cyanide, ammonia, siderophores and hydrolytic enzymes like lipase, amylase and protease were detected in all the mutants as well as in the parent strain, but fungal cell-walldegrading enzymes,  $\beta$ -1, 3-glucanase and chitosanase were not detected in

any of the mutants as well in the parent strain.

Partial characterization of extracellular diffusible antifungal metabolites by thin layer chromatography and SDS PAGE revealed cyclic lipopeptides belonging to iturin group of peptide antibiotics that are hydrophobic, methanol soluble and exhibit stability at a wide range of temperature and *p*H. HPLC and LC MS analyses revealed presence of iturin, fengysin and surfactin in cell-free extracts; iturin operon presence was also confirmed by the PCRbased assay.



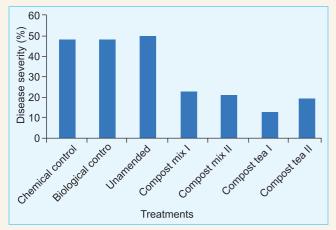
Bioassay of *Bacillus subtilis* RP24 and its negative mutant (NM), and ethyl acetate extracts of RP24 **(a)** and its mutants (**b** PM1, **c** NM, **d** PM3, **e** PM9) against *Macrophomina phaseolina* 

This study revealed broad antifungal spectrum of *Bacillus* strain; and this needs to be exploited as an effective biocontrol agent and as a rich source of antifungal compounds.

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### Novel microbe amended disease-suppressive composts

Efficacy of the microbial antagonists' amended paddystraw compost and compost-tea (extract of compost) for suppressing diseases caused by plant-pathogenic fungal consortium – *Fusarium oxysporum*, *Pythium debaryanum*, *P. aphanidermatum* and *Rhizoctonia solani* – was evaluated on the tomato seedings. Comparative performance of the fungi-challenged treatments revealed superiority of both amended compost and compost-tea in enhancing seed germination, seedling length and biomass; with 40-50% enhancement in plant parameters. The most effective control of the diseases was obtained by compost-tea obtained after amendment with synergistic cyanobacteriumbacterium combination (compost-tea I – Anabaena



Influence of different treatments on disease severity of fungi-challenged tomato-seedlings

oscillarioides and Bacillus subtilis). The amended compost and compost- tea led to significantly better control; 29-37 % reduction in disease severity was observed over biological control (*Trichoderma* formulation) and chemical control (*Thiram*-Carbendazim). This study revealed for the first time, promise of novel microbe–fortified composts in the sustainable and organic agriculture.

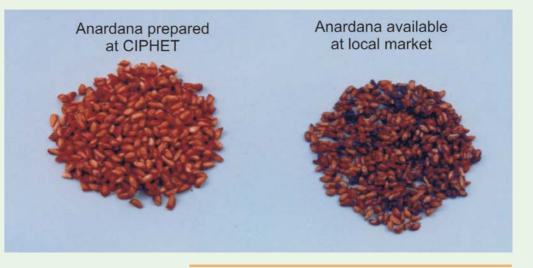
#### Radha Prasanna, Vidhi Chaudhary, Ajinath S. Dukare, Lata and Anil Kumar Saxena Division of Microbiology, Indian Agricultural Research Institute, New Delhi 110 012

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# Novel value-added products from wild pomegranate

# Anardana and its powder

Wild Pomegranate (Punica granatum) fruits are found in abundance in sub-mountainous and outer Himalayas of Himachal Pradesh, Jammu and Kashmir and in Uttarakhand, up to an altitude of 1,800 metres above mean sea level. The conventional utilization of wild pomegranate fruits lies



in drying seeds along with pulp (arils), which constitute *"Anardana"*.

An improved technology to process wild pomegranates into quality *anardana* having maximum shelf-life has been developed.

**Technology for quality** *anardana*. After separation from wild fruits, arils are subjected to steam blanching for 5 minutes. They are dried in cabinet driers at optimum temperature of 55° C to yield best quality *anardana* and its powder. This *anardana* can be stored up to 6 months at the room temperature and 8-10 months at low temperature ( $8 \pm 2^{\circ}$ C) when packed in the LDPE 200g bags and glass bottles. The cabinet dried *anardana* at 55° C retains desirable acidity, maximum vitamin C and appealing characteristic red colour, and is much superior in quality to the local traditional product.

#### Anardana nutritional composition

	· · · · · · · · · · · · · · · · · · ·
Acidity	7.0%
Ascorbic acid	16.0 mg/100g
Reducing sugar	36.0 %
Zn, ppm	30
Fe, ppm	168
Cu, ppm	489
Mn, ppm	11
Proteins (%)	1.47
Total phenols (%)	9.3

The dried anardana is made into powder in heavy duty grinder. The final product i.e. *anardana* powder after sieving in 60 mess (size: 0.190mm) and packed in 150 gauge LDPE can be stored up to 6 months.

#### Anardana hazmahazham

**Procedure for preparation.** For *anardana hazmahazham* preparation, following formulation can

be used:*anardana* powder (size: 0.19 mm, 250g) +sugar (250g)+glucose (50g).Along with the above mixture, following ingredients are added (for 1,000g of *anardana* powder).

Sindhi salt	150 g	
Nimbu satava	120 g	
Safed zira	160 g	
Kali mirch	160 g	
Pipali	160 g	
Dhania	90 g	
<i>llaichi</i> (Big and small)	160 g	
Navasadar	40 g	

After grinding these ingredients into fine powder, they are mixed well with *anardana* powder. Sugar and glucose are also to be mixed till a homogenous mixture is prepared. The final form of the product is produced by projecting mixture to extra duty single stroke Multipunch Tablet/slab making machine. The product tablets should be packed in 200g metalized poly propylene (MPP) pouches. The *anardana hazmahazam* tablets can be stored for 6 months in appropriate package without spoilage.

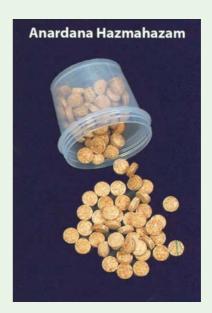
### Anardana ready-to-mix chutney

The prepared *anardana* powder is used for making ready-to-mix *anardana* chutney of acceptance quality and texture.

Anardana chutney	composition
Moisture	5.0%
Acidity	8.2%
Protein	1.23%
Vit. C(mg/100 g)	19.5
Reducing sugar	27.7%

The formulation for *chutney anardana* (50.00g) + *dhania* (10.0g) + *pudina* (10.0g) + *chilli* (5g) + onion (2.5g) has shown better texture, attractive colour, appealing flavour and smell, and acceptable taste, refreshing mouth-feel, and within acceptable microbial load, least browning and spoilage during storage.

All the ingredients including salt are ground to uniform slurry in a blender for 3-5 minutes. Citric acid is added to avoid any microbial spoilage during longer drying period of chutney. The slurry is spread uniformly on the aluminium trays of 15cm x 30 cm (200g/tray), and dried in cabinet-dryer at  $65\pm2^{\circ}$  C for 10-12 hours. The prepared powder form of *chutney* is packed in 100 gauge HDPE (Polyethylene film) and 100 gauge Metalized Poly Propylene (MPP) pouches. This *chutney* can be stored up to 6 months without spoilage in its original qualities. For retention of its typical quality characteristics, the product



must be kept in cool and dry place. For immediate consumption, the powder form of *chutney* is mixed with luke warm water to form a paste.

#### Preparation of rind powder

Mature and healthy pomegranate fruits are washed and cut manually to separate seeds and rind. Rind cut into small pieces (10-20 mm) is dried in an air circulatory tray dryer at 60°C for 48 hr. Dried pieces are cooled and powdered in a heavy-duty grinder, sieved using a 60-mesh sieve and packed in 200 gauge High Density Poly Ethylene bags.

#### Waste utilization

Pomegranate waste like rind, leaves and flowers can be processed for value-added products having medicinal, industrial and cosmetic value.

The study revealed that wild pomegranate has maximum potential of producing nutritional and mineral rich rind powder having maximum potential for retaining its quality for more than six months when stored in a low temperature. Rind powder containing high concentration of proteins and phenols will enhance absorption and retention of  $\beta$ -carotene, minerals and other flavonoids when consumed in different forms for nutritional, medicinal and *ayurvedic* purpose.

The rind powder of wild pomegranate and variety Mridula have been found to contain maximum nutritional and mineral contents, and retain colour and quality for more than six months when tightly packed in 200g polyethylene bags.

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### Zinc-phosphate solubilizing Aspergillus flavus

A fungus was isolated on tris-minimal agar medium supplemented with insoluble zinc-phosphate compound from rhizosphere soil of wheat grown under soybean-based cropping systems in Malwa region of Central India. Based on the morphological characterization and molecular analysis using partial sequencing of the ITS regions (including full ITS1 5.8S and ITS2 regions) of rDNA, this fungus was identified as Aspergillus flavus ZFS-2 with Accession no. NFCCI 1780). It is noteworthy that A. flavus produced highest halo diameter of zinc solubilization zone on trisminimal agar medium supplemented with insoluble zinc-phosphate compound as compared to other zincsolubilizing fungi viz., Scytalidium sp. ZSF-7, Fusarium oxysporum ZSF-3 and Penicillium citrinum ZSF-4 used in the study. While evaluating in broth (liquid), on the

same medium, the fungus was found to release considerable amount of soluble Zn (2.89  $\mu$ g /ml broth) and P (50.489  $\mu$ g/ ml broth) in broth; significantly higher than uninoculated control. A concomitant reduction in *p*H of broth from 6.8 to 3.54 inoculated with *A. flavus* was noted, indicating a mechanism of solubilization. The fungus was found potentially active producer of acid- and alkaline- phosphatases and phytase enzymes in Czapeck's dox broth.

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# Heat, light and storage stable azadirachtins



Sensitivity of the azadirachtin molecules to acid, base, moisture, heat, and light and their instability in storage and field has necessitated improvements in stabilization for shelf-life and bioactivity.

An improved process has been developed for

preparing storage-stable reduced azadirachtins comprising dihydro- and tetrahydroazadirachtin, and

that has been formulated to 1-20% a.i. azadiractin. Reduced azadirachtins display higher degree of antifedent and growth inhibitory activity against *Spodoptera litura* and *Spilosoma obliqua*.

Reduced azadirachtins based formulated products can be applied from 20 to 200 g/ha to control insect-pests on agricultural crops and stored grain commodities.

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# Low-cost weaning infant food for combating malnutrition

Weaning period is very crucial phase in child's development, which if not properly taken care of, leads to malnutrition and other complications at the early preschool age children. Ready-mix foods available in the market are costly.

Weaning foods prepared for children in coastal Odisha are especially deficient in protein and vitamin A, though protein -rich pulses and vitamin A- rich sweet -potato are abundant in the state. Some of the weaning foods adopted by the mothers in the state are as follows — The most preferred is roasted *chura* (flakes) powder +milk + sugar and soft-boiled rice + boiled potato; this is deficient in protein, fat, vitamin A and the other nutrients; Muslim families are not able to give colostrum to the newborns as customary, they breast- feed infants after three days, and only give milk up to an age of one year; Most of the families prefer lactogen (powdered milk) to cow's milk with the belief

that cow's milk causes cough and cold in infants. In most of the families hygiene was not maintained during preparation and handling of foods that caused diarrhoea in infants.

Common weaning foods in the coastal Odisha and some combinations of the mixes of the foods locally available were tried. For preparation of mixes, sweet-potato (Gourie variety, rich in vit A), green leaves (amaranths), and potato were dehydrated, kept for quality storage in airtight containers and in polythene bags. It was observed that sweet-potato chips and potato-chips can be kept in good condition up to a year in the air- tight containers, and green leaves up to 6 months in polythene bags.

Roasted flake powder + roasted wheat powder + pulses (mung *dal* + chickpea *dal*) + sesame seeds powder were selected as the base for weaning mix. After standardization of the base, weaning mix was tried out with various combinations of dehydrated sweet-potato powder. Five combinations were tried and tested for their acceptability by mothers on the basis of taste, flavour, texture and appearance, and base + dehydrated sweetNutrient composition of sweet-potato based weaning mix (per 100g)

Nutrients	Percentage
Moisture	5.81
Crude protein	10.86
Crude fat	5.89
Total ash	1.95
Crude fibre	2.78
Energy value (Kcal/ 100g)	456
Beta-carotene (mg/ 100g)	84.54
Dry matter	94.19

potato powder in 75:25 proportion was selected finally.

Low-cost weaning mix technology is a boost for farmwomen in the rural areas, and dissemination of this technology on a large scale will lead to healthy development of children in the rural India.

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### Value-added products from banana pseudostems

Sap

Total

Banana fibres extraction with the existing fibre-extracting unit was tedious, time-consuming and non-profitable. The Navsari Agricultural University, Navsari, with the assistance from the CIRCOT, Mumbai, a consortia partner, modified Raspador unit and standardized process for fibre extraction. Besides fibres, vermicompost, sap and candy are developed from banana pseudostems.

The products for which economics was calculated are fibre extraction, vermicompost preparation and sap as liquid fertilizer and candy from central core.

Product	Particulars	Amount (₹)
Fibre	(1.1) Variable cost (₹/ha)	
(587 kg/ha)	- Labour (@₹100/day/labour)	18,255
	- Electricity charges (@ ₹7.0/unit)	7,170
	(1.2) Fixed cost (₹/ha)	
	- Raspador machine (cost ₹100,000	1,292
	with lifespan of 15 years + mainter	nance
	and repairing @ 2% + upset value	
	10% + interest @ 12%)	
	Total cost for fibre (1.1+1.2)	26,717
Vermicompost	(2.1) Variable cost	
(12,000 kg/ha)	- Scutcher, 30 tonnes/ha	7,500
	(@ ₹0.25/kg labour charges)	
	- Dung, 12 tonnes/ha (@ ₹0.75/kg)	9,000
	- Earthworm	700
	- Labour cost (bed preparation,	9,000
	watering, harvesting, processing)	
	- Bagging	1,200
	(2.2) Fixed cost	
	- Nethouse (@ ₹300/m²)	4,400
	- Maintenance and repairing (@ 1.09	
	Total cost for vermicompost (2.1+2.	.2) 32,200
Sap	Labour charges (@ ₹0.10/litre)	1,500
(15,000 litres/ha	) 15,000 litres sap/ha	

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Economics of products from banana pseudostems (on hectare basis)					
Product	Yield (kg or litres/ha)	Total cost (₹ha)	Selling price (₹kg or	Gross income (₹/ha)	Net income (₹/ha)
			litres)		
Fibre	587	26,717	85	49,895	23,178
Vermicom	post12,000	32,200	5	60,000	27,800

124,895 Employment generated (man days/ha) 183

15,000

13,500

64,478

1,500

15,000

**Note: 1.** Biomass yield of pseudostem 68,000 kg/ha i.e., 3,400 plants/ ha (spacing 2.4m 1.2 m) considered. **2.** The standardization of processes for preparing yarn, fabrics and handmade paper on large scale are in progress. Similarly, the processes for preparing MCC and mordant have been developed on the laboratory scale only.

Cost of production of candy (10 kg) from central core of pseudostem

Particulars Amo	ount (₹)
Cost of sugar, potassium metabisilphite, citric acid	490
Labour charges (cleaning, cutting, dicing, processing)	100
Electricity consumption	315
Gas and other miscellaneous expenses	10
Packaging	33
Total cost for candy	948

Economics of candy production on a small scale (10 kg)

Product	Total cost of production (₹/10 kg)	Selling price (₹/ 10 kg)	Net income (₹/ 10 kg)
Candy	948	1,800	852

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### Canine spinal injuries and paraplegia treated successfully

A three-and-a-half years old male Dachshund dog suffered from hind-limb paralysis following dog-fight that resulted in compression of spinal cord and fracture in T12 and T13 vertebrae; dog could move the forelimbs very fast but dragged the hind-quarters.

Bone-marrow cells from the iliac crest of a dog were aspirated and mesenchymal stem cells were isolated, cultured in the laboratory and transplanted (allogenically) in the dog at the injured site. After oneand-a-half months, the animal showed some improvement, and it started putting some weight on its right hind-limb when brought to standing position; however, the animal was not able to get up on its own. After 40 days of a second injection, the animal was able to get up for a few seconds and after another five days was able to walk a short distance. Thereafter, the animal improved faster, and in the next 15 days, the animal was able to walk and move freely and all the wounds on the hind-limb also healed.

In another case, a 10-year- old Spitz dog had fallen from a height and suffered fracture of the vertebrae and disk protrusion. The owner agreed to try one last time with the hope that the new stem-cell technology may cure the animal and relieve it from suffering. The animal was earlier treated with the attending veterinarians with numerous drugs including methyl calcium prednisolone acetate, preparations, meloxicam, nervine tonics, gabapin, pregablin and electroacupuncture for about 11 months. Though some improvement was seen in the initial period and the animal was able to get up and walk for short distances with great difficulty but no further improvement could be observed in the last 5-6 months. This dog was given allogenic mesenchymal stem cell therapy, and it showed some improvement after a month's time. Incoordination of hind-limbs had reduced and animal was able to get up and sit down without difficulty. It walked without dragging limbs and used hind-limbs even for scratching. Proprioception was almost normal and patellar reflexes were normal in both the hindlimbs. The animal was again treated with stem cells at the same site and examined after 20 days and was found to have improved tremendously. The overall recovery was considered satisfactory.

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### HACCP principles in shrimp hatchery for disease risk-mitigation

Hazard Analysis Critical Control Point (HACCP) can be applied to shrimp production, starting from hatcheries, with the emphasis on preventing diseases and ensuring food safety. To follow HACCP principles, it is mandatory for hatchery to prepare a HACCP manual, based on the hatchery's requirements and environment.

In view of the need for the preparation of specific manuals for shrimp hatcheries in the country, an electronic guide in CD-ROM has been developed on the applications of HACCP principles. This will serve to prepare a HACCP manual for each shrimp hatchery and also serve as an awareness tool to highlight potential of HACCP in tackling food safety and diseases' risk.

The guide contains pre-requisites and HACCP procedures, seven principles of HACCP(hazards, Critical Control Points (CCPs), monitoring procedures, requisites for HACCP certification, good hatchery practices (GHPs), procedures after spawning, sanitation standard operating procedures), HACCP plan for shrimp hatchery operations, chemicals and treatments commonly used in shrimp aquaculture, antibiotics and pharmacologically active substances banned for use

in aquaculture, quarantine / maturation tank daily datasheet, spawning/hatching tank daily data-sheet and larval-rearing tank daily data-sheet.

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### WAY FORWARD

Here and the sector of the sec

Hilly areas provide congenial environment for the growth of variety of high-value fruits, flowers, especially orchids, vegetables and forest products. And these areas also support diverse livestock species; mithun and yak are endemic to them. In the NE Region, the potential of livestock has not been harnessed fully; and returns from livestock, especially dairying and mixed farming, in small and medium holdings can be highly sustainable. Development of this sector, therefore, will result in balanced rural economy and also improvement in the socio-economic status of the poor people associated with livestock.

The ICAR research institutes and SAUs cater to the technological needs of the hills. Despite the generation of a large number of suitable agricultural technologies, majority of the hill-farmers still follow age-old practices, thus lowering of productivity. Hence, taking appropriate technologies to farms and development of related infrastructure like strengthening inputs' supply system, conservation and judicious utilization of natural resources and marketing network can provide much needed boost to agricultural growth in the hilly areas. In hills, most of the areas face scarcity of water, despite adequate rainfall. Focused efforts are needed for water-conservation measures. LDPE filmlined tank is an inexpensive water-conservation device. In this, public-public, public-private-partnership and involvement of NGOs can be of immense help.

Seed replacement rate is also low in hills and so use of improved seeds is likely to result in production gains up to 20-40%. Promoting cultivation of quality maize protein, a rich source of lysine and tryptophan, can help in alleviating malnutrition in hills without sacrificing yields. Strengthening of seed and quality planting material chain with the active involvement of KVKs' and farmers' participation is a must.

Diversification of agriculture is also of paramount importance for hill agriculture. Hills have advantage of producing off-season vegetables during summer. Cultivation of cut-flowers and mushrooms and apiculture can also greatly uplift livelihood security of hill-farmers. Livestock is an important socio-economic component of culture and agriculture that serves as



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

the pillar of rural food, nutrition and livelihood security. However, the need for quality fodder in adequate quantity is a perpetual constraint in harnessing the potential of livestock production in hills. Programmes of fodder production can be initiated on wastelands, forest floors and terrace-risers, and by utilizing dual-purpose (green fodder-cum-grain) crop varieties. The concept of 'feed-fodder bank' will be of great help for livestock-rearing in hills.

And with the existing fish production of about one lakh tonnes in Himalayan states, cold-water fishes, particularly trout, snow-trout, mahseer, common carp, can add significantly to income of hill-farmers.

Serious efforts are also needed for mechanization of agriculture in hills to reduce drudgery. Vivek Thresher-1, a barnyard and fingermillet thresher, and paddle-driven paddy thresher are consistently used in hilly areas. Similarly, mechanization in handling of horticultural produce – picking, grading and packing – for enhanced profits can give necessary thrust to horticulture. It is equally important to create processing and value-addition units near to or at the sites of production.

The technology backstopping provided by the ICAR institutes and agricultural universities located in the hill region to farmers has enhanced productivity, profitability and their socio-economic status. With the existing infrastructure and proven technologies, hill agriculture is on the threshold of making a paradigm shift. But the challenges call for concerted efforts from all stakeholders to make hill agriculture sustainable as well as profitable, besides generating additional employment opportunities for youth.

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