

Functional Plant Science and Biotechnology

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Cover photos: Top: Stalk propagation of ScYLV infected and uninfected plants (Vasconcelos *et al.*, pp 31-35); Bottom: Solid phase microextraction procedure (Guedes de Pinho *et al.*, pp 1-15).

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CONTENTS

Paula Guedes de Pinho, David M. Pereira, Rui F. Gonçalves, Patrícia Valentão, Fátima Fernandes, Marcos Taveira, Paula B. Andrade (Portugal) Headspace-Solid Phase MicroExtraction and Gas Chromatography Mass Spectrometry Applied to Determination of Volatiles in Natural Matrices	1
Werayut Pothitirat, Pimolpan Pithayanukul, Mullika Traidej Chomnawang, Roongtawan Supabphol, Wandee Gritsanapan (Thailand) Biological and Chemical Stability of Mangosteen Fruit Rind Extract	16
Sandra M. Scagliusi (Brazil), Saikat K. Basu (Canada), Jorge Alberto de Gouvea, Jorge Vega (Brazil) Physiological Alterations in Brazilian Sugarcane Varieties Infected by <i>Sugarcane yellow leaf virus</i> (ScYLV)	19
Sandra M. Scagliusi (Brazil), Saikat K. Basu (Canada), Jorge Alberto de Gouvea, Jorge Vega (Brazil) Comparison of Two Diagnostic Methods for Evaluation of <i>Sugarcane yellow leaf virus</i> Concentration in Brazilian Sugarcane Cultivars	26
Antônio Carlos M. Vasconcelos, Marcos C. Gonçalves, Luciana R. Pinto, Marcos G. A. Landell, Dilermando Percin (Brazil) Effects of <i>Sugarcane yellow leaf virus</i> on Sugarcane Yield and Root System Development	31
Jun-jie Shan, Feng-xia Ren, Geng-yuan Tian (China) Structure Characterization and Hypoglycemic Activity of a Glycoconjugate from <i>Atractylodes macrocephalae</i> Koidz	36
Manal Al-Kandari, Amina Redha, Patrice Suleman (Kuwait) Polyamine Accumulation and Osmotic Adjustment as Adaptive Responses to Water and Salinity Stress in <i>Conocarpus lancifolius</i>	42
Appaji Nanda, Hiregouja M. Prakasha, Yelugere L. Krishna Murthy, Hebbalalu S. Suresh (India) Seasonality, Flowering and Fruiting Patterns in a Tropical Dry Deciduous Forest of Bhadra Wildlife Sanctuary, Southern India	49
Shital B. Pokharkar, D. M. Mahajan, T. D. Nikam, V. R. Gunale (India) Assessing Impacts of Habitat Modification on Plant Diversity of an Urban Wetland	55
H. T. Raghavendra Gowda, Vijaya Kumara (India) Overview of Some Wetlands in the Lakkavali Range of Bhadra Wildlife Sanctuary, Mid Western Ghat Region Karnataka: Threats, Management and Conservation Issues	60
Sondeep Singh, Bavita Asthir (India) Biochemical and Physiological Parameters: Swift Tools for Screening High Temperature Tolerance in Barley	70

Paula Guedes de Pinho, David M. Pereira, Rui F. Gonçalves, Patrícia Valentão, Fátima Fernandes, Marcos Taveira, Paula B. Andrade (Portugal) Headspace-Solid Phase MicroExtraction and Gas Chromatography Mass Spectrometry Applied to Determination of Volatiles in Natural Matrices (pp 1-15)

ABSTRACT

Invited Review: Headspace – Solid Phase MicroExtraction (HS-SPME) combined with Gas Chromatography-Mass Spectrometry-Ion Trap Detector (GC-MS-IT) has been used to characterize volatile compounds in several natural matrices. A great number of fibres with different polarities are commercially available, allowing the screening of the highest possible number of components. HS-SPME has several advantages, such as the condensation of extraction, concentration and sample introduction into a single step. In this extraction technique, a medium polarity fibre for flavours (Carboxen/Polydimethylsiloxane or Divinylbenzene/Polydimethylsiloxane) is usually used in order to achieve equilibrium between non-polar and polar compounds, thus increasing the screening range. The traditional methodologies using organic solvents, namely dichloromethane (DCM), allow the determination of a considerable number of volatile and semi-volatile compounds in natural matrices. In this review, the main theoretical points involved in GC-MS analysis will be discussed. The application of HS-SPME and other extraction methodologies (hydrodistillation, Soxhlet and solvent extraction), coupled to GC-MS-IT, to several natural matrices (macroalgae, *Rumex induratus*, *Brassica oleracea* L. var. *costata* DC., *Catharanthus roseus* and mushrooms, with particular emphasis in these last two) will be referred. With this the usefulness of these methodologies in the screening of volatile and semi-volatile compounds will be demonstrated.

Werayut Pothitirat, Pimolpan Pithayanukul, Mullika Traidej Chomnawang, Roongtawan Supabphol, Wandee Gritsanapan (Thailand) Biological and Chemical Stability of Mangosteen Fruit Rind Extract (pp 16-18)

ABSTRACT

Short Communication: The stability of α -mangostin, antiradical activity against DPPH radical and antibacterial activity against bacteria-inducing acne, i.e. *Propionibacterium acnes* and *Staphylococcus epidermidis* of *Garcinia mangostana* fruit rind extract were studied. The extract was kept in an amber glass vial and in an aluminium foil bag at different temperatures (4-8, 25-28 and 45°C) for 120 days. The content of α -mangostin in the extract stored at each condition was analyzed by the validated RP-HPLC method, while free radical scavenging and antibacterial activity were performed using the DPPH scavenging assay and broth microdilution method, respectively. The results revealed that packaging material and storage temperature had no effect on the content of α -mangostin, antiradical and anti-acne activities of the *G. mangostana* fruit rind extract. However, a small change in color and characteristic of the extract was observed when the extract was kept at 25-28 and 45°C. These data indicate good chemical and biological stabilities of *G. mangostana* fruit rind extract for appropriate antioxidant and anti-acne raw materials in pharmaceutical preparations.

Sandra M. Scagliusi (Brazil), Saikat K. Basu (Canada), Jorge Alberto de Gouvea, Jorge Vega (Brazil) Physiological Alterations in Brazilian Sugarcane Varieties Infected by *Sugarcane yellow leaf virus* (ScYLV) (pp 19-25)

ABSTRACT

Original Research Paper: The objective of this study was to verify the physiological alterations caused by *Sugarcane yellow leaf virus* (ScYLV), one of the most important viral diseases of sugarcane (*Saccharum officinarum* L.). We analyzed the levels of soluble sugars and the activity of peroxidases in different Brazilian sugarcane varieties, with different levels of resistance and susceptibility to the virus. Comparison was also carried out between the leaf blade and the main ribs of +1 and +4 leaves, from healthy and infected plants. Soluble sugars levels (especially sucrose) were found to be higher in leaves of symptomatic infected plants, but not in healthy or asymptomatic infected plants. There was a significant increase in the content of soluble sugars in the main ribs of infected plants, followed by the leaf blade of +1 leaves. The activity of peroxidases was significant higher in symptomatic infected plants compared to healthy or infected plants without symptoms.

Sandra M. Scagliusi (Brazil), Saikat K. Basu (Canada), Jorge Alberto de Gouvea, Jorge Vega (Brazil) Comparison of Two Diagnostic Methods for Evaluation of *Sugarcane yellow leaf virus* Concentration in Brazilian Sugarcane Cultivars (pp 26-30)

ABSTRACT

Original Research Paper: *Sugarcane yellow leaf virus* (ScYLV) is one of the main virus diseases infecting sugarcane (*Saccharum* sp.) in major sugarcane-producing areas around the world. The virus belongs to the *Luteoviridae* family and is transmitted by different aphid species. This work was carried out to evaluate ScYLV concentration in different tissues of infected plants (with or without symptoms), and to compare a serological (DAS-ELISA) and a molecular (RT-PCR) method of detection. Both tests were highly specific and their sensitivity was very similar. Both methods of detection revealed the presence of ScYLV in asymptomatic and symptomatic sugarcane plants, suggesting different levels of tolerance or resistance. To better understand some aspects of virus distribution, virus concentration was also evaluated in younger and older leaves and in two parts of the leaf (the midrib and the leaf blade). Virus concentration was significantly higher in the younger leaves compared to the more mature leaves, and there was a significant increase in virus concentration in the midribs. There was no relationship between virus concentration in infected plants and intensity of symptoms. These results provide information on the most appropriate method for routine ScYLV detection and identify the best plant tissue to be used for a reliable diagnosis.

Antônio Carlos M. Vasconcelos, Marcos C. Gonçalves, Luciana R. Pinto, Marcos G. A. Landell, Dilermando Percin (Brazil) Effects of *Sugarcane yellow leaf virus* on Sugarcane Yield and Root System Development (pp 31-35)

ABSTRACT

Original Research Paper: Sugarcane yellow leaf syndrome (YLS) causes significant yield losses in susceptible sugarcane varieties. In Brazil, YLS was not recognized as economical importance until early 1990s. However, with the drastic epidemics of the viral form of the disease in variety SP71-6163, breeders began to take into account its occurrence during the selection stages and its effects on vegetative development. The objective of the present work was to evaluate the effects of the *Sugarcane yellow leaf virus* (ScYLV), its main causal agent in Brazil, on sugarcane yield and root system development. The experiment was conducted in Ribeirão Preto, SP, Brazil, on Typic Hapludox soil, in variety IAC89-2135 during the plant cane cycle. ScYLV diagnosis was assayed by DAS-ELISA and RT-PCR for discrimination between infected and uninfected plants. Plants grown from ScYLV infected stalks showed typical infection symptoms as a consequence of virus perpetuation in the stalk. The infected plants showed significant reduction on roots dry weight and fresh weight of the above plant parts and an increase in Brix and sucrose content in the stalks. Although infected plants maintained regular root system vertical architecture, root dry weight was reduced and negatively correlated with fresh weight and stalk number, showing that alterations in root and vascular systems constitute important effects from ScYLV infection.

Jun-jie Shan, Feng-xia Ren, Geng-yuan Tian (China) Structure Characterization and Hypoglycemic Activity of a Glycoconjugate from *Atractylodes macrocephalae* Koidz (pp 36-41)

ABSTRACT

Original Research Paper: *Atractylodes macrocephalae* Koidz is a traditional medicinal plant in China. We previously reported that a complex-polysaccharide fraction (AMP-B) isolated from the root of this plant showed potent hypoglycemic activity in alloxan-induced diabetic rats after oral administration, so we further isolated and purified the active component from AMP-B to study its structure and hypoglycemic activity. Using DEAE-cellulose and Sepharose CL-6B gel filtration chromatography, we obtained an active glycoconjugate (AMP-2) from AMP-B. The molecular weight of AMP-2 was estimated to be 56660 Da by MALDI-TOF-MS. AMP-2 contains 80.9% (w/w) carbohydrate and 19.5% protein. It is composed of L-rhamnose, L-arabinose, D-mannose, D-galactose, D-glucose and D-galacturonic acid in a molar ratio of 1.0: 3.0: 1.0: 3.5: 2.1: 3.0. Its structural features were elucidated by reduction of carboxyl-groups, enzymatic degradation and reductive alkaline degradation, methylation analysis, ¹H-NMR and ¹³C-NMR. The results suggest that AMP-2 has the following residues: L-1,5-linked and rich terminal arabinose, D-1,2-linked, 1,4-linked and terminal galactose, L-1,2,4-linked rhamnose, D-1,2-linked and D-1,6-linked glucose, terminal D-mannose, and D-1,4-linked and terminal galacturonic acid. The protein fractions may be linked with L-1,2,4-linked rhamnose and D-1,6-linked glucose. AMP-2 showed a remarkable hypoglycemic activity in alloxan-induced diabetic rats after oral administration at a dose of 50 mg/kg. In conclusion, a highly branched carbohydrate-conjugate obtained from the root of *Atractylodes macrocephala* has shown marked hypoglycemic activity, which may provide a practical quality control protocol for this herbal medicine.

Manal Al-Kandari, Amina Redha, Patrice Suleman (Kuwait) Polyamine Accumulation and Osmotic Adjustment as Adaptive Responses to Water and Salinity Stress in *Conocarpus lancifolius* (pp 42-48)

ABSTRACT

Original Research Paper: *Conocarpus lancifolius* is an ornamental plant that flourishes under the semi-arid conditions of Kuwait. The adaptive response of this species to drought, salinity stress and the relationship of free polyamines in leaves, leaf osmotic potential, chlorophyll content and fluorescence were determined. These were done with single shoots (14-15 leaves) in a controlled environmental chamber at 25°C and RH of 45-50%. The leaves of plants treated with ≥ 1.37 M NaCl had a relative water content (RWC) of 65% or lower. Shoot elongation and leaf development ceased just after leaf osmotic potential (ψ_s) of ≥ 3.32 MPa and the threshold for salt damage in the leaves appeared to be at 1.03-1.37 M NaCl after 10 days. The level of salt tolerance indicated that *C. lancifolius* may be a xerohalophyte. The chlorophyll (chl) content index increased by the formation of green islands and declined with ultrastructural changes of the chloroplasts in 1.37-1.71 M NaCl. The minimal Chl fluorescence (F_0) increased with increase in salinity and drought but the electron transport rate ETR, photochemical quantum yield (Y), photochemical quenching (qP) and variable to maximal fluorescence (F_v/F_m) declined after 10 and 4 DAT with increase in salinity and drought, respectively, an indication of some damage to PSII. Putrescine (Put) was the predominant polyamine during the early stages of drought and salinity stress. Although Put was the most abundant polyamine in 0.17-0.34 M NaCl, at higher concentrations spermidine (Spd) and spermine (Spm) were more abundant. The titer and type of polyamine accumulated in *C. lancifolius* appeared to be related to the nature, intensity and duration of environmental stress. Understanding its response to drought and salinity stress will assist in the management and longevity of this species.

Appaji Nanda, Hiregouja M. Prakasha, Yelugere L. Krishna Murthy, Hebbalalu S. Suresh (India) Seasonality, Flowering and Fruiting Patterns in a Tropical Dry Deciduous Forest of Bhadra Wildlife Sanctuary, Southern India (pp 49-54)

ABSTRACT

Original Research Paper: We examined 24 months' data of flowering and fruiting phenophases of 277 individuals of 45 species at the community level. We determined the timing of the phenophases in relation to seasonal rainfall and temperature. Regression analysis was performed to examine how variations in rainfall and temperature influenced the peaks and troughs of phenology cycles. We also investigated seasonality of various phenophases to understand their cyclicity and strength. Flower initiation begins in November and continues until June with a peak during April to June. Flower opening of pollination peak occur in April and May, respectively. Fruiting initiation peaked twice, in February to April in the dry season and after rainfall in September to October. Fruit maturity peaked in March in the dry season and after rainfall maturation in August and November. Deviations from phenology cycles were largely attributable to short-term fluctuations in rainfall and/or temperature. Overall our study suggests that the response of reproductive phenophases is an indication of regional environmental changes.

Shital B. Pokharkar, D. M. Mahajan, T. D. Nikam, V. R. Gunale (India) Assessing Impacts of Habitat Modification on Plant Diversity of an Urban Wetland (pp 55-59)

ABSTRACT

Research Note: Wetland plant species composition is highly sensitive to habitat modification. It is influenced by climate change, eutrophication, and other anthropogenic activities. The present study emphasizes impacts of habitat modification of an urban wetland (Bhosari Lake) on plant species composition in rapidly developed Pune suburban area. The influence of urbanization and subsequent beautification of lakes on an aquatic ecosystem was assessed to know the transformation of plant species and composition. Intensive field surveys of this area resulted into documentation of more than 100 plant species. After analyzing the results, it was observed that the species composition has been significantly altered. The main factor responsible for species alteration was habitat modification and subsequent invasion of introduced and naturalized alien species. Though the species diversity increased, most of the naturally occurring aquatic species had disappeared. The plants that disappeared were sensitive to habitat modification: *Coix lacryma-jobi* L., *Najas indica* (Willd.) Cham., *Nymphoides hydrophylla* (Lour.) O. Ktze., *Ottelia alismoides* (L.) Pers., *Vallisneria spiralis* L. and *Caesullia axillaris* Roxb. Species like *Ipomoea carnea* Jacq., *Eichhornia crassipes* Solms., *Persicaria glabra* (Willd.) Gomez, *Alternanthera philoxeroides* (Mart.) Griseb invaded the wetland area. There was major loss in typical submerged, rooted and marshy macrophyte species, whereas the number of free-floating, facultative and obligate upland plants increased significantly. Upland and fringe plant species occupied the newly modified wetlands area. This signifies the impacts of habitat modification. In the process of recreation and beautification, lake vegetation has changed from marshy to upland.

H. T. Raghavendra Gowda, Vijaya Kumara (India) Overview of Some Wetlands in the Lakkavali Range of Bhadra Wildlife Sanctuary, Mid Western Ghat Region Karnataka: Threats, Management and Conservation Issues (pp 60-69)

ABSTRACT

Original Research Paper: The Lakkavalli range comprises an endangered habitat with several endemic species that can be found in many countries, mainly in the Lakkavalli range of the Bhadra Wildlife Sanctuary, but that are rapidly disappearing. For designing optimal conservation and management strategies for this ecosystem, appropriate characterization and classification of these wetlands is necessary based on the different types of habitats and their varying environmental conditions. This paper presents the current ecological status and threat to Lakkavalli range wetlands of Bhadra Wildlife Sanctuary and summarizes some management and conservation issues based on the existing experience on a regional level. The study reveals that most of the water and Soil quality parameters are in the normal range.

Sondeep Singh, Bavita Asthir (India) Biochemical and Physiological Parameters: Swift Tools for Screening High Temperature Tolerance in Barley (pp 70-75)

ABSTRACT

Original Research Paper: High temperature (HT) stress is a wide-spread problem seriously affecting barley production and quality. Development of heat-tolerant cultivars is of prime importance; however, the progress is hampered by a lack of swift and efficient tools for selection of tolerant germplasm that can be used as a source of candidate genes. This study was conducted to explore the thermotolerance behaviour of barley with respect to germination, dry mass accumulation, membrane thermal stability, TTC (2,3,5-triphenyl tetrazolium chloride) cell viability, lipid peroxidation and chlorophyll content. Twenty genotypes of barley were subjected to a brief heat shock (HS) episode of 45°C for 2 h followed by transfer to a normal temperature (25°C) for five days. HS reduced the germination, dry mass accumulation and damaged the integrity of cellular membranes as indicated by increased electrolyte leakage after exposure to HS. Genotypic testing, using TTC reduction as a measure of tissue viability, following HS treatment, confirmed the thermal responsiveness of seedlings. The inhibitory effect of HS was reflected by increased lipid peroxidation and a decline in chlorophyll content. The tolerant genotypes registered less reduction in germination, dry mass accumulation, TTC cell viability and chlorophyll content under HS conditions compared to susceptible ones. Oxidative damage, in terms of generation of malondialdehyde, was markedly higher in susceptible genotype. These results suggest that the biochemical and the physiological parameters can be used as tools for screening HT tolerance in barley to facilitate the development of HT-tolerant germplasm.