



Plant Biotechnology Division

Jammu & Kashmir state is rich storehouse of medicinal, aromatic and other economic plants due to the wide variation of temperature (-20°C to 45°C) and altitudes (300 to 8,600 meter above the sea level) found throughout the state. The Plant Biotechnology Division at IIIM, taking advantage of rich biodiversity of India in general and J&K in particular, uses cutting edge molecular biology and genetic tools to engineer metabolic pathways and produce elite varieties with desired chemoprofiles and for production of novel phytochemicals.

Missions & Goals

To prepare a detailed database of medicinal plants, their active principals and medicinal activities.

To enrich the existing herbarium and drug repositories with newer varieties.

Discover new phytochemicals as well as newer uses of existing phytochemicals in (a) Medicine (b) Health foods (c) Aromatics (d) Culinary (e) Beauty care (f) Oral care (g) Baby care (h) Life style ailments (like obesity).

Conservation of plant varieties with desired properties and development of agrotechniques for their large scale propagation.

Gene prospection, pathway elucidation and gene-expression and regulation analysis via systems biology approach encompassing transcriptomics, proteomics, metabolomics, genome annotations and bioinformatics.

Develop tissue culture and genetic transformation methods for medicinal plants.

Genetic engineering of medicinal plants for making them resistant to diseases, to produce novel phytochemicals, to alter the metabolic pathways in order to overproduce desired metabolites and suppress production of toxic compounds.

Competencies

A strong herbal resource base with huge biodiversity
Extension farms with different agroclimatic niches suitable for cultivation of varied medicinal and aromatic plants. These experimental farms are being used for large-scale field trials.

Large scale tissue culture facility and related expertise for micro-propagation of endangered varieties

A large herbarium collection, Repository of raw and synthetic drugs derived from plant sources and a database of all medicinal and aromatic properties associated with major Indian medicinal plants. This rich resource is used not only for experimental work, but the knowledge is used for certification and quality estimation of various plant based drugs sold in open markets.

State-of-the-art facilities and scientific expertise for chemical (GC-MS, HPLC etc) and molecular (DNA markers for genetic diversity) evaluation and quality assurance conforming to global standards

Facility and scientific expertise for modern day molecular biology techniques like EST library preparation, DNA sequencing, Full length cDNA cloning, heterologous protein expression and purification, in vitro enzymatic activity characterization, Transgenic overexpression of a gene, use of RNA interference for down regulation of genes in transgenics etc.

Facilities

Janaki Ammal Herbarium houses more than 21500 specimens representing 3254 species, 1,152 genera and 218 families. This facility is used by scientists from academia & herbal drug industry as ready reference for authentication of plants. It is recognized internationally and is registered in *Index herbariorum* at New York, U.S.A under the acronym RRLH.

Herbal Drug Repository contains 3600 crude drug samples of authenticated parts of the plants, used as medicine. This referral facility is accessible to pharmaceutical industry,

traders, medicinal practitioners, natural product chemists, students and academics and provides ready material for laboratory scale experiments.

Experimental Farms at different altitudes in Jammu, Srinagar and Yarrikha offer unique opportunity to conduct field trials on tropical, subtropical and temperate climate plants.

Molecular Biology facility contains 96 slot gradient PCR machines, Real-Time PCR, High throughput automated DNA sequencer used for sequencing large scale EST libraries, Gel Documentation systems and other such lab equipment for molecular biology work make it possible to plan and execute any mol-bio based project in the lab. We also have the facility to do differential expression analysis using 2D protein gels. The spots are eluted and then handed over to the analytical division of IIMM for identification. Softwares for automated analysis of DNA marker polymorphism and other routine molecular biology work are also available.

Plant tissue culture and Hardening unit is used for *in-vitro* regeneration and transgenesis. The facility is well equipped with laminar air hoods, plant growth chambers and culture rooms. Large green house is used for acclimatization of tissue culture raised plants before transferring them to the fields.

Cell Biology and Cytogenetics studies like Karyotyping and Fluorescence *in-situ* hybridization (FISH) is carried out using sophisticated microscopes and softwares for automated analysis.

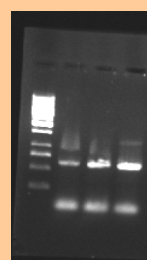
Current Research

Ethylene modulation via antisense transgenics in *Pelargonium graveolans*

Rose scented geranium (*Pelargonium graveolens*) is an important worldwide essential oil bearing plant. Under tropics and subtropics, geranium crop is highly susceptible to wilting due to production of ethylene on account of environmental stresses imposed by thermal shock, high humidity and variation in available water amount. To modulate the production of ethylene via biotechnological interventions, a partial length sequence encoding a key regulatory enzyme 1-aminocyclopropane-1-carboxylate synthase (ACC Synthase) has been cloned. To supplement and complement these efforts, an efficient *in vitro* regeneration system has also been developed for the introduction of antisense or polynucleotide of ACC synthase for transgenic development.



An efficient *in vitro* regeneration protocol via organogenesis and forced axillary bud induction; Field view of luxuriantly growing *in vitro* regenerated plants.



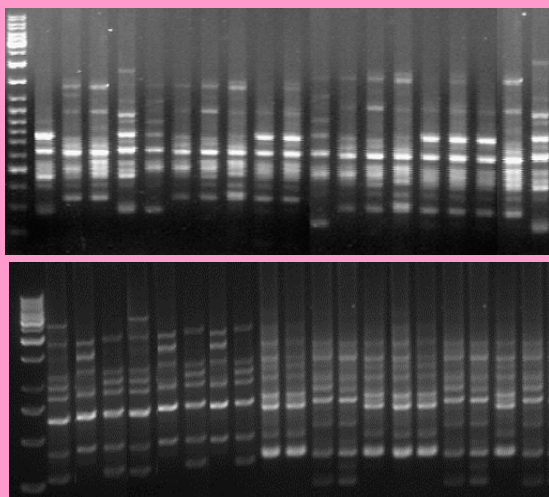
Amplification of ACC synthase gene in *Pelargonium graveolens*

← 630bp

Molecular tagging, reproductive biology and development of commercial cultivars in medicinal and aromatic plants

AFLP, ISSR and RAPD profiles of *W. somnifera* (Ashwagandha), and *Andrographis paniculata* (Kalmegh) from different geographical regions of India were developed to discriminate between different morphochemical stocks. DNA based molecular markers were successfully used to resolve the correlation of molecular data with the presence of secondary metabolites.

M 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19

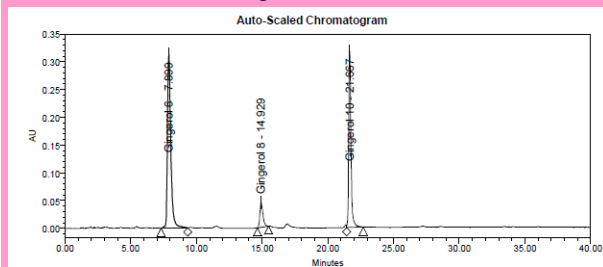


Amplification profiles of *W. somnifera* by RAPD (OPC-11) (a) and ISSR (TGG)₄ (b) Lane numbers 1-19 represent the accessions as: AGB 002, AGB 003, AGB 005, AGB 006, AGB 012, AGB 013, AGB 015, AGB 021, AGB 024, AGB 025, AGB 030, AGB 032, AGB 035, AGB 039, AGB 044, AGB 045, AGB 046, AGB 062, AGB 058. M, Molecular Weight Markers (10 kb DNA ladder). *J Plant Biology* 2008, 35 (2): 107-113 ; *Genetic Resour crop Evol* 2008. 55 (1): 33-43

DNA fingerprinting, proteomics and chemo profiling of traditional ginger cultivars of Northwestern Himalayas



Field view of Ginger



HPLC chromatogram showing 6,8,10 gingerol

Ginger rhizome its oil & oleoresin are medicinally & economically important crop. It is grown all over India in different agro-climatic zones. Lots of variation in terms of morphology and chemistry exists in the germplasm. However the real potential of the crop has not been explored at maximum due to lack of study in this area. This project on ginger variability studies, will assess genetic diversity using retrotransposons and genic markers. Chemical evaluation of oil and oleoresin of all the lines collected from the Northwestern Himalayas is being analyzed in fresh as well as dried ginger. The fiber content and other important quality parameters will be correlated with each other for selecting elite lines.

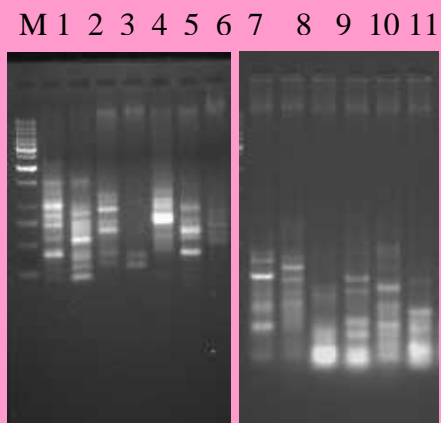
Rapid plant regeneration and analysis of genetic fidelity of in vitro derived plants of *Chlorophytum arundinaceum* baker- an endangered medicinal herb

An efficient *in vitro* multiplication system via multiple shoot bud induction and regeneration has been developed in *Chlorophytum arundinaceum* using shoot crown explants. High multiplication frequency, molecular, cytological and phenotypic stability ensures the efficacy of the protocol developed for the production and conservation of this important endangered medicinal herb.



Development of hardy and drought tolerant variety *kalam* - a citral rich lemongrass released for commercial cultivation

Hardy, drought tolerant strain of lemongrass (RRL F2-38) christened as KALAM has been developed through hybridization and rigorous screening of the segregants in F₂ generation. High survival coefficient (80-90%) coupled with high stability and wider adaptability are some of the essential attributes of the variety developed. Herb and oil yield averages 40 tons and 170 L/ ha respectively. The citral percentage averages around 81%. The returns from KALAM lemongrass are two and half times higher than the traditional crops grown under rainfed conditions.



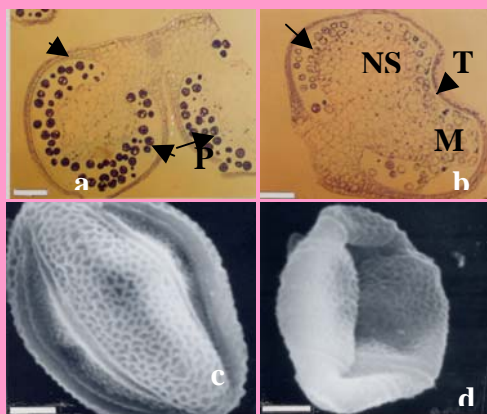
DNA fingerprint of variety KALAM using different arbitrary primers (Operon Technology)-lane 1-7 (S116, S119, S107, S111, S113, S120, S164) and SSR (National Bioscience, Inc.) - lane 8-14 (11-12, 21-22, 23-24, 27-28, 37-38, 39-40), M-Molecular Weight Markers (2 kb)



Release of hardy and drought tolerant variety 'KALAM' by Hon'ble. Ex-President of India Dr. A. P.J. Abdul Kalam

Genetics and mechanism of induced male sterility in *andrographis paniculata* (burm. F.) Nees and the significance thereof

Andrographis paniculata is predisposed for selfing due to its floral architecture and overlap of male and female phases. On account of its obligate inbreeding nature, small flower size, intimate proximity of minute reproductive parts and their vulnerability to mechanical injuries during manual emasculating, it is extremely tedious and laborious to emasculate the flowers to harness the benefits of intervarietal hybridization. To obviate some of these bottlenecks, the present study was aimed to explore the possibility of inducing male sterility in order to emasculate the flowers genetically with a twin objective of optimizing its genetic amelioration, and also to enrich our germplasm resource base.

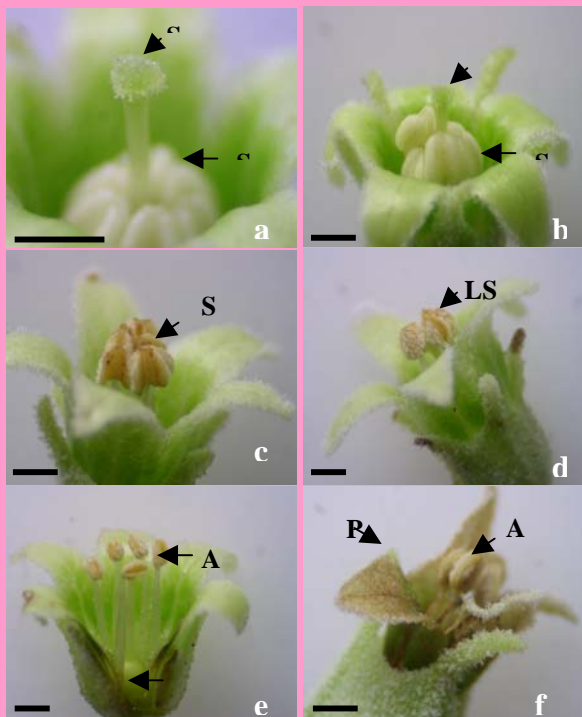


Semithin sections of bisporangiate fertile and sterile anthers: Stained viable pollen (P) and intact tapetum (T) of male fertile anther (Bar = 100 µm) (a), unstained aborted microspores (M), hypertrophied non-sporogenous tissue (NS) invading the locule of male sterile anther (Bar = 100 µm) (b), scanning electron micrographs of fertile pollen (Bar = 5 µm) (c) and of sterile pollen (Bar = 5 µm) (d)

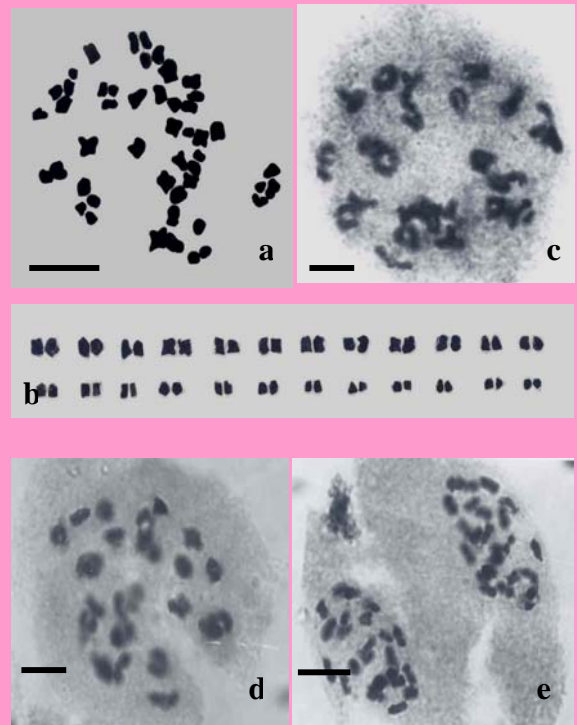
Versatile sexual mechanism ensures robust genetic and chemical polymorphism in *Withania somnifera*

Withania somnifera presents a versatile sexual polymorphism of mixed mating. Experiments indicate that that individual flowers exhibit partial temporal dichogamy of protogynous type, under which receptive stigma remains exerted beyond the undehisced staminal cone to receive cross pollen through insect vectors. In a probable situation of non-receipt of pollen through insect pollinators,

autonomous fertilization is guaranteed by the upward staminal increase to form a cone connivent about the receptive stigma. Functional dimension and floral configuration suggests that crossing and autonomous selfing are mutually exclusive as the self-pollen arrives late during the floral ontogeny. Seed set efficiency and fruiting success are not influenced by pollen genotypes (self / cross) under different pollination treatments (autogamy, geitonogamy and xenogamy).



A flower of *Withania somnifera* just after anthesis showing papillate, globose, exerted stigma S beyond the staminal cone SC (a), relative position of receptive stigma S and staminal cone SC after two days of anthesis (b), staminal cone SC connivent about the receptive stigma after 3 days of anthesis (anther dehiscence stage) (c), arrow showing pollen presentation through longitudinal slit LS on outer of the stigmatic side (d), ovary O after fertilization and reflexed-dehiscent anthers A (e), abscising petals P and anthers A after 6-7 days of anthesis (f). Bars = 1 mm.



Somatic metaphase spread showing 48 chromosomes (a), the karyoidiogram thereof depicting 10 M + 9 SM + 5 ST (b), diakinesis (c), meiotic metaphase-I presenting 24^{II} (d), late anaphase-I showing normal 24:24 disjunction (e). Bars = 10 µm.

Curr. Sci. 2007, 92(10): 1390-1399.