Combined Proceedings IPPS 69:438-518. 2019.

Message from Convener

Rakhi Chaturvedi

Department of Biosciences & Bioengineering Indian Institute of Technology Guwahati, Assam, India

rakhi_chaturvedi@iitg.ac.in

Abstract

Namaskar! On behalf of the Organizing Committee of "Next Generation Plant Production and Utilization Technologies (NGPPBUT-2019)", it is my privilege and honor to welcome you all to Indian Institute of Technology Guwahati (IITG), the most dynamic and beautiful educational campuses in the country . This year IIT Guwahati has completed it 25 years of journey and this conference is being organized to celebrate its silver jubilee.

NGPPBUT-2019 also marks the first meeting of the International Plant Propagators' Society in India. It endeavors to provide a unique forum to more than 250 Indian/International attendees for three days of intensive presentations, beneficial discussions and field tours. It will bring together leading academic institutions and plant biotechnology industries, making the conference a platform for aspiring academicians, plant biotechnologists, leading industrialists, technocrats, students, entrepreneurs and plant growers to promote information exchange, networking and strategic alliances. The conference will establish the best possible model for livelihood transformation, plant production technologies, bioresources management and utilization in the changing world. The scope of the conference will include all aspects that are pertinent to successful development and commercialization of quality plants and their products.

We once again welcome you to International Conference on NGPPBUT 2019 and are confident that your engagement, scientific contribution and ideas will pave a way to provide solutions to challenges in the field of Plant production technology. IIT Guwahati, with its idyllic setting in a region tremendously rich in flora and fauna, provides the perfect background to the engagements and possible solutions that we seek to have.

Advances in Nursery Production Technology

Alan M. Jones

Manor View Farm, 15601 Manor Road,
Monkton, Maryland, USA

ajones@manorview.com

Keywords: Nursery, Industry

Plenary speaker

Abstract

This talk will review a new, unique, and different technology that is being utilized in the nursery industry in the USA and around the world.

The talk will also look at research that will inspire new and different uses for the technology within the industry.

Designing Crop Plants for the Future: Molecular Genetic Enhancement of Rice Yield and Resilience

Prakash P. Kumar

Department of Biological Sciences,
National University of Singapore,
Singapore

prakash.kumar@nus.edu.sg

Keynote speaker

Keywords: Avicennia officinalis, Casparian bands and suberin, lamellae, rice plants, salinity tolerance

Abstract

Salinity stress affects growth and yield of crops. Genetic variations cause different plant species to exhibit different degrees of stress tolerance. Hence, we need to understand how plants respond to stresses in order to design plants with improved tolerance to salinity without compromising yield. We will discuss our attempts to apply findings from *Arabidopsis* and the mangrove *Avicennia officinalis* to improve rice plant. A study of *OsTPS8* of the trehalose-phosphate-synthase (*TPS*) gene family showed that *OsTPS8* controls salinity-stress tolerance and key agronomic traits in rice. *OsTPS8* belongs to class II TPS, whose involvement in salinity tolerance was not studied previously. Loss-of-function *ostps8* mutant was salt sensitive, and ectopic expression of *OsTPS8* confers salt tolerance in rice without any undesirable effects. Another aspect to be discussed is application of findings from *Avicennia* that grows in saline environment with the help of specific adaptations, including NaCl exclusion (ultrafiltration) at roots. *Avicennia* roots exclude ~95% salt by developing enhanced hydrophobic barriers (Casparian bands and suberin lamellae in the endodermis and exodermis). Cytochrome P450s play a key role in biosynthesis of suberin precursors. We identified several *Cytochrome P450* (*CYP*) genes that were differentially expressed upon salt treatment in *Avicennia* roots. Using an *Arabidopsis* mutant, *atcyp86b1*, we characterized the function of *CYP86B1* in regulating suberin biosynthesis. When treated with salt, the mutant plant roots exhibited reduction in growth, suberin lamellae, and Casparian bands. We identified specific WRKY

transcription factors as the upstream regulators of *CYP* genes.

We exploited this molecular regulatory mechanism, and by heterologous expression of the *Avicennia CYP* genes in rice, higher salinity tolerance was conferred to transgenic rice plants. Hence, besides contributing to basic knowledge of the underlying molecular regulatory mechanism, our findings provide fresh approaches for generating abiotic-stress-tolerant crop plants in the future.

Plant Nursery of the Future

Dharam P. Sharma

Dry Creek Labs, Duarte Nursery,
Hughson, California 95326

dharam@drycreeklab.com

Keynote speaker

Keywords: Aquaponics, carbon mitigation, environmental controls, hydroponics, plant breeding, plant nursery

Abstract

Plant propagation has come a long way since the times when man learnt how to sprout a seed or stick a shoot in soil and make it grow into an independent tree. In 2018, the total turnover of global seed market was US $52.69 billion with 35% contribution from U.S. alone (www.mordorintelligence.com). Nursery and garden stores in the same year in the United States had a market share of US$ 48 billion (www.ibisworld.com), and the sales are projected to grow more than 7% every year until 2025. However, the constantly increasing population pressures are limiting the availability of land, water, and other inputs besides adversely impacting the environment. New innovations and technologies are driving agriculture to adopt cultural practices to conserve precious natural resources and nursery industry is no exception.

Carbon mitigation, hydroponics, aquaponics, vertical farming, biocontrol and automated gadgets to control environmental variables such as light, temperature, humidity, and so forth are increasingly being used to address such concerns and may see a further surge in the coming decades. The changing consumer demands, political and regulatory pressures, emerging marketing, and advertising strategies and availability of labor shall further shape the nursery industry in future. The product development and plant breeding strategies will be dictated by efficiency, quality, and consumer preferences. The online shopping is gradually taking over visiting garden centers or nurseries and the trend may increase in future. Overall, we shall witness micro-managed, more automated nursery industry with increased environmental controls.

Application of *Vitis* Regeneration System Production and Genetic Improvement

Sadanand A. Dhekney[1] and Dennis J. Gray[2]

[1]Department of Agriculture, Food and Resource Sciences, University of Maryland, Eastern Shore, Princess Anne, Maryland, USA
[2]Mid-Florida Research and Education Center, University of Florida, Apopka, Florida

sdhekney@umes.edu

Keynote speaker

Keywords: Grapevine, *in vitro* regeneration, micropropagation

Abstract

Grape is among the most ancient crops cultivated worldwide and is highly valued for its value-added products such as wine, juice, jelly, and jam. Fungal and viral pathogens affecting grapevine can result in significant decrease in productivity of vineyards worldwide. Availability of clean, disease-free planting material is an important prerequisite for ensuring sustainable productivity and long-term vineyard health. Micropropagation serves as an alternative to traditional propagation methods for rapidly increasing the amount of clean, disease-free planting material of elite grape cultivars. Cultures initiated from shoot apical meristems proliferate on a medium containing cytokinins and can be maintained by transfer to another fresh medium at regular intervals. Rapidly proliferating shoots can be regenerated following growth on a culture medium containing auxins. Grapevine somatic embryogenesis, which utilizes asexual tissues for plant regeneration, has wide applications for clonal propagation and genetic improvement. Grapevine somatic embryos are ideal target tissues for insertion of desired traits of interest and recovery of modified grapevines. Grape species and cultivars widely vary in their embryogenic response, which necessitates the optimization of protocols for individual cultivars. Factors influencing the production of embryogenic cultures including explant type, development stage, growth media, and culture conditions have been optimized for a large number of grape species and cultivars. The development of efficient grapevine micropropagation and somatic embryogenesis systems can enable large-scale production of clean planting material while advancing efforts for grape genetic improvement using precision breeding.

The Enhancement of Functional Ingredient in the Vegetables and Future Issues

Kenta Nakanishi[1] and Emiko Yanase[1, 2]
[1]Graduate School of Natural Science and Technology, Gifu University, Gifu, Japan
[2]Faculty of Applied Biological Sciences, Gifu University, Gifu, Japan

e-yanase@gifu-u.ac.jp

Keynote speaker

Keywords: Brassicaceae, glucosinolate, isoflavone, legumes, secondary metabolites, sprouts

Abstract

Vegetables are important sources of many nutrients, especially secondary metabolites. Some of secondary metabolites in vegetables, such as polyphenols, are known for improving our health and have attracted the attention as the functional components. “Sprouts” are one of the vegetables, artificially germinated by seeds and then eaten by consumers. Sprouts are said to be rich source of vitamins, minerals, and phytochemicals due to the metabolism with the germination. Legumes and Brassicaceae are two well-known sprout types in Japan; isoflavone and glucosinolate are their known as functional compounds.

The productivity of these secondary metabolites is known to be easily affected by growing conditions, such as drought, temperature, and elicitor. Conversely, by increasing the productivity of secondary metabolites, there is a possibility that functionality of vegetables can be enhanced. In this research, to develop the high-value-added vegetables by enhancement of functional ingredients, we investigated effects of plant-growing conditions on the content of phytochemicals, focusing on the soybean and broccoli sprouts. Furthermore, as various metabolites are contained in vegetables, we also investigated how plant-growing conditions affect the production of other metabolites.

Propagating Indigenous Flora for Eco-Restoration

Todd Jones

Fourth Corner Nurseries, Sand Road,
Bellingham, Washington, USA

toddjones57@hotmail.com

Keynote speaker

Keywords: Anthropocentric, native plants, next-generation restoration, propagation

Abstract

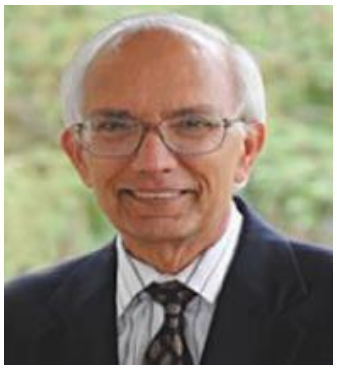
Choosing indigenous plant material for eco-restoration is an evolving science with several schools of thought. This can often make the propagation of plant material for restoration a challenge, requiring completely different methods than those used in ornamental landscape plant production. Understanding the purpose of the installation, and the goals the restoration hopes to achieve, is a critical part of the propagation process. A biocentric approach to habitat restoration considers the importance of biodiversity in the landscape and recognizes that plants do not grow in isolation from other living things around them. With the rapid reduction of wild, undisturbed ecosystems the need to preserve genetic diversity is of primary importance. This type of restoration requires the use of carefully sourced native flora; yet, the definition of what is native is not universal and can add an additional level of complexity to the propagation process. An anthropocentric approach to restoration utilizes both natural and social sciences and may not be as concerned with what is and is not native flora, its objectives may be quite different than the latter approach. Mitigating effects of human disruption to offset health dangers and concerns related to climate change are two common focus areas that utilize native plantings to achieve their objective. This type of installation may require a different plant production approach. As a long-time propagator and grower of native flora, I will outline protocols I have developed to address these differences and share some of the important elements I believe a successful next-generation restoration nursery needs in order to address this rapidly changing industry.

Soil-Centric Approach to Advancing Food Security and Improving Environment

Rattan Lal

Carbon Management and Sequestration Center, The Ohio State University, Columbus, OH 43210, USA

lal.1@osu.edu



Keynote speaker

Keywords: Climate change adaptation and mitigation, conservation agriculture, eco-intensification, soil degradation, soil organic carbon sequestration, yield gap

Abstract

India has made a commendable progress in increasing food production since 1960s. However, there are second-generation problems related to degradation of soil, contamination and depletion of water resources, pollution of air, and extinction of biodiversity. These problems are exacerbated by increase in population, rapid urbanization, and growing demands of the ever-affluent population. Despite the increase in yield of food crops and other agronomic commodities, there exists a large scope and the necessity for improvement. Important among options to narrow the yield gap is the strategy of restoring soil health. Soils of agroecosystems are prone to degradation by accelerated erosion, depletion of soil organic carbon (SOC) and plant nutrients, secondary salinization, and decline in aggregation amount and strength leading to crusting, compaction, and hard setting. Concentration of SOC in soils of some agro-ecoregions is as low as 0.05% compared with the desired range of 1.1–1.5% or more. Thus, the use efficiency of inputs (e.g., improved varieties, fertilizers and other amendments, and irrigation) is low and soils are a major source of greenhouse gases including carbon dioxide, methane, and nitrous oxide .Yet, these degradation trends must be reversed through restoration of degraded soils and ecosystems by conversion to a restorative land use and adoption of recommended practices of soil, water, crop/trees, and livestock management. A system-based conservation agriculture (CA), involving a judicious combination of no-till, retention of crop residue mulch, incorporation of a cover crop in the rotation cycle, and integration of crops with trees and livestock, can create a positive soil/ecosystem carbon budget, making soils a sink of atmospheric carbon dioxide with an attendant increase in soil organic carbon concentration and stock along with gradual improvement in soil health (physical, chemical, biological, and ecological). Adoption of CA can be promoted by payments to farmers and land managers for provisioning of ecosystem services and advancing sustainable development goals of the United Nations.

Engineering Optimization Protocols for Plant Cell/Hairy Root Cultivations

Ashok Kumar Srivastava

Department of Biochemical Engineering and Biotechnology, Indian Institute of Technology Delhi, Hauz Khas, New Delhi, India

ashokks@dbeb.iitd.ac.in

Keynote speaker

Keywords: Azadirachtin production, biopesticide, bioreactor cultivation, engineering optimization protocols, hairy root cultivation, plant cell cultivation

Abstract

Plants are considered as the versatile source of strategic secondary metabolites (flavors, pesticides, drugs, etc.) for society. Presently, these compounds are produced by large-scale cultivation of natural plants, wherein plants are uprooted, which is followed by (solvent) extraction of bioactive compounds from different plant parts. This traditional production process features several disadvantages, for example, inadequate, sustained availability of the source plant part throughout the year, low product yield arising from the slow growth of natural plant, and so forth. Sometimes the overexploitation of plant parts (particularly roots) for recovery of secondary metabolite leads to extinction of plant. Therefore, there is a desperate need to establish alternate *in vitro* plant cell/hairy root bioreactor cultivation technologies and to develop innovative bioreactor designs to not only eliminate dependence on natural plants but also significantly increase the availability of bioactive compounds for growing demands of society.

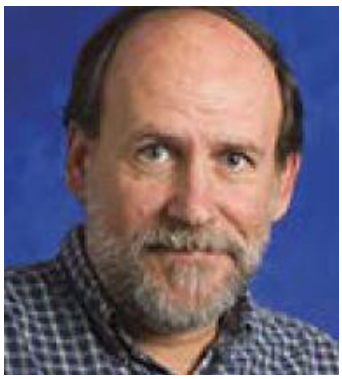
It is invariably observed that plant cell cultivation is a relatively new and faster production protocol to cultivate specialized plant cells (developed from different plant parts) and mass produce bioactive compounds but yet there is a huge scope of use of engineering optimization protocols and development of innovative bioreactor designs to significantly increase the concentration, yield, and productivity of the desired secondary metabolite not only in minimum time but also with least experimental trials. The above-mentioned methodology(ies) and some newer bioreactor designs will be presented for the mass production of biopesticide (azadirachtin) using plant cell/hairy root cultivations in a bioreactor.

Advances in Nursery Production

Robert Geneve

Department of Horticulture, University of Kentucky, Lexington, Kentucky, USA

rgeneve@uky.edu



Keynote speaker

Keywords: Automation, cuttings, plant propagation, robotics, seeds

Abstract

Nurseries produce high-value crops for the ornamental industry. They rely on seeds and vegetative propagation to provide the initial liners or plugs that initiate a production cycle. Propagation has traditionally involved a significant amount of hand labor. Recent efficiencies in work flow have reduced labor costs throughout the industry. Additional efficiencies in production have relied on computer-assisted production practices. Many of these practices reduce or eliminate traditional hand labor operations. These include environmental control systems, computer-aided irrigation control, mechanical conveyance of plant material, and robotic systems for cutting and seed propagation. This presentation will endeavor to illustrate some of the recent advances in nursery production technology with an emphasis on innovations related to plant propagation.

Designer Plants to Survive Global Warming and Climate Change

Puthiyaparambil Josekutty

Skybury Coffee Pty Ltd., Mareeba, Queensland 4880, Australia

josekutty964@gmail.com

Keywords: Breeding, climate change, gene editing, genetic diversity, genetic engineering, global warming

Abstract

Global warming and subsequent climate change are impacting agriculture and horticulture across the world. Impact of climate change affects plants to a greater extent than animals because of their confinement to the environment they are born for their entire life. Natural adaptations do occur in the plant kingdom and the fittest species and individuals survive adversities, changes to the environment arising from climate change. Abiotic stresses such as extreme heat, cold, drought, flood, sea level rise, and biotic stresses such as new diseases and pests as well as weeds/super weeds are major impacts of the climate change. Various methods "conventional and modern" for developing "Designer Plants" with capabilities to survive adversities caused by the climate change will be discussed in this paper.

Subcellular Bioengineering of *Artemisia annua* L. for Enhanced Biosynthesis and Accumulation of Artemisinin

M. Z. Abdin

Department of Biotechnology, School of Chemical and Life Sciences,
Jamia Hamdard, Hamdard Nagar, 110062, New Delhi, India

mzabdin@jamiahamdard.ac.in

Keywords: Active pharmaceutical ingredient, *Artemisia annua* L., Artemisnin biosynthesis, Secondary plant metabolism

Abstract

Plants synthesize a vast array of secondary plant metabolites through a network of complex metabolic pathways regulated by endogenous and environmental factors. Although these compounds are believed to be non-essential for plants to live, they play an important role in interaction of plants with the environment ensuring their survival in the ecosystem. Some of these secondary metabolites are also of immense medicinal importance because of their therapeutic value. These are referred as active pharmaceutical ingredients (APIs). The concentrations of these compounds in medicinal plants are, however, very low, limiting their commercial exploitation. Artemisinin is one of these APIs isolated from aerial parts of *Artemisia annua* L. It is a potent antimalarial drug against drug-resistant malaria. In recent times, the demand for artemisinin is exponentially increasing with the increased incidence of drug-resistant malaria throughout the world, especially African and Asian continents. However, the commercial production of artemisinin-based combination therapies has limitation due to the presence of low concentration of artemisinin in plants. Therefore, we employed bioengineering approach to develop transgenic lines of *A. annua* L., overexpressing HMG-Co A reductase (hmgr), amorpha-4, 11-diene synthase (ads), and cytochrome P450 monooxygenase (cyp71av1) genes to enhance artemisinin content. The selected transgenic lines were found to accumulate 1.29% to 1.44% artemisinin. Thus, results obtained in these studies, clearly indicate that the synthesis of APIs in medicinal plants is tightly regulated, and bioengineering approach can be used in modulating plant metabolism to improve their biosynthesis, so that drugs manufactured from these APIs could be available at cheaper rates to the public.

Containerisation of a Plant Production Nursery Affords a Wide Range of Benefits in Propagation and Disease Management

David O. Cliffe

Narromine Transplants Pty Ltd., Narromine, NSW, Australia 2821

david@transplants.com.au

Keywords: Irrigation, nursery, plant production, terracotta containers

Abstract

During the last 60 years there has been a transformation in many plant production nurseries worldwide as they took advantage of the availability of new plastics that would partly replace the practices of traditional in-ground growing and the use of terracotta containers. Coupled with a proliferation of container shapes and sizes and the advent of more modern substrates that better suit the use of containers, a revolution in the production and management of plants has taken place.

These changes have also allowed closer management of nutrition, irrigation application, closer attention to nursery hygiene, and significant changes in nursery infrastructure. This paper attempts to describe the evolution that has occurred over this period and the ongoing benefits that have accrued to those that have commenced or continued to practice best management principles.

In vitro plant propagation and commercial cultivation in the Micronesian region: challenges and measures for sustainable commercial pepper production

Virendra M. Verma

Micronesia Plant Propagation Research Center, Kosrae Agricultural Experiment Station (USDA Landgrant Program), College of Micronesia-FSM, Kosrae, Micronesia, USA

vmv_vmv@hotmail.com

Keywords: Best management practices, commercial propagation, organic fertilizers, pepper, soil amendments, sustainable cultivation

Abstract

Black pepper (*Piper nigrum* L.), a flowering vine of the Piperaceae family, is valued for its dried berries called peppercorns, which are known for their health benefits and used as a spice and seasoning. Native to the humid jungles of the Malabar Coast of Southwestern India, the plant is cultivated in the tropics worldwide. In the Micronesian region, it is gaining commercial importance as an important cash crop because of the premium price of peppercorns. However, the limited availability of disease-free black pepper seedlings and the trunks of the native tree fern (*Cyathea nigricans*), which are used as supports for black pepper vines, are becoming limitations for sustainable commercial black pepper cultivation in the region. Therefore, to ensure the year-round availability of uniform, disease-free, and high-quality planting material in Micronesia, an efficient micropropagation and acclimatization protocol was developed for a local commercially important black pepper cultivar (*P. nigrum* cv. Srilanka). Shoot apical meristems were used as explants for culture establishment. Best culture initiation was observed on Murashige and Skoog medium augmented with 5 µM 6-benzylaminopurine (BAP). For further growth and subsequent multiplication, the established cultures were transferred on 7.5 µM BAP and 5 µM indole-3-acetic acid (IAA). The number of multiple shoots produced from each explant after two subcultures varied from eight to 20. Best rooting was observed on 2 µM indole-3-butyric acid (IBA). Plantlets were acclimatized with 68% survival rate in 10 weeks. Research trials for sustainable commercial black pepper cultivation were designed, implemented, and vigorous vegetative growth was observed. To overcome the limitations of live tree-fern supports, nonliving supports such as reinforced cement–concrete standards were specifically designed and used to support the vines for commercial cultivation. First harvesting was done after 12 months of planting and data collection and analysis are being continued. Outcomes of this analysis would be used to provide assistance to the regional farming communities to promote sustainable commercial cultivation of black pepper in the region.

Commercial Aspects of Micropropagation and Hydroponics, The Future of Farming

Shweta Narasimhan
Plant Tissue Culture, HiMedia Laboratories Pvt. Ltd., Mumbai, Maharashtra, India

nshweta@himedialabs.com

Keywords: Conservation, conventional propagation techniques, hydroponics, plant tissue culture

Abstract

Food and Agriculture Organization's vision is of a "world free from hunger and malnutrition, where food and agriculture contributes for improving the living standards of all, especially in an economically, socially, and environmentally sustainable manner." Gains in productivity and technological advances have contributed to more efficient resource use and improved food safety. Around 795 million people still suffer from hunger, and more than two billion from micronutrient deficiencies or forms of undernourishment. Plant biotechnology holds a promise to resolve the problem and is indeed a blessing to achieve global prosperity. Understanding basic biology of plants is a prerequisite for proper utilization of the plant system or parts thereof. Plant tissue culture and hydroponics-growing plants without soil, have emerged as promising tools to increase farming outputs and grow plants in artificial habitats. As emerging technologies, both have a great impact on agriculture and industry by enriching plant population, needed to meet the ever-increasing world demand. It has made significant contributions to the advancement of agricultural sciences in recent times and today they constitute an indispensable tool in modern agriculture. With modernization in technology, currently several engineering techniques (robust, automated, and computerized) such as hydroponics have been applied to micropropagation with the objective of providing optimum environmental conditions to *in vitro* plant stock at a larger level. With the increasing demand in the technology, the demand for supply of resources has increased over a period of time allowing us to develop a wide range of products for both the techniques of farming helping growers to meet their needs.

Sustainable Farming Technologies Focused on Micro-Irrigation to Double the Irrigable Command Area and Farm Income

M. S. Ramesh

Jain Irrigation Systems Ltd., New No. 15, Old No. 10, Lakshmi Nivas, West Avenue Road, Kodambakkam, Chennai, Tamil Nadu, India

ramesh.ms@jains.com

Keywords: Crop and farm productivity improvement, drip irrigation, doubling the irrigable area with a given volume of water, integrated irrigation systems, sustainable farming practices

Abstract

In the Asian region, the irrigation is done through flow irrigation thro' canals and wells. In the canal system of irrigation, problems include (i) wastage of water thro' percolation (up to 40%) and evaporation (up to 20%); (ii) excessive use of water especially at the head end of the delivery point; (iii) unreliable water supply due to poor control over the entire system; and (iv) non uniformity of water application due to flow irrigation, rotational water supply (once in a few days), and seasonal water supply (either for a season or for 4 months in a year). Hence, the utilization is less than the irrigation potential created and there is an increasing gap between these two.

"Resource to Root™" (meaning delivering water directly from water resource to roots of individual plants), a revolutionary concept that provides effective solutions to above-mentioned challenges. While it provides an easy, precise, reliable control for the distribution of water, it also delivers water straight to roots of a plant, thus increasing the "Water Use Efficiency" by nearly 50%.

As water is distributed under pressure through closed pipe network, it results in uniform and equitable distribution to all stakeholders. Water delivered directly at root zone of plants thro' drip irrigation system ensures optimal utilization of water by crops, thus reducing water losses thro' percolation and evaporation during water conveyance from the source and also during farm-level irrigation. The saving of water in the whole project is up to 50% compared with traditional ways, thus doubling the irrigable command area. The proven benefits of "Resource to Root™" with very high overall project efficiency and water factor productivity result in doubling the irrigated area, increase crop yields, and ultimately generate more income and profits for farmers. Deliverables make these projects economically viable, thus contributing to increased share of agriculture sector in overall GDP of the nation.

Nanocarriers-Mediated Smart Delivery Applications for the Next-Generation Seed and Horticultural Products

J. Pon Samuel

Single Cell Technologies, ETT Core Technology, SIBS (Systems Innovations for Breeding Seeds product) R&D, Corteva agriscience™, Agricultural Division of DowDuPont™ (ADOD), Johnston, Iowa, USA

pon.samuel@corteva.com

Keywords: Cell-penetrating peptides (CPPs), nano-conjugates, nanoparticles, quantum dots, smart delivery, subcellular internalization

Abstract

Producing food for a healthy tomorrow calls for key enabling transformational technologies for cell manipulation by defending the genome integrity and at the same time to develop innovative future product concepts. The recent development of smart delivery applications across the intact cell walls using nanocarriers including quantum dots (QDs) and plant-derived cell-penetrating peptides (CPPs) have been facilitating the development of sensitive fluorescence biosensors due to their high quantum yield, narrow and tunable emission spectrum, and good photostability. We first introduced the use of QDs and plant-derived CPPs, their preparation and functionalization approaches for smart delivery into intact plant cells, tissues, and organs. In this presentation, we describe methods for introducing a molecule of interest into plant cells with intact cell walls by using QD-peptide linked to CPPs. The use of QDs or polystyrene nanoparticles with improved biocompatibility further promotes biological applications and we summarize QDs-based fluorescent biosensing for proteins and nucleic acids, and QDs-based applications in cellular and *in vitro* and *in vivo* subcellular targeting and imaging. Last but not the least, we envision the potential of such smart non-invasive delivery and tracking technologies for future seed products.

Iron Biofortification in Bananas by Expression of *Oryza sativa* Nicotianamine Synthase Genes

M. Mayil Vaganan, C. Sivagandhi, S. Ganesan, M. Kumaravel, I. Ravi, K. J. Jeyabaskaran, S. Backiyarani, and S. Uma

ICAR National Research Centre for Banana, Thayanur Post, Thogamalai Road Tiruchirappalli 620102, Tamil Nadu, India
mmayilv@gmail.com
Keywords: Grand naine, iron deficiency, OsNAS1, OsNAS2, rasthali, transgenics

Abstract

Iron is an essential micro nutrient for plant growth and human health. Iron deficiency is one of the most prevalent micro nutrient deficiencies in the world, causing anemia to several billions people especially in developing countries. The production of iron-biofortified staple crops will help to ameliorate iron-deficiency anemia. Banana is one of the staple crops of Asian and African population. Two commercial banana genotypes, Rasthali and Grand Naine, are biofortified by introduction of two rice (Oryza sativa) nicotianamine synthase genes, OsNAS1 and OsNAS2, driven by Zea mays ubiquitin promoter to increase the iron content in fruit pulp. Agrobacterium-mediated transformation of the constructs carrying these genes and selectable marker, nptII, was carried out with embryogenic cell suspension (ECS) of the bananas. One hundred independent transgenic events of each banana genotype for each gene construct were generated and being field grown along with untransformed control plants in iron-sufficient soils under confinement in transgenic net houses equipped with all biosafety standards as per guidelines of Department of Biotechnology, Government of India.

The transformants, before planting, were confirmed for transgene presence and selectable marker with specific primers using polymerase chain reaction (PCR) and checked for Agrobacterium contamination using Vir-C gene-specific primers to eliminate false positives. Presently, transgenic lines are 9 months old and in shooting and fruit-bearing stages. Estimation by Inductively coupled plasma atomic emission spectroscopy (ICP-OES) of iron element content of 25 representative lines of each genotype at vegetative stage is promising with seven Rasthali lines and six Grand Naine lines having average iron content of 24.5 mg/100 g dry wt. as compared with non transgenic lines with maximum of 11.2 mg /100 g. As the increase in iron concentration was 2.2 times in the transgenic lines, the results offer promise to effectively increase the level of iron in the fruit. Transgene integration and the copy number of the transformed events are being performed.

Biochar Utilization in Benefiting Plant Productivity

Chien-Teh Chen, [1] Irene Kosasih, [2] San-Gwang Hwang,[3] Ying Yong,[3]
Yi-Jun Chen,[3] and Keng-Tung Wu[4]

[1]Department of Agronomy, National Chung Hsing University, Taiwan
[2]International Master Program of Agriculture, National Chung Hsing University, Taiwan
[3]Department of Horticulture, National Chung Hsing University, Taiwan
[4]Department of Forestry, National Chung Hsing University, Taiwan

ctchen41@nchu.edu.tw

Keywords: Agricultural waste, biomass seed shell, *Caliandra*, sweet potato vines, tomato stem, and leaves (RTS)

Abstract

Normally, the agriculture waste is mixed with soil as fertilizer. However, some of agro-wastes are not suitable to be dumped into soil. For example, seed shells of oil tea (SST) are not allowed abandoned openly because of having considerable amount of alkaloid; sweet potato vines (SPV) if abandoned on the field may cause the root rot disease in the next season; residual of tomato stems and leaves (RTS) are unfavorable for continuous tomato cultivation because of having high content of phenolic compounds. The main objective of this study is to convert the eco-unfriendly agricultural waste into biochar for further utilization. Our results showed that the biochar from SST inhibited growth of rice seedling even as low as 1% in soil. SPV can be added into soil for 10% without inhibition to rice. RTS can be added into soil for 50% without inhibition to rice. It was noticed that the biochar of RTS showed significant benefits on the growth of tomato seedlings.

Biochar could be the byproduct of biomass gasification power plant and has been widely known for benefiting plant productivity. *Caliandra calothyrsus* recently utilized in biomass power plant in Indonesia, the effect of biochar on nursery of *C. calothyrsus* seedlings was conducted. Experiments were processed on the Island of Kundur, Riau Islands Province, Indonesia. A half-year of nursery planting for *C. calothyrsus* was performed. Seedlings that meet the qualification for field planting were counted for the number, height, and diameter. Results showed biochar amendment increased the qualified seedling by 18%. In 1-year trial of field planting, biochar amendment increased wood and leaf yield of *C. calothyrsus*. It is concluded that the application of biochar in *C. calothyrsus* nursery planting could be included in circulating system for biomass power plant.

Biomass Utilization for Development of Value-Added Products for Food Packaging Applications

Vimal Katiyar

Department of Chemical Engineering, Indian Institute of Technology Guwahati, Guwahati 781039, Assam

vkatiyar@iitg.ac.in

Keywords: Biodegradable polymers, biofiller, cellulose nanocrystal, packaging, silk nanocrystal

Abstract

The utilization of biomass for the development of various tailor-made sustainable polymers including poly (lactic acid) (PLA), poly (ε-caprolactone) (PCL), polyhydroxyalkanoates (PHA), and so forth targeting food packaging application is the main focus of the presentation. It is noteworthy to mention that both plant- and marine-based biomass can be transformed into value-added chemicals including nanocellulose (NC), silk nano crystals (SNCs), functionalized nanochitosan (FNC), nano-gums (NG), and so forth, which are components of both edible and no edible food packaging materials. Further, the application of proper strategies for fabricating the mentioned biomass-derived materials can help to deliver tuned properties in the concerned application. Thus, the formulation of biocomposites by incorporating the addressed biomass-derived materials (NC, SNCs, FNC, and NG) into the above-mentioned biodegradable polymers (PLA, PCL, and PHA) is a way to deliver tuned properties as food packaging materials. Further, the development of bio-composites of sustainable polymers has found to offer improved properties in terms of thermal, mechanical, and barrier properties, which are an important criterion for its uses in food packaging. In addition to above, the addressed chemicals are also considered a promising candidate in developing edible food packaging with enhanced thermo mechanical properties and health benefits. The application of herbal bioactive agents to biopolymers for acting as edible food packaging materials will further improve shelf life of perishable fruits, which allow consumers a value-added food product with reduced plastic-based waste. In this regard, chitosan has the properties of nontoxicity, biocompatibility, biodegradability, and antimicrobial activity. Further, chitosan provides many health beneficial properties including antioxidant property, antidiabetic property, weight-reducing activity, anticancer activity, cholesterol-lowering activity, and others, which make it a promising agent for edible films and coatings with added benefits. Moreover, residual biomass of green algae is another excellent source of various bioactive compounds such as protein, lipids, antioxidant, vitamins, minerals, and so forth. Thus, the utilization of algae for the extraction of nutraceuticals would be a great source of value addition of this class of biomass.

Further, it can be utilized for the production of bio-energy through microbial fuel cell technology, which will also be discussed during the talk. These sustainable materials can further be utilized as emerging candidates as a catalyst for various processes, additives for tailoring of polymers properties, efficient energy storage and power-generation medium, bioactive medium in biomedical applications, and so forth.

Biotechnological Intervention for Boosting *Citrus* Industry

Ashok K. Sharma

Tissue Culture Laboratory, CSIR National Botanical Research Institute, Lucknow, Uttar Pradesh, India

ashok_nbri@hotmail.com

Keywords: Citrus fruit, cloning, germplasm preservation, tissue culture

Abstract

Citrus may be considered as the number one fruit of the world and number three fruit of India after mango and banana in view of its nutritional and medicinal value as well as its production. Despite Northeastern India being the native place of *Citrus* and also having a huge orchard area, rich germplasm, and favorable agroclimate, the *Citrus* fruit production in India has been declining at an alarming rate during the past several decades mainly due to the destruction of orchards by diseases (particularly viral) and general neglect toward replenishing the declining orchards with certified healthy and high productive varieties of *Citrus*. The poor state of *Citrus* industry obtained in the country can greatly be alleviated or even changed to a situation in which India becomes a major *Citrus*-producing country in the world with the application of biotechnology, precisely plant tissue culture. At National Botanical Research Institute (NBRI), Lucknow, the first *Citrus* tree in the world through *in vitro* culture has been produced in 1972 and later remarkable success has also been achieved in various important aspects of *Citrus* tissue culture research, such as shoot meristem culture, unpollinated pistil culture, and micrografting for virus elimination; haploid production through androgeneis for genetic improvement; production of cloned plants of several commercially important scion species through nodal stem segments; and production of cloned as well as disease-free rootstocks by exploiting nucellar polyembryony. Under multilocational field trials, the shoot meristem-regenerated plants of *C. aurantifolia* exhibited better performance. Besides, efficient *in vitro* processes for germplasm preservation of several *Citrus* spp., including *C. indica* (an endemic threatened wild relative of *Citrus*), have been developed for establishing "Germplasm Repositories" of *Citrus* spp. growing in diverse agroclimates.

Integrating biotechnology and ecology for threatened plant conservation of Sikkim Himalaya

Mithilesh Singh, and Devendra Kumar

G.B. Pant National Institute of Himalayan Environment and Sustainable Development, Sikkim Regional Centre, Pangthang, Gangtok, 737101, Sikkim, India

singmithilesh@gmail.com

Keywords: Himalaya, MaxEnt modeling, propagation, threatened plants

Abstract

Sikkim Himalaya that forms a part of the "Himalayan Biodiversity Hotspot" is one of the biologically richest areas in the Eastern Himalayas. The region encompasses more than 4400 species of plants including those of medicinal value and otherwise economically important ones. However, rise in human population with demand on land for farming, construction of roadways, hydropower projects and allied works, and of late the tourist influx have collectively resulted in building up of considerable pressure on the survival of important plant species in Himalayan region. Declining in the number of species and significant changes in their natural habitat, as well as in some cases, complete population annihilation in the wild have become strong issues of concern, and a compelling reason to start work on the conservation of threatened plants of the region. In this study, very strong efforts have been made to improve conservation status of two threatened plans of Sikkim Himalayan Region, namely Rhododendron leptocarpum and Phoenix rupicola. Maximum entropy (MaxEnt) based distribution modelling algorithm was used to identify suitable habitats for plant reintroduction. Biotechnological interventions have been made for producing large number plants of R. leptocarpum and P. rupicola. For reintroduction, about 1000 plants of R. leptocarpum and 5000 plants of P. rupicola were produced through micropropagation and macropropagation techniques, respectively. Combined approach of ecological niche modeling and biotechnological techniques used in this study is ideal for the conservation of endangered plant species of Himalaya.

The Majesty of Plant Secondary Metabolites: Unlocking the Futuristic Trends of Bioresource Augmentation

Suchitra Banerjee

Plant Biotechnology Division, CSIR-CIMAP, Lucknow, Uttar Pradesh, India

suchitrabanerjee07@yahoo.com

Keywords: Bioresource augmentation, disease-pest infestations, MAPs, nutraceuticals, plant secondary metabolites

Abstract

Plants have been used for centuries to cure diseases throughout history owing to their ability to synthesize a fascinating class of phytochemicals, called "plant secondary metabolites (PSMs)." The vast array of chemically distinct PSMs of "Medicinal and Aromatic Plants' (MAPs)" origin represent diverse/novel molecular scaffolds, which are unachievable through synthetic chemistry leads. Their huge ranges of pharmacological activities have revolutionized the global history of blockbuster drug-development process by their direct use or as templates for semisynthetic derivatization. Recent advancements in the area of combinatorial chemistry and computational drug designing processes have accelerated their demand in drug discovery through identification of novel drug targets. Apart from pharmaceuticals, PSMs find multifaceted uses in nutraceuticals, fragrance / perfumery, cosmaceuticals, herbal remedies, and bio-insecticides. Additionally, current insights into the multifunctionality of plant volatiles in raising healthy livestock and aquaculture have further propelled their rapid commercial need. In light of the escalating global demand for PSMs, pharmaceutical companies are finding it difficult to comply with the year-round supply of biochemically consistent raw materials of MAPs because of their chronic limited supplies owing to multiple impediments. Indiscriminate wild harvesting without strategic cultivation practices, habitat loss due to rapid urbanization/industrialization, climate change, disease–pest infestations, and so forth not only radically jeopardized the sustainable use of MAPs but also imposed an endlessly mounting threat of extinction on a huge list of already endangered natural MAP resources. Creating the situation of assured affordability of commercially desirable MAPs' raw materials epitomizes the supreme challenge of this century. The concept of growing MAPs along with conventional crops for sustenance of their adequate resources is slowly changing the value of agriculture to the rapidly developing perception of entrepreneurial opportunities. Moreover, combining and refining the abilities of plant biotechnology with the major regulatory challenges of the pharmaceutical industry are also progressively drawing global attention by complying with the mandatory promise of more environmentally sound, economical, and effective PSM-delivery processes under controlled environment. Credible examples illustrating such strategies will be discussed to unlock the futuristic trends of PSMs' bioresource augmentation.

Role of Secondary Metabolites in Crop Protection

Ajeet Singh Tomar

International Marketing Development Department, Gharda Chemicals Ltd., Mumbai, Maharashtra, India

astomar@gharda.com

Keywords: Crop protection, microbial secondary metabolites

Abstract

Globally there is an increasing concern regarding the food production to sustain the exponentially increasing human population under a frequently changing environment (global warming). To meet the growing food demand, without affecting the human health and damaging the environment, compounds derived from microbes "secondary metabolites" can play an important role in crop protection as alternatives to chemical pesticides. Numerous bacterial and fungal secondary metabolites have been isolated and evaluated for fungicidal, insecticidal, and herbicidal activity. Of these, 33,500 microbial metabolites, about 12.5%, are metabolites of unicellular bacteria and cyanobacteria, 41% are products of Actinomycetes fermentations, and about 47% are of fungal origin. most effectively commercialized bacterial metabolites as bio-insecticides are endotoxins produced by *Bacillus thuringensis*, among soil microbes, actinomycetes produce wide-spectrum biologically active substances already commercialized as kusagamycin (bio-fungicide), abamectin (bio-insecticide), spinosad (bio-insecticide), and streptomycin (antibiotics). Thus, there is a huge potential for screening of new secondary metabolites that can be applied in crop protection, which would have least impact on food and environment and would also be benign to other non-target organisms.

Identification and Characterization of Known and Novel Cyclotides in the Indian Medicinal Plant *Viola odorata* and Its *In Vitro* Cultures

M. Narayani, Anju Chadha, and Smita Srivastava

Department of Biotechnology, Indian Institute of Technology Madras, Chennai, Tamil Nadu

smita@iitm.ac.in

Keywords: Cyclotides, cytotoxic, hemolytic, *in vitro* cultures, mass spectrometry, *Viola odorata*

Abstract

Cyclotides are a class of cyclic plant proteins with a unique topology that is responsible for their exceptional chemical, thermal, and enzymatic stability. This makes them a potential candidate for diverse commercial applications such as agrochemicals, pharmaceutical scaffolds for drug delivery, and therapeutic agents. Currently, cyclotides are obtained only via direct extraction from limited plants. In this study, known and novel cyclotides were identified for the first time in the Indian variety of the medicinal plant *Viola odorata* using liquid chromatography and Fourier transform mass spectrometry. Specific protocols were developed for successful identification and characterization of cyclotides in the plant that included confirmation based on their mass (2.5–4 kDa), hydrophobic nature, disulfide bonds, circular structure, and amino acid sequence. A total of 71 known and 98 putative new cyclotides were identified in the Indian varieties of *V. odorata*. Among the 98 putative new cyclotides, amino acid sequences of eight cyclotides have been established using *de novo* sequencing approach.

This study revealed that the production of cyclotides in plants varies with geographical location and the type of the plant tissue, hence cannot serve as a reliable source for the production of cyclotides. Moreover, owing to extensive wild crafting of *V. odorata* for several commercial applications, it is categorized as an endangered species in parts of India. Hence, to establish an alternative and sustainable production platform for cyclotides, cell (callus, cell suspension) and organ cultures (somatic embryo and shoot cultures) of *V. odorata* were developed to investigate the production of known and new cyclotides. Furthermore, the somatic embryos (rich in cyclotides) demonstrated equivalent and in some cases superior biological activities (cytotoxic, hemolytic, and antimicrobial) than the natural plant, suggesting it as an alternative source for several therapeutic applications.

Commercial Micropropagation—It's About Time!

Gayle R.L. Suttle

Microplant Nurseries, Inc., Gervais, Oregon, US

gayle@microplantnurseries.com

Keywords: Tissue culture, Industry impact

Abstract

The speaker will discuss her personal journey over the past four decades growing plants in culture for profit. She will highlight some of the technical challenges her company has overcome, discuss the impact micropropagation has had on our horticultural industry, and share her perspective of the opportunities ahead.

Advances in Agricultural Tools and Technology: A Transition from Traditional to Conventional Agriculture Practices in India

Pankaj Rawat

Bhanu Biotech Pvt. Limited, Bank Enclave, Laxmi Nagar, New Delhi 110092, India

bhanubiotech1@gmail.com

Keywords: Automation, farming, high tech green house, IoT

Abstract

Greenhouse technology and protected cultivation have proved to be an advantage for researchers/farmers by providing favorable climatic conditions in agricultural sector to harness improvements in marketable yields, significant reductions in the control of pests and weeds, increased density of crops, efficient utilization of water, and the accommodation of the use of different cultivation methods. This type of farming system has long been in the background and has not yet explored its maximum capability. However, recently observations have been made about the use of greenhouse technology in the agriculture sector. Plant growth modules (high tech green houses, tissue culture laboratories, cold rooms, and plant growth chambers) and allied scientific equipment cater the needs of Science community at a large in agriculture from automation to technology directed with the Internet of Things (IoT). The company design and implement environment monitoring system along with plant tissue culture laboratory establishment with various wireless sensor to control and monitor the plant propagation.

Our latest innovative technology includes front-end data acquisition, data processing, data transmission, and data reception and at the same time, researchers may view, analyze, and store data that provide real-time statistics for agricultural greenhouse/cold room/plant growth chamber/clean room facilities, and other weather control equipment, thus achieving real-time weather updates. The company provides latest drone-based agriculture solutions for pesticide spraying and crop monitoring. Additionally, observations made indicate that a lack of knowledge and experience in these new technologies are the most pressing challenges faced by Indian researchers/greenhouse producers and most traditional farmers. Thus, latest agricultural tools/farming techniques and their systems are applicable apparatus for developing and improving the agricultural produce of India.

Ex Situ Conservation of *Coelogyne ovalis* Lindl. Through Asymbiotic Seed Germination and Assessment of Genetic Variation for Reintroduction

Nutan Singh and Suman Kumaria

Department of Botany, Plant Biotechnology Laboratory, North-Eastern Hill University, Shillong-793022, Meghalaya, India

sumankhatrikumaria@gmail.com

Keywords: Coelogyne ovalis, conservation, genetic fidelity, reintroduction

Abstract

For conservation of an endangered, ornamental, and medicinally important orchid *Coelogyne ovalis* Lindl., an efficient protocol was developed via asymbiotic seed germination. Eight months old capsules of *C. ovalis* were inoculated aseptically on different media viz., Gamborg (B5), Knudson C (KC), and Mitra, with and without supplementation of plant growth regulators (PGRs) to check their effect on the growth of seedlings. Among the studied media, the best seed germination response was found in KC medium. The highest germination percentage was found in KC medium supplemented with 6-benzyl-aminopurine (BAP). For shoot induction, seedlings were further cultured in KC medium supplemented with PGRs viz., BAP and 1-naphthaleneacetic acid (NAA). The best shooting was observed in KC medium supplemented with 15 µM of BAP and 5 µM of NAA. Optimum rooting frequency of regenerated shoots was achieved in KC medium augmented with 15 µM NAA and 30 µM phloroglucinol. Well-developed plantlets were acclimatized in a compost mixture. Genetic variation of *in vitro* raised regenerated plantlets was ascertained using start codon targeted polymorphism (SCoT) and inter simple sequence repeat (ISSR) markers. The present report on *in vitro* generated *C. ovalis* insures rapid propagation of plantlets for conservation purposes and then their reintroduction in fields.

An Efficient Method of *In Vitro* Propagation and Hardening of Plum (*Prunus salicina*) Cultivar Santa Rosa

Vishal Sharma, Niharika, Manu Vivek, and Manisha Thakur

Department of Biotechnology, Dr Y. S. Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh, India

vishalshrm073@gmail.com

Keywords: Plum, *In vitro* propagation, hardening

Abstract

In the present investigation, a technique for *in vitro* propagation and hardening of plum (*Prunus salicina* L.) cultivar Santa Rosa has been developed. Treatment of explants with 1% sodium hypochlorite for 10 min was found to be the best as it gave the maximum number of uncontaminated buds and buds' survival. Maximum *in vitro* establishment (75%) of explants was achieved on Murashige and Skoog (MS) medium fortified with 0.5 mg/L 6-benzylaminopurine (BAP) and 0.05 mg/L indole-3-butyric acid (IBA), in the month of February and March. The highest multiplication rate (1:8) was obtained on MS medium fortified with 0.5 mg/L BAP, 0.1 mg/L gibberellic acid (GA_3), and 0.1 mg/L IBA. Shoot multiplication rate and shoot length showed an increase with the increase in passages, which increased to a maximal of 1:10 and 4 cm, respectively, and showed a decline in further passages after the fourth passage.

Two procedures for rooting, that is, single step and two steps were adopted, and the maximum rooting (70%) was observed on ½-strength MS medium supplemented with 0.5 mg/L IBA following the single-step approach. In two-step procedure, 30% rooting was observed after 48 h dark incubation in ½-strength liquid MS medium fortified with 0.5 mg/L IBA followed by transfer to semisolid ½-strength MS basal medium within 4 weeks of culture. *In vitro* rooting efficiency increased with the increase in passages, which increased to 70% during the ninth passage. Best hardening was observed by following hydroponic approach in which the plantlets were dipped in liquid MS medium without sucrose and myo-inositol for 15 days with 66.6% survival after 4 weeks of transfer to cocopeat. Drenching of potting mixture with 15 mL Jeevamrit (3%) showed the survival of 60.33%. *In vitro* regenerated plantlets showed no morph-ological variations when compared with mother trees.

In Vitro Propagation of Plum (*Prunus salicina* L.) Cultivar Frontier Through Control of Shoot Tip Necrosis

Manisha Thakur and Vishal Sharma

Department of Biotechnology, Dr Y. S. Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh

drmanisha72@yahoo.com

Keywords: Plum, shoot tip necrosis, tissue culture

Abstract

The present investigation was carried out to develop an efficient protocol for *in vitro* propagation of plum (*Prunus salicina* L.) cultivar (cv.) Frontier and to control shoot tip necrosis (STN). High yielding trees of cv. Frontier, indexed against *Prunus* necrotic ring spot virus, cherry leaf roll virus, and apple chlorotic leaf spot virus, were selected for the study. Shoot apices and axillary buds were collected from mother plants and cultured on Murashige and Skoog (MS) medium containing different concentration and combination of phytohormones. MS medium supplemented with 0.1 mg/L 6-benzylaminopurine (BAP), 0.1 mg/L kinetin, and 0.05 mg/L indole-3-butyric acid (IBA) was found to be the best and gave highest percentage (79.50%) of shoot bud establishment after 4 weeks of culture; however, all shoots showed necrosis and died after the first subculture. Necrosis appeared with yellowing of shoot tips within 7 days of the first subculture and gradually increased downward, resulting in yellowing of leaves followed by death of shoots.

Among various reported methods tested to control STN, combination of fructose (0.55 mM) and calcium chloride (1.0 mM) proved 100% effective in STN control. Highest *in vitro* shoot multiplication of 1:5 was achieved on MS medium supplemented with 0.5 mg/L BAP, 0.1 mg/L gibberellic acid (GA_3), 0.05 mg/L IBA, 0.55 mM fructose, and 1.0 mM calcium chloride. For rooting, a two-step rooting procedure was followed, where microshoots were dipped in half-strength MS broth containing 0.5 mg/L IBA and incubated in dark for 48 h before transferring to the same strength basal MS medium, resulting in 85.00% rooting. Rooted plantlets were successfully hardened in cocopeat with 70–80% survival after 10 weeks and after 1 year, these plants were transferred to the field with 100% success. Regenerated plants showed no morphological variation when compared with mother trees.

Formulation of Agro-Waste-Based Bacterial Biofertilizer and Its Plant-Growth-Promoting Effects

Gargi Chakravarty
Department of Botany, D. K. College, Mirza, Assam, India

gargichakravarty2008@gmail.com
Keywords: Agro waste, *Bacillus subtilis*, biofertilizer, plant growth-promotion, *Pseudomons fluorescens*

Abstract

The main purpose of this research is to find a possible way for efficient utilization of agricultural wastes for sustainable plant production systems. Agro wastes were used as substrates for the growth of plant growth-promoting bacteria *Pseudomonas fluorescens* and *Bacillus subtilis* and production of biofertilizers. Altogether nine substrate combinations were studied, namely fruit wastes, vegetable wastes, fallen leaves, and rice straw each one inoculated with *Pseudomonas fluorescens* (C1–C4), again each of the four organic wastes inoculated with *Bacillus subtilis* (C5–C8), and the final substrate C9 containing a mixture of the four agro wastes inoculated with both strains of *P. fluorescens* and *B. subtilis*. Control (C10) consisted of a mixture of the four agro wastes inoculated with any of the two bacterial strains. Enzyme activities and plant growth-promoting traits of the selected bacterial strains were tested. Temperature and pH during the composting period were measured at regular intervals up to 28 days, and chemical analysis including organic carbon, nitrogen, phosphorus and potassium concentrations of the various inoculated substrates were carried out.

The viable bacterial population of the decomposed substrates was evaluated at 15 days interval up to 90 days. The three best bioformulations depending on bacterial load were selected for study of their effects on shoot length, leaf surface area, total leaves and branches emerged in the plant, and fruit yield in the test plant *Solanum melongena*. The carbohydrate, protein, and chlorophyll content of the treated and control plants were also recorded. The formulation with a mixture of the four agro wastes and both bacterial strains (C9) showed the best result to be used as a potential and effective biofertilizer, meeting the need of environment-friendly food production systems and providing balanced nutrient supply and waste recycling.

Artificial Seed Technology for Short/Medium-Term Germplasm Storage in *Aquilaria malaccensis* Lam.; a Commercially Important Tree

Seram Devika Devi, Meera C. Das, and Suman Kumaria

Plant Biotechnology Laboratory, Centre for Advance Studies in Botany,
North-Eastern Hill University, Shillong 793022, Meghalaya, India

seram.devika@yahoo.com

Keywords: *Aquilaria malaccensis*, artificial seed, recalcitrant, regeneration, response, storage

Abstract

Aquilaria malaccensis is a tropical tree of high commercial value due to the production of a non-timber forest product called "agarwood". Storage is highly problematic in the species due to the recalcitrant nature of seeds. Artificial seed technology is known to be the best alternative for short/medium-term storage of germplasm in such plant species that possesses seeds with very short shelf life. The study therefore explored the effect of two temperatures, that is, 4°C and 23± 2°C on storage potential of *A. malaccensis* zygotic embryos and *in vitro* derived nodal buds. Maximum responses were observed in liquid full-strength Murashige and Skoog's medium from both the explants, while regeneration was observed only from *in vitro* nodal buds in the same medium. Encapsulated zygotic embryos could be stored for only 20 days at both the studied temperatures; in case of nodal buds as explants, storage was possible for 60 and 50 days at 4°C and 23±2°C respectively. Meanwhile, encapsulated zygotic embryos failed to regenerate into shoots in addition to its inefficiency for storage while all survived encapsulated nodal buds regenerated into shoots. Shoot development from the encapsulated nodal buds stored at 4°C was found to be better than that of 23 ± 2°C.

A Comparative Study of Selective Macrophytes for *In Situ* Bioconcentration From Sediment–Water Continuum as Self-Remediative Lacustrine Function

Umar Nazir, A. Salim Khan, and Anisa B. Khan

Department of Ecology and Environmental Sciences, Pondicherry University, Puducherry 605014, India

salimamu@gmail.com

Keywords: Autotrophy, biofiltration, nitrate, phytofiltration, remediation, senescence

Abstract

Macrophytes perform optimization and self-purification functions; otherwise, the evident water quality could get worse. They behave as a living link between abiotic water–sediment interfaces as depicted by the seasonal nutrient regeneration fluctuations. Tissue concentrations of nutrients and trace elements differ species wise but well correlate with ambient water and sediment media. The peak biomass values (gm^{-2} dry weight basis) measured are 880.2 in case of *Myriophyllum aquaticum*, 678.4 in *Nelumbo nucifera*, 182.4 in *Ceratophyllum demersum*, and 45 in *Salvinia natans*. Biomass parameters such as dry weight, productivity, NPP and specific growth rate establish similar variations in the experimental species but species turnover is highest in case of *S. natans* and lowest for *C. demersum*. N and P are intimately related to biological productivity of aquatic ecosystems. Higher temperature favor bioproduction and elevate carbonate-driven pH significantly. The diminishing conductivity at improved temperature conditions suggest higher bioaccumulation rate of nutrients. The peak nutrient uptake and bioconcentration coincide with peak biomass in summer and autumn. BCF criterion indicates hyperaccumulation for most of the metals in case of *C. demersum* and *S. natans*. Although the quantum of removal potential for different elements in the analyzed species is divergent but the pattern is related, which suggests unselective absorption. The elemental turnover rates in selected macrophytes closer to the reference value of 1 has significance. Although the emergent macrophyte proved efficient in mineral and metal retention, but, submerged ones provide a better biofilters' option in terms of spontaneous occurrence and site aesthetics. Based on the outcome of the study, lake ecological restoration is possible by limiting human perturbations, practising periodic dredging and sediment trapping, scaled-cum-selective deweeding, and construction of vegetation buffer strips for decelerating cultural eutrophication. Therefore, internal structure, external inputs, compartment cycling, and resource exploitation together quantify changes in inland waters.

Recovery of Lignin from Agro-Waste

M. Arjun, A. K. Harigovind, R. Ananyanjali, Maryl Ann, N. Gazliya, and K. Aparna

Department of Chemical Engineering, National Institute of Technology, Calicut, India

gazliya.pn@gmail.com

Keywords: Agro waste, alkaline treatment, banana stalk, lignin, lignocellulose

Abstract

India contributes to 29.19% of banana production, being the world's leading producer. Banana occupies 20% of the total area under crop production in India, which covers almost the entire country. Lignocellulose biomass is gaining increased attention currently; because it is renewable and sustainable in the form of agro-waste. The agro-waste that is produced after harvest is plant biomass consisting of energy-rich lignocelluloses that can be efficiently used to make value-added products such as lignin, which in turn, can be used for the production of chemicals, fuel, and electricity. Banana stalk is an unused part of the plant, which is discarded in local markets and packing centres, where residues are dumped into the open and water resources.

The stalk consists of ~16% lignin under dry biomass weight. The present work is undertaken to recover lignin from black liquor, produced by the alkaline treatment of banana stalk. Under peroxide-assisted, mild-alkaline, and ambient-temperature conditions, 6.6% lignin yield was obtained from 25 g of dry biomass, which accounts for 41.25% of the total lignin content. The lignin is recovered by acid precipitation and characterized by Fourier Transformed Infrared spectroscopy (FTIR), Differential scanning calorimetry (DSC), Thermo-gravimetric Analysis (TGA), X-ray diffraction (XRD), and molecular-weight analysis

Standardization of an *In Vitro* Regeneration Protocol in Khasi Mandarin

Panchashree Das

Department of Agricultural Biotechnology, Assam Agricultural University, Jorhat, India

panchashreedas@gmail.com

Keywords: Regeneration, root induction, shoot induction, zygotic seedlings

Abstract

Citrus species are the most widely grown fruit crops within the whole world. India is the fourth largest producer of orange in the world. North-Eastern India is considered as one of the centres of origin of many citrus species. Among them Khasi Mandarin is the most widely grown citrus species. According to Ministry of Agriculture and Irrigation, Govt. of India, the yield of Khasi Mandarin is declining day by day drastically due to biotic and abiotic stresses. Conventional breeding for overcoming this problem is restrictive due to non-availability of resistant sources. Recent advances in plant tissue culture have made it possible to develop abiotic and biotic stress-resistant cultivars. For developing such cultivars a suitable *in vitro* regeneration protocol is prerequisite. Citrus cultivars vary in their response to *in vitro* organogenesis. This results in the need for cultivar-specific optimization of *in vitro* regeneration protocol. The present study was conducted in 2015–2017 at Department of Biotechnology, Assam Agricultural University, Jorhat. In the present investigation, *in vitro* regeneration of Khasi Mandarin was optimized using zygotic seedlings as explants. Modified Murashige and Skoog (MS) medium containing 1 mg/L 6-benzylaminopurine (BAP), 0.5 mg/L 1-naphthaleneacetic acid (NAA), and 0.4 mg/L kinetin shows the best result for multiple shoot induction with an efficiency of 68%. The average number of multiple shoots developed was 5. The modified MS medium containing 0.25 mg/L BAP, 0.5 mg/L NAA, 0.5 mg/L indole-3-butyric acid shows the best result for rooting of explants with an efficiency of 82% and average root length of 4 cm. These results suggest that standardization of these factors can help in development of a commercially viable tissue culture system for Khasi Mandarin.

Bio-Oil Generation and Characterization from a Woody Biomass as Sustainable and Renewable Energy Source

Harshal Kawale and Nanda Kishore

Department of Chemical Engineering, Indian Institute of Technology Guwahati, Guwahati, India

nkishore@iitg.ac.in

Keywords: Biomass, bio-oil, gas chromatography–mass spectroscopy, ^{1}H-NMR, pyrolysis

Abstract

The petroleum depletion over a period of time and the need of value-added petrochemicals drives a force to develop fruitful technologies. The recent trends in renewable energy production are yet to qualify the energy needs. Thermal processes for biofuels from biomass results biofuels in the form of solid, liquid and gas, which can be utilized as fuel and value-added products. Among these technologies, pyrolysis and catalytic pyrolysis are cost effective technologies on pilot scale as well as these can be delivered to form a biorefinery. We have conducted pyrolysis experiments at variable-temperature ranges to get biofuels from a lignocellulosic biomass. The source of biomass is available at tremendous amount around the far region of land but scarcely researched. Main product obtained was pyrolytic bio-oil, which was characterized using Fourier transform infrared spectroscopy (FTIR), and the analysis showed the number of functional groups present in it. Gas chromatography – mass spectroscopy (GC-MS) results in large number of components present in the bio-oil. ^{1}H-NMR spectroscopy analyzed to show various functional groups present in the sample as well as the amount of aromatic carbon content in the bio-oil. Thus, these characterization techniques revealed the fuel potential of the biomass to be used as an energy crop for the production of energy in a sustainable manner.

Metal Uptake Potentiality of *Salvinia cucullata*, Roxb. to Paper Mill Effluent – A Phytoremediation Approach

Kisholay Mazumdar and Suchismita Das

Department of Life Science and Bioinformatics, Assam University, Silchar, Assam, India

drsuchismita9@gmail.com

Keywords: Bioconcentration, hyperaccumulation, paper mill effluent, phytoremediation, translocation

Abstract

In the human development, paper mill industry plays a significant role but simultaneously it causes health hazard by emitting various heavy elemental constitution in the form of effluent. A novel fern *Salvinia cucullata* has the potentiality to resolve the problem by hyperaccumulating trace elements from contaminated sites. To acknowledge the phytoremediation or metal uptake potentiality of the plant, experiments were begun by treating *S. cucullata* with different concentrations of paper mill effluent (25%, 50%, 75%, and 100% v/v) for 28 days. It revealed that the plant has the ability to accumulate 10 different heavy metals such as Cd, Cu, Cr, Ni, Pb, Mg, Mn, P, Fe, and Zn at different effluent concentration treatments. However, the plant scored both translocation factor and bioconcentration factor values >1 at all the four effluent concentration treatments, which reflects metal hyper-accumulation potentiality of the plant. *S. cucullata* however undoubtedly a suitable plant for phytoremediation of paper mill effluent.

Phytoremediation of Heavy-Metal-Contaminated Soil using *Pongamia pinnata*; a Biofuel Plant

Pallabi Borah,[1,2] Tanmoy Karak,[3] Latha Rangan,[4] and Sudip Mitra[2*]

[1]Department of Environmental Science, Tezpur University, Assam, India
[2]Centre for Rural Technology, Indian Institute of Technology, North Guwahati, Assam, India
[3]Upper Assam Advisory Centre, Tea Research Association, Dikom, Dibrugarh, Assam, India
[4]Department of Biosciences and Bioengineering, Indian Institute of Technology, North Guwahati, Assam, India

sudipmitra@iitg.ac.in/sudipmitra@yahoo.com

Keywords: Bioaccumulation factor, heavy metals, phytoremediation, *Pongamia pinnata*, translocation factor

Abstract

The aim of the study is to assess the phytoremediation potential of *Pongamia pinnata*, a biofuel plant. Both, pot and field experiments were conducted in soil contaminated with paper mill wastes and municipal wastes. The pot experiment was conducted with different combinations of contaminated and control (forest) soil @ 0%, 25%, 50%, 75%, and 100%. The pot study showed its effectiveness in removing heavy metals (HMs) (Cd, Cr, Cu, Fe, Mn, Ni, and Zn) from the contaminated soil. The plants grown in 25% contaminated soil exhibited highest uptake capacity of the studied HMs. High average concentration of HMs were observed in roots except for Cd and Mn, whose concentration were highest in stems. Cu, Cr, and Ni were effectively removed by *P. pinnata*, with more than 50% removal efficiency. The average bioaccumulation factor values were found to be <1 for all the selected HMs, thus belonging to the excluder category. The translocation factor values exceeded 1 for Cd, Cr, and Mn in paper mill soil combination but in municipal wastes soil it exceeded 1 only for Cd and Mn. In the field experiment, *P. pinnata* showed accumulation of Zn and exclusion of other heavy metals (Cd, Cr, Cu, Fe, Mn, and Ni). Thus, the present study suggests that combination of soil @ 25% polluted soil with 75% control increases the uptake potential of *P. pinnata*.

In Vitro Studies of Histological and Biochemical Induction of Rooting of *Bacopa monnieri* (L.) Wettst.

Aanchal Goel[1] and Anil Kumar[2]

[1]University Institute of Biotechnology, Chandigarh University, Gharuan, India
[2]Department of Biotechnology, TIFAC CORE, Thapar University, Patiala, India

anchalgoel.biotech@cumail.in

Keywords: Alzheimer's disease, meristemoids**,** polyphenol oxidase, primordia, rhizogenesis

Abstract

Bacopa monnieri (L.) or Brahmi has its importance in ayurvedic medicine for treating nervous system disorders such as Alzheimer's disease and anxiety and psychiatric disorders such as improving memory. The present study was carried to investigate histological and biochemical changes during adventitious rooting of microcuttings of *Bacopa monnieri* (L.) Wettst. Histological studies were conducted to identify different phases of rooting in these microcuttings. The root meristemoids with distinct polarity become visible after 3 days and mark the beginning of *in vitro* root initiation phase. Biochemical studies were also conducted from basal portions of microcuttings. Total carbohydrate content was lower during initial periods (up to day 1) and was found to increase during root initiation and primordia development, which reflects high energy demands for active cell divisions. A significantly higher level of phenols was observed in microcuttings on medium supplemented with indole butyric acid (IBA). Polyphenol oxidase, peroxidase (POX), and ascorbate peroxidase activities were also found to vary during different phases of rhizogenesis. Early phases were also marked with the lower activities of POX and indole acetic acid oxidase (IAAO). This study reveals significant roles of enzymes, sugars, and phenols during different phases of rooting.

Dedifferentiated *In Vitro* Cell Lines: A Bioresource Utilization Method for Enhanced Spilanthol Production by Optimizing Media Constituents using Response Surface Methodology from *Spilanthes paniculata*

Radhika Rajendran[1] and Rakhi Chaturvedi[1]

[1]Department of Biosciences and Bioengineering, Indian Institute of Technology Guwahati,Guwahati-781039, Assam, India

rakhi_chaturvedi@iitg.ac.in

Keywords: In vitro dedifferentiated cultures, response surface methodology, *S. paniculata*, spilanthol

Abstract

Spilanthes paniculata is a medicinal herb with rich source of therapeutic constituents. *In vitro* callus cultures were established from leaf-disc explants of *S. paniculata*, which is an alternative method of cell biomass utilization and conservation of natural plant resources. Additionally, response surface methodology (RSM) was performed to maximize the production of *N*-alkylamide, Deca-2*E*, 6Z, 8E-trienoic acid isobutylamide (spilanthol) from *in vitro* cell cultures. In the first step of optimization, with Plackett-Burman design (PB), Murashige and Skoog (MS) major salts, sucrose, 2,4-dichlorophenoxyacetic acid (2,4-D), N6-benzylaminopurine (BAP) were found to be the important factors affecting spilanthol production significantly. In the second step, a 2^4 full factorial central composite design was applied to determine the optimal concentration of each significant variable. A second-order polynomial was determined by the multiple regression analysis of the experimental data. Optimum values for the critical components were obtained as MS (1.5), sucrose (5%), 2,4 –D (1.8 µM) and BAP (4.82 µM), with a predicted value of maximum spilanthol production of 3.72 mg/g dry weight (DW). Under the optimal conditions, the experimental value of spilanthol production was 2.81 mg/g DW. The coefficient of determination (R^2) was 0.9922, which ensures adequate credibility of the model. Furthermore, the higher production of spilanthol was achieved by statistical model as compared with that of non-optimized media constituents. Before optimization, callus cultures and leaves from parental plant (control) yielded 1.75 and 0.26 mg/g DW spilanthol, respectively. Thus, RSM is an effective tool for optimizing the media combinations, which uses quantitative data from an appropriate experimental design to simultaneously solve and determine multivariate equations on maximizing the production of therapeutic compounds. Current work is unique information on statistical optimization and production of spilanthol from leaf-disc callus cultures of *S. paniculata*.

A Comparative Biochemical and Molecular Approach to Understand the Factor Determining Salt Tolerance in Rice Cultivars

Amber Gupta[1] and Birendra Prasad Shaw[1]

Plant and Microbial Biotechnology Unit, Institute of Life Sciences,
Nalco Square, Bhubaneswar, 751023, Odisha, India

bpshaw@ils.res.in

Keywords: Active transport, antiporters, energy gradient, H^++ ATPase, NHX, salt tolerance

Abstract

Among the abiotic stresses high soil salinity, contributed largely by Na^+ and often compounded with drought, is the main factor that adversely limits growth and productivity of major crop plants, including rice. Salt (NaCl) affects a plant in two ways. First, it lowers the water potential of the environment, resulting in reduction in turgor pressure. Second, its accumulation in cytoplasm has toxic effect on cellular processes, leading to inhibition of growth and development. As a counter measure, plant adapt a number of strategies. Low concentration of Na^+ ions in the cytoplasm is apparently achieved by regulation of Na^+/K^+ selectivity of antiporters/channels across the membranes. The functioning of secondary transporters is activated by proton pumps, which are the primary active transport systems in membranes. The free energy gradient ($\Delta\mu H^+$) produced by the plasma membrane and vacuolar H^+ ATPase is presumed to provide the driving force in regulating the functioning of secondary transporters (housed in respective membranes) for the maintenance of ionic balance. Majorly two secondary transporters SOS1 (plasma-membrane-bound Na^+/H^+) and NHX (vacuole-membrane-bound Na^+/H^+) have also been reported to play important role in salt tolerance in plants. However contribution of biochemical processes in salt tolerance is not well defined hence the current study was conducted to understand their functioning in rice cultivars; tolerant and sensitive one to salinity. The present study was planned to go for a comparative study and to find out few of the most potential and universal salinity-stress-responsive gene in two salt-tolerant (Nona Bokara and Pokkali) versus two salt-sensitive (IR29 and IR64) rice varieties, so as to get a holistic picture of their importance and involvement in salt tolerance.

Phytochemical Diversity and Micropropagation of *Paris Polyphylla* Sm. Rhizomes from Northeast India

Khumuckcham Sangeeta Devi,[1] Pramod Tandon,[2] Suman Kumaria,[2] Ashiho Asosii Mao,[3] Nripemo Odyuo,[1] Deepu Vijayan,[1] Ashutosh Pathak[1]

[1]Botanical Survey of India, Eastern Regional Centre, Shillong, Meghalaya, India
[2]North-Eastern Hill University, Mawlai, Shillong, Meghalaya, India
[3]Botanical Survey of India Headquarter, Kolkata, West Bengal, India

khsangbotany@gmail.com

Keywords: HPLC, khonoma, micropropagation. *Paris polyphylla*, saponins

Abstract

Identification of medicinal plant for elite genotypes requires the assessment of phytochemical diversity across different populations. Such study is fundamental for further scale of plant resources as well as subsequent drug development for the treatment of human ailments. The present study was taken up to assess the total steroidal saponins diversity in *Paris polyphylla* across the northeastern region of India. Nine populations from four northeastern states of India were assessed in the present study. Quantification of the steroidal saponins in the selected population was carried out by comparing against standard saponins using high-performance liquid chromatography (HPLC). From the study, it was found that *P. polyphylla* populations from Khonoma showed the highest total saponins content, recording an average of 32.06 mg/g dry weight in comparison with all other populations under study Micropropagation of Khonoma populations was carried out for large-scale propagation of this elite zonal chemotype. Efficiency of two cytokinins with different sucrose concentrations on minirhizome induction was studied, and it was found that 6-benzylaminopurine 0.5 mg/L + 6% sucrose and 2-isopentyladenine (2iP) 1.0 mg/L + 6% sucrose resulted in the best response giving 88.6% and 89.2% with 1.27 ± 0.02 g fresh weight and 1.36 ± 0.10 g fresh weight of minirhizome, respectively.

DNA Barcoding of Commercially Important *Vanda* Species (Orchidaceae) of Nagaland

Joyrison Kamba and Chitta Ranjan Deb

Department of Botany, Nagaland University, Lumami, Nagaland, India

debchitta@gmail.com

Keywords: DNA barcoding, molecular markers, orchids, recognition tag

Abstract

Orchids are known for their ornamental and medicinal importance, besides their beautiful looks and attractive color that has led to indiscriminate collection and smuggle extensively in the wild, leading to rare, endangered, and threatened many important species without proper documentation. Moreover, they can be crossed within the same genus or with different genera, leading to the production of various types of new hybrids, having similar morphological traits in nature. So, in the present work, we established barcodes for *Vanda* species, which are used for rapid identification at any stage of life and detection of species that relies on short DNA sequence variation and provides a unique recognition tag to a species. For these, six *Vanda* species, that is, *Vanda coerulea*, *V. bicolor*, *V. stangeana*, *V. ampullacea*, *V. testacea*, and *V. alpina* were tested with three molecular marker system (*matK*, *rbcL*, and ITS) from cytoplasmic and nuclear regions. The species identity generated from BLAST results obtained by these markers system correctly matched morphological traits, which indicate that DNA barcoding can be used for the rapid identification and detection of any unknown biological sample at the molecular level for species and for variety certification for protection and conservation purposes.

In Vitro Induction of Cormlet in *Crocus sativus* and its Future in Genetic Transformation

Mona Hamal Thakuri and N. K. Chrungoo

Centre for advance studies in Botany, North Eastern Hill University,
Shillong 793022, Meghalaya, India

hamalthakurimona@gmail.com

Keywords: Agrobacterium, cormlet, genetic transformation, saffron, sterile triploid

Abstract

Crocus sativus (saffron) is a triploid sterile plant belonging to family Iridaceae. Its dry stigma is considered to be the world's most expensive spice, having diverse medicinal properties. The plant has an active growth period in the autumn and winter along with flower and leaf formation. Corms multiply very slowly producing only four to five daughter corms per mother corm. They survive for only one season, forming daughter corms and perish. Genetic improvement in saffron is not possible due to its triploid nature and male sterility. Crop improvement is confined to the evaluation and selection of naturally occurring clonal variants. To overcome these challenges, application of tissue culture methods offers great potential for mass multiplication and genetic improvement. In the present study, we have induced microcorms from different somatic tissues that were grown on Murashige and Skoog (MS) half-strength medium supplemented with 0.5 mg/L 6-benzyl amino purine (BAP) + 0.1 mg/L α-naphthalene acetic acid (NAA). Formations of cormlet-like structure from these embryos was observed in MS medium supplemented with 2 mg/L thidiazuron (TDZ) and 1 mg/L indole acetic acid (IAA). Germination of cormlets could be achieved on MS medium containing 1 mg/L BAP and 0.5 mg/L NAA. Alternately, transformants containing key regulatory genes involved in enhancing the phytochemical property of saffron or genes involved in inducing increased number of cormlets can be carried out. The need of the hour is to develop a transfer mechanism for the genetic transformation, for which calli and embryos were cocultivated with *Agrobacterium* and direct *in planta* cocultivation of mother corms were carried out using the native expression vector. Screening of the transformant was done using green fluorescent protein and β-glucuronidase (GUS) analysis. The development of a method for gene transfer will help in designing an effective method for the development of disease-resistant and high-yielding saffron plants.

Tissue-Culture-Mediated Propagation of Some Medicinal Plants for Conservation and Sustainable Utilization

Soumendra K. Naik
Department of Botany, Ravenshaw University, Cuttack, India

sknuu@yahoo.com

Keywords: ISSR, medicinal plants, micropropagation, RAPD, synthetic seeds

Abstract

Plants in general and multipurpose medicinal plants in particular are under severe stress in their wild habitat and depleting rapidly due to various anthropogenic activities. A number of such plants have limitations in propagation through conventional means. Thus, in our laboratory, we have developed various tissue-culture-mediated plant propagation systems for few such medicinal plants species, namely *Paederia foetida*, *Bacopa monnieri*, *Withania somnifera*, and *Operculina turpethum*. Protocols for the production of synthetic seeds using axenic nodal segments by encapsulation method have also been developed for plant propagation and germplasm exchange of *P. foetida* and *B. monnieri*. In some of these protocols, clonal fidelity of micropropagated plants with that of the mother plant was confirmed using various molecular markers including Random amplified polymorphic DNA (RAPD) and Inter simple sequence repeat (ISSR). Plant propagation methods of these plant species have the potential to provide constant supply of planting materials independent of season, thus useful for their reintroduction in wild and/or providing raw materials with the uniform quality for manufacturing therapeutics and other uses in a sustainable manner without disturbing wild populations.

Iron Biofortification In Bananas by Expression of *Oryza sativa* Nicotianamine Synthase Genes

M. Mayil Vaganan, C. Sivagandhi, S. Ganesan, M. Kumaravel, I. Ravi, K. J. Jeyabaskaran, S. Backiyarani and S. Uma

ICAR National Research Centre for Banana, Thayanur Post, Thogamalai Road Tiruchirappalli 620102, Tamil Nadu, India

mmayilv@gmail.com

Keywords: Grand Naine, iron deficiency, *OsNAS1*, *OsNAS2*, Rasthali, transgenics

Abstract

Iron is an essential micronutrient for plant growth and human health. Iron deficiency is one of the most prevalent micronutrient deficiencies in the world, causing anemia to several billion people especially in developing countries. The production of iron-biofortified staple crops will help to ameliorate iron-deficiency anemia. Banana is one of the staple crops of Asian and African population. Two commercial banana genotypes, Rasthali and Grand Naine, are biofortified by introduction of two rice (*Oryza sativa*) nicotianamine synthase genes, *OsNAS1* and *OsNAS2*, driven by *Zea mays* ubiquitin promoter to increase the iron content in fruit pulp. *Agrobacterium*-mediated transformation of the constructs carrying these genes and selectable marker, *nptII*, was carried out with embryogenic cell suspension (ECS) of the bananas. One hundred independent transgenic events of each banana genotype for each gene construct were generated and being field grown along with untransformed control plants in iron-sufficient soils under confinement in transgenic net houses equipped with all biosafety standards as per guidelines of Department of Biotechnology, Government of India.

The transformants, before planting, were confirmed for transgene presence and selectable marker with specific primers using polymerase chain reaction (PCR) and checked for *Agrobacterium* contamination using *Vir-C* gene-specific primers to eliminate false positives. Presently, transgenic lines are 9 months old and in shooting and fruit-bearing stages. Estimation by Inductively coupled plasma atomic emission spectroscopy (ICP-OES) of iron element content of 25 representative lines of each genotype at vegetative stage is promising with seven Rasthali lines and six Grand Naine lines having average iron content of 24.5 mg/100 g dry wt. as compared with nontransgenic lines with maximum of 11.2 mg /100 g. As the increase in iron concentration was 2.2 times in the transgenic lines, the results offer promise to effectively increase the level of iron in the fruit. Transgene integration and the copy number of the transformed events are being performed.

Long-Term Seed Storage and Acclimatization of *In Vitro* Derived Plantlets of *Paphiopedilum villosum* – A Threatened Commercial Slipper Orchid

Reema Vareen Diengdoh, Meera Chettri Das, and Suman Kumaria

Plant Biotechnology Laboratory, Department of Botany, Centre for Advanced Studies, North-Eastern Hill University, Shillong 793022, Meghalaya, India

rimavareen@gmail.com

Keywords: Acclimatization, *Ex situ*, orchid seeds, SEM, storage, sustainable utilization

Abstract

In today's times, orchids are appraised for their beauty and discussed for their scanty population. Heterogeneous orchid seeds exhibit, by nature, features like minute seed size and ample availability per capsule. Cryopreservation is an efficient *ex situ* strategy, but orchid seeds conservation is hampered by poor storage conditions and *in vitro* regeneration with effective acclimatization protocols need to be standardized. The advantages of seed storage are indefinite storage period, genetic stability, reduced infrastructure, and the stored genetic material does not require manipulation. Stored seeds of *Paphiopedilum villosum* showed the best result on storage at –196°C with no significant variation in *in vitro* germination (81.5%) as well as viability analysis (80.1%) up to 360 days. Mature seeds stored at –196°C were appropriate for long-term storage, having germination of 81.3 ± 1.5% with similar viability percentage 80.1 ± 1.9%, over 360 days storage supported by scanning electron microscopic (SEM) studies. Plantlet growth *in vitro* was best recorded in Murashige and Skoog medium containing 20 µM 6-benzylaminopurine + 5 µM indole-3-acetic acid with high cumulative response of 84.5 ± 3.1%. While for acclimatization in two experimental locations, the combination of compost mixture comprising charcoal + brick stone + soil + layer of moss litter in the ratio 1:1:1:1 was found with good survival of 59.1 ± 3.1% with average shoot length of 11.8 ± 0.8 cm. *Ex situ* conservation offers safer security backup system for the conservation, allowing accessibility for research evaluation, commercial propagation, and ultimately lead to sustainable utilization for the future.

Efficient *In Vitro* Propagation of *Hedychium coronarium* J. Koenig using Rhizome Segment and Assessment of the Genetic and Biochemical Fidelity of Micropropagated Plants

Shashikanta Behera,[1] Pradeep K. Kamila,[2] Kedar K. Rout,[3] Durga P. Barik,[1] Pratap C. Panda,[2] and Soumendra K. Naik[1]

[1]Department of Botany, Ravenshaw University, Cuttack, Odisha, India
[2]Plant Taxonomy and Conservation Division, Regional Plant Resource Centre, Bhubaneswar, Odisha, India
[3]Siddhi Binayak Science College, Angul, Odisha,India

sknuu@yahoo.com

Keywords: Hedychium coronarium, HPTLC, ISSR, medicinal plant, micropropagation

Abstract

Hedychium coronarium J. Koenig (Zingiberaceae) is an important medicinal plant with ornamental values. The rhizome and flower of the plant also possess volatile compounds and thus useful in perfume industries. *H. coronarium* has already been enlisted as a medicinal plant of conservation concern in various states including Madhya Pradesh, Odisha, Kerala, and Karnataka due to unsustainable harvesting to meet the demand. Tissue-culture-mediated plant regeneration has the potential to produce large number of plants, overcoming limitations of conventional propagation methods, thus fulfilling the demand of the plant and plant materials in a sustainable manner. Therefore, the present study was undertaken to develop an efficient protocol using rhizome segments for clonal propagation of *H. coronarium*. Best shoot proliferation was recorded on Murashige and Skoog (MS) medium containing 0.8 mg/L thidiazuron (TDZ), followed by their subculture on MS augmented with 1.0 mg/L gibberellic acid (GA_3). Upscaling of shoots was carried out using axenic stem segments derived from primary *in vitro* shoots. Simultaneous root development from shoots was observed during shoot multiplication. This phenomenon eliminates the requirement of an additional step of rooting, thus reducing both the cost and time of the plant propagation. The present protocol is an efficient one and could produce ~540 plantlets starting from a single explant within 14 weeks. *In vitro* plantlets were successfully acclimatized and eventually established in the field. Monomorphic banding profile of micropropagated plants viz-à-viz mother plant obtained by inter simple sequence repeat (ISSR) confirmed the clonal fidelity of tissue-culture-raised plants while bio-chemical stability of micropropagated plants was also ascertained using various quantitative phytochemical analyses including high-performance thin-layer chromatography (HPTLC). This protocol could be useful for commercial-scale propagation and conservation of *H. coronarium*.

Evaluation of Nutritive Value, Dietary Antioxidants and *In Vitro* Antioxidant Activity of Some Edible Flowers from Ethnic Sources: Exploration of Lesser Known Food Sources of North East India

Phunumani Baruah, Kangkon Saikia, Khanjan Das, Karabee Dutta, A. K. Handique

Department of Biotechnology, Gauhati University, Guwahati, Assam, India.

phunumanibaruah@gmail.com

Keywords: Antioxidant activity, dietary antioxidants, edible flowers, HPLC-DAD, nutritive value, polyphenols

Abstract

Certain flowers are use as food supplement which is an old food culture for many ethnic communities in the world and is part of traditional knowledge system. However, information about their basic food chemistry is very limited and so there is a big knowledge gap between traditional knowledge, food culture, and scientific scrutiny to validate the traditional knowledge. The present study was focused on evaluation of nutritive and nutraceutical values of edible flowers that are traditionally consumed by various ethnic communities of North-East India. Seven flower samples were selected for study namely, *Moringa oleifera, Nyctanthes arbor-tristis, Musa balbisiana, Phlogacanthus thyrsiformis, Carica papaya, Adhatoda vasica* and *Curcuma angustifolia*. The objectives of the present study are: (1) To study food value in terms of major nutritional parameters; (2) To quantify major dietary antioxidants namely, total phenolics and flavonoids along with profiling of major polyphenols using high-performance liquid chromatography coupled with diode array detection (HPLC-DAD), (3) To evaluate antioxidant efficacy by *in vitro* antioxidant assays. The present study revealed that the flowers are nutritionally rich with protein content of 10-20%, carbohydrate 8-14%, dietary fibre 3-17% and lipid 1.5-4%. Total mineral content was 6-17% with major content of K, P, Na, Ca, Fe and Zn. Among dietary antioxidants, content of total phenolics was 0.3–3.4 mg GAE/g and total flavonoids were 1.9-57.1 mg RE/g. The major polyphenols found were quercetin, naringin, rutin, chlorogenic acid, vanillic acid, sinapic acid, *p*-coumaric acid and gallic acid. Significant variation in antioxidant activity was observed among the flowers. The findings will help to eliminate the misconception that non-conventional and ethnic food are "poor man's food". Being excellent source for nutritional components as well as dietary antioxidants edibles flowers can be considered as a non-conventional source to fight nutritional security.

Role of Elicitors in Eugenol Production in Hairy Root Cultures of *Ocimum tenuiflorum* L.

Swati Sharan,[1,2] Kunal Mukhopadhyay,[2] and Anjana Rustagi[3]*

[1]School of Life Sciences, Jawaharlal Nehru University, New Delhi, India
[2]Department of Bio-Engineering, Birla Institute of Technology, Mesra, Ranchi, India
[3]Department of Botany, Gargi College, University of Delhi, Delhi, India

anjana.chaurasia@gmail.com

Keywords: Eugenol, hairy root cultures, methyl jasmonate, Ocimum tenuiflorum, salicylic acid, yeast extract

Abstract

Ocimum tenuiflorum is an annual aromatic herb highly prized for its culinary and therapeutic values. Eugenol is one of the major active components of *O. tenuiflorum*, which has been recommended for the treatment of different ailments under different systems of medicines. In the present study, we report different tissue culture methodologies and synergistic effects of elicitors for enhanced biomass production and eugenol accumulation in hairy root cultures of *O. tenuiflorum* and their subsequent quantitative estimation using high-performance liquid chromatography. The concentration of elicitors, age of cultures, and exposure time were studied for optimization. Our investigations suggest that the determination of the right stage of culture for addition of elicitors is one of the important parameters for enhanced production of biomass and eugenol accumulation. A sigmoid growth curve was obtained for increase in dry weight (DW) and/or fresh weight (FW) versus accumulation of eugenol. Roots of *in vitro* cultured plants were inoculated in a liquid culture in flasks and multiplied to the maximum biomass, which was 6.63-fold higher than that of initial inoculum after 25 days. Eugenol accumulation in hairy roots versus time graph was a sigmoid curve. Different concentrations of various elicitors, yeast extract, methyl jasmonate, and salicylic acid, were added to 17-day-old (exponential phase) and 22-day-old (stationary phase) hairy root cultures for different exposure times (4, 8, and 12 days). Results suggest that 8 days of exposure to yeast extract is the optimum condition for the maximum biomass production and the accumulation of eugenol in 17-day-old hairy roots cultures. These optimum conditions led to a 6-fold increase in eugenol production. This study recognizes the potential of hairy roots and role of different elicitors for enhanced biomass production and the accumulation of secondary metabolites under *in vitro* conditions.

In Vitro Culture Establishment and Shoot Multiplication in *Buchanania lanzan* (Chironjii)

Fatima Shirin, Deepti Bhadrawale, Sonam Singh Baghel, Sushma Marawi, Trilok Gupta, and Mukesh Sonkar

Genetics and Tree Improvement Division, Tropical Forest Research Institute, P.O. RFRC, Mandla Road, Jabalpur 482 021, India

fatimashirin1997@gmail.com

Keywords: Benzyl adenine, *Buchanania lanzan*, indole acetic acid, *in vitro*

Abstract

Buchanania lanzan (family Anacardiaceae), known as char or chironji, is a commercially useful tree species of India. Its seeds are used as dry fruits and sold at the rate Rs. 600–2000/kg. Seeds are the major source of natural regeneration. The major problem in the reforestation of *B. lanzan* is the low-percentage germination of seeds due to fungal contamination associated with storage of seeds. Seeds exposed to sunlight fail to germinate and soon lose their viability. In forest biotechnology, micropropagation is a promising choice for mass propagation of superior forest tree genotypes. The present research work was undertaken with a view to solve the above-mentioned problem. Ripe fruits were collected from three different healthy trees of chironji located in Tropical Forest Research Institute campus. The hard seed coat was removed manually, and seeds were collected after depulping of fruits. They were surface sterilized by using 0.1% mercuric chloride solution ($HgCl_2$) for 4–5 min followed by rinsing with sterile distilled water. Nodal segments of germinated seedlings were used as explants. Axillary shoot proliferation through nodal segments was tried on Murashige and Skoog (MS) medium supplemented with different concentrations of benzyl adenine (BA; 1, 3, and 5 mg/L) and indole-3-acetic acid (IAA; 0, 0.1, and 0.5 mg/L). The maximum sprouting was obtained on MS medium supplemented with 3 mg/L BA and 0.5 mg/L IAA. Around three to four shoots of 2–3 cm shoot length were formed after 20–30 days of inoculation. These results will be very helpful in shoot multiplication of this commercially important tree species.

Flexibility in Genetic System Directing Diversification and Migration of Medicinally Important Genus *Artemisia* L. Inhabiting North West Himalayas, India

Uma Bharti, Eshan sharma, and Namrata Sharma

Department of Botany, University of Jammu, Jammu, Jammu and Kashmir, India

umabotany786@gmail.com

Keywords: *Artemisia*, chromosome, polyploid, population, rootstock, variability

Abstract

Cytological and morphological variations are usually found in taxa having a wide distribution range; it helps them to survive and adapt to different kinds of environmental conditions. Such variations occur both at inter- and intrapopulation levels and reflect the adaptability as well as future survival that can help the plant diverge its habitat in apparently random directions. *Artemisia* L., a taxonomically complex, medicinally important, and cytologically flexible genus of family Asteraceae, is an adequate example of the same. *Artemisia* L. has a long history of use in herbal medicine especially in matters connected to digestive system and treatment of worms. Present work is based on four species of this genus that are *Artemisia nilagirica* (C.B. Clarke) Pamp, *Artemisia scoparia* Waldst. & Kit, *Artemisia maritima* L., and *Artemisia tournefortiana* Rchb. forming populations at variable altitudinal regimes (332–3350 masl) of J & K state, India.

Cytological details reveal that *A. nilagirica* is the most complex with four cytological races and chromosome numbers as $2n = 18, 32, 34,$ and 54; others are cytologically stable. Abnormalities in male meiotic tract of *A. nilagirica* affect the genetic constitution and viability of male gametes. Such gametes result in to the origin of aneuploids, polyploids, and species complexes with derived/new chromosome numbers in this genus. Besides, these are also responsible for reduced reproductive success through seeds, that is, reduced percentage of healthy seeds. Although little amount of variability survives, it is maintained and ramified through alternate means using rootstock. The rootstock hence acts as the main organ for plant propagation and it can be used for large-scale multiplication of favorable genotypes, if the need arises in the near future, keeping in mind the economic value of various species of this genus.

Defense-Related Gene Expression in *Musa paradisiaca* var. Kachkal against *Odoiporous longicollis* Infestation

Sudarshana Borah,[1,2] Dipsikha Bora,[2] and Priyadarshini Bhorali[1]

[1]Department of Agricultural Biotechnology, Assam Agricultural University, Jorhat, Assam, India
[2]Department of Life Sciences, Dibrugarh University, Assam, India

sborah151@gmail.com

Keywords: Defense, pseudostem weevil, resistance, transcriptome

Abstract

Banana pseudostem weevil, *Odoiporus longicollis*, is an important pest of banana, which causes significant yield loss. In Assam, it is a serious constraint to banana production and management is highly dependent on the use of chemical pesticides. In the present study, we investigated the defense transcriptome of a banana cultivar resistant to *O. longicollis* infestation, Kachkal, in an attempt to monitor the defense-related transcriptomic changes taking place in the resistant plant at molecular level and to identify genes potentially involved in conferring resistance. We artificially introduced the pest to plants raised in controlled environment. The insect causes extensive tunneling to the core of pseudostem and leaf sheath forming a lethal condition for the host. Transcriptome sequencing of uninfested and infested plants depicted a differential pattern in terms of defense-related genes specifically modulated in response to the pest infestation. The transcriptome data show that important defense-regulated genes such as ones encoding pathogenesis-related proteins, chitinase, lipoxygenase, lectin, and so on, showed high fold change upon insect infestation. Important defense-related genes highly expressed in response to infestation were further validated through quantitative polymerase chain reaction (qPCR). In the present research, we are presenting the analysis of the transcriptomic changes taking place in Kachkal specifically against pseudostem weevil infestation, and validation of expression of important defense genes that could be potentially utilized for generating resistant banana lines against the pest.

Characteristics of Biomass Briquettes Prepared in a Low-Power Screw-Press Machine using Wild *Colocasia esculenta* Tuber as Binder

Anjali Narzary[1] and Amarendra Kumar Das[2]

[1]Centre for Rural Technology, IIT Guwahati, Guwahati 781039, Assam
[2]Department of Design, IIT Guwahati, Guwahati 781039, Assam

anjalinarzary@iitg.ac.in

Keywords: Binder, briquette, calorific value, *Colocasia esculenta* (taro)

Abstract

Biomass briquetting is a very promising technology to tackle problems such as agriculture and forest waste management and also generate high-density, good burning characteristics fuel energy that could be a great substitute for fire wood used for cooking purposes in rural areas, and this could in turn solve the problem related to deforestation. Biomass briquettes were made using dry leaves, wood charcoal, and straw. The binder used in this work was wild *Colocasia esculent* tuber (Taro). The following two methods were used for the preparation of the binder: gelatinization and starch extraction. The result shows that wild Taro can make a good substitute for other complicated binders used in different studies. Charcoal and dry leaves were mixed in three different ratios to find the best results. Among all the ratios, 3:1 dry leaves and charcoal briquettes showed the highest volatile matter content of 83.21%, low ash content of 8.45%, high bulk density of 0.605 g/cm^3, and high calorific value of 5414.5 MJ/kg. Binder was tested for two different percentages, 20% and 40%, for dry leaves briquette, and the 20% binder showed better results such as 82.03%, 7.63%, 0.532 g/cm^3, 4239.98 MJ/kg, and 9.03% of volatile matter content, moisture content, bulk density, calorific value, and ash content, respectively. The comparison was also done for gelatinization and starch extraction procedure, out of which the latter showed better results.

Evaluation of Nutritional Parameters, Antioxidant Potential and Polyphenol Profile of *Moringa oleifera* Lam. using HPLC/ESI MS-MS.

Bhanita Talukdar[1], Kangkon Saikia[2], Durlav N. Singha[1]

[1]Advanced Level Institutional Biotech Hub, Dakshin Kamrup College, Mirza, Assam, India.
[2]Department of Biotechnology, Gauhati University, Guwahati, Assam, India.

bhanitatalukdar13@gmail.com

Keywords: Antioxidants, HPLC-DAD, HPLC-ESI-MS/MS, nutritive value

Abstract

Moringa oleifera has been used widely as local food, in traditional medicine and in industrial applications. Leaves of *M. oleifera* are considered to be rich source of vitamins, minerals and exhibit strong antioxidant activity. In the present study, assessment of major nutritional parameters is carried out for protein, carbohydrates, minerals, lipid and dietary fibre; *in vitro* assessment of antioxidant potential was done by examining radical scavenging capability, ferric reducing antioxidant power, metal ion chelating ability and total antioxidant capacity. High-performance liquid chromatography (HPLC) coupled with electrospray ionization mass spectrometric (ESI-MS) detection in positive and negative ion mode has been used to identify the phenolic compounds. Photodiode-array detection (DAD) has been used for screening of different classes of phenolic compounds, whereas MS-MS fragmentation data were employed for their structural characterization.

The *M. oleifera* leaves are found to be rich in protein and carbohydrate content with abundant composition of minerals like calcium (Ca), potassium (K), magnesium (Mg), phosphorous (P), sodium (Na), iron (Fe), zinc (Zn), copper (Cu). The high calcium content (2275 mg/100g) is essential as a part of human nutrition for growth and development. Among the nutraceutical parameters, *M. oleifera* was found to be rich in hydroxycinnamic acids. Major constituent being cholorogenic acid and p-coumaric acid, which is remarkable with total phenolics content of 14.89 mg GAE/g. Leaves are also found to be rich in flavonoid content (28.75 mg RE/g) where, majority of them were characterized as quercetin, kaempherol, apigenin and with some of their derivatives. The polyphenolic composition reflecting the antioxidant activity can be recognized as strongest among various conventional and non-conventional food plants.

Biochar for Soil Moisture Conservation

Kanku Deka[1], P. P. Hazarika,[2] and B. K. Medhi[3]

[1]Krishi Vigyan Kendra, Baksa, Assam, India
[2]Department of Soil Science, Assam Agricultural University, Jorhat, Assam, India
[3]AICRP on Irrigation Water Management, Assam Agricultural University, Jorhat, Assam, India

prarthana96780@gmail.com

Keywords: Biochar, Gravimetric water, pyrolysis

Abstract

"Biochar" is a relatively new term, yet it is not a new substance. Biochars, product of thermal decomposition or incomplete combustion of biomass or bio-wastes under limited oxygen supply, are fine-grained highly porous charcoal substances that are distinguished from other charcoals in its intended use as soil amendments. Recent estimate suggests that nearly 16, 12, 2.78, 58, and 188 lakh tons of rice straw, rice husk, *toria* stover, and bamboo leaves, respectively, remain unutilized annually and these farm wastes have the potential of further reutilization through production of biochar, which may effectively be used in sustainable production system. Characterization of biochar with respect to physico-chemical properties determines the suitability of biochar to conserve soil moisture, which is again regulated by kind and source of feed stock materials. Keeping these aspects in view, a study on characterization of biochars prepared from four different feed stocks, namely rice straw, rice husk, toria stover, and bamboo leaves was conducted at Assam Agricultural University during 2014–15 and 2015–16 to validate its efficiency. After determining the physicochemical properties of the four biochars, a set of pot culture experiment in poly house taking *toria* as test crop was conducted with four biochars. Four hundred gram of soil (preferably light textured) in 500 g capacity of plastic pot replicated thrice was designed statistically (*factorial* CRD) with four doses of biochars (0, 0.5, 1.0, and 1.5% wt/wt). Initially, a moisture level at field capacity was maintained and periodical volumetric soil moisture content (upto 70 days) was monitored to evaluate their efficiency. Gravimetric soil moisture content decreased significantly with the progress in days of experimentation irrespective of types of biochar used. However, increase in biochar doses increased the soil moisture content significantly over the one where no biochar was applied. Highest efficiency to conserve soil moisture over the days of study period was due to the application of bamboo leaves biochar.

In Vitro Antioxidant Activity of Some Non-Conventional Leafy Vegetables and Modelling of Isothermal Degradation of Dietary Antioxidants upon Cooking Time

Kangkon Saikia, Jharna Thakuria, Karabee Dutta, A. K. Handique

Department of Biotechnology, Gauhati University, Guwahati, Assam, India.

kangkonsaikia@gmail.com

Keywords: Antioxidant activity, dietary antioxidants, flavonoids, polyphenols, thermal degradation, HPLC-DAD.

Abstract

Use of non-conventional food plant particularly non-conventional leafy vegetables appears to be a relics of pre-historic age when human beings were hunter gatherer. Over millennium, the practice continued and is mostly common in tropical and subtropical countries in the world which are rich in biodiversity. Five different non-conventional leafy vegetables were selected as plant materials, undertaking major objectives of determination of antioxidant activity, estimation of dietary antioxidants and quantification of eight polyphenols namely, quercetin, naringin, rutin, chlorogenic acid, vanillic acid, sinapic acid, *p*-coumaric acid and gallic acid by high-performance liquid chromatography (HPLC) coupled with a photodiode-array detection (DAD) system. The degradation kinetics was studied for dietary antioxidants namely, total polyphenols and flavonoid content in correlation with different *in vitro* antioxidant assays namely, radical scavenging assay, ferric reducing antioxidant power, metal ion chelating ability and total antioxidant capacity. The kinetic model was designed to resemble the process of cooking (at 100 ^{o}C) within the time interval of 30 to 120 minutes. The fitting of experimental data in first order kinetic equation and the degradation rates were discussed. Significant variation in antioxidant content and antioxidant activity were observed among plant species. The kinetic model revealed that the total phenolics and flavonoid content along with their antioxidant activity increases within first 30 minutes and then reduces marginally after prolonged cooking (i.e. after 60 minutes) as a subject of degradation or loss of its antioxidant potential. However, marginal decrease in antioxidant capacity of some standard antioxidants were also observed within the same conditions but for some standards the decrease was insignificant. The primary conclusion drawn from the present study is that the heat treatment initially liberates some low molecular weight phenolic compounds depending on its intercellular occurrence which when subjected to prolonged heat treatment results in isothermal degradation and hence loss in antioxidant capacity.

Development of Broad-Spectrum Bacterial Blight Resistance in Traditional Basmati Rice Cultivar

Meenakshi Raina[1], Romesh Kumar Salgotra[1], and Deepak Kumar[2]

[1]School of Biotechnology, Sher-e-Kashmir University of Agricultural Sciences & Technology of Jammu, Jammu and Kashmir, India
[2]Department of Botany, Central University of Jammu, Rahya-Suchani (Bagla), Samba 181143 Jammu & Kashmir, India

raina.kesar592@gmail.com

Keywords: Bacterial blight, Basmati, marker-assisted backcross breeding, semi-dwarf

Abstract

The basmati rice variety "Ranbir Basmati" is very popular among farmers of Jammu region due to its palatable taste and short duration to fit in rice–wheat cropping system. However, the variety has recently succumbed to bacterial leaf blight (*Xoo*) caused by *Xanthomonas oryzae* pv. *oryzae* (*Xoo*) and prone to lodging. The severity and significance of damage caused by both the disease and lodging have necessitated the development of effective strategies for their management. Two major bacterial blight (BB) resistance genes and a semi-dwarf gene were introgressed into an Indian Basmati through marker-assisted backcross breeding. A high-yielding introgressed line PAU148 carrying *xa13*, *Xa21*, and *sd1* genes was used as a donor parent. Marker-assisted backcrossing was continued till BC2 generation wherein gene-specific markers specific for resistance genes were used for foreground selection, and a set of parental polymorphic microsatellite markers was used for the background selection at each stage of backcrossing.

In BC_2F_2 population, 19 plants were found to be positive for all three genes, whereas the maximum genome recovery of Ranbir Basmati in BC2F2 was 86.9% in introgressed line SBTIL121. Introgressed lines carrying resistance genes were further evaluated for BB resistance. Genotypes carrying both resistance genes exhibited very high level of resistance against BB, whereas lines containing either *Xa21* or *xa13* gene alone showed moderate resistance. Pyramided lines were also analyzed for agro-morphological characters in randomized block design with two replications. All lines were found to be significant for all agro-morphological traits. Newly introgressed lines in the background of basmati will also be a unique genetic stock and a source for BB resistance genes along with semi-dwarfing gene. These lines can be used as semi-dwarf BB resistance donors for further basmati improvement program.

Production of Androgenic Haploid Plants Through *In Vitro* Anther Culture of *Camellia assamica* Ssp. *assamica* (Masters)

Vijay Kumar Mishra[1] and Rakhi Chaturvedi[2]

[1]Department of Bio-Sciences, Rajiv Gandhi University of Knowledge Technologies, Nuzvid, Krishna District 521202, Andhra Pradesh, India
[2]Department of Biosciences and Bioengineering, Indian Institute of Technology Guwahati, Guwahati 781039, Assam

vijay@rguktn.ac.in

Keywords: Androgenesis, anther culture, *Camellia assamica*, embryogenesis, haploids

Abstract

Tea, *Camellia assamica* ssp. *assamica* (Masters) (family Theaceae), is one of the most important commercial crop, which is consumed daily as a nonalcoholic beverage worldwide. Available *Camellia* species are genetically highly heterozygous due to cross-pollination nature with long gestation periods, which poses a hurdle in the production of elite clones or pure breeding/homozygous lines. In the present report, anthers cultures have been opted for the production of haploids. Androgenic haploid embryos were developed through callus formation from microspores during early-to-late uninucleate stages in anther cultures. Maximum callus induction (96%) was obtained on Murashige and Skoog's (MS) medium having 6% (w/v) glucose, supplemented with 5 μM 2,4-dichlorophenoxyacetic acid (2,4-D), 5 μM 6-furfurylaminopurine (kinetin), 800 mg/L L-glutamine, and 200 mg/L L-serine (callus-induction medium). Proliferation of callus occurred when glucose was replaced with 3% (w/v) sucrose in the callus-induction medium. Embryogenesis of nodulated callus was obtained in 85% of the androgenic callus cultures on MS medium containing 10 μM 6-benzylaminopurine (BAP), 3 μM gibberellic acid (GA_3), 800 mg/L L-glutamine and 200 mg/L L-serine (embryo-induction medium). Maturation of embryos occurred when the concentration of growth regulators and adjuvants present in the embryo-induction medium was reduced by 10-fold. Germination of embryos into the complete plantlets took place when the MS medium was supplemented with 10 μM BAP, 1 μM indole-3-butyric acid (IBA), 0.5 μM GA_3, 80 mg/L L-glutamine, and 20 mg/L L-serine. The chromosomal constitution of *in vitro* developed plantlets was confirmed as $2n = x = 15$ by cytological squash preparation of root tips. Flow cytometric analysis of leaves from these *in vitro* developed plantlets confirmed the ploidy status as haploid.

Unraveling the Role of Aldose Reductase Gene from a Resurrection Plant for Methylglyoxal Detoxification and Abiotic Stress Alleviation in Blackgram

Deepak Kumar,[1] Preeti Singh, [2] and Neera Bhalla Sarin[2]

[1]Department of Botany, Central University of Jammu, Rahya-Suchani (Bagla), Jammu, Jammu & Kashmir, India
[2]School of Life Sciences, Jawaharlal Nehru University, New Delhi, India

deepakkumar@cujammu.ac.in

Keywords: Aldose reductase, drought stress, methylglyoxal

Abstract

Drought and salinity exert osmotic stress on terrestrial plants causing water deficit, which consequently affects plant growth and development. Therefore, these stresses are recognized among the most serious challenges to crop production worldwide. Different environmental stresses imposed on plants may result in similar responses at the cellular and molecular levels. This is due to the fact that diverse environmental stresses often activate similar cell signaling pathways and cellular processes, such as the production of stress proteins, upregulation of antioxidants, and accumulation of compatible solutes. It has been demonstrated that the level of methylglyoxal (MG), a cytotoxic compound, increases upon exposure of plants to various abiotic stresses, in addition to Reactive oxygen species (ROS). Methylglyoxal (MG) is mainly catabolized by two major enzymatic pathways. The first is the ubiquitous detoxification pathway, the glyoxalase pathway. An alternate pathway involves aldose reductase that converts MG into acetol in a Nicotinamide adenine dinucleotide phosphate (NADPH)-dependent two-step reaction.

It is this pathway that we have exploited in our study. Aldose reductase belongs to the aldo–keto reductase super family of enzymes and plays numerous roles in the metabolism of steroids, sugars, and other carbonyls in plants and animals. A detailed functional validation of aldose reductase homologue *ALDRXV4* was first carried out in a model plant, tobacco, and subsequently used for the transformation of a recalcitrant pulse crop, *Vigna mungo*. Studies with the model plant and crop plant revealed that overexpression of *ALDRXV4* in transgenics were more tolerant not only to osmotic stress but also to salinity stress. The increased aldose reductase activity, higher sorbitol content, and less accumulation of the toxic metabolite, MG,in the transgenic lines under nonstress and stress conditions resulted in increased protection through maintenance of better photosynthetic efficiency, higher relative water content, and less photo-oxidative damage. Together, these findings suggest the potential of engineering aldose reductase levels for better performance of agriculturally important crop plants growing under stress conditions in future.

Chrysanthemum morifolium Ramat. as Antigenotoxic Agent in *Allium cepa* Test System

Sakshi Bhushan[1], Jatinder Kaur Katnoria[2], and Saroj Arora[2]

[1]Department of Botany, University of Jammu, Jammu 180006, Jammu & Kashmir, India
[2]Department of Botanical and Environmental Sciences,Guru Nanak Dev University, Amritsar 143005, Punjab, India

bhushan.sakshi8@gmail.com

Keywords: *Allium cepa*, antigenotoxic, *Chrysanthemum*, secondary metabolites

Abstract

The demand for clear and safe environment has amplified over last few decades due to the accumulation of different pollutants such as pesticides, inorganic fertilizers, heavy metals, poly aromatic hydrocarbons, and so on. Among these pollutants, heavy metals have the tendency to persist in the environment and to cause several ill effects in human beings on exposure. Asteraceae, a family consisting of about 300 species, has a great importance in the field of medicine due to the presence of compounds that possess therapeutic properties such as anti-inflammatory, antimutagenicity, and anticarcinogenicity. Similarly, one of its species, *Chrysanthemum morifolium*, has been proved to be a rich source of secondary metabolites such as pyretheroids, sesquiterpenoids, flavonoids, coumarins, triterpenoids, steroids, phenolics, purines, lipids, aliphatic compounds, and so forth. Considering this, the present study was planned to explore the antigenotoxic potential of aqueous, butanol, and ethyl acetate extracts of *C. morifolium* Ramat. against nickel-induced genotoxicity in *Allium cepa* root chromosomal aberration assay. It was observed that all three extracts showed potential to reduce physiological as well as clastogenic aberrations in cells with significant percentage inhibition. Thus, ability of C. *morifolium* to act as antigenotoxic agent revealed its potential to act as an interesting candidate for future drug discovery using natural bioresources.

Floral Artifice in *Trichosanthes cucumerina* L.: Possible Implications and Effects

Mamata Jamwal and Namrata Sharma
Department of Botany, University of Jammu, Jammu 180006, Jammu & Kashmir, India

jamwal.mamta@gmail.com

Keywords: Anthesis, diurnal, fimbriae, monoecious, nocturnal

Abstract

Trichosanthes cucumerina L. is a herbaceous annual climbing vine valued for its medicinal importance. Plants of the species are monoecious and thus need a vector to transfer pollen from male to female flowers for successful fruit and seed set. Both male and female flowers in the species are small, tubular, and white in color with deeply fimbriate petals. Species is nocturnal, and flower opening is initiated between 20:00 and 20:30 hrs. Flowers of both the sexes remain open for full night and a major part of the next day, with their closing occurs around 18:30 hrs on this day. These flowers adopt interesting strategy during anthesis. Both the types show full expansion of fimbriate petals by 22:30 to 3:00 hrs, thus providing a suitable platform for the landing of nocturnal pollinators especially moths, which visit these flowers in ample numbers. After 3:00 hrs, the fimbriae of petals start retracting and retraction is complete by 5:00 hrs. Thereafter, flowers are seen visited by small diurnal visitors that include butterflies, ants, and *Ceratina* sp. Folding of petals give these pollinators better chance to probe flowers and help in pollen transfer. Dual, that is, both nocturnal and diurnal, pollination results in high fruit (64.2 ± 2.01) and seed (14.6 ± 0.29) set in the species. The presentation will elaborate in detail on this phenomenon.

Evaluation of Nutritional Value and Yield Characteristics of Different Species of *Pleurotus* species by Utilizing Various Agro-Forestry Wastes

Durlav Nr. Singha[1], Bhanita Talukdar[2], Manabendra Kalita[2], Nilakshi Gohain[2] and Gunajit Kalita[3]

[1]Department of Botany, Dakshin Kamrup College, Mirza, Assam, India
[2]Advanced Level Institutional Biotech Hub, Dakshin Kamrup College, Mirza, India
[3]Department of Botany, Nalbari College, Nalbari, Assam, India

durlavns@gmail.com

Keywords: Bagasse, bioconversion, ligno-cellulose, malnutrition, nutraceutical, spawn

Abstract

Mushrooms are organisms of fungal lineage whose popularity and nutraceutical properties have been realised in recent years and presently, identified as excellent food source to cope malnutrition in developing countries like India. Cultivation of edible mushroom through biotechnological intervention involves the bioconversion of various ligno-cellulosic rich agro-forestry wastes into proteinaceous foods. In the present study, cultivation of different *Pleurotus* spp. is carried out using low-cost eco-friendly technology wherein locally available agro-forestry wastes such as straw, saw dust, bagasse, husk and leaves. are utilized as raw material. Moreover, effectiveness of locally available substrates for spawn production were also examined and recorded for spawn running, pinhead formation, fruit body formation and mean yield.

The experimental setup consist of complete randomized design with three replicates of each *Pleurotus* spp. and also substrate for spawn and mushroom production. The abiotic factors such as temperature, humidity and light and so forth, also plays an important role in production of mushrooms. Significant differences in yield characteristics was observed among different *Pleurotus* spp. growing in different substrates. Different substrates so chosen for cultivation of mushroom when supplemented with different carbon and nitrogen sources also shows enhanced nutrient content of mushroom. Mushroom cultivation, therefore, generates sufficient employment opportunities not only for unemployed youth but also provides adequate financial support to women folks and other weaker sections of the society.

Commercial Production of Micropropagated *Coccinia indica* (Ivy Gourd)—A Success Story

Daksha Bhatt, Prashant Bhatt, and Dipak Jagdale
Aranya Agri Biotech, Mogar, Gujarat, India

technical@aranyaagri.in

Keywords: *Coccinia indica*, commercialization, ivy gourd, medicinal value, micropropagation

Abstract

Coccinia indica, or ivy gourd, is a common vegetable, which belongs to the cucurbitaceae family. It is a perennial, vine-producing parthenocarpic fruit and propagated by vegetative cuttings during the rainy season. Apart from its use as a food, recent research highlights its excellent medicinal value. Considering the poor survival of vegetative propagation, followed by low yield, an alternative was sought to micropropagate selected mother plants of a locally cultivated variety. Here, we describe the first successful commercialization of micropropagated *C. indica*. Micro-propagation was standardized using modified Murashige and Skoog (MS) medium fortified with different concentrations of N6-furfulryladenine (kinetin) and indole-3-acetic acid (IAA). After hardening, field planting was done. The first trials were taken in our own field in year 2009 when 100 plants were planted to monitor their performance. In subsequent years, large-scale plantations were done each year; till date, we have planted around four lac plants.

On an average, the farmers harvested every third day between 1000 and 2500 kg vegetable per acre as compared with 150–250 kg from the conventional plantation. Moreover, the uniformity of fruit shape and size is 70% as compared with 30% in the conventional plantation. A typical plantation of 1 ha gives a total yield of 90–200 tons per year. Plants produced by micropropagation outperformed the conventionally propagated plants by a factor of 10. This method of production of micropropagated plants has now become a common practice for this crop, fulfilling demands not only in Gujarat but all over India and world too. On an average, we produce and sell about one lac tissue culture tindora plants per annum. Details on micro-propagated plants versus conventionally propagated plants are discussed.

Regulation of Growth, Photosynthetic Parameters and Sugar Metabolism in Rice (*Oryza sativa* L.) Seedlings by Arsenic and Their Possible Alteration by Silicon

Susmita Das and Asok K. Biswas

Plant Physiology and Biochemistry Laboratory, Centre of Advanced Studies, Department of Botany, University of Calcutta, Kolkata 700019, West Bengal, India

sdbot_rs@caluniv.ac.in

Keywords: Arsenic, arsenate reductase, carbohydrate metabolism, photosynthesis, rice, silicon

Abstract

Arsenic toxicity is a global concern owing to ever-increasing groundwater contamination, crops irrigation in many regions of the world including Bangladesh and West Bengal, India. Arsenic contaminated soil adversely affects economic development of Bengal due to its profound effects on growth and physiological activities of agriculturally important crops, and possess human health risk. Arsenate and arsenite are two important inorganic species of arsenic of which arsenite prevails in paddy soils. The effect of arsenate with or without silicate on growth, estimation of photosynthetic parameters by using LI-6400XT Portable Photosynthesis System and carbohydrate metabolism in rice (*Oryza sativa* L. cv. MTU-1010) seedlings were investigated. In the test cultivar, arsenic toxicity significantly decreased growth parameters while increased the level of oxidative stress markers. Rate of arsenate accumulation and its conversion to arsenite by arsenate reductase were significantly enhanced in all arsenate treated seedlings while in jointly treated seedlings with arsenate and silicate, arsenate accumulation and its conversion to arsenite decreased. Arsenate exposure hampered all chloroplast pigment content, namely, chlorophyll-a, chlorophyll-b, carotene and xanthophyll as well as photosynthetic parameters, namely intercellular-CO_2-concentration, net-photosynthesis, transpiration-rate and stomatal-conductance in rice seedlings. Arsenic toxicity increased the accumulation of sugar contents but decreased starch contents indicating major adaptive mechanisms of plants under arsenic stress that contributes to osmoregulation and provides protection of biomolecules. Activities of sucrose phosphate synthase, acid invertase, and starch phosphorylase were increased, sucrose phosphate synthase, activity was decreased. Co-application of silicate and arsenate showed significant alterations on all the examined parameters compared to arsenate treatment alone due to less accumulation of arsenic in tissue leading to better growth and productivity in rice seedlings. Such studies will help to develop a cost effective and farmer-friendly way to overcome this threat by the application of silicon-enriched fertilizers in arsenic-contaminated rice fields.

Development of Micropropagation Protocols of Apple Rootstocks for High-Density Plantation

Manju Modgil

Department of Biotechnology, Dr Y.S. Parmar University of Horticulture and Forestry Nauni, Solan 173230, Himachal Pradesh, India

manju_modgil@yahoo.com

Keywords: Apple, clonal rootstocks, commercial propagation, hardening, micropropagation, rooting

Abstract

As a result of collaborative efforts of University of Horticulture and Forestry and State Department of Horticulture, Shimla, dwarf and semi-dwarf clonal rootstocks of apple (M&MM series) have been recommended for commercial cultivation for high-density planting after testing. For large-scale production and to cater needs of growers, *in vitro* multiplication is of great use and application. Therefore, repeatable protocols have been developed for the micropropagation of recommended rootstocks using axillary buds/shoot meristems successfully for the production of quality planting material and their demonstration. Different-sized explants were excised and initiated to shoot proliferation on Murashige and Skoog (MS) medium supplemented with benzyl adenine (BA) and gibberellic acid (GA_3), with or without an auxin. Following establishment phase, small shoots emerged from clean explants were subcultured on multiplication medium. Shoot multiplication was influenced by cytokinin type, its concentration, and genotype. Of the cytokinins tested, BA was found superior to others.

For rooting, vigorously growing shoots were either given a short pre-treatment in an auxin-containing medium or exposed to an auxin throughout the rooting phase. Rooted plantlets were hardened under controlled conditions and successfully transferred to field. Tissue culture (TC) raised plants were randomly tested for trueness to type and found genetically stable. Procedures were further refined for *in vitro* rooting and hardening of plants for their mass production. Nurseries were raised at different locations of the university where high survival, plant uniformity, and better growth have been observed and the plants were distributed to farmers. The methods standardized here are commercially viable and have provided the basis for rapid bulking up of plants. Therefore, *in vitro* shoot cultures /technologies were passed on to entrepreneurs/industry for further commercialization who are selling TC-raised rootstock plants to farmers of Himachal Pradesh and Jammu & Kashmir, and to Horticulture Department of Arunachal Pradesh.

Production and Characterization of Bio-Oil Derived from Banaba Seeds

Riturani Tamuly[1,2] and Prasenjit Saikia[1,2]

[1]CSIR North East Institute of Science and Technology, Jorhat 785006, Assam, India
[2]Academy of Scientific and Industrial Research, 110020, New Delhi, India

rituranitamuly2012@gmail.com

Keywords: Banaba seeds, bio-char, bio-oil, FTIR, pyrolysis

Abstract

One of the main thermochemical processes to develop useful and valuable bio-fuel such as bio-oil and bio-char is pyrolysis. This paper represents a study on the characterization of the bio-oil derived from banaba seeds (*Lagerstroemia speciosa*, a member of the family Lythraceae) through thermal pyrolysis method at various terminal temperatures from 350 to 650°C with a heating rate of 10°C/min. Maximum bio-oil yield (38%) was obtained at a pyrolysis temperature of 550°C. The product was characterized by Fourier Transform Infrared Spectroscopy (FTIR), Nuclear Magnetic Resonance (NMR; ^{1}H and ^{13}C), and Gas Chromatography–Mass Spectroscopy (GC-MS), which confirmed the presence of various oxygenated hydro-carbons and alcohols. Fuel properties were studied by measuring flash point, pour point, calorific value, and rheological properties.

Quantitative Estimation of Lysine, Cadaverine, Piperideine, and Piperine in *Piper longum* L. Using Reverse-Phase High-Performance Liquid Chromatography

Mrinalini Prasad and Prem Kumar Dantu

Department of Botany, Dayalbagh Educational Institute (Deemed University), Dayalbagh, Agra 282005, Uttar Pradesh, India

premdantu@gmail.com

Keywords: Cadaverine, lysine, piperine, *Piper longum*

Abstract

India seems to be the center of origin for about 110 species of family Piperaceae, and genus *Piper* in India has 50 species. Use of this genus in folk medicines is because of the presence of secondary metabolites. This study shows phytochemical analysis of *Piper longum* (pippali or long pepper). It is widely distributed in tropical and subtropical regions of the world. Its fruit is very important and used in spices after drying. Both fruit and roots of *P. longum* are rich in alkaloids and the key component is piperine. In Ayurveda, it is a good rejuvenator, stimulates the appetite, dispels gas from intestines, and cures respiratory diseases. The main purpose of this study is to emphasize on the recent pharmacology and pharmacognosy research on *P. longum*. The plant is an accepted source of drugs pippali and pippalimulam throughout India. We analyzed the spatio-temporal study of plants in different seasons of India by using chromatography. Main compounds of the study were lysine, cadaverine, piperideine, and piperine in different parts and seasons. All compounds were found to be higher in spikes followed by roots, petiole of the first leaf, and the first leaf. In case of the prespike stage, the maximum quantity of all compounds were found in roots and then in petiole of the first leaf. Interestingly, piperidine accumulation was high in all parts during all the three stages. Cadaverine and piperine contents were minimum in the second leaf and petiole, third leaf and petiole, lower leaf, and internode. While cadaverine was present at reasonably good quantity in nodes during all the three stages, piperine was perceptibly low in this part during all the three stages. Almost all the data show the same pattern in lysine, cadaverine, piperidine, and piperine in three different stages except nodes of the plant.

Characterization of *Areca catechu* Husk Char and its Utilization for Heavy Metal Adsorption

Silvia Saikia[1] and Prasenjit Saikia[2]

[1]Tezpur Central University, Tezpur 784028, Assam, India
[2]North East Institute of Science and Technology, Jorhat 785006, Assam, India

silviasaikia22@gmail.com

Keywords: Areca catechu, char, thermochemical conversion

Abstract

The current investigation deals with the evaluation of properties of char obtained from thermochemical conversion of *Areca catechu* (Ac) husk. Properties of *A. catechu* husk were determined in terms of physico-chemical, biochemical, ultimate, and thermogravimetric (TGA/DTG) and Fourier transform infrared spectroscopic (FTIR) analyses. The thermochemical conversion of *A. catechu* husk was performed using four terminal temperatures namely, 350°C, 450°C, 550°C, and 650°C, with a heating rate of 10°C/min. Characterization and properties of char obtained were evaluated by proximate/ultimate analysis, TGA/DTG, FTIR, X-ray diffraction (XRD), scanning electron microscopy (SEM), and so forth. The heavy metals' adsorption property of *A. catechu* is also reported here.

Effect of Synchronized Sound Waves in the Form of Indian Classical Instrumental Music (Strings and Closed Pipes) on Fruit Ripening Aspect of *Psidium guajava* And *Manilkara zapota*

Sanhita Padhi, P. P. Rout, and R. Swain

Department of Botany, Ravenshaw University, Cuttack, Odisha, India

san_puri9828@rediffmail.com

Keywords: Consonants, dissonant, harmonic frequencies, music therapy, octaves, sound wave technology

Abstract

Harmonic frequencies of octaves in Indian classical music influence the growth of plants starting from the germination to the fruit ripening. It can either enhance or restrict the growth of plants depending on the type of consonants or dissonant being played. Today, the sound waves technology has gained much popularity in this field. It has been studied that sound waves at different frequencies, amplitude, intensity/sound pressure levels; exposure periods; and distances from the sound source influence plants' growth. In recent times, music therapy has become more effective and popular. Soothing and rhythmic harmonic frequencies play a vital role on the basic physical and physiological processes of the living organisms such as plants and animals. Human-played harmonious melodic frequency of instrumental music is proved to exert extraordinary and magical influence on the fruit ripening and many biochemical processes of the experimental species of the present research work: grafted fruits-bearing plants, *Psidium guajava* (guava) and *Manilkara zapota* (sapodilla). In the present research work, experiments were conducted to study the effects of Indian classical *Ragas* (instrumental) having different harmonic frequencies of octaves on fruit-bearing plants. Observations inferred that plants exposed to "dose-dependent" and "time-dependent" soothing, harmonic frequencies of octaves of Indian instrumental classical music showed an earlier fruiting and fruit ripening. Moreover, other biochemical analyses of some primary metabolites in the ripened fruits treated with harmonic, melodious classical music showed astounding results of an increased concentration of metabolites such as reducing sugar, carbohydrates, and proteins inferring the certain development of fruit qualities.

Subcellular Bioengineering of *Artemisia annua* L. for Enhanced Biosynthesis and Accumulation of Artemisinin

M. Z. Abdin

Department of Biotechnology, School of Chemical and Life Sciences, Jamia Hamdard, Hamdard Nagar, 110062, New Delhi, India

mzabdin@jamiahamdard.ac.in

Keywords: Active pharmaceutical ingredient, *Artemisia annua* L., artemisnin biosynthesis, secondary plant metabolism

Abstract

Plants synthesize a vast array of secondary plant metabolites through a network of complex metabolic pathways regulated by endogenous and environmental factors. Although these compounds are believed to be nonessential for plants to live, they play an important role in interaction of plants with the environment ensuring their survival in the ecosystem. Some of these secondary metabolites are also of immense medicinal importance because of their therapeutic value. These are referred as active pharmaceutical ingredients (APIs). The concentrations of these compounds in medicinal plants are, however, very low, limiting their commercial exploitation. Artemisinin is one of these APIs isolated from aerial parts of *Artemisia annua* L. It is a potent antimalarial drug against drug-resistant malaria. In recent times, the demand for artemisinin is exponentially increasing with the increased incidence of drug-resistant malaria throughout the world, especially African and Asian continents. However, the commercial production of artemisinin-based combination therapies has limitation due to the presence of low concentration of artemisinin in plants. Therefore, we employed bioengineering approach to develop transgenic lines of *A. annua* L., overexpressing HMG-Co A reductase *(hmgr)*, amorpha-4, 11-diene synthase *(ads)*, and cytochrome P450 monooxygenase *(cyp71av1)* genes to enhance artemisinin content. The selected transgenic lines were found to accumulate 1.29% to 1.44% artemisinin. Thus, results obtained in these studies, clearly indicate that the synthesis of APIs in medicinal plants is tightly regulated, and bioengineering approach can be used in modulating plant metabolism to improve their biosynthesis, so that drugs manufactured from these APIs could be available at cheaper rates to the public.

A Case Study of Organic Farming in Sonapur, Assam

Kunal Swargiary

Department of Environmental science, Gauhati University, Guwahati, Assam, India

swargairykunal@gmail.com

Keywords: Fertilizer, OFAA, organic cultivation, pesticide

Abstract

Sonapur area with the richness of soil qualities, vegetation, and other suitable requirements is a future hub for organic cultivation. Current study was undertaken to investigate the existing organic farms, both private and government, including interaction with the local farmers with their views and opinions about welcoming the organic sector in their respective regions. The condition is favorable because of the support from the agriculture department toward farmers' efforts in cultivating organic crops. Farmers are in dire need of management skills and development programs. A recent study by Indian Council for Agriculture Research and Assam Agriculture University revealed that the soil is virgin and untouched by chemical fertilizers or pesticides, so can be categorized as organic by default. In 2006, Organic farmers and farms in Assam (OFAA), Sonapur belt was accorded "organic certification" by SKAl International of the Netherlands. The Indian government has also sanctioned a huge amount of money under organic farming schemes for the development of farms and research areas. Different schemes of the government are left unnoticed by the local people due to the lack of awareness. Awareness camps should be organized by government officials in selected areas. An emphasis on the both private and government organic farms could be the leading direction toward the future of organic farming in Assam.

Comparative Assessment of Morphological and Molecular Markers for Describing Genetic Relationships in Some Non-Commercial Banana Cultivars (*Musa* L.) of Assam, India

Debanjali Saikia, Kongkona Borborah, S. K. Borthakur, and Bhaben Tanti

Department of Botany, Gauhati University, Guwahati 781014, Assam, India

btanti@gauhati.ac.in

Keywords: Genetic variation, ISSR markers, morphometrics, non-commercial banana cultivars

Abstract

This study aimed to compare the genetic diversity of 14 non-commercial banana (*Musa* L.) cultivars of Assam, India, by using 48 morphological traits and inter simple sequence repeats (ISSR) markers. Results showed that the accessions of banana samples exhibited a significant amount of variation for their morphological studied parameters. The morphological data were presented in the form of hierarchical clusters and principal components. Furthermore, genetic variability among accessions using ISSR markers, showed the average percentage of polymorphism. Total seven primers were selected to evaluate the genetic diversity among the experimental banana samples. Each primer could generate polymorphism among the accessions, which may be due to a mutation at priming sites. A total of 62 bands were detected, of which 56 bands showed polymorphism. The highest number of bands was detected with the primer UBC-843, and the lowest number of bands was observed with UBC-848.

The genetic similarity index was prepared using Jaccard's similarity coefficient, and the range of genetic similarity was from 0.28 to 0.77, with an average of 0.51. Dendrogram produced from the cluster analysis showed the clear division of the genotypes into two distinct clusters. Seven accessions namely *Bharatmoni*, *Assamiya-malbhog*, *Gobin tulashi*, *Bokmoni*, *Bangali-malbhog*, *Katiya-jahaji*, and *Abor-malbhog* were clustered in the first group. Second group contains the accessions namely *Jatikol*, *Adeel*, *Guwahatia-kol*, *Fessa-monohar*, *Athiya kol*, *Ximalu-monohar*, and *Bogi-monohar*. Relationships between morphological traits and ISSR markers variation were estimated using Mantel test. Morphological characters are good markers for overall genetic variation, whereas ISSR markers cannot resolve plant groups defined by visible traits up to interspecies level.

Isolation and Identification of Endophytic Bacterial Communities Associated with *Piper logum* L., An Important Medicinal Plant using Metagenomic Approach

Mubashar Nazir Mintoo and Prem Kumar Dantu

Plant Biotechnology Lab, Department of Botany, Dayalbagh Educational Institute, Dayalbagh, Agra 282005, Uttar Pradesh, India

premdantu@gmail.com

Keywords: Bioinformatics, endophytic bacteria, phenotypic characterization, *Piper longum* 16S rRNA

Abstract

Endophytic bacteria were isolated from nodes, internodes, petiole, and root segments of *Piper longum*. These bacteria were cultured on specific culture medium. The aim of this work was to isolate bacterial endophytes from *P. longum* and identify them by using both conventional and metagenomic approaches. These bacteria were first differentiated on the basis of morphological parameters. Six different colonies were isolated, purified, and selected for further analysis based upon morphological and colony characterization. These isolated bacteria were used to compare the phenological characterization with molecular identification on the basis of analysis using 16S rDNA sequences. 16S rDNA gene is highly diverse in different bacteria and serves as a phylogenetic marker for identification. In these bacteria, the conserved 16S rRNA was amplified using specific primers and amplicons sequenced. Sequences have been deposited with National Center for Biotechnology Information (NCBI), and accession numbers were obtained. Bioinformatics of the sequences identified bacteria as endophytic bacteria 135L-3, *Enterobacter* sp. SQ6-43, *Bacillus casamacensis* strain TN3, *Alishwanella* sp. JS-30, *Bacterium* B28, and *Enterobacter ludwigii* strain g45 belonging to two different bacterial groups, that is, γ-proterobacteria and firmicutes. These bacteria were present in different branches within a tree, suggesting that these clusters showed different phylotypes. These bacteria were known to perform beneficial roles in plant growth promotion and several other processes of plant metabolism. The present study is the first report about the endophytic bacterial population in *P. longum* using 16S rRNA technique through which six endophytic bacteria have been identified and characterized. The identified bacteria belong to γ-proterobacteria and firmicutes groups.

Purification of Biogas by Pressure Swing Adsorption Process with Mitigation of Methane Gas Emission

Goldy Shah,[1] Virendra Kumar Vijay,[1] and Kamal Kishore Pant[2]
[1]Centre for Rural Development and Technology, Indian Institute of Technology Delhi, Hauz Khas, New Delhi

[2]Department of Chemical Engineering, Indian Institute of Technology Delhi, Hauz Khas, New Delhi

goldyshah@gmail.com

Keywords: Biogas upgradation, methane loss, pressure swing adsorption

Abstract

Multi-fold increase in energy demand worldwide has led to a situation where quest for alternate energy sources has become unavoidable. Beside many other problems, the world is challenged by global warming, and the target is to reduce greenhouse gas (GHG) emissions. Moreover, biogas has potential as a renewable source of energy for rural areas because the majority of the rural population is agrarian, thereby generating substantial agricultural waste throughout the year. Interestingly, the biogas application at commercial scale may help in getting rid of biomass waste and reducing GHG emissions. The upgraded biogas can be utilized in many ways but local use is still the most common option and the most economically viable. However, compressed biogas is considered to be a potential alternative to compressed natural gas (CNG) because of its compositional similarity to CNG. Pressure-swing-adsorption (PSA) based biogas upgradation technology has the remarkable potential to produce bio-CNG as an alternative for natural gas. Over the decades, PSA performance has been described by different mathematical models, but there is limited work done in the field of process simulation, where adsorbent models can be incorporated with other unit operations using commercially available simulator. A major issue faced in the majority of the upgradation plant is that an off-gas stream with a significant methane (CH_4) content is produced and released directly into the atmosphere, which requires to be treated further to avoid emission into the environment. Therefore, this work aims to evaluate parameters and off-gas treatment to develop an efficient model for reducing CH_4 loss. Effects of the parameters such as flow rate, cycle time, and CH_4 concentration on CH_4 and carbon dioxide (CO_2) concentrations were optimized. The optimum conditions for the maximal CH_4 concentration (90.82%) and the lowest CO_2 concentration (7.5%) were 17.56 m^3/h flow rate, 51.89 min duration, and 53.04% CH_4 concentration in raw biogas.

Leaf-Waste-Based Biochar as a Promising Adsorbent for Effectual Hydrogen Sulfide Removal from Biogas

Shivali, Virendra Kumar Vijay, P. M. V. Subbarao, and Ram Chandra

Centre for Rural Development and Technology, Indian Institute of Technology Delhi, Hauz Khas, New Delhi

shivalisahota@gmail.com

Keywords: Adsorption, biogas, H_2S removal, leaf waste biochar

Abstract

Biogas is arguably a more versatile renewable energy source due to its determinate energy value and abundant biomass availability. The main hurdle for implementing the technology for vehicular application is that biogas contains certain concentration of impurity of hydrogen sulfide (H_2S), which leads to the corrosion of expensive metal parts inside plants and engines due to its high corrosiveness and toxicity. Previous literature studies have shown that activated carbon is an effective carbonaceous sorbent for H_2S removal due to its high surface area and porosity for sorption. But the high cost of activated carbon is still a competent challenge.To overcome such problems related to existing technologies, biochar has been found to be one of the best ways to remove H_2S at lower cost. Moreover, biochar-based adsorbents are eco-friendly and relatively 10 times cheaper than commercially available activated carbon. Especially, application of leaf-waste-based biochar will be beneficial for rural applications, where small, independent, robust and decentralized units for biogas production and subsequent upgradation can be installed. Thus, the present study is aimed to evaluate the suitability of leaf-waste-based biochar for H_2S removal from biogas for decentralized rural units. Leaf waste was carbonized at different temperatures to study its effectiveness for H_2S removal from biogas in an adsorption tower. Moreover, freshly prepared biochar and saturated biochar were characterized using attenuated total reflectance Fourier transform infrared spectroscopy, X-ray diffraction, scanning electron microscopy, and energy dispersive spectrometer to develop an insight into the adsorption mechanism. These observations show that leaf biochar can play two in one role as an H_2S absorbent and a nutrient-rich amendment for sulfur-deficient soils along with several factors affecting H_2S adsorption such as surface area of biochar, higher alkaline pH, carbonization temperature, and mineral elements present on the surface of biochar.

In Vitro Optimization Studies for the Production and Scale-Up of Artemisinin—An Important Antimalarial Drug

Rajendra Adak and Rakhi Chaturvedi

Department of Biosciences and Bioengineering, Indian Institute of Technology Guwahati, Guwahati 781039, Assam

rakhi_chaturvedi@iitg.ac.in

Keywords: Antimalarial, *Artemisia annua*, artemisinin, elicitors, secondary metabolites

Abstract

Artemisia annua L. is a diploid cross-pollinating species, which belongs to the family Asteraceae. It is also known as Qinghao, Wormwood, and Sweet Annie. This herb is a hub of several medicinally important secondary metabolites such as artemisinin, asarteannuin, absinthin, myricetin, quercetin, caffeic acid, and gallic acid. Although each one of these metabolites is important, artemisinin is in high demand owing to its wide therapeutic applications in malaria, cancer, and peptic ulcer. Additionally, the compound is reported to have potent antibacterial and anti-inflammatory properties. Essential oil obtained from *A. annua* is known to poses powerful insect-repelling properties. Despite being a high-value crop, extracting consistent amount of the metabolites from this plant becomes difficult due to occurrence of high variability within the existing genera. In such a scenario, *in vitro* culture technique would serve as the best method for developing high-metabolite-producing cell lines independent of genetic and seasonal variations. The present review touches aspects for the best *in vitro* regeneration protocol to obtain elite clones in *A. annua* and further inspect into yield enhancement strategies using biotic (jasmonic acid and salicylic acid) and abiotic physical elicitors (UV-B, salinity, and temperature) to scrutinize and attain ideal parameters for scale-up studies of artemisinin production using a bioreactor.

Plant Regeneration Via Repetitive Secondary Embryogenesis from Androgenic Embryos in Suspension Cultures of *Camellia assamica* ssp. *assamica* and Clonal Fidelity Assessment using RAPD Marker

Ruchira Bajpai and Rakhi Chaturvedi

Department of Biosciences and Bioengineering, Indian Institute of Technology Guwahati, Guwahati 781039, Assam, India

rakhi_chaturvedi@iitg.ac.in

Keywords: FESEM, histology, hyperhydricity, RAPD, Recurrent embryogenesis, two-step protocol

Abstract

Rapid, recurrent embryogenesis was attained from *in vitro* developed androgenic haploid embryos of *Camellia assamica* ssp. *assamica* using a liquid medium. Primary globular embryos from an embryo multiplication medium, Murashige and Skoog (MS) medium supplemented with 10 μM 6-benzylaminopurine (BAP), 3 μM gibberellic acid (GA_3), 800 mg/L L-glutamine, and 200 mg/L L-serine, were subjected to pretreatment with 18 μM abscisic acid (ABA) either alone or in combination with either of the osmoticum, 25 g/L mannitol, 30 g/L polyethylene glycol (PEG-400) or 1 g/L glycine betaine. Higher multiplication with a fivefold increase in secondary embryogenesis was achieved after pretreatment of primary embryos on MS medium consisting of 18 μM ABA and 25 g/L mannitol for 30 days, followed by transfer to an embryo maturation medium (control), MS with 1 μM BAP, 0.3 μM GA_3, 80 mg/L L-glutamine, and 20 mg/L L-serine. Effect of physical state of the medium on embryo germination leading to shoot differentiation was also studied. An increased germination rate of 66.6% in secondary embryos was obtained when cultures were initially kept for 20 days in a liquid medium followed by transferred to a semisolid medium. In comparison, a liquid–solid medium favoured 44.4% embryo germination in 30 days, whereas a semisolid medium resulted in only 25% embryo germination in 45 days. The continuous immersion in a liquid medium led to hyperhydricity in cultures. Therefore, a two-step protocol, involving combination of liquid and semisolid medium transition was chosen for attaining large-scale multiplication of this elite clone in less time. Histology and field emission scanning electron microscopic (FESEM) analyses were performed to determine ontological stages of embryo development. Clonal fidelity of plants attained from liquid–semisolid medium transition was assessed using random amplified polymorphic DNA (RAPD) markers to rule out any somaclonal variations occurred during multiple transfer cycle.