

## Conference Abstracts

### South African Association of Botanists

Abstracts of papers and posters presented at the 31<sup>st</sup> Annual Congress of the South African Association of Botanists held at the University of the Free State, Bloemfontein, 10–14 January 2005

*The presenter of multi-authored papers is underlined*

#### ★ Awards made to students

#### Plenary Lectures

##### Reactive oxygen species often lie at the heart of plant stress

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Reactive oxygen species (ROS) are an important group of free radicals. All are derivatives of molecular oxygen, with one or more unpaired electrons, and are associated with plant stress as primary elicitors, as products and propagators of oxidative damage, or as signal molecules that initiate mechanisms of defence or adaptation. Like any other living organism, a plant constantly produces and extinguishes ROS. A complex system of enzymes and other ROS-scavenging molecules, such as polyphenols, keep the concentration of free radicals very low, but this balance can be disturbed by increased radical production or by decreased antioxidant capacity. External factors may induce free radical production, the scavenging of which may exhaust the antioxidant system, for example, by oxidising the ascorbate and glutathione pools if these cannot be restored fast enough. Adverse temperatures, drought or disease may decrease the activity of antioxidant enzymes. In particular, photosynthetic electron transport is a major site of oxidative stress by visible or ultraviolet light, high or low temperatures, or herbicides and pollutants. In their adaptation/acclimation plants produce antioxidants in response to certain stresses. These antioxidants have an important function in the removal of free radicals and therefore in conferring tolerance to both abiotic and biotic stress factors. Direct monitoring of ROS is therefore a necessary and informative aspect of stress experiments, even under conditions where certain physiological parameters may initially seem to be unaffected.

##### Defence against aphids in crop plants

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Aphids are serious pests on cereals, causing economic loss due to direct effects and as vectors of viral diseases. Information about natural plant defence systems is useful for alternative strategies for pest control, crop production systems, and breeding strategies. The report contains studies on a constitutive defence system acting against aphids in rye (*Secale cereale*) and on aphid-induced defence reactions in barley (*Hordeum vulgare*). The constitutive defence found in rye, and also in wheat and maize, consists of cyclic hydroxamic acid glucosides and  $\beta$ -glucosidases. The components are separated in the intact plant but come in contact and activate the defence upon tissue wounding. The  $\beta$ -glucosidases have been localised at tissue and subcellular level by immunochemistry. Their

kinetic and molecular characteristics are examined by cloning, expression in *E. coli*, and studies of the recombinant proteins. In barley, the reactions induced by the bird cherry-oat aphid (*Rhopalosiphum padi*) are studied. Using specific antibodies, it has been shown that pathogenesis-related proteins, such as chitinase and  $\beta$ -1,3-glucanase are induced. To obtain a broad picture, subtracted cDNA libraries from infested and non-infested barley plants have been constructed and screened for differentially expressed clones. Two clones, identified as the *R. padi* virus and an O-methyltransferase (OMT), have been further investigated. The *R. padi* virus has been shown to be widely spread among *R. padi* populations in Sweden. The putative OMT has been expressed in *E. coli* and purified. It acts on substrates other than those previously reported and is here proposed to be involved in the biosynthesis of defence chemicals. Recently we started differential expression profiling using DNA microarray techniques. Preliminary results from this ongoing study will also be presented.

#### Papers

##### Sink – source transition in relation to plastochron index in *Pisum sativum*

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Based on translocation studies, three groups of leaves are widely known to occur at the same time: source, transition and sink. As leaves unfold and start to expand, the transport of assimilates change from being sink orientated to bidirectional (transition) and at full maturity, source orientated. We investigated the sink to source transition in *Pisum sativum* L. leaves at different plastochron ages in non-nodulating plants, using the phloem-mobile symplasmic fluorescent marker, 5,6-carboxyfluorescein. Our results demonstrate that young leaves remain strong sinks up until LPI 0.5, after which sink-source transition occurs between LPI 1.0 and 2.0 and leaflets become strong source systems between LPI 2.5 and 3.0. There are reports which state that sink-source transition commences after leaf unfolding, but in this study, unfolded leaflets at LPI 0.5 are still strong sinks and transition does not commence until much later, at approximately LPI 1.0. Our results suggest that the use of the plastochron index allows for easier prediction of the sink/source status of leaves.

##### Phloem transport in compound leaves – a requirement for assimilate balance?

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The phloem pathway has been demonstrated to involve the movement of photoassimilate from sites of production (sources) to sites of utilisation (sinks), no matter how weak the sink might be. Application of the phloem-mobile symplasmic fluorescent marker, 5,6-carboxyfluorescein, revealed that in compound leaves, the fluorochrome was first transported from the leaflet to which 5,6-CF was applied, to its opposite leaflet in the pair – in all leaves, irrespective of the application leaves being source or sink. In source leaflets, fluorochrome balance occurs between leaflet pair before export to the axis. In sink leaflets, fluorochrome is transported between paired leaflets, but without any transfer out of the leaf system. The results indicate that an assimilate balance occurs between paired leaflets, irrespective of the leaflets being exporters or importers of assimilate. At the light microscopy level, we noted a well developed vascular connection between paired leaflets which joined with that of the plant axis. Activity within the vascular connection between paired leaflets is obviously under a kind of regulatory (manifold) pressure, which is independent of source to sink movement.

### The successful re-introduction of indigenous African vegetables to a community in South Africa

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A great deal of research is being conducted into plants that have the potential to become 'new' crops in South Africa. These species may be exotics that have the potential to be cultivated in the country, or indigenous species that have been used by local people in the past. Much of this research is aimed at providing emerging farmers with potential niche market crops. All of this research, however, will come to naught unless the crops are successfully introduced into the farming system. Between 1999 and 2001 an international project was conducted on the domestication and improvement of three indigenous African vegetables. The project looked at the use of amaranths, cucurbits and the Livingstone potato (*Plectranthus esculentus* N.E.Br. – Lamiaceae) as potential crops in Tanzania and South Africa. Genetic material was collected, multiplied and re-introduced into three communities in South Africa. The amaranth species used were *Amaranthus hybridus* L., *A. hypochondriacus* L. and *A. tricolor* L., while the main cucurbits in use were found to be *Cucurbita maxima* Duch., *C. pepo* L. and *Cucumis melo* L.. Evaluations of two other useful cucurbits (*Momordica balsamina* L. and *Acanthosicyos naudiniana* (Sond.) C.Jeffrey) were also carried out. This paper reports on the successful re-introduction of some of these plants into the Mamobolo community in the Limpopo Province. The processes and procedures followed are presented, together with problems that were encountered and lessons that were learned during the project. The successful conclusion of this project resulted in a model that could be followed in introducing new crops into communities. The lessons learned are easily applicable to other projects of this nature, and could prevent costly mistakes in other projects where new crops are to be introduced.

### Identification of genes uniquely expressed in resistant wheat infected by leaf rust

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Transcriptional control of the expression of stress-responsive genes is a crucial part of the response of plants to various biotic and abiotic stresses. By means of DD-RT-PCR, an attempt was made to identify genes from Thatcher+Lr34 wheat which are involved in the resistance response against leaf rust caused by *Puccinia triticina*. Thirty differentially expressed cDNA fragments were identified and cloned. The expression of each clone was tested in infected resistant and susceptible wheat using Reverse Northern Blots. Three clones were uniquely expressed in infected resistant plants shortly after infection. Two of the three clones shared homology with genes encoding a monosaccharide transporter and transcription factor respectively, while the third is unknown at this stage.

### Micropropagation of *Watsonia* – getting to the root of the problem

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The genus *Watsonia* comprises 52 species indigenous to southern Africa, 31 of which are endemic to the Cape and 18 to the summer-rainfall regions. The remaining species are widely distributed. Relative to other members of the Iridaceae, less attention has been paid to improving existing species and developing new *Watsonia* hybrids. There are as yet no indications that species from this genus have been successfully propagated *in vitro*. Seeds from species growing in winter rainfall areas (*W. laccata* and *W. vanderspuyiae*) germinated *in vitro* only in response to a three week cold treatment, while species from summer rainfall areas (*W. gladioloides* and *W. lepida*) germinated at room temperature. Seedlings were divided into three explant types: root, hypocotyl and leaf to induce adventitious shoot formation. Surprisingly, both an auxin and a cytokinin are required for shoot multiplication, and this occurs simultaneously with root formation. Shoots were subcultured for multiplication and then propagated in a liquid-shake culture. Shoots regenerated from the liquid culture were used for corm induction experiments as well as rooting.

### Ecological and environmental impacts of bulk extraction from the Table Mountain Group (TMG) Aquifer System

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Faced with increasing water demand the City of Cape Town has begun investigating the potential of the TMG aquifer system as a means of increasing the cities water supply. Abstraction from the aquifer may result in the drying of seeps and wetlands. Vulnerability curves (a plot of percent loss conductivity versus water stress) can be used as a means of comparing the vulnerability of different species to increased drought stress. Vulnerability curves can be used to identify seep species that are most sensitive to water stress and therefore limited to areas that are permanently wet. Such species are most likely to be dependent on aquifer fed wetlands and therefore most at risk from abstraction. A method to identify ground water fed wetlands and the species most dependent on them is presented as a first step to assessing the potential impacts of abstraction from the TMG aquifer.

### The effect of plant growth regulators on somaclonal variation in Cavendish banana (*Musa* AAA cv. 'Zelig')

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The effect of the type and concentration of plant growth regulators and sub-culturing on somaclonal variation were studied in Cavendish banana cv. 'Zelig' obtained from African Biotechnologies Ltd., South Africa. *In vitro* grown plants at the fourth multiplication cycle were used for the investigation. Auxins (IAA, IBA and NAA) and cytokinins (BA and TDZ) were used to multiply shoots for ten generations. Bands generated through RAPD-PCR were scored according to whether they were present (1) or absent (0) to determine the extent of somaclonal variation. Results were then analysed using cluster analysis. The relationship between multiplication rate and somaclonal variation was assessed using correlation analysis. Results indicated that treatments with higher multiplication rates produced more variants; sometimes as high as 72%. Dwarf off-types accounted for 87.7% of the variation. A dwarf-specific band, about 1 500kb in size, was amplified by the primer OPC-15. The band appeared consistently in normal plants but was absent in all dwarf plants.

## The involvement of the lipoxygenase and cyclooxygenase pathways during the resistance response of wheat to the Russian wheat aphid

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Plants that are under attack from either pathogens or herbivores are known to undergo changes in the oxylipin signature. The processes leading up to oxylipin synthesis was studied during conditions of infestation with the Russian wheat aphid (RWA), *Diuraphis noxia* (Mordvilko) and non-infestation in the resistant ('Tugela DN') and the susceptible near-isogenic wheat cultivar ('Tugela'). The aim was to gain information leading up to oxylipin synthesis and ultimately resistance. Lipoxygenase (LOX), which catalyses the first step in oxylipin metabolism, was selectively induced in the resistant wheat after infestation with the RWA. An increase in lipid peroxidation coincided with the increase in LOX activity in the infested resistant wheat. Several lipid-like products were newly induced after infestation and the synthesis of other lipids was enhanced after infestation in the resistant wheat. Many of these lipid products were identified as hydroxyl- and keto-fatty acids. These are important signalling molecules during a defense response. A cyclooxygenase (COX) protein was selectively induced after infestation in the resistant wheat. This COX protein has homology to mammalian COX, which is responsible for prostaglandin biosynthesis. To understand to role of prostaglandin in plants under aphid attack, the biosynthesis of prostaglandins were inhibited with indomethacin. LOX and peroxidase (POD) activities were measured. Inhibition of prostaglandin biosynthesis resulted in a decrease in LOX and POD activities. The results emphasise the importance of two lipid biosynthesis pathways via LOX and COX that are essential for a successful defense response to sucking insects.

## The identification of a putative protein kinase gene involved in the early interaction between sunflower and leaf rust

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Sunflower (*Helianthus annuus*) is a commercially important crop plant. Susceptible plants infected with leaf rust (*Puccinia helianthi*) can lead to decreased yields. The aim of this study was therefore to identify protein kinase genes (from a resistant sunflower cultivar) that are involved in the resistance response when the plant is infected with leaf rust. Putative protein kinase genes were amplified using RT-PCR with primers specific for the conserved subdomain VIb of protein kinases. The cDNA fragments were cloned into a plasmid vector and the expression thereof confirmed using a reverse northern blot. One of the identified clones showed a 2½ fold induction of gene expression in infected resistant sunflower, while the expression in the susceptible infected cultivar was constitutive. When sequenced, the cloned cDNA fragment was found to encode a putative protein kinase gene. The induction profile of the gene was re-confirmed using RT-PCR in both infected susceptible plants and resistant plants.

## The Gulliver strategy in savanna trees

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Tree species in frequently burnt savannas face formidable recruitment problems. Seedlings have to acquire the ability to resprout very early to survive dry season fires. Saplings suffer repeated 'top-kill' (death of stems) and may be trapped for many years in the flame zone of grass-fuelled fires. Here we describe a distinctive life history strategy, the 'Gulliver' strategy, that seems to be common in many mesic savannas. This is the combination of

large woody storage organs below ground and a pole-like architecture above-ground which increases the probability of sapling escaping from the fire trap. Allocation patterns represent an intriguing problem for the plant which we explore in this talk.

## Plasmodesmal ultrastructural curiosities: Monocotyledonous leaves revisited

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Knowledge of plasmodesmal structure remains fundamental to efforts directed towards unravelling processes involved in cell to cell regulation of the symplasmic assimilate transport pathway in leaves. There are fundamental differences in anatomy and the related pathways followed by assimilates in C<sub>3</sub> and C<sub>4</sub> grasses and sedges. In Cyperaceae, there is an increase in the number of cell layers across which transport of assimilate to the phloem must occur. Up to three sheath layers have to be crossed in C<sub>3</sub> and C<sub>4</sub> species, before entry to the phloem complex is achieved via vascular parenchyma (VP) cells. Excluding the interface between the mesophyll (Mes) and the outermost sheath, these are the bundle sheath (BS), a mestome sheath (MS) and a border parenchymatous sheath (BP) along a BS>MS>BP and BS>MS>KMS>VP, in C<sub>3</sub> and C<sub>4</sub> species respectively. Apart from this complex array of sheaths, physical barriers to symplasmic transport, such as a compound middle lamella, that may or may not contain suberin and other waxy deposits, may be present along the pathway, and have to be. Where present suberization forces a symplasmic route through this barrier, via functional plasmodesmata. In regions of suberization, plasmodesmata are restricted to about 50nm total diameter, and the effective trafficking pathway to about 2nm. This paper explores aspects of plasmodesmal biology relative to the unique barriers and anatomies presented by C<sub>3</sub> and C<sub>4</sub> grasses and sedges.

## Functionality within plasmodesmata: roles for callose, actin and myosin?

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Plasmodesmata form unique microchannels that are involved in the transport (trafficking) of small and large molecules including virions, between living cells within all plants. This symplasmic transport system is known to be a finely-regulated one, in which various other constituents are involved. For example, calcium regulates the deposition of β-1, 3 glucan, (callose), which in turn, regulates the opening and closing of the apertures to the plasmodesmata. This gating process is also thought to be under the control of proteins such as actin, centrin, fimbrin and myosin as well. Even though these quasi-organelles have been widely studied, we are not yet able to characterise their functionality. This paper highlights intriguing aspects of the substructure of plasmodesmata in monocotyledons, and focuses particularly on the neck region of plasmodesmata which are intimately involved in movement across the various interfaces which exist from the mesophyll to the phloem tissue. It is this neck region which is implicated in control and regulation of the plasmodesmal orifice, gating them open or closed, or in the regulation the flow rate of materials through the confined space available for transport. Our particular interest is in the electron-dense and electron-lucent regions which exist within the neck regions of plasmodesmata, and regions of electron-dense material which occur outside of the neck region, as these are implicated in regulation, via sphincter-like activity. We explore the functional substructure of these regions of the plasmodesmata, using conventional fluorescence microscopy, histochemical techniques, and dual antibody systems, to localise callose (β-1, 3 glucan) and examine important movement-controlling constituents such as actin and myosin using fluorescently-tagged secondary antibodies.

## Regulation of sucrose accumulation in sugarcane: insight from metabolic studies and analysis of transgenic clones

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Previous work in sugarcane has shown that sucrose accumulation is characterised by a shift of carbon away from the insoluble matter and respiratory intermediates. Evidently this carbon shift is not a one-directional process, but involves a rather complex network of metabolic cycling within the stem parenchyma. This is illustrated by the failure of altering sucrose accumulation by down regulation of soluble and cell wall invertase, aldolase and enzymes involved in regulation for fructose-2,6-bisphosphate, and the success by reducing the levels of cytosolic pyrophosphate: D-fructose-6-phosphate 1-phosphotransferase (PFP, EC 2.7.1.90) activity in transgenic clones. Labelling work indicated that a cycle of sucrose synthesis and breakdown occurs, even in fully mature internodes. Data suggests that the rate of sucrose synthesis is similar between low and high storing genotypes but the hydrolysis of sucrose is three times higher in the low sucrose storing genotype. There are reasons to believe that the neutral invertase, rather than acid invertase, is important. In addition, kinetic modelling also indicates a fairly high metabolic flux coefficient (0.7) for the NI catalysed reaction step. Analysis of transgenic lines with reduced PFP expression indicated that this genetic change resulted in a major shift in the hexose-phosphate to triose-phosphate ratio in young internodes. Coinciding with the increased hexose-phosphate levels is a general increase in the levels of sugars. Interestingly, sugars such as galactose and maltose, which are normally undetectable in the sugarcane culm, are also significantly higher in the transgenic lines. This suggests that sugar levels, including sucrose, are dependent on the prevailing hexose-phosphate levels. The implication of these findings, and potential future strategies to enhance sucrose accumulation, will be discussed.

## The efficacy of outreach nurseries in enhancing the asset portfolios of community participants

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Outreach nurseries have been widely implemented internationally – with varying results – in attempts to achieve a range of natural resource management and socioeconomic goals. The Sustainable Livelihoods framework, which is based on five types of capital assets that people can accrue and/or draw on viz. financial, social, human, natural, and physical, is used to assess changes to the portfolios of community participants through outreach nurseries in South Africa. Ten outreach nurseries with a range of objectives, activities and settings were assessed in six provinces. Natural capital had increased in some projects through the greening of home gardens and communities, but there were limited improvements in medicinal plant nurseries, with harvesting levels remaining constant in two projects and increasing in a third due to beneficiation activities. Physical capital in the form of infrastructure had been enhanced in all but one project in which the institution had not provided promised support. However, problems were often experienced through lack of water and transport. At an individual level, financial capital decreased rather than improved over the project time period, with only 30% of the projects having started to generate sufficient funds to pay participants, and only 20% consistently. It had taken 5–10 years to reach this point, despite one project having received intensive institutional support and considerable funding over the five years of its existence. All but 30% of the projects depended strongly on external funding and

50% were still struggling to break even, with most still being subsidised through inputs such as water and 'free' labour. Human capital had grown in terms of environmental education and horticultural skills, but business skills were limited or lacking in all the projects. Within the community, social capital was sometimes increased as people built up linkages and trust. However, it was also diminished at different levels in all the projects through conflicts, vandalism, thefts, jealousies and other forms of erosion of social networks.

## A vegetation description of the farms Ingleside and Welgedacht of the Mountain Zebra National Park, Eastern Cape

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Because ecosystems react differently to different management practices, it is important that a description and classification of the vegetation of an area be done. As part of a vegetation survey programme for the newly acquired farms incorporated into the Mountain Zebra National Park, the vegetation of the Ingleside and Welgedacht sections were investigated. From a TWINSPLAN classification, refined by Braun-Blanquet procedures, 11 plant communities, which can be grouped into seven major groups, were identified. A total of four plant communities not previously described in the Park were identified. A classification and description of these communities, as well as a vegetation map, are presented. Descriptions of the plant communities include diagnostic species as well as prominent and less conspicuous species of the tree, shrub, herb and grass strata. This study proves that the extended land incorporated into the Park contributes to the biological diversity of the Park by adding more vegetation communities and habitat types to the Park.

## Isolation, characterisation and functional analysis of a ripening-related promoter from *Vitis vinifera* L. (grapevine) cv. Merlot

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A major stumbling block in the genetic manipulation of grapevine remains the availability of promoter to regulate transgene expression specifically in the fruit-tissue of post-*véraison* berries. This study focuses on the identification of a fruit-specific, ripening-related gene that is specifically expressed during the post-*véraison* stages of berry development, and the isolation and characterisation of the promoter element of this gene. Due to the low genetic transformation efficiency, slow regeneration (about 18 months) and long reproductive life cycle of grapevine, tobacco and strawberry was evaluated as alternative systems to verify the functionality and specificity of this promoter in stably transformed plants. To study promoter activity in a non-destructive manner, the putative promoter was fused to the green fluorescent protein (GFP) reporter gene. GFP expression analysis showed that in both tobacco and strawberry, the promoter was functional, tissue-specific and developmentally-regulated. In tobacco abundant levels of GFP were visualised in the nectary tissue of the developing flower, while GFP expression in transgenic strawberry plants were specific to the ripening fruit. These transgenic strawberries represent the first fruit tissue in which a promoter from grapevine can be evaluated.

## Assessment of water use by alien plants in selected Eastern Cape catchments

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Exotic invasive plants have been shown to severely affect mountain catchment areas by displacing indigenous vegetation and substantially reducing surface runoff. South Africa is a drought-prone, water-poor region with a mean annual rainfall of 502mm and a mean annual runoff of only 42mm. Consequently, it is imperative that its water supplies be managed efficiently. The Working for Water (WfW) Programme is a management strategy that where removal of water-consuming alien vegetation is aimed at restoring stream flow in rivers. Much work has been done of the benefits of alien removal in the Western Cape, but this study explores the case for such clearing in six selected mountain catchments in the Eastern and South-eastern Cape. The primary benefit is water yield. Water saving was calculated for areas cleared by WfW after completion of clearing. Water savings of between 500 000m<sup>3</sup> and 4 150 000m<sup>3</sup> were estimated for the first year after clearing. Integrated water saving over a 20-year period ranged from 2 million m<sup>3</sup> in the small catchments (such as Pott River and Kat River) to 1 300 million m<sup>3</sup> in the high rainfall Tsitsikamma catchment. This data was compiled with a non-water benefit profile and known costs provided by WfW into a cost-benefit analysis. Excluding non-water or ecosystem benefits, none of the catchments showed a cost benefit for the WfW Programme. Benefit cost ratios ranged from 0.03 to 0.75 with an overall of 0.59 (i.e. for every R1 spent on the WfW, 59c was gained in water and other quantifiable benefits). Improved management and a slightly higher price for water could make the Kouga and Tsitsikamma catchment Programmes beneficial.

## Effect of different bio-stimulants on the yield and quality of hydroponically grown tomatoes

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The use of bio-stimulants from plants, with the potential to increase yields, may be part of the solution to decrease the large amounts of inorganic fertilisers and pesticides used in modern agricultural practices. These bio-stimulants must not only promote growth and yield, but must also be biodegradable and environmental friendly. ComCat®, a new bio-stimulant, is currently extensively evaluated on different crops under different field conditions, both locally and internationally. It is not a fertiliser substitute, but a biological enhancer which apparently increases yield by enhancing vegetative- and root growth, improve flower bud formation and fruit development, induces resistance to pathogens and improve product quality without leaving residues in the crop. Moreover, another bio-stimulant with huge potential (Seed Suspension) was identified and isolated locally at the University of the Free State and is also extensively evaluated locally. It appears that these bio-stimulants have a marked impact on crop production under field trials. The aim of this investigation was to evaluate the yield improving potential of the above-mentioned bio-stimulants under optimum growth conditions where the crop is not exposed to various stress factors. For this reason the effect of both ComCat® and Seed Suspension on the quality and yield of hydroponically grown tomatoes was investigated. The results obtained will be discussed.

## Antimalarial diterpenoid from *Hyptis suaveolens*

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*Hyptis suaveolens* is a perennial herb found in dense clumps along roadsides, over-grazed pastures and around stockyards across the tropics. It is an erect, fast-growing herb with branched, semi-woody stems up to 2m tall with quadrate hairy stems. Petroleum ether crude extracts of the leaves showed high anti-malarial activity (IC<sub>50</sub> 2.5g/ml). Through bioassay-guided separation, a polar compound – identified as a diterpenoid – that displayed high antimalarial activity, was isolated.

## Ecology of urban areas: importance and application of vegetation studies

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Maintenance of civilisation in cities is still regarded as a battle against nature. Results of urban ecological studies have, however, resulted in a wider awareness of the importance of the preservation of urban biota and their habitats. Different definitions exist for the terms 'urban' and 'urban vegetation' depending on the research question. In our current scenario, 'urban' refers to all open areas within the limits of the city, and 'urban vegetation' include all types of spontaneous vegetation. Increasing urbanisation, including an increase in informal settlements on the urban fringe is a major threat to the preservation of biodiversity in South African cities. These trends of sub-urbanisation, have resulted in fragmentation and sprawling of cities. Natural vegetation in and around South African cities is destroyed at an alarming rate and immense areas of ecologically significant open spaces are cleared for persistent lateral growth. In this presentation an overview of projects conducted by the author and co-workers to gain more information on the ecology of urban areas, will be given. These projects involve basic studies such as phytosociology and vegetation dynamics as well as more applied studies such as biotope mapping. The mapping of urban biotopes is an attempt to put basic ecological information in a format that is easily accessible, understandable and applicable to managers and policy-makers. Additionally, preliminary results of studies on organic matter breakdown and plant functional types, following an urbanisation gradient approach will be given. Emphasis will also be placed on an integrated approach towards urban ecological studies which is the only way in which patterns and processes of urban and human-occupied ecosystems should be studied.

## Population biology and the effects of harvesting on *Pelargonium reniforme* populations Grahamstown and surrounding areas, Eastern Cape, South Africa

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*Pelargonium reniforme*, an East Cape near endemic plant has been harvested for use as a traditional medicinal plant for the treatment of respiratory infections amongst other ailments. Recent commercial interest in both *P. reniforme* and the closely related *P. sidoides* by pharmaceutical companies has led to an increase in the demand for the plant. The result has been an increase in the harvesting of both species. As *P. reniforme* is abundant around Grahamstown, and is being heavily harvested in the area, there is concern for the survival of the species. The species has been little studied. Any population information that could give an idea of the response of the plant to harvesting as well as general life history and population ecology is essential for making recommendations for managing harvesting. A study on the population biology of *Pelargonium reniforme* in and around Grahamstown, Eastern Cape, South Africa, was conducted. Six sites, three of which were previously harvested and three of which were unharvested, were studied. Five permanent quadrats in each of the sites were laid out. All of the *Pelargonium reniforme* plants in each of these quadrats were removed as entirely as possible. The plants were measured in order to determine whether there was a significant difference between both harvested and unharvested sites for plant number, total biomass and root fraction. Also, regression analyses determined relationships of above-ground measures of below-ground size, to make

it possible to study populations without having to dig plants up and potentially contributing to a sustainable harvest plan. Although harvesting decreased the total biomass of populations, there was no significant difference in root fraction between harvested and unharvested sites. The size of the subterranean tuber could not be effectively predicted by a number of above-ground measures although the size may be estimated from a certain point. Future studies will have to be conducted by digging up the plants if accurate tuber size information is needed. Pieces of tuber remaining in the soil do resprout as early as five months after harvesting, in some cases even increasing the number of shoots in the quadrat. Future study of the permanent quadrats will determine the rate of re-growth of any tuber pieces remaining after the harvesting event.

### The effects of aphid feeding on resistant and susceptible wheat varieties: resistance or tolerance?

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Development of crops that are resistant to insect pests has become increasingly important. Many of these crops are genetically engineered to be resistant to specific insects, and although the addition of certain genes has conferred resistance to certain crop species, the mechanisms of this resistance is often unknown. This study determined the effects of aphid feeding on resistant and susceptible varieties of wheat (*Triticum aestivum*); Betta and Betta Dn respectively. Wheat of each of the varieties as well as the control cultivar SST825 was grown with or without aphid colonies. Each of the experiments were examined specifically for the formation of wound callose using Aniline blue and fluorescence microscopy. Our results indicate that aphid feeding does stimulate the formation of wound callose in susceptible varieties, suggesting that the plant's response to aphid feeding is a long term, wounding response in which callose deposition plays a central role. However, callose formation is comparatively reduced in the resistant strains. Application of 5,6-carboxyfluorescein diacetate (5,6-CFDA) to attached leaves, examined 4 hours after application, showed little evidence of phloem transport of the cleaved and fluorescent 5,6-carboxyfluorescein (5,6-CF), below known aphid probed sieve tubes. Low levels or absence of 5,6-CF suggests that either the aphids have successfully redirected sap to themselves, or that the phloem is no longer functional. In Betta-Dn, 5,6-CF transport was still evident below sites of aphid probing, suggesting that the phloem was still capable of long-distance transport. Thus callose deposition is reduced and, as a result, transport not affected by aphid feeding in resistant wheat varieties. This indicates that the 'resistant' wheat variety may in fact be tolerant to aphid feeding by ignoring the nutrient drain that they impose on the system.

### *Syncarpha recurvata* (Asteraceae, vulnerable): phenology and conservation status

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*Syncarpha recurvata* (L. f.) B. Nord. is one of the most attractive everlastings in the Eastern Cape. It is an Eastern Cape endemic listed as 'vulnerable' with a range extending from the Maitlands River in the west to Alexandria in the east at elevations between 0 and 200m asl. Of the eight populations known to have existed only five remain, one of which is under immediate threat of extinction. Two further populations are under threat from *Acacia cyclops* infestations, while another is in imminent danger due to mining. A population not recorded before has been located in a conservation area. Two of the nine populations are not in immediate danger. Phenological patterns for populations within a 50km radius of Port Elizabeth varied substantially. Environmental factors controlling this are assessed. The plants reproduce by seeds and trial field sowing after mine rehabilitation has been successful. Field germi-

nation triggers are specific, but as yet unknown. Transplanting of young individuals was successful, but mature individuals did not transplant well. *S. recurvata* meets with 75% of the criteria that give a plant species a high conservation priority.

### Free State plants and their associated threats

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The Free State Province is a relatively flat area with a rainfall gradient that ranges from 250mm in the west to 750mm per annum in the east. Due to this broad rainfall gradient, relatively flat topography, gradual change in geological formations and soils, the habitat diversity is relatively low in comparison to other parts of the country. The vegetation units reflect broad transition zones between biomes. Of the 3 000 plant species present in the Free State relatively few are Red listed. The typical threats such as habitat destruction due to agricultural activities, urbanisation, road and dam construction, overgrazing and the subsequent erosion of topsoil, overexploitation of medicinal plant resources, etc. are also putting pressure on the plant species populations of the Free State. A study of the impact of urbanisation on a unique succulent dwarf shrub community in the 'Valley of the Seven Dams' near Bloemfontein has been conducted. A large number of protected species were noted. In light of the mounting pressure of urban expansion in the 'Valley of the Seven Dams' area near Bloemfontein, the conservation of the succulent dwarf shrub communities constitutes a high priority due to their habitat specificity, restricted distribution and high species richness.

### Getting communities involved in conservation and monitoring of threatened species

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Custodians of Rare and Endangered Wildflowers (CREW), a CEPF funded CAPE project, is one and half years into its implementation. The project focuses on involving civil society groups within priority areas of the CAPE lowlands to conserve threatened plants and habitats. We are working with 7 groups in 6 areas. The main project activities so far include getting civil society to:

- Work with local authorities to conserve critical sites.
- Surveying Renosterveld fragments for threatened plant populations specifically collecting data on threats to the populations, population size and area of extent.
- Building local awareness among landowners.
- Contributing to the management of conservation worthy sites.

CREW recognises the value of volunteer involvement and aims to facilitate their integration into conservation initiatives in the lowlands of the CFR.

### Investigation of anti-inflammatory activity and potential mutagenic effects of some useful trees in South African traditional medicine

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Aqueous, ethyl acetate and ethanolic extracts of different parts of 10 trees used in South African traditional medicine for treating ailments of inflammatory nature, were screened for anti-inflammatory activity using cyclooxygenase assays (COX-1 and COX-2) and investigated for potential mutagenic effects using the Ames test. With the exception of leaf extracts from *Trichilia emetica* and *Ficus sur*, all organic and aqueous extracts of the investigated plants showed considerable activity against COX-1, while 70% of these extracts inhibited both COX-1 and COX-2. Strong

inhibitory effects for both COX-1 (87–95%) and COX-2 (88–97%) activities, were obtained from different parts of *Acacia nilotica* subspecies *kraussiana*. Organic solvents inhibited COX enzymes most followed by a water extract. Exceptions were *Acacia* sp and *Faidherbia albida*. Non of the investigated plants showed any mutagenic effects in the *Salmonella* /microsome assay.

### Development and application of a technique to determine MIC values of plant extracts for facultative aerobic acidogenic oral bacteria

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During orthodontic treatment it is difficult to protect teeth against tooth decay bacteria and this frequently leads to white spots where the enamel has been decalcified or pink spots where chlorhexidine has coloured the teeth. Efforts are underway to search for alternative more efficient antibacterial compounds. Herbal preparations have been used to combat tooth decay bacteria. Agar diffusion methods used for assaying activity of plant extracts however, give poor results. We developed a method to evaluate the activity of tooth decay bacteria qualitatively by bioautography and quantitatively by a serial microplate dilution method in a carbon dioxide enriched environment. These techniques were applied to different extracts of five non-toxic widely used herbal medicines that have good activity against nosocomial bacteria from our earlier work and a plant species with little activity as negative control. Fluoride and chlorhexidine were used as positive controls. The bacteria selected for evaluation were: *Streptococcus mutans*, *Streptococcus sobrinus*, *Actinomyces naeslundii* and *Lactobacillus casei*. The bioautography technique did not work well with the lactobacillus but the in all other cases the techniques worked well. One of the extracts gave an activity substantially better than chlorhexidine. The use of this extract has now been patented. Studies are underway to isolate the antibacterial compound in this extract due to interest of an international company to develop this product. A useful technique for studying antibacterial activity of facultative anaerobic organisms led to this product.

### Sleep movements in *Oxalis* flowers

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For many *Oxalis* species flowering is timed to coincide with the onset of spring, bringing a profusion of blooms to the Namaqualand landscape. Species differ in the length of their flowering period; producing flowers asynchronously. *Oxalis* species exhibit a rhythm in flowering, opening fully between 10h00 and 15h00 and closing in the late afternoon. Not all species, however, open and close at the same time. This diurnal rhythm follows daily fluctuations in both light and temperature. Throughout the morning as light increases, flowers start to unfold, but close up for the night as light intensity decreases. And, while flowers open as the temperature increases to between 17 and 20°C, they start to close as temperatures drop. On cold days, however, when temperatures remain below 15°C, the flowers continue to sleep. Temperature provides an important environmental cue in regulating circadian rhythms. In controlled environments, flowers remain closed at a constant temperature of 15°C. But when plants are exposed to higher temperatures (25°C) flowers continue to open and close on a daily basis. The rhythm appears to alter, however, when plants are kept at a constant temperature of 20°C. Movements in other spring ephemerals like Namaqualand daisies are remarkably similar. Here too, it seems that temperature, rather than light, provides the cue for flowering. This pattern in diurnal flowering may be to protect pollen or, by housing potential pollinators, to increase their chances of receiving pollen.

### The impacts of climate change on *Aloe dichotoma* (the Kokerboom): Implications for the conservation and management of threatened plants

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Anthropogenic climate change has recently been identified as one of the greatest threats to the future of global biodiversity. Although a growing number of biological indicators of climate change have been detected, studies exploring impacts in the southern hemisphere, Africa, covering entire species ranges or of long-lived plants are rare. As atmospheric temperatures warm, the climatic 'envelopes' to which organisms are adapted shift toward the nearest pole and, where available, to higher altitudes. We examined population level mortality and recruitment in 53 *Aloe dichotoma* populations throughout the species' range. We present six pieces of evidence that clearly show that *Aloe dichotoma*'s mortality patterns are consistent with the early stages of a poleward range shift. Assessment of AFLP variation shows that resulting range loss will impact on those populations characterised by the highest levels of genetic variation. We discuss the implications of these findings for South Africa's threatened plants and explore conservation and management approaches for adapting to inevitable biodiversity changes ahead.

### Egg hatching success and life-cycle of the powdery stinkbug, *Atelocera raptoria* (Pentatomidae), on pistachio in South Africa

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Pistachio, *Pistacia vera* (Anacardiaceae), is an important new nut crop to South Africa. The powdery stinkbug, *Atelocera raptoria*, is a major pest on this crop, causing severe tissue necrosis to individual nuts on nut clusters. *A. raptoria* egg batches were sampled from pistachio trees at IDC – Green Valley Nuts (GVN), near Prieska in the Northern Cape Province. These egg batches were used in a variety of experiments aimed at developing a degree-day model for *A. raptoria* and included investigations into life-cycle strategies (with emphasis on the number of nymphal instars), egg-hatching success at fluctuating temperatures and egg parasitoid loads. Approximately 407 *A. raptoria* specimens of varying sizes were collected at GVN and these were used to determine the number of instars by modifying and applying Dayr's Rule as an analysis index. The influence of temperature on tolerance and hatching of eggs were conducted at temperatures ranging from 10°C to 40°C. After hatching, nymphs were removed and placed in terrariums to develop and time spans between moults up until adult eclosion were noted. Willow (*Salix babylonica*) leaves were provided for sustenance. Parasitoid loads of egg clusters were also counted and noted. The results show that there are 5 nymphal instars before adulthood is reached. High loads of the egg parasitoid, *Trissolcus basalis* (Scelionidae), were also recorded, especially at mid-range temperatures, whilst successful hatching also appeared to peak at mid-range temperatures. Approximate upper and lower tolerance thresholds could also be determined. The study is ongoing.

## Impacts on Wetlands in South Africa

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Despite having world class legislation protecting wetlands, South Africa's wetlands, including pans, vleis, and riverine systems, are in a very bad state. Primary impacts on wetlands are summarised as pollution, invasion, and destruction, and individually detailed, with destruction including both removal, development and conversion. Some wetlands are able to cope with some impacts while being severely affected by others. Examples are given of wetlands under stress or in danger, including pans in the Lake Chrissie panfields, and peatlands in Kosi Bay.

## Are nutrient-limited coastal dune plants better able to survive sand-burial by maintaining a low biomass?

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The growth of coastal foredune plants is affected by gradients of disturbance and low resource availability. Disturbance (salt spray, sand movement and mechanical damage) decreases with distance inland, while resources (particularly nutrients) remain uniformly low. Plant biomass increases and production decreases with distance inland, while changes in species composition results in visible zonation. We hypothesise that in continually disturbed environments with limited resources, high turnover and growth is required and that low biomass ensures an adequate supply of resources to sustain this growth. We have shown that the growth and biomass of plants inhabiting different zones in the dunes, respond differently to experimental nutrient additions, and hypothesise similar responses to sand additions. We suggest that foredune plant species may maintain a low biomass in order to have sufficient nutrients to respond to sand burial.

## The tree with too many names

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Describing 'natural' plants from garden specimens is fraught with the danger of making foolish errors. What if the garden records are in error or missing? We examine one such case. It has been suspected for some time that *Eugenia incerta* Dümmer, described from Durban Botanic Garden, is not African. Consideration of the anatomy of the plant and the probable historical circumstances of its introduction to the Garden led to a search of similar species in English-speaking countries of the Caribbean area. Phenetic studies show that the name *E. incerta* is, in fact, a synonym of *E. oerstediana* Berg, and that the Durban plant probably comes from Belize. Historical studies show a very plausible train of events by which this could have occurred.

## Spatial distribution of *Isoglossa woodii* and its effects on subtropical coastal dune forest regeneration

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*Isoglossa woodii* is a dominant understorey herb in subtropical coastal dune forests. High levels of competition from *I. woodii* may limit recruitment of tree seedlings in these habitats. We examined spatial patterns of *I. woodii* cover in relation to topography, soil fertility, and canopy vegetation and found that *I. woodii* covered

65–95% of the forest understorey. The occurrence of *I. woodii* was not associated with differences in soil fertility or tree community composition but did vary according to topographic position and tree density. Reduced height of *I. woodii* occurred in areas with high tree density and a binary logistic regression model found that tree density was a better predictor of *I. woodii* cover than topographic position. We assessed the effects of *I. woodii* cover on early and advanced regeneration and found that seedling number and seedling species diversity were reduced under *I. woodii* but there were no apparent effects of *I. woodii* on the sapling community. Effects recorded at the early regeneration stage were likely due to differences in light availability, which was significantly reduced under *I. woodii* cover. Given the abundance of *I. woodii* in the understorey, this species may play a critical role in coastal dune forest regeneration by limiting the tree community to species that are either extremely shade-tolerant at the seedling stage or that can survive through the persistence niche by producing multiple stems.

## Ozone is a strong localised stressor in plants that induces systemic resistance

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Plants have evolved a number of strategies to defend themselves against various stressors and some of these defence mechanisms are inducible. The expression of resistance could be local or of the entire plant, where the latter responses are referred to as systemic acquired resistance (SAR). Ozone cause mainly oxidative stress in plants and resembles pathogen-like stress reactions. Since ozone is a major air pollutant in the troposphere and major crop losses have been attributed to critical levels being exceeded in industrialised countries, we investigated chlorophyll fluorescence and the photochemical response of several plant species to ozone stress with the aim to determine the extent of the damage, and whether the injuries and responses are only local or also systemic. Should systemic priming occur this could be important to curb the potential damage that ozone can cause to natural and crop vegetation. Our findings are the first to indicate that ozone is a strong local stressor, but that definite systemic priming occurs.

## The use of remote sensing sensors as a monitoring tool for rehabilitated wetlands

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Millions of rands are spent every year on wetland rehabilitation projects so it is of great importance to measure their success. To do this it is crucial to determine the most cost-effective procedure for monitoring rehabilitated wetlands. The aim of this study was to evaluate various types of remotely-sensed imagery to detect rehabilitation structures and remote sensing-based indicators that were identified for the purpose of monitoring rehabilitated wetlands. Various airborne and satellite-based sensors were evaluated. Ten of the eleven selected indicators could be derived from remotely-sensed imagery, the exception being water quality. Issues related to mapping these indicators are: the optimum time of year, the bands required and the spatial resolution to produce accurate maps versus the cost of data and time to process the data. The best results were obtained from high spectral resolution images. For mapping vegetation, multispectral data with bands ranging from green to near infrared (0.52 to 0.90:μm) was found to be of great importance to highlight vegetation activity. In order to monitor wetland vegetation over a long-term period, the comparable images must represent the same season but from different years. The choice between the different remote sensing sensors depends largely on the application of the sensor, state of the rehabilitation structure and the vegetation response to the rehabilitation measures. This study gave a general overview of the different sensors, their capabilities and limitations, as



well as highlighting their benefits and effectiveness as a monitoring tool. The results have value in formalising the procedure for monitoring the biophysical conditions, wetland utilisation and structural rehabilitation work of rehabilitated wetlands. Indicators must, however, be monitored over time. It is recommended that future studies include the analysis of vegetation dynamics linked with the hydrology to investigate the change in wetland vegetation after rehabilitation.

### The role of spiders as predators of insect pests on pistachio in South Africa, with special emphasis on *Heliophanus pistaciae* (Araneae: Salticidae)

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Pistachio nuts are a new crop under development in the Prieska district of the Northern Cape Province, South Africa. As part of a larger biomonitoring program of the arthropod fauna of the orchards, a study was conducted to determine the community composition of spiders (Arachnida: Araneae) in three orchards of different ages. The tree canopy fauna was sampled using insecticide knockdown from January 2000 to December 2001. The ground cover fauna was sampled using a sweep net, and the ground-dwelling fauna using pitfall traps, from July 2000 to July 2001. Spiders were abundant in all three strata sampled, and may constitute the most abundant and diverse order of generalist predators in the tree canopies and ground covers. Qualitative field observations on three species, namely *Heliophanus pistaciae* Wesolowska (Salticidae), *Neoscona subfusca* (C.L. Koch) (Araneidae) and *Cheiracanthium furculatum* Karsch (Mituridae), showed that these species consumed a broad spectrum of pest prey, including minor pest false cinch bugs, aphids and leafhoppers. A focused study on the predation of the false cinch bug *Nysius natalensis* Evans (Hemiptera: Lygaeidae) by *H. pistaciae* was carried out in laboratory and field trials. Results of feeding tests comparing predation rates of female and male *H. pistaciae* on *N. natalensis* and vinegar flies *Drosophila melanogaster* Meigen (Drosophilidae) are presented. Various factors affect predation rate, including spider sex and size, prey size and mobility, and prey palatability. Field trials found female *H. pistaciae* ( $n = 20$ ) to kill a mean of 1.05 *N. natalensis* in a 24-hour period. The experiments indicate that *H. pistaciae* may have a restricted role as a biological control agent of *N. natalensis*, but a combination of predation and effective orchard management should be able to suppress *N. natalensis* populations below damaging levels in pistachio canopies.

### Molecular physiology of thermoinhibition in *Tagetes minuta* L. (Kakibos)

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Achenes of *Tagetes minuta* become thermoinhibited when imbibed at temperatures in excess of 35°C. This process appears to be actively controlled by the embryonic axis. Previous research has shown that a small subset of thermoinhibition-specific proteins is expressed under thermoinhibitory conditions. Differential display was used to isolate genes which are expressed specifically in thermoinhibited achenes of *Tagetes minuta* L. Thermoinhibition-specific cDNAs were cloned and their specificity confirmed by reverse-northern hybridisation analysis prior to sequencing. Electronic homology searches have thus far resulted in the putative identification of ten of the cDNA sequences obtained. Of these, four may play a role in the control of thermoinhibition, whilst the others probably serve a protective function in the embryo under the heat stress conditions which accompany thermoinhibition. These results are consistent with the hypothesis that thermoinhibition in *T. minuta* achenes is under positive genetic control, in a manner analogous to embryo dormancy.

### A taxonomic revision of the genus *Gazania* (Asteraceae: Gorteriinae)

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*Gazania* is a small genus of the subtribe Gorteriinae, tribe Arctoteae, that contains 16 species. The genus was last revised in 1959 by Roessler, who noted that 'the formation and delimitation of the species [of *Gazania* can be] extraordinarily difficult'. These small herbs and subshrubs are popular with horticulturalists and gardeners due to the large size and bright colour of their flowers, as well as the ease with which they hybridise to generate novel character combinations. Other molecular studies have indicated that *Gazania* is monophyletic, but embedded within a paraphyletic *Hirpicium*. Here we report on a molecular phylogenetic study of the genus using both nuclear and chloroplast DNA sequence data to determine species relationships and limits.

### The importance of economic analysis in new crop development

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The globalisation of agriculture combined with increasing costs of production and regulatory pressures have influenced profit margins for many agricultural commodities. Therefore, farm operators generally are constantly searching for new or alternative crops that will help them diversify their crop mix and improve their revenue stream. Although other considerations (ecological or political) are important in determining the eligibility of crops, a new crop irrespective of its scientific achievements will fail unless the returns for producing it are greater than the costs involved. The development of new crops carries large risks if the producer is ignorant of production and marketing issues. Production costs and returns studies, assessments of consumer preferences, assessments of marketing and distribution channels and analysis of alternative returns across these channels, as well as production and market risks analysis are all necessary to inform the decision-making in new crop development. The outcome of such analysis will determine whether investments in new crop development will culminate in improved income to producers, lower and higher value commodities to consumers, and added wealth to investors.

### Evidence for a CO<sub>2</sub> controlled mechanism to explain tree invasion of African savannas and adjacent grasslands

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A widespread increase in woody plant cover has been reported since the LGM, and interpreted as a response to a moistening climate, especially in savanna ecosystems in Africa. However, atmospheric CO<sub>2</sub> rose from levels that severely constrain C<sub>3</sub> photosynthesis over a similar time period. Here we test experimentally if the impacts of LGM to 5 x LGM CO<sub>2</sub> levels on photosynthesis translate to changes in carbon allocation and growth rate in woody plants, and we model how these allocation changes might alter woody plant dominance in fire-dependent mixed tree-grass systems. Experimental results for three woody species revealed growth and photosynthetic stimulation accompanied by reduced stomatal conductance, increased water use and nutrient use efficiency at increasing CO<sub>2</sub>. In *A. karoo* and *A. nilotica* especially, root mass and carbohydrate content of roots increased in trees grown at high CO<sub>2</sub> levels, facilitating a significant increase in

resprouting after simulated fire or grazing damage. Higher CO<sub>2</sub> levels were also associated with greater investment in physical and chemical defenses in *A. karoo*. Greater resprout rates drove modeled increases in tree dominance in mixed tree-grass ecosystems, which match paleorecords for a South African savanna. These findings suggest a positive response by trees to increasing atmospheric CO<sub>2</sub> since the last glacial maximum, and suggest CO<sub>2</sub>, rather than climate change *per se*, has and will play a significant global role in woody plant success.

### The African plant checklist and database project

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The African Plant Checklist and Database Project (APCD) is a joint project between the National Herbarium, Pretoria (PRE) and Conservatoire et Jardin Botaniques de la Ville de Genève (CJB), Switzerland. The aim of the project is to combine the existing datasets of Flora of Southern Africa (FSA) and Enumération des plantes à fleurs d'Afrique tropicale (EPFAT) to produce an angiosperm checklist for sub-Saharan Africa. Merging the two datasets is not a straightforward process and there are many discrepancies between the two lists. It is important to bear in mind that the aim of the APCD is not to solve taxonomic problems, but rather to highlight discrepancies, providing useful information about problematic taxa where revision and further study is necessary. The end product of the APCD project will be a hardcopy checklist, an electronic version accessible through the Internet, and possibly a CD. The APCD is a first important step towards a more comprehensive database that can include other information such as maps, illustrations, ecological information, etc. Having an updated index of current names and synonyms and exact information on the number of angiosperm taxa for the African flora South of the Sahara will enhance, amongst others, conservation and biodiversity management on the continent.

### Understanding and influencing starch metabolism

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Starch is an important transient carbohydrate store in many different plant organs. It is also extracted from plants and used in a range of industrial applications. We have tried to further our understanding on the regulation of starch turnover, as well as factors determining starch structure and functionality in order to optimise crop yield and quality or to adapt starch functionality to make post-harvest treatments obsolete. We have achieved this through reverse genetic approaches when we downregulated the expression of genes encoding starch biosynthetic enzymes using antisense RNAs or RNAi technology. Starch is composed of linear  $\alpha$ -1,4 glucans (amylose) as well as  $\alpha$ -1,4 glucans that contain ca. 4% of  $\alpha$ -1,6 glucosidic branchpoints (amylopectin). Amylopectin in addition also contains covalently bound phosphate that resides either at the C-6 or C-3 position of the glucose monomers. It has a remarkably ordered structure, as its branches are arranged in clusters that give crystallinity to the polysaccharide. The biosynthesis of starch proceeds from ADP-glucose that is utilised by different isoforms of the starch synthases to produce linear  $\alpha$ -1,4 glucans. These glucans are randomly branched by branching enzymes and subsequently trimmed by debranching enzymes in order to establish the clusters. Other enzymes such as hydrolases, transglucosidases, or dikinases are also involved in determining starch structure. When the carbohydrates that are stored

within starch are needed for metabolism, starch is mobilised through the combined action of different hydrolases and glucosyltransferases. Different approaches to influence starch metabolism through manipulating different enzymatic activities will be discussed with respect to the effects on starch quantity, quality, as well as turnover will be discussed.

### Smoke as a promoter of seedling vigour in medicinal plants

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The use of smoke to germinate the seeds of species from the fire climax areas, such as California (chaparral), Australia (kwongan) and South Africa (fynbos) has generated a wide interest in smoke-stimulated seed germination. Research has shown that, in general, species from fire-prone areas respond to a germination cue present in smoke. Smoke-stimulated seed germination is not limited to species from regions which experience regular fires as certain species from fire-free regions also respond to smoke treatments. In the present study, an attempt has been made to examine the effect of aerosol and aqueous smoke on seed germination and seedling vigour of three South African indigenous medicinal plants – *Albuca pachychlamys*, *Merwillia natalensis* and *Tulbaghia violacea*. Although no significant ( $P < 0.05$ ) improvement in germination was recorded, the seeds of all three species subjected to the aerosol smoke and smoke extract solutions showed a high seedling vigour index. *Albuca pachychlamys* and *T. violacea* seedlings which were germinated and grown together with a diluted smoke solution showed significantly ( $P < 0.05$ ) higher seedling growth than untreated seedlings. Seeds exposed to aerosol smoke exhibited higher seedling survival percentages for *A. pachychlamys* and *T. violacea* than non-smoked seeds, while no significant effect was observed for *M. natalensis* seedlings. This study indicates that although smoke treatment may not necessarily have an effect on germination, it may have an effect on post-germination processes. Consequently, this investigation shows that the application of smoke technology can be adopted to produce seedlings with improved vigour.

### Fruit and feed characteristics of the most commonly planted cactus pear [*Opuntia ficus-indica* (L.) Mill.] cultivars in South Africa

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In South Africa, cactus pear is grown mainly for fruit production. The cladodes are utilised as a by-product for animal feed. Selection of cultivars with good fruit- and animal fodder potential would assist in spreading risk, especially for farmers in semi-rural areas. The aim of this study was to determine fruit and cladode characteristics of the 10 most commonly grown cactus pear cultivars in South Africa under rain-fed, subtropical conditions, to identify the best dual type cultivar. The cultivars were evaluated for plant- and fruit characteristics, as well as chemical characteristics of cladodes that are related to feed potential. Skinners Court favoured vegetative growth, while Turpin and Meyers were good fruit producers with poor feed characteristics. The fruit of Gymno Carpo had a very low total soluble sugars (TSS) content which makes it undesirable for human consumption. Roedtan had a cladode yield above average, while the feed characteristics of the cladodes ranked very high. It also had a good fruit yield and fruit mass, and very important, a very good TSS. It can therefore be used as a dual purpose cultivar.

## ***Drimiopsis*, *Resnova* and *Ledebouria*: the case for three genera**

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Provisional results of a cladistic analysis of *Drimiopsis* and sister taxa in the Hyacinthaceae, based on morphological data, as well as a small scale total evidence analysis, produce results contrary to those recently published based solely on DNA data. Whereas the authors of the latter analysis have jumped to taxonomic conclusions in sinking *Drimiopsis* and *Resnova* in *Ledebouria* due to a lack of hierarchy in their data, our results suggest three well defined clades corresponding to the mentioned genera. A total evidence analysis on a subset of taxa and combined morphological and DNA data result in trees congruent to the morphological data. Apart from the better known characters pertaining to the Ledebouriinae, we report on a number of hitherto uncoded characters and diagnose their distribution on resultant trees. We conclude that it is as yet ill-advised to adopt the newly proposed classification system.

## **Survey of utilisation and nutritional value of indigenous green leafy vegetables consumed by the Basotho; In search of new crops**

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The country Lesotho has large areas of low agricultural potential, so that food security is problematic. In a survey it was found that edible wild plants are still crucial sources of food in Africa, not only in the pre-harvest, so-called 'hungry season', but also because they are traditionally desired foods. It is mainly the rural people of Lesotho that make use of these indigenous plants as food sources, because they have easier access to the sources, and because they have not lost the custom of the usage. Due to seasonal variation in maturation, some plants are always available throughout the year. Lesotho bears many types of edible plants, but the nutritional properties are mostly unknown. Some of the most used plants were analysed for the nutritional composition, and it was found that the nutritional status is similar, or even higher, than that of many exotic vegetables. Furthermore, the optimal harvest time is important, and the nutritional value is dependent on the locality where the plants grow. The greatest advantage of indigenous plants is that they are better adapted to the harsh climatic conditions of the area and resistant to pests. Depending on the biomass production, at least two plants could possibly be considered for development as cash crops.

## **Commercial trade in medical plants in Maseru markets, Lesotho**

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Medicinal plants play an important role in the health care system in most developing countries. In many of these, especially in sub-Saharan Africa, poverty, unemployment and the scourge of HIV/AIDS are high. Although there is high prevalence of opportunistic infections associated with the high incidence of HIV/AIDS, many households are unable to access medical services provided by hospitals and clinics because of cost. Lesotho is no exception to this problem, with an estimated HIV/AIDS infection rate at 28.9% (ages 15–49), under-five mortality rate at 87 per 1 000 live births and new born mortality rate at 64 per live births (UNDP, 2004). The solution for many households is to resort to natural resources that are found within the commonage. As communal resources, they are easily accessed with no rules of exclusion. This regrettably engenders overexploitation which threatens the very same livelihoods in the long run. Because of demand, unreg-

ulated commercial markets for medicinal plants have developed in almost all major urban towns in Lesotho. The aim of our study was to make an inventory of species traded in Maseru markets, estimate the amounts sold and income derived by the sellers. We found 42 sellers in Maseru alone. We surveyed 30 of those representing 71% of the entire sample. A semi-structured questionnaire was used and a digital balance was used to measure quantities sold per unit price in order to match amounts sold to revenue obtained per species. A total of 151 species were traded in the markets representing 36 families. The commonest families exploited were Asteraceae, Asphodelaceae and Fabaceae. The commonest species were *Aloe ferox*, *Hypoxis hemerocallidea*, *Dicoma anomala* and *Bulbine narcissifolia*. For most species, the subterranean material was used. Income derived ranged from R250–R970 per seller per month. The study also documented the various ailments for which these plants are used.

## **Size class distribution, use and regeneration of *Leucosidea sericea* (Rosaceae) in protected and communal areas in Tsehlanyane, Lesotho**

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This study investigated the availability, spatial distribution, ecological status, regeneration and use of *Leucosidea sericea* Eckl. & Zeyh. in Tsehlanyane, Lesotho. *L. sericea* is a small frost resistant evergreen tree that produces soft durable wood that burns slowly and generally occurs in fairly homogenous stands. The Tsehlanyane forest was divided into two parts in the 1990s as a result of the activities of the Lesotho Highlands Water Project (LHWP). One portion was protected from continued human harvesting for subsistence needs while the other was left in the communal domain for continued use. Four monitoring plots (two in a communal area and two in a protected area) were established in 2000, and increased to 10 for 2001–2002 (five in each area). Both stands (communal and protected) displayed inverse J-distribution curves in basal stem diameter, indicating continued recruitment and hence potentially stable populations. Density of live trees was higher in the protected area ( $23\,575 \pm 1\,425$  trees ha<sup>-1</sup>) than in the communal area ( $12\,150 \pm 1\,075$  trees ha<sup>-1</sup>). Density of stumps was higher in the communal area ( $4\,950 \pm 425$  stumps ha<sup>-1</sup>) than in the protected area ( $825 \pm 200$  stumps ha<sup>-1</sup>). The protected stand had a wide range of basal stem diameter sizes while the communal stand had few trees >10cm in diameter. *L. sericea* resprouts vigorously with ≥83% of stumps having resprouted in the protected area and ≥94% in the communal area over the study period. Standing biomass in the protected area was higher ( $99\,138 \pm 1\,014$  kg ha<sup>-1</sup>) than in the communal area ( $62\,270 \pm 874$  kg ha<sup>-1</sup>). However, annual stem diameter growth rates were higher in the communal area ( $0.52 \pm 0.24$  cm yr<sup>-1</sup>) than in the protected area ( $0.49 \pm 0.13$  cm yr<sup>-1</sup>), but more biomass was accumulated in the protected area ( $1\,015 \pm 69$  kg ha<sup>-1</sup> yr<sup>-1</sup>) than in the communal area ( $516 \pm 32$  kg ha<sup>-1</sup> yr<sup>-1</sup>). A mean biomass of  $1\,521 \pm 92$  kg ha<sup>-1</sup> yr<sup>-1</sup> was harvested in the communal area, resulting in an over-harvest of  $1\,005$  kg ha<sup>-1</sup> yr<sup>-1</sup>. The increase in biomass, as well as density in the protected area, indicates positive growth of the stock, while the communal stand has a negative trajectory in both biomass and density, raising concerns about sustainability and availability in the future.

## **Variation in the rooting pattern of *Terminalia sericea* (Combretaceae)**

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An investigation into the roots of *Terminalia sericea* revealed variation in their rooting pattern. *Terminalia sericea* found in ecosystems of relatively low precipitation formed lateral woody roots of length varying from 3m to 8m, whilst those in relatively high precipitation ecosystems were found forming woody tap roots of length that varied from 0.5m to 1.5m.

### An investigation into the antibacterial activity of *Maytenus senegalensis*

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*Maytenus senegalensis* (Lam.) Exell. is widely used in traditional medicine for a variety of ailments. Due to its popularity in traditional medicine and coupled with the destruction of habitat, populations of *M. senegalensis* are reportedly on the decline in Eastern Africa. Stem bark extracts of *M. senegalensis* have been identified as having antibacterial activity. The work presented here focuses on the attempted isolation and identification of compounds responsible for the observed antibacterial activity. Root cultures of *M. senegalensis* were also established and the media found to exhibit antibacterial activity after 3 weeks of culture growth. This raised the question as to the nature of those compounds responsible for the observed antibacterial activity and the feasibility of root cultures being used as a possible alternative source of antibacterial compounds. These questions are addressed herein.

### Sanitation as a tactic in holistic plant health management of cactus pear orchards in South Africa: A case study

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Cactus pear, *Opuntia ficus-indica* (Cactaceae), is an important dual purpose new crop in South Africa that is utilised for animal fodder, as well as for food and beverage products. Ever since this crop started receiving wider recognition amongst farmers, there has been an increasing demand for the development of pest and disease management strategies. Hence, this study, which forms part of a larger Holistic Plant Health Management program on cactus pear in South Africa. Samples of fallen rotting cactus pear cladodes, with soil samples below each fallen cladode, were taken weekly over a 14 week period in three different orchards on the farm Glen de Arg near Bloemfontein in the Free State Province. Samples of the fallen fermenting fruit were also taken, but only for three weeks, due to limited availability. A total of 626 insects of 27 morpho-species and 6 orders were collected from fermenting fruit. From the rotting cladodes and soil samples a total of 1 552 insects of 65 morpho-species and 10 orders were recorded. In a separate study numerous fungal pathogens have been isolated that adversely effect the health of cactus pear plants, whilst other species have been recorded that cause fruit deterioration. Insect species that are confirmed vectors of many of these pathogens were identified from the mentioned datasets. These include vinegar flies (Diptera: Drosophilidae: *Drosophila melanogaster* and *D. hydei*) and dried fruit beetles (Coleoptera: Nitidulidae: *Carpophilus hemipterus* and *Urophorus humeralis*). Important in a different sense, is cochineal soft scale (Homoptera: Dactylopiidae: *Dactylopius opuntiae*), that survive well on fallen cladodes and can thus re-infect the crop under these conditions. The results of this study emphasise the importance of efficient and regular orchard sanitation in the management of insect and fungal pest species in agroecosystems.

### The morphology and anatomy of the genus *Arctopus* (Apiaceae)

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*Arctopus* is an anomalous genus of the family Apiaceae, endemic to the Cape region of South Africa. As a first step towards a revision of this genus a survey of morphological and anatomical characters was done. The three species differ mainly in their reproductive morphologies and can easily be distinguished by the involucre bracts that surround the female pseudo-anther. The hitherto poorly known *A. dregei* can also be identified by the ciliate, rather than spinescent leaf margins, which are found in the well known *A. echinatus* and *A. monacanthus*. The similarities and differences in the anatomy of the fruit and leaves (petiole, midrib and lamina) are reported for the first time.

### The anatomical and morphological investigation on *Salvia stenophylla* (Lamiaceae) and its tissue culture propagation

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*Salvia* is the one of the well-recognised genera within the Lamiaceae family. Most of its species are widespread and – as a result of their commercial value – are documented in the pharmacopoeias of many countries throughout the world. However, some species of this genus need to be domesticated and promoted as alternative crops after value addition. *Salvia stenophylla* belongs to this category. It is an odorous herb which is widely distributed in areas of the Eastern Cape, KwaZulu/Natal and Gauteng province. It is regarded as one of the plants species that contains a high percentage of α-bisabolol and manool in their essential oil product. It is, therefore, important to conduct research on value addition and optimisation of precious secondary metabolites of *S. stenophylla*. As preliminary work to this objective, anatomical investigation and tissue culture propagation of this species will be discussed in this paper.

### Population structure, genetic diversity and conservation of two endemic *Barleria* species, *B. argillicola* and *B. greenii* (Acanthaceae)

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Allozyme variation and differentiation, pollination biology, management strategies and red data assessment were studied in two *Barleria* species (*B. argillicola* and *B. greenii*) restricted to the midlands of KwaZulu-Natal at Estcourt. These two species of *Barleria* are sympatric although ecologically separated in different habitats. Their populations occur in areas with different fire and grazing regimes; a two year burning cycle on privately owned land and four year burning cycle in the Weenen Game Reserve. Livestock grazes on the privately owned land but there are only very low stocking rates in the Nature Reserve. The proposed Gongolo Reserve will cover most areas in the privately owned land. Allozyme variation and differentiation was studied using starch gel electrophoresis. The relationship between the observed levels of allozyme diversity and the mating systems is discussed. The effects of the management regimes were studied in eight 15m by 15m quadrats from three populations in the four year burning cycle, three populations in the two year burning cycle and one population from the annual burn in *B. greenii*. In *B. argillicola*, three populations from eroded areas and two from the non-eroded flat area were studied. The study suggested that inbreeding is more favoured in *B. argillicola* than in *B. greenii*, possibly as a result of occurring in very fragmented habitats, eroded areas, low genetic diversity, being visited by a variety of insects and existence of anthers and stigmas at the same level during flower opening. The level of biodiversity seems to be maintained by two different burning regimes: four year burning cycle with no grazing favours *B. argillicola* whereas *B. greenii* seems to favour a two year burning cycle with grazing. It is recommended that conservation measures should favour *B. argillicola* because it is more threatened than *B. greenii*.

## The genus *Salvia*: emerging as a model for medicinal plant biotechnology

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*Salvia* species as targets for medicinal plant biotechnology have been limited to species growing in China, where extensive research on this genus has been conducted. Those species endemic to Africa, particularly southern Africa, have been neglected for study in this manner. Even though the transformation of these medicinal plants into a high value commodity and increasing destructive harvesting has a potential to lead to rapid decline in populations in the wild in regions where these species are distributed, little to no attention has been paid to their conservation. The limited phytochemical analysis performed on the southern African species has revealed that some species produce essential oils that are potentially useful for the essential oils industry, as well as alkaloids and other secondary metabolites. It is therefore important to increase the research efforts on South African *Salvia* species for pharmaceutical, cosmetic and food utilisation. This study aimed at evaluating whether the response of the other members of the same genus *in vitro*, which has proven ideal or has ideally met expectations, would be mimicked by the African species. The ease-of-establishment of cultures of *S. africana-lutea* concomitant with the rapid production of putative transgenic roots from germinating seedling has indicated the potential for this species to become a model plant for ethnomedicinal-biotechnological studies in our laboratories. From our research effort, it may be concluded that *S. africana-lutea* is highly regenerative in tissue culture and is amenable to genetic transformation by *Agrobacterium rhizogenes*, displaying a typical 'hairy root' phenotype upon gene transfer.

## How savanna grasses decompose: photodegradation versus microbial decomposition

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Decomposition studies typically use litterbags placed on the soil. The fact that decomposition starts while plant litter is still standing in the field, experiencing breakdown by the sun, has seldom been considered. Here I report a comparative study of rates, and correlates, of decomposition of ten species of savanna grasses in the sun and on the soil. Rates of photodegradation of grass litter were significantly slower than microbial breakdown. The variables that best predicted decomposition in the sun were also different from those that determined decomposition on the soil. These results have important implications for which grasses accumulate fuel, standing dead litter, in savannas.

## Isolation and identification of bioactive compounds from *Indigofera daleoides*

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*Indigofera daleoides*, a herb belonging to the Fabaceae family, is often used by traditional healers for the treatment of diarrhoea in the Limpopo Province, South Africa. The present phytochemical study was carried out to isolate and identify the possible bioactive compounds in this species. The ethanol extract of the whole plant was partitioned with hexane, ethylacetate and butanol separately and these phases were spotted on thin layer chromatography (TLC) plates, eluted with ethylacetate: methanol (9:1) and sprayed with a 24 hour old bacterial culture of *Staphylococcus aureus*. Zones of inhibition in the ethylacetate fraction indicated good antibacterial activity and this fraction was therefore further purified on a silica gel column. Semi-pure fractions obtained from this column

were subjected to Sephadex column chromatography which resulted in the isolation of two pure compounds. The chemical structures of these compounds and their bioactivity against diarrhoeal-causative microorganisms (*Vibrio cholera*, *Escherichia coli*, *Staphylococcus aureus*, *Shigella* spp, and *Salmonella typhi*) will be reported.

## Identification of bioactive compounds from *Pelargonium sidoides*

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*Pelargonium sidoides*, a species belonging to the family Geraniaceae, is known for the aromatic and herbal properties. South Africans and some Europeans use the roots of this plant to treat wound-infection, diarrhoea, stomach problems, coughs, colds and respiratory problems. A bioassay guided fractionation of the ethanol extract of the roots of this plant led to the isolation of two coumarins, namely 'Umckalin' and 'Scopoletin'. Fresh root material of *P. sidoides* was homogenised with ethanol, followed by liquid partitioning with ethyl acetate and butanol. Butanol extract was subjected to sephadex column chromatography twice using 100% methanol as eluent. Semipure fractions obtained from the sephadex column were then subjected to silica gel column chromatography and lastly compounds were purified using preparative thin layer chromatographic plates (TLC). The identification of the pure compounds, umckalin and scopoletin was confirmed using their <sup>1</sup>H-NMR and <sup>13</sup>C-NMR spectral data. Both compounds were spotted on a TLC plates and eluted with 10% methanol in ethylacetate. The developed TLC plates were sprayed with 24hrs old culture of *Bacillus cereus* and *Pseudomonas aeruginosa*. Only umckalin, indicated antibacterial activity in a thin layer bioautographic assay. The antituberculous activity of purified compounds will be reported.

## Vegetation and habitat types of the Umkhanyakude Node, KwaZulu-Natal, South Africa

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The aim of this study was to identify and classify the woodlands, shrublands and grasslands of the Umkhanyakude Integrated Sustainable Rural Development Node (also known as Maputuland) into broad management units to assist with the natural resource audit and monitoring programme initiated by the Department of Agriculture. The Node is situated in KwaZulu-Natal Province at the extreme northern border between South Africa and Mozambique. Following 225 surveys, conducted during March–July 2002, the ecological data were analysed using multivariate ordination and classification techniques. Four broad plant communities within two geographically separated vegetation types were described. The Coastal Sandveld occurs on the coastal plain on recent arenaceous sediments. The Clay Thornveld occurs in the interior and is associated with a variety of terrain types and geological formations but predominantly rhyolite, basalt, shale and mudstones. Differentiation of communities within the major vegetation types was mainly based on climate (temperature and precipitation differences associated with differences in elevation and topography) and anthropogenic disturbances such as old fields, settlements and deforested plantations. The two major plant communities were subdivided into two sub-communities that can be used as management units since they have approximately the same physiognomy, grass composition and habitat characteristics. The four broad management units are the *Acacia nilotica* – *Acacia karroo* – *Dichrostachys cinerea* Community (Clay Thornveld); the *Cissus rotundifolia* – *Enteropogon macrostachyus* (Valley Bushveld) Community; the *Panicum maximum* – *Brachylaena discolor* Community (Coastal Bushveld) and the *Themeda triandra* – *Urelytrum agropyroides* Community (Grass – Palmveld).

## The major vegetation types of the Soutpansberg Conservancy and the Blouberg Nature Reserve as part of the Soutpansberg Centre of plant endemism

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This study presents an ecological account of the major vegetation types representing the Soutpansberg Conservancy and the Blouberg Nature Reserve. TWINSPLAN was used to classify the vegetation data, while table-sorting procedures based on the Braun-Blanquet approach was used for further refinement. The classification lead to nine major vegetation types, interpreted as *Blepharis subvulbilis* – *Sclerocarya birrea* Blouberg Arid Mountain Bushveld, *Indigofera rhytidocarpa* – *Acacia nigrescens* Blouberg Dry Mountain Bushveld, *Rhus leptodictya* – *Euclea divinorum* Blouberg Mesic Mountain Bushveld, *Rhynchosia vendae* – *Englerophytum magalismontanum* Blouberg Moist Mountain Bushveld, *Adansonia digitata* – *Acacia nigrescens* Soutpansberg Arid Northern Bushveld, *Catha edulis* – *Flueggia virosa* Soutpansberg Moist Mountain Thickets, *Diplorhynchus condylocarpon* – *Burkea africana* Soutpansberg Leached Sandveld, *Rhus rigida* var. *rigida* – *Rhus magalismontanum* subsp. *coddii* Soutpansberg Mist Belt Vegetation and *Xymalos monospora* – *Rhus chirendensis* Soutpansberg Forest Vegetation. Plant communities of each major vegetation type are described, while diagnostic and threatened or rare species are highlighted. Vegetation-ecology and -patterns in the Soutpansberg Conservancy and the Blouberg Nature Reserve are driven mainly by the dramatic rain-shadow-effect of the east-west running ridges of the mountain complex, and its influence on the distribution of precipitation. The underlying geology seems to play only a minor role in the region's phytosociology. High topographical diversity within this small area, together with its connectedness with the surrounding centres of endemism, resulted in the high alpha and beta diversities observed. The Soutpansberg – Blouberg complex should be regarded as an important botanical hotspot of diversity, with vegetation including arid Kalahari sandveld, Afromontane forest, moist mist belt vegetation, arid bushveld, wetlands and high lying grasslands.

## Afrotemperate forests of South Africa in global perspective

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Afrotemperate (also known as 'afromontane') forests of South Africa were classified using plot-based floristic data. The floristic composition and a number of biogeographically important genera (*Ilex*, *Ocotea*, *Podocarpus*, *Prunus*, *Rapanea*/*Myrica* etc.) suggest close links between these forests and the evergreen sclerophyllous forests of various warm-temperate regions experiencing similar climatic patterns of the World (southeastern China, southernmost regions of Korean Peninsula, southern Japan, laurisilva of the Canary islands, *Podocarpus* forests of northern Argentina and the evergreen forests of southeastern USA etc.). The relationships between the afrotemperate forests, their analogons on other continents and so called montane tropical rainforests are discussed in palaeoecological, evolutionary perspective and in view of their extant distribution. A research agenda into origins and past dynamics of the afrotemperate forests using molecular-phylogeny methods and studies into life-history syndromes of the dominant species of the afrotemperate forests is outlined.

## The effect of evaporative cooling on yield of pistachio

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Pistachio (*Pistacia vera* L.) was introduced to South Africa in 1990 and currently 1000 ha of this plant is under intensive cultivation near Prieska in the northern Cape. A trial was conducted on the cultivars, Ariyeh, Shufra and Sirora in commercial pistachio orchards. Three cooling regimes using overhead irrigation were

used to evaluate the possible positive effect of evaporative cooling on nut yield. Orchard temperature was maintained at 18°C, during early spring, 2003. An additional treatment was implemented in Sirora utilising Budbreak as a rest breaker to investigate possible interactions. Trees were monitored on an individual basis for flowering and yield data. No significant differences ( $P < 0.05$ ) were observed between cooling treatments for Ariyeh. The cooling treatment for Shufra differed significantly from the control treatment with 1 203g/tree being recorded for the former. Sirora's unsprayed August–October treatment differed significantly from all other Sirora treatments and with 2 664g/tree recorded from the uncooled, unsprayed treatment.

## Woody plant species diversity, richness and distribution in a semi-deciduous tropical rain forest reserve, north-western Uganda

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Tree and shrub species alpha-diversity, richness, distributions and ecological importance were investigated in Budongo Forest Reserve (BFR). Plots were laid to capture the main historical management practices, forest sub-types, and environmental gradients. Thirty-two 50x100m (0.5ha) plots, each consisting of five contiguous 50x 20m plots (0.1ha) were sampled, and all individuals with a  $\geq 2.0$ cm diameter at reference height measured and identified. A total of 36 468 individuals representing 269 species from 171 genera and 51 families were recorded. The most speciose families were Euphorbiaceae, Fabaceae, Rubiaceae, Moraceae, Meliaceae, Rutaceae, Annonaceae, and Flacourtiaceae, accounting for 147 (54.6%) of the species. The most important families in terms of Familial Importance values (FIV) were Fabaceae, Euphorbiaceae, Ulmaceae, Meliaceae, Rhamnaceae, Apocynaceae, Moraceae, Rubiaceae, Violaceae, and Sapotaceae. In this regard, BFR's woody plant communities are quantitatively similar to the other Albertine Rift, Guineo-Congolese and Amazonian forests. The most important species in relation to importance value index (IVI) were *Cynometra alexandri*, *Lasiodiscus mildbraedii*, *Celtis mildbraedii*, *Senna spectabilis*, *Acalypha neptunica*, *Acalypha ornata*, and *Funtumia elastica*. Species richness was highest in previously logged sites and lowest for those with higher densities of *Cynometra alexandri* and *Senna spectabilis*. About 63.6% of the species were sparsely distributed, with some having restricted habitats. Fisher's alpha-diversity ranged 4.45 to 30.59 (for stems  $\geq 2.0$ cm dbh) and 3.07 to 29.7 (for stems  $\geq 10$ cm dbh), and was highest for previously disturbed sites and lowest for sites managed strictly as nature reserves since the forest was gazetted in 1932. The woody species diversity and richness of BFR is lower than for Amazonia and other rainforests receiving higher rainfall. Results revealed that a  $\geq 2.0$ cm diameter cut-off and 0.5ha plots are suitable for species diversity studies in this type of forest where most woody species rarely exceed the more commonly employed  $\geq 10$ cm diameter.

## The effects of cryopreservation of recalcitrant seed axes on the vigour of recovered seedlings

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Cryopreservation at liquid nitrogen temperatures is the only feasible route for the long-term preservation of recalcitrant seeds, and considerable attention has been paid to understanding and optimising freezing and thawing protocols. However, the long term objective is the re-introduction of viable plants into the field, and so it is important to assess the vigour of plantlets derived from embryonic axes subjected to cryopreservation. Data will be presented on several amaryllid species showing that axes that have been subjected to cryopreservation produce seedlings that are less vigorous than those from control axes. Germination and establishment are lower, and such seedlings are more susceptible to water stress. Methods to alleviate this problem will be discussed.

## An assessment of transformation technologies in *Pinus patula* Scheide et Deppe

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★ Awarded van Staden Prize for best oral presentation by an PhD student

The combined tools of biotechnology and genetic engineering have facilitated the genetic modification of *Pinus patula*, one of South Africa's most important softwood species for pulp production. Pre-existing *in vitro* and cryopreservation protocols were a platform on which a biolistic protocol for embryogenic suspensor masses (ESM) was established for *P. patula*. Stable integration of the bombarded pAHC25 plasmid was elucidated by positive PCR analysis of both GUS and *bar* transgenes – the first report in this species. Production of transgenic *P. patula* holds potential for manipulation of the lignin biosynthetic pathway, amongst other applications for the South African forestry industry. An *Agrobacterium*-mediated approach is being formulated in response to disadvantages of a biolistic transfer method where fragmented or multicopy events of the transgene are frequent. The most efficient mode of gene transfer is currently being investigated.

## Spatial variability in *Azorella selago* (Apiaceae) on sub-Antarctic Marion Island: climate change implications

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A number of stations in the Antarctic and on sub-Antarctic islands have reported rapid environmental warming along with changes in precipitation patterns over the last 30 to 50 years. One such station is sub-Antarctic Marion Island. On this island, *Azorella selago* Hook. f. (Apiaceae) is the most abundant and widely distributed vascular plant, dominating fellfield habitats. Fellfield communities are considered amongst the most vulnerable of Marion Island's habitats to climate change. The occurrence of fellfield communities over a broad altitudinal range, together with changes in mean temperatures with altitude, provides a unique opportunity to study the relationship between *A. selago* and environmental variables, including climate change. This study examines changes in the morphology and epiphyte load of *A. selago* in fellfields across the altitudinal gradient on Marion Island to quantify the response of this species to altitude and island-related environmental change. *Azorella selago* plants were sampled along four altitudinal transects (two easterly and two westerly transects) on the island, with three altitudinal bands with a plot of 50 plants in each band per transect. Temperature loggers were inserted into three plants per plot, and hourly readings taken over a two-month period in winter and in summer. The western side of the island was shown to experience more moderate temperatures than the east. Although plant growth rates were not significantly different between the two sides, plants on the east were generally larger with higher stomatal densities than those on the west. Trichome densities were also higher on the western side of the island. The temperature decline with altitude, and from east to west, matched the epiphyte density decline, suggesting that temperature may be a factor limiting the distribution of epiphytes on the island. These results provide insight into the likely impact of climate change on *A. selago* and fellfield habitats.

## Nectar volume and sugar concentration in *Aloe* species

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*Aloe* species are remarkably uniform in their nectar sugar com-

position. *Aloe* nectar is known to be hexose (95%, with equal amounts of fructose and glucose) dominant with only small amounts of sucrose (up to 5%). A study was done to determine if nectar volume and sugar concentration are also conservative characters in *Aloe*. Nectar from 34 species, including 97 plants and a total of 291 samples was analysed. The data recorded included nectar volume, flower tube length, sugar concentration and sugar composition. The conservative nectar sugar composition was confirmed with a maximum of 13% sucrose. Plant to plant and population to population variation within species were found to be remarkably invariable. The sugar concentration varied from 6–24% Brix and the nectar volumes from 3–129 µl. The data does not support the hypothesis that large flowers produce a more dilute nectar and small flowers a concentrated nectar as was found in other genera.

## Classification and ordination of the woody and herbaceous vegetation on Welgevonden Private Game Reserve in the Waterberg of Limpopo Province

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Welgevonden Private Game Reserve is a recently developed (1994) conservation area situated in the savanna environment of the Waterberg region of the Limpopo Province of South Africa. Management objectives include the maintenance of biodiversity and utilisation for non-consumptive wildlife tourism. Managing for the long-term maintenance of biodiversity requires an understanding of ecosystem function and seeking correlations between vegetation pattern and the variation of environmental factors is an important first step in this regard. Data on total aboveground woody biomass, absolute percentage herbaceous aerial cover and numerous site factors were collected and classification (TWINSPAN) and ordination (CANOCO) techniques were used to determine and describe vegetation pattern on Welgevonden and to elucidate, as far as possible, the associated environmental correlates. At a landscape level, the entire reserve could be viewed as *Burkea africana* broadleaf savanna. At the reserve scale, however, distinct associations of species and differences in structural features were revealed by the TWINSPAN analyses and four woody and six herbaceous communities were identified. Differentiation between the woody communities was mostly on the basis of structural rather than compositional criteria, and although subtle differences between the woody communities were observed for certain soil chemical and physical properties, the primary environmental correlates determining woody vegetation pattern were identified as rock cover (soil depth) and topographic position. Structural differences between the herbaceous communities were not as evident as for the woody layer, and differentiation between the herbaceous communities was therefore mostly on the basis of compositional criteria. As with the woody layer, rock cover and topographic position were identified as important environmental correlates in determining herbaceous vegetation pattern, but evidence suggests that other physiographic and soil chemical properties are also important with regard to the distribution of herbaceous species across the reserve.

## The response of the invasive alien *Chromolaena odorata* to elevated atmospheric CO<sub>2</sub> concentration at two levels of nutrient supply

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*Chromolaena odorata* (L.) R.M. King and H. Robinson is a pan-tropical C<sub>3</sub> weed that has become a serious problem in the warm mesic parts of South Africa. The response of C<sub>3</sub> plants to increased atmospheric CO<sub>2</sub> concentrations is known to be influenced by the availability of other resources and the inherent growth potential of the species. As an invasive weed, *C. odorata*



has a high maximum growth rate and is expected to respond positively to CO<sub>2</sub>. To test this, *C. odorata* plants were grown in open top chambers at ambient and 700 µl l<sup>-1</sup> atmospheric CO<sub>2</sub>, at high and low nutrient supply, and in competition with two grass species. The results showed that under these growth conditions, except for a slight decrease in specific leaf area, neither the growth nor photosynthetic characteristics of *C. odorata* responded to increased atmospheric CO<sub>2</sub> at either nutrient level. There was, however, a marked response to nutrient availability. It is possible that this unexpected lack of responsiveness to atmospheric CO<sub>2</sub> concentrations, particularly at high nutrient availability, was a consequence of restrictions on growth imposed by the open top chambers.

### The new IUCN system and the Red List of the Free State flora

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The first attempt to record the state of Earth's biodiversity was in the 1960s, through the publication of Red Lists of species believed to be at risk of extinction. Over the years increasing numbers of organisms have been assessed and several improvements to the methods for highlighting those species under threat have been made. The 1994 Red List criteria were a move from a qualitative to a quantitative system, and in 2001 the World Conservation Union (IUCN) issued new quantitative criteria for assessing the risk of extinction, that have been globally accepted. Red lists increase the awareness of the problem of depletion in abundance and possible extinction, of taxa, and are largely attributable to the development of the Red Data Book and Red Data List of the World Conservation Union (IUCN). Five different criteria of a taxon's life and history traits were considered to assess the threat of becoming extinct in the near future. The new and updated criteria include information on population size, distribution range, rate of decline and the probability of extinction (quantitative analysis). The Red List for the Free State flora will be presented for discussion and peer-review.

### Effect of the axis and testa on oxygen consumption and protein metabolism of germinating cucumber and squash seedlings

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Plant growth and development depends on successful mobilisation of the cotyledonary reserves during germination and seedling establishment. Both cucumbers and squash (Cucurbitaceae) contain mostly lipids as reserve material in their seeds. Both the presence of the axis, which acts as sink tissue, and oxygen availability is a prerequisite for optimum lipid mobilisation in these seeds. Total oxygen consumption ( $V_T$ ) in germinating cucumber and squash seeds is the result of mitochondrial activity ( $V_{MITO}$ ) and residual oxygen uptake ( $V_{RES}$ ), which is mostly represented by the  $\beta$ -oxidation of fatty acids in the glyoxysomes during active lipid mobilisation. However, the protective testa apparently restricts the diffusion of oxygen into the seed, making it a rate limiting factor for efficient lipid mobilisation during seedling growth. The aim of this investigation was to determine the effect of the testa on respiratory- ( $V_T$ ) and protein metabolism of germinating cucumber and squash seeds. A secondary aim was to determine the effect of the testa on the capacity of  $V_{MITO}$  and  $V_{RES}$  during germination and subsequent seedling establishment. Both manometry and polarography as techniques were also evaluated in an attempt to find the most applicable technique to determine  $V_T$ ,  $V_{MITO}$  and  $V_{RES}$  in intact seed tissue. The results obtained will be discussed.

### Isolation of a triterpenoid with anti-HIV activity from *Cassine papillosa*

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HIV/AIDS threaten more than 40 million people worldwide and more than 5 million in South Africa alone. There is no cure for the disease yet, and novel drugs need to be discovered to make any progress in combating the disease. Twelve extracts from indigenous South African plants were made, of which one of them, *Cassine papillosa*, showed exceptionally good inhibition of the functioning and production of HIV promoters, enzymes and a recombinant HIV strain. This extract showed inhibition in the HELA-TAT-luc assays and also against a VSV-pseudotyped recombinant virus. Inhibition of up to 80% was observed in the active fractions of the crude chloroform extract. The active compound was isolated, and identified by NMR as a triterpenoid. The isolated compound showed the same activity as the active fractions with inhibition of 80% against the recombinant virus and HELA-TAT-luc assays at concentrations of 100 ng/ml and 250 ng/ml.

### An ecological assessment of the sustainable utilisation of the woody vegetation in the Lowveld Bushveld, Mpumalanga Province

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This study was done in the communal area surrounding Makoko village and in the adjacent conservation area within the Kruger National Park, Mpumalanga province, South Africa. The structure of the woody vegetation within the two areas were compared in terms of species diversity, density, size structure distribution and biomass to determine the impact of fuel wood harvesting within the communal area upon the woody vegetation. There was no difference in woody plant species richness between the conservation and communal areas, but there was a difference between the uplands and lowlands. The conservation area uplands had the highest woody plant density and woody plant biomass. There were differences between areas in terms of the woody size class structure. The socio-economic status of Makoko village was determined by interviewing 100 households within the village. The use of fuels including wood, paraffin, candles and electricity was determined. Community and Kruger National Park issues such as advantages and disadvantages of living adjacent to the Kruger National Park were also noted. The demand for fuel wood within Makoko village was 338.9 kg per person per year, but the supply of fuel wood in the communal area was only 54.6 kg per person per year, if harvested sustainably. A conservation area of equal size could provide 270.0 kg of fuel wood per person per year on a sustainable basis. Management recommendations were made towards achieving sustainability in the use of the woody plant resources.

### Responses of cycads with different life histories to the impact of plant collecting: simulation models to determine important life history stages and population recovery times

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Collection of plants and seeds from wild populations threatens a large number of cycad species. We investigated to what extent



individual life history stages contribute to population growth, and compared two species with major differences in life histories in the African genus, *Encephalartos*: *E. cycadifolius*, a highly persistent grassland species that resprouts after fire, and *E. villosus*, a relatively fast growing, non-sprouting forest species. Several harvesting scenarios impacting different sized individuals were simulated to determine the sensitivity of the two functional types to harvesting. Both species were most sensitive to changes in abundance of adult plants. The harvesting of seeds had minimal impact on population growth rates, whereas harvesting of adult plants led to rapid population declines. This response from two very different functional types suggests that the conservation of adult plants is critical for all cycad species. Despite similar responses to adult mortality, the two species had substantially different population growth rates. This determined recovery time after harvesting of adult individuals. *E. cycadifolius* is typical of highly persistent plant species associated with low levels of recruitment and unable to recover from even small losses of adults within a reasonable conservation time frame (<100 years). Our results suggest that the ability to recover from loss of individuals is an important factor that should be considered when assessing the vulnerability of wild populations to threats.

### Mutagenic and antimutagenic screening of South African plants

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Dichloromethane and 90% methanol extracts from 46 South African plant species used in traditional medicine were evaluated for mutagenic and antimutagenic properties using the Salmonella/microsome mutagenicity assay (Ames) against tester strains TA98 and TA100, with and without S9. Vitotox®, Umu-C, Comet assay and the Micronucleus assay were tests also used for selected extracts only. For mutagenicity, all dichloromethane extracts produced negative responses. The 90% methanol extracts from the whole plant of *Helichrysum simillimum*, *H. herbaceum* and *H. rugulosum* produced positive responses. Five of the 46 species screened indicated antimutagenic properties, all in the presence of S9. The 90% methanol extracts were of *Bauhinia galpinii* (leaf), *Clerodendrum myricoides* (leaf), *Datura stramonium* (seed pod and leaf), *Buddleja saligna* (leaf) and *Sutherlandia frutescens* (leaves and stem), and the dichloromethane extract of *Bauhinia galpinii* (leaf). *C. myricoides* indicated good antimutagenicity and hence possible anticarcinogenic properties and was further fractionated to help identify the active components.

### Comparative morphology and chemical aspects of essential oils in *Lippia* (Verbenaceae) in southern Africa

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*Lippia* (Verbenaceae), established by Linnaeus in 1753, comprises about 200 species. These occur mainly in the new World (tropical and temperate regions of America) with a few species in

the Old World. Six species are found in southern Africa, including Namibia, Botswana, South Africa, Swaziland and Lesotho. These species are widely spread in the region, except in Namibia, and the Northern and Western Cape Provinces of South Africa. They inhabit savanna and grassland, mainly in open places. *Lippia* is characterised by bracteate, capituliform spike-like inflorescences. The phyllotaxis, form, size and indumentum of the bracts are important characters of taxonomic significance. The strongly aromatic, opposite leaves give off a lemon smell when crushed. The trichome complement of the stems, leaves and bracts consists of unbranched, standard trichomes, usually with 1-layered, multicellular bases and unbranched, sessile, glandular trichomes. It is in the glandular trichomes that essential oils are located. Initially, the chemical analyses of the essential oils, extracted by steam distillation, focused on *L. scaberrima*. The major terpenoids present in the oil from a bulk sample were quantified by GC-FID and their identities confirmed by GC-MS. In a consecutive study, the chemical profiles of the oils obtained from 37 individual specimens of the same species were compared. These specimens were harvested from five localities in North-West Province. The results indicate that there is variation between specimens within the same and different localities. This finding is consistent with that of other investigators involved in studies on *Lippia* species.

### Differential drought regulation of photochemistry in C<sub>3</sub> and C<sub>4</sub> subtypes of *Alloteropsis semialata*

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Physiological differences between C<sub>3</sub> and C<sub>4</sub> photosynthetic subtypes confer characteristics that result in differential responses to drought. Potential differences include the following: rates of transpiration and hence of declining soil water content; leaf water potential; stomatal conductance; photosynthetic rates and photoinhibitory responses. These responses were quantified for two photosynthetic subtypes of *Alloteropsis semialata* and showed that during soil drying, stomatal conductance responded differentially, with photosynthetic consequences. Photochemical energy utilisation under well-watered conditions was different, implicating differences in photorespiratory rates and alternative sinks for reductant. These differences in energy utilisation between subtypes remained apparent during drought, pointing to the operation of different mechanisms under photoinhibitory conditions.

### An investigation into possible heavy metal contamination of the central Karoo vegetation

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Extensive uranium trial mining by foreign companies was undertaken during the late 1970s in the uranium belt of the central Karoo. Due to the drop of the international uranium price these companies withdrew from South Africa without rehabilitating the mining areas. Heavy metals leaching out of ore tailings poses the potential of polluting the immediate environment. The aim of this pioneering study was to determine whether the local vegetation was polluted by heavy metals, whether these plants are able to bio-accumulate these heavy metals, the potential toxic effects of these heavy metals on selected plants and the possible bio-magnification thereof in the food chain as well as heavy metal phyto-extraction potential of the vegetation sampled. Vegetation and soil samples on two different affected farms in the Beaufort West district were analysed for U, Pb, Mo, Cu, Cd and As contents by means of ICP-OES (Inductively Coupled Plasma Optical Emission Spectrometry). In addition, protein concentrations and profiles of selected plants were also determined.

## Can the season of burning affect the growth of *Dichrostachys cinerea* populations? A hypothesis of phenology

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A hypothesis was put forward that summer burning would constrain the population growth of *Dichrostachys cinerea* as at this stage in the plant's phenology carbon reserves are lowest after the initial spring flush and therefore post-burn re-growth would be stunted. Initial slow re-growth would affect the time taken to reach maturity and the size of the plant in the next burn, thus indirectly affecting both fecundity and mortality. The hypothesis was tested by collecting data pertaining to the different fecundity, growth and persistence levels of *D. cinerea* populations in long term experimental burn plots that have been burnt in different months and at different frequencies in the Kruger National Park. The change in population density of *D. cinerea* in the plots over the last half-century was investigated to test whether any short term effects of the season of fire on a particular generation have long-term repercussions on the population density of the species. It was found that the average size of plants and their fertility levels are generally lower for plants burnt in mid-summer compared to plants burnt in winter. On all the burn plots investigated *D. cinerea* population numbers have increased in the last half century but averaged across the study areas, these increases are most pronounced on winter triennial burns.

## Pistachio production in South Africa: establishing a new agro-industry

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The unique nutritional value of pistachio nuts is the main driving force for this expanding industry. World production increased from  $\pm$  50 000 tons in the 70s to more than 400 000 tons currently, of which more than 70% is sold in the snack market. The main pistachio producing areas of the world are Iran, USA, Turkey and Syria. In South Africa, Green Valley Nuts is a project of the Industrial Development Corporation that is aimed at stimulating the formation of a local commercial pistachio industry. A 1 000ha planting is currently being established near Prieska, which will include a processing factory with an optimum capacity to process about 3 000 tons/year, mainly for export purposes. Pistachio nut cultivation requires long hot summers and cold winters. Culture practices such as nutrition, pest and disease management and tree canopy management pose major challenges. An adequate water supply, fertilisation, pest and disease management, weed control and pruning are thus required to optimise production. Budded trees are spaced at 333–432 trees/ha, of which male trees constitute 4–10% of the orchard. Production commences after a minimum of 5 years and trees should be in full production 10 years after establishment with a production potential of between 2–3 tons/ha. The new pistachio industry in SA will hopefully create new economic viability in an area of the country desperate for alternative agricultural crops.

## Basic research and the breeding of *Lachenalia* (Hyacinthaceae)

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Various basic chromosome numbers (i.e. 5, 6, 7, 8, 11 and 13) have been observed in the approximately 120 species forming the genus *Lachenalia*. The aim of this study was to compare the basic chromosome numbers with a molecular phylogeny and to determine whether this data may influence the planning of breeding programmes. Chromosome numbers were determined from mitotic and/or meiotic material by squashes in 2% acetocarmine. The molecular data were obtained from sequences of the *trnL-F* region.

Sequences were aligned with the aid of CLUSTAL X and manually corrected. Cladograms were obtained using PAUP 4.0b10. Sequences of 64 taxa were obtained from Genbank and 137 taxa were sequenced during this study. Chromosome numbers of 54 taxa were determined and compared with published chromosome numbers. These numbers were superimposed on the cladograms and this indicated that the majority of basic chromosome numbers are monophyletic and form a logical descending aneuploid series. Deviations from this pattern indicate reticulate evolution where the phylogenetic pattern is obscured by the maternal inheritance of the chloroplast. This study suggests that the basic chromosome number of the family Hyacinthaceae can be used as an indication of the phylogenetic affiliation of the specific taxon. This study further suggests that breeding programmes should be restricted to a certain clade to obtain viable hybrids.

## Seaweeds as a source of plant hormones for agriculture

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The use of seaweed-derived concentrates (SWCs) to improve growth and yield in agricultural crops is a well-established practice. Plant growth regulators (cytokinins and auxins) and betaines are considered the active principles. Recent advances in analytical methods now allows for more detailed and accurate analysis of plant-derived samples. When 31 South African seaweeds from the Chlorophyta, Rhodophyta and Phaeophyta were analysed, 12 isoprenoid and 6 aromatic cytokinins were identified. The endogenous cytokinins profile was very consistent with high levels of isopen-tenyl- and zeatin-type cytokinins and no dihydrozeatin-types being detected in all the species. The ribotide and free base conjugates were predominant while no N-glucoside conjugates were detected. Two SWCs made from the kelps *Ecklonia maxima* and *Macrocystis pyrifera* using the Cell Burst Method, were analysed for cytokinins and auxins using the same techniques. Eighteen cytokinins and 8 indole conjugates including IAA, were identified. Although the cytokinins occurred at 10x lower concentrations than the indoles, they increased when the SWCs were stored at an elevated temperature of 54°C for 14 days (accelerated ageing) while the indole concentrations decreased. These SWCs were also biologically active when tested in the mungbean rooting (auxin-like activity) and soybean callus (cytokinins-like activity) bioassays. As the cytokinins and auxin types found in the SWCs are similar to those found in higher plants, these products can serve as a rich source of growth biostimulants for higher plants.

## A holistic approach to new crop development

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The cultivation of a new crop in a monoculture system introduces it to new environments and management systems, which inevitably bring about new risks and challenges related to pest and disease management. Plant species that are grown in a new ecosystem are often particularly susceptible to endemic insects or pathogens present on other related plant hosts and capable of being transferred to a new food source. Co-evolved pests can be inadvertently introduced with the new crop and may then exploit their new environment, free from natural enemies. New management and culture practices sometimes also favour attack by pests and diseases particularly where sub-optimal or stress conditions for the host plant are created. In the light of the expanding new crop industry in South Africa, it is imperative that novel crop pro-

tection strategies, with particular emphasis on plant health, be adopted. This will enable growers to act less therapeutically and more proactively in crop health management by concentrating on optimising the health of crops instead of combating associated pests and diseases. The New Crop Pathology Programme (NCP) and the Insects on New Crops Programme (INCroP) at the University of the Free State in Bloemfontein were established independently for the purpose of serving new crop growers with information on sustainable methods to manage or control actual and potential pest and disease problems. The NCP and INCroP have consequently adopted an umbrella Holistic Plant Health Management (HPHM) strategy which encompasses a blanket analysis of all biotic and abiotic variables in a particular agro-ecosystem and their interactions with each other. The strategy, which is based on a decision based model that provides as much information as possible, is ultimately directed towards the development of a sustainable information and consultation framework for new crop development in southern Africa.

### Population dynamics, structure and life-history of *Oldenburgia grandis* (Asteraceae)

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*Oldenburgia grandis* is a rare, endemic species restricted to Witteberg quartzite outcrops in the Eastern Cape, South Africa. The aim of the study is to examine the dynamics of five populations in terms of their structure and stability. A stage-structured matrix model is used to estimate present population growth rates and to project future population dynamics from stage-specific vital rates. The contribution of each stage class to overall population growth rate is determined by elasticity analyses. A comparison of population growth rates and matrix transitions between sites is used to establish the possible threats to the species and to recommend possible management strategies. This paper reports preliminary population dynamics data and outlines proposed further research to complement the demographic analyses. This includes GIS analysis to determine current species distribution and repeat photography to investigate past population changes in different areas.

### Grassland productivity and global climate change. The role of rainfall timing in determining the annual above-ground productivity of two South African grasslands

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Annual rainfall has been shown to determine the net above-ground productivity of grasslands throughout the world. However, little is known of the effect of the distribution of rainfall events through the growing season on productivity. Models of global climate change predict that rainfall events may become larger but less frequent for many parts of the world covered by grasslands and savannas. Understanding the effect of rainfall timing, as well as rainfall amount, is therefore important for predicting ecosystem level responses of grass-dominated systems to climate change. Productivity and daily rainfall data was obtained from long-term experiments conducted at two grassland sites: Ukalinga in KwaZulu-Natal, with a high average rainfall and low inter-annual variability in total rainfall, and Sydenham in the Free State, with lower total rainfall but higher inter-annual variability. The average number of days between rainfall events – a measure of the variability of rainfall events through the growing season – can explain as much of the variation in annual productivity as total rainfall. However, the importance of timing was found to be greater for the drier site and depended on grass species composition.

### Factors affecting thermoinhibition in *Tagetes minuta* L.

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Thermoinhibition in *T. minuta* is tightly and rapidly regulated. Endogenous embryo factors control thermoinhibition, as at high temperatures radicle elongation and, to a lesser degree, hypocotyl elongation is prevented. Phytohormones play an important role, both in the imposition of thermoinhibition and the release thereof. Abscissic acid and ethylene appear to play critical roles in the control of germination, but minor roles for gibberellins, cytokinins and auxins can not be excluded. The increase in ABA levels at thermoinhibitory temperatures and the prevention of germination by exogenous ABA application at 25°C suggests a role for ABA in the imposition of thermoinhibition, whilst the requirement for ethylene during normal germination suggests that any factor impacting on ethylene production will prevent germination. The inability of ABA biosynthesis inhibitors to alleviate thermoinhibition implies that the absolute concentration of ABA is not the controlling factor but rather the sensitivity of the achenes to ABA, as ABA sensitivity was found to increase in *T. minuta* achenes in response to high temperatures. These hormones do not appear to act alone and it is the indirect interaction between ABA and ethylene that is critical for germination in *T. minuta*, as at high temperatures a combination of treatments that both reduced ABA levels and increased ethylene levels resulted in normal germination.

### New diseases of kenaf (*Hibiscus cannabinus*) in South Africa

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Kenaf (Malvaceae: *Hibiscus cannabinus* L.) is a new commercial crop in South Africa owing to the high quality cellulose fibres which it produces. Six fungal pathogens, four of which have previously been reported on kenaf in other countries, have been associated with diseased kenaf plants in South Africa since 2001. A high incidence of rust, caused by *Aecidium garkeanum* was observed on foliage of mature plants on one experimental site in the KwaZulu-Natal province and clear differences in susceptibility between cultivars were evident. In March 2001, at least 50% of 4- to 5-month-old kenaf plants were observed as having powdery mildew (*Leveillula taurica*). Symptoms were observed on Everglades 41, Cuba 108, El Salvador, SF459 and Tainung 2 but no differences in disease severity were noted between these cultivars. Grey mould (*Botrytis cinerea*) caused lodging on 25% of mature plants in three experimental sites and the relative susceptibility of kenaf cultivars was determined in field and glasshouse studies by means of artificial inoculation. *Sclerotium rolfsii* was isolated from 33% of plants showing symptoms of severe wilting accompanied by white mycelial strands and small dark brown sclerotia on stems. Two of the six pathogens that have been observed are first reports for kenaf, worldwide. Of these, the most significant is *Pythium* group G identified as the cause of large, black sunken lesions (10–20cm long) at the base of the stems, and severe root rot. Initially it resulted in 20% mortality of 4-month-old kenaf plants on one site but has since been reported from all experimental sites. *Fusarium verticillioides* was isolated from seedlings with damping-off on two sites. Glasshouse studies confirmed the pathogenicity of all pathogens and indicated significant differences in susceptibility between cultivars. From this survey, it is evident that numerous fungal pathogens pose a threat to kenaf cultivation in South Africa. An IPM program taking cognizance of cultivar susceptibility to disease must therefore be implemented if commercial cultivation is to be successful.

### Beauty in the eye of the share-holder: moving South African Iridaceae into the commercial sector

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★ **Awarded SAAB Certificate for best young scientist oral presentation at the Conference**

South African Iridaceae are typically floriferous, with comparatively large and attractive flowers with good longevity. Such plants exhibit economic potential, especially in the Northern Hemisphere, where a niche exists for dwarf geophytes (<50cm in height, including the inflorescence) suitable for container or garden planting. Unfortunately, this size limit precludes most *Watsonia* and *Gladiolus* species from being developed commercially. Chemical dwarfing and polyploidisation can negate these size constraints, without resorting to long-term and costly breeding programs. Chemical dwarfing treatments that examined the mode and timing of application of the anti-gibberellin Paclobutrazol, revealed that a single corm application can reduce *Watsonia* height by up to 75%. Dwarfism in these species was also retained following the cold storage of treated corms, which permits global marketing. Seed and emergent seedlings of *Watsonia* and *Gladiolus* species were investigated (*in* and *ex vitro*) to determine the timing and duration of exposure to 0.01% colchicine which most effectively produced stable polyploids. Seedlings were morphologically indistinguishable and the efficacy of induction was assessed as a function of the level of ploidy achieved and the disadvantageous presence of chimeras. The effect of increased ploidy on marketable characters in these genera will be assessed in due course, but it is hoped that mature polyploids will display, over and above dwarfism, larger corms and increased flower size with enhanced colour intensity.

**Rhizotron studies on *Zea mays* L. to evaluate biocontrol activity of *Bacillus* spp.**

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*Bacillus* spp. that originated from rhizosphere-containing roots of vegetable crops with promising biocontrol and growth stimulating abilities were isolated from the Tala valley of KwaZulu-Natal. They were tested in rhizotrons in a growth chamber with relative humidity 60% with a 12h photoperiod and day and night temperatures of 24°C and 18°C respectively. *Rhizoctonia solani* Kühn caused pre-emergence damping-off in the untreated maize seeds. Weak and soft roots were however observed through the Perspex rhizotrons. *Bacillus* B81 achieved biocontrol of 35% as regards shoot dry biomass and 48% with root dry biomass. *Pythium* sp. caused pre- and post- emergence damping-off of the untreated seeds. Root rot of the maize seedlings was effectively controlled by *Bacillus* sp. B81 and B69. *Bacillus* B81 had biocontrol activity of 11% as regards root area while B69 achieved 18%. Image analysis was used very successfully to quantify the effects of *Bacillus* treatment on *Pythium* sp. infected seedlings. For the biocontrol of *Fusarium* sp. *Bacillus* B77, B69, B81 were better biocontrol agents in relation to the control. B69 and B81 achieved biocontrol activity of 48% and 33% respectively.

**Conservation biology of *Ceropegia decidua* subsp. *pretoriensis*, a critically endangered plant, endemic to the Magaliesberg, South Africa**

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*Ceropegia decidua* subsp. *pretoriensis* is a perennial herb endemic to the Magaliesberg range in Pretoria. In 2002, the Gauteng Directorate of Conservation listed this subspecies as Critically Endangered with only 1069 plants having been located in nine subpopulations. This study surveyed 6 of those subpopulations in 2003. Subpopulation size varied from 4 to 875 plants, totaling a population size of 1104. Assessments of abiotic and biotic

habitat characteristics were made. This subspecies grew predominantly at the mid-slope position on the SSW aspect of quartzite/andesite ridges. It was associated with high % grass and litter cover. Plants only grew in full shade. Population size structure (stem length) showed a high proportion of small plants (≤100mm) producing a negative J-curve. The low percentage of reproductive adults (44%) was attributed to the then current (2002/3) drought conditions. The percentage of flowering and fruiting plants per subpopulation varied between 0–25%. Percentage germination (within 28 days) was low (21%). It was concluded that the primary threat is habitat loss through urbanisation. Existing threats, small subpopulation sizes and its restricted distribution confirms that this species is Critically Endangered B1ab (ii, iii, iv, v) + 2ab (ii, iii, iv, v).

**Fungal pathogens of pigeonpea in South Africa**

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Pigeonpea (*Cajanus cajan* (L.) Millspaugh) is a versatile legume widely cultivated in the tropics and subtropics and considered a new crop in South Africa. Diseases observed in experimental field trials in 2001, included powdery mildew caused by *Oidiopsis taurica* (Lev.) Salmon, leaf spot caused by *Passalora cajani* (Hennings) U. Braun & Crous and *Cercospora apii* Fresen, and rust caused by *Uredo cajani* Syd. Studies on *P. cajani* and *C. apii* focused on characterisation of the species in terms of certain physiological requirements *in vitro* and fungicide sensitivity in addition to molecular analysis. *Passalora cajani* and *C. apii* isolates differed significantly ( $P < 0.05$ ) in their ability to utilise four culture media. Temperature studies revealed that 25°C was most conducive to growth of all *C. apii*, and most *P. cajani* isolates. All five fungicides tested inhibited growth significantly at concentrations higher than 5 µg a.i./ml. Cluster analyses of physiological and molecular data were consistent. Germination of *U. cajani* spores and formation of infection structures were also examined using fluorescence and scanning electron microscopy. Results indicated significant differences ( $P < 0.05$ ) in colony size within and between medium- and long-duration pigeonpea types. Distinct host variation was observed in both duration types for susceptibility to *U. cajani*. A disease rating system for the pigeonpea-rust pathosystem was developed and used in a field trial to determine differences between six varieties. All varieties in this trial were rust susceptible, with variety ICP 6927 being most susceptible and variety ICEAP 0020 the least susceptible. In unsprayed plots, disease severity increased significantly ( $P < 0.05$ ) from the first assessment to the second, with ratings varying from 3 to 5.7 on ICPL 87119 and ICP 6927 respectively in April, and 9.2 and 11.9 on ICEAP 0020 and ICP 6927 respectively in May 2002. However, no significant differences in terms of yield or protein content between fungicide treated plots and untreated plots were recorded.

**A potent germination cue from plant-derived smoke**

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Fire is known to play an integral role in many ecosystems. In particular, smoke is an important trigger for the germination of many species. Using bioactivity-guided fractionation, a highly active compound, 3-methyl-2H-furo[2,3-c]pyran-2-one, that stimulates seed germination was isolated and identified from smoke

water. The identification of this natural molecule, the major germination cue from plant-derived smoke, should lead to a more comprehensive understanding of the role of smoke as a promoter of seed germination.

### A pilot study of the phytosociology of the pans in the Lake Chrissie district

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Characteristics particular to wetlands in South Africa are the result of interplay between rates of precipitation, evaporation and geology. Some of the oldest rocks in the world have been exposed to erosion in this area for millions of years with the consequence that suitably broken land where water may be trapped into lakes and rivers is scarce. Pans may be formed on geologically susceptible surfaces when they are eroded by grazing animals, salt-weathering and swirling winds. The lack of integrated drainage systems such as rivers, cause these circular or oval shallow basins to fill with standing water. Pans may be formed in fossil riverbeds to form linear associations, as is seen in the Lake Chrissie district. Endorheic pans (pans without any outlet) in the Chrissie panveld form the habitat for a large number of migratory and other birds and animals. Vegetation communities identified in this study clearly support the classification of pans in this system into three types as distinguished by Allan *et al* (1980). Moreover, a distinct relationship is shown between these pan types and elevation, suggesting that succession of plant species and therefore productivity of pans is related to their age.

### Phylogenetic development in Apocynaceae

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The two families Apocynaceae and Asclepiadaceae were recently grouped together into one family, the Apocynaceae s.l. Within the family Apocynaceae s.l. five subfamilies are recognised: Rauvolfioideae Kostel. (rather than Plumerioideae K. Schum.), Apocynoideae Burnett, Periplocoideae R. Br. Ex Endl., Secamonoideae Endl. and Asclepiadoideae R. Br. Ex Burnett. If one considers the pollen structure in the Apocynaceae s.l., there is a phylogenetic line of development that can be followed. The evolutionary trends that can be followed are 3 or 4-colporate grains in Rauvolfioideae, which are more primitive than the 3-porate grains in Apocynoideae. Tetrads in nine genera of Apocynoideae can be regarded as more advanced than 3-porate grains. Within the Periplocoideae the tetrads of *Raphionacme*, having numerous pores, could be regarded as more advanced than tetrads of genera with four to six pores. Pollinia in Periplocoideae can be regarded as more advanced than single tetrads. Of the Periplocoideae with pollinia, the multiporate tetrads in the pollinium of *Schlechterella* differ from the nearly inaperturate tetrads in the pollinia of all other taxa studied. This suggests that pollinia may have arisen twice within the Periplocoideae: once in Asia, and once in Africa. Pollinia in Periplocoideae and Secamonoideae are similar in that the pollinium consists of tetrads and is not covered by a pollinium wall, as it is in the Asclepiadoideae. In the Secamonoideae, however, the pollinia are attached to a corpusculum. In *Fockea*, belonging to the basal most tribe in the Asclepiadoideae, the pollinium also consists of tetrads, and is not covered by a pollinium wall. In this respect *Fockea* is more similar to Periplocoideae and Secamonoideae. The number of pollinia, however, is reduced to two and the distal wall is typical of that found in the Asclepiadoideae. In the Asclepiadoideae, the most advanced subfamily, the pollinium is covered by a pollinium wall, and single pollen grains, which lack apertures, are present. The pollinia are reduced to two per anther and are attached to a corpusculum by a caudiculum.

### Could At-RLK3 act as the elusive redox sensor in *Arabidopsis*?

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At-RLK3 is a novel receptor-like protein kinase from *Arabidopsis*. The expression of the gene was previously shown to be rapidly induced upon various treatments that all include the production of H<sub>2</sub>O<sub>2</sub>. The enzyme is an active protein kinase that is serine/threonine specific. Furthermore, the enzyme is plasma membrane bound, making it an ideal candidate to act as a receptor protein. Using transgenic plants expressing an antisense copy of the gene, the response of wild type and transgenic plants was found to differ substantially when exposed to H<sub>2</sub>O<sub>2</sub> and salicylic acid. Using this unique phenotype of the transgenic plants, a role for At-RLK3 as redox sensor will be proposed.

### Potential for the wild harvest and cultivation of *Pelargonium sidoides* with high bioactive compound content

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Extracts from the tubers of two *Pelargonium* species, *P. reniforme* and *P. sidoides*, are used in the Eastern Cape as traditional medicines for the treatment of respiratory tract and gastro-intestinal infections and globally as herbal treatments for bronchitis, asthma and as an immune system booster. Following the exploitation of wild populations by illegal harvesters for Western pharmaceutical companies, these species were awarded a protected status. Further research is now needed to develop an adequate management strategy for these species. This study attempts to provide some solutions to the problem of *Pelargonium* over-exploitation by quantitatively comparing the bioactive metabolite profiles of harvested and cultivated *Pelargonium* tubers and secondly by developing methods for the cultivation and sustainable-harvesting of these tubers with high bioactive compound content. The isolation of a documented bioactive compound and the development of a routine chemical assay for the quantification of this compound in extracts made from the tubers used in this study are fundamental to the success of this investigation. Progress with regards to the above and the assessment of the sustainability of wild plant harvest will be presented.

### Ecology and seed biology of the invasive alien plant, *Campuloclinium macrocephalum* (Asteraceae, Pompomweed), in Gauteng Province, South Africa

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The pompomweed, *Campuloclinium macrocephalum* (Less.) DC (Asteraceae), is a weed/non-native plant, especially prominent within the Gauteng Province. It originates from South America and is showing evidence of becoming one of the most aggressive alien invader plants in South Africa. Pompoeweeds invade disturbed sites such as roadsides, but also natural grasslands and wetlands. The pompomweed is an herbaceous plant with perennial rootstock and regrowth occurs every season from the rhizome. Plant density, seed production, seed banks and seed germination and viability

were studied in two populations within each of 2 sites, Groenkloof Nature Reserve and Voortrekker Monument Nature Reserve, in the Pretoria region. Furthermore, field and greenhouse experiments were established to test the effects of burial depth, light conditions, and water availability on seed viability and germination over a 30 month period (May 2002 to November 2004). Finally the thermotolerance of the seeds was tested in terms of soil surface temperatures, both during- and post-fire, as well as cold temperature tolerance (frost is prevalent on the highveld). We also determined the allometric relationship between plant size (height, canopy, above-ground biomass) and seed production potential. Pompoeweed has prolific seed production, and produces a potentially very large persistent seed bank. It has the seed biology of a typical weed – i.e. germination stimulated by high light conditions typical of disturbed environments. Its spread is aided by light-weight wind-dispersed seeds produced in large numbers. It also has the ability to grow from the below-ground fleshy rhizomes/tubers. In terms of seed biology, pompoeweed is the highveld chromolaena!

## Posters

### The African Plants Initiative: An electronic resource for continental taxonomy

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Herbaria are vast stores of information, knowledge, and expertise on the plant diversity of individual countries, floristic regions, or a continent. Alpha-taxonomic studies using herbarium collections remain a cornerstone of modern biology. Access to types (that is physical specimens) that fix the names of taxa is imperative if plants are to be correctly named and knowledge about species is to be collated and disseminated. However, information on the existence, status, location and condition of such specimens is often lacking and is a serious impediment to the study of plant diversity. This difficulty is especially acute in Africa, where resources and facilities for taxonomic research are limited. In addition, the type specimens of African plants are mostly held by herbaria situated in the 'north', making their location and accessibility an acute problem particularly for African taxonomists. A new project, the African Plants Initiative (API), funded by the Andrew W. Mellon Foundation, aims to aggregate data on African plants from various regions of the world and to build and support an online database of these resources. This facility will be developed and maintained by Aluka, a not-for-profit subsidiary of, among others, the Mellon Foundation. Several major herbaria in Africa, Europe and America are participating in this initiative. During the initial phase of the project the participating herbaria will scan all their type specimens and make the images and associated information available to Aluka. The South African National Biodiversity Institute (SANBI) also participates in the initiative and all the types held in the three major herbaria of SANBI will be scanned, databased, and the images provided to Aluka. Digitising type material will enhance access to material that is dispersed around the world and often difficult to access. Digitisation will also help preserve irreplaceable material that are in danger of being lost when shipped across the globe. The process of imaging, data basing, and the envisioned end product of this initiative will be presented.

### Producing doubled haploid plants from wheat: How responsive are South African genotypes?

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Despite the enormous potential of using doubled haploid technology in commercial breeding programs, little is known concerning the response of South African genotypes to anther and microspore culture. Anther and microspore culture was performed on two cultivars previously reported to be responsive (cv Chris and cv Pavon 76) as well as several South African inbred and hybrid lines. Treatments used to induce androgenesis included heat and cold shock, induction with 2-hydroxynicotinic acid, nitrogen and carbohydrate starvation. In addition to screening South African germplasm, several published media formulations were assessed for their ability to regenerate microspore-derived embryoids. The number of embryo-like structures were counted after 30 days in culture, and these were allowed to regenerate on a suitable medium. Ploidy of regenerated plantlets was determined by flow cytometry.

### The effect of ReTain® (aminoethoxyvinylglycine, AVG) on the postharvest quality of minimally processed cantaloupe melons

**JS Boatwright and CS Whitehead**

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The effect of ReTain® (active ingredient aminoethoxyvinylglycine (AVG)), on the postharvest quality of fresh cut cantaloupe melons was studied. It was found that a postharvest application of AVG (an inhibitor of ethylene synthesis) in the form of a dip as opposed to a preharvest spray was more effective in reducing ethylene production in whole melons and melons cut into punnets. This reduction in ethylene synthesis resulted in a delay in ripening and deterioration of cut fruit.

### Biochemical and histological aspects of resistance to *Puccinia striiformis* f. sp. *tritici* in wheat

**JM Bower, AJ van der Westhuizen and ZA Pretorius**

Department of Plant Sciences, University of the Free State, Bloemfontein, 9300

Yellow rust, caused by *Puccinia striiformis* f. sp. *tritici*, is one of the major pathogens of bread wheat (*Triticum aestivum*). In South Africa the disease was first detected in the Western Cape in 1996 and has since limited crop yields in epidemic years. To investigate biochemical aspects of resistance, activities of  $\beta$ -1,3-glucanase, chitinase and peroxidase were investigated in the near-isogenic lines Avocet-Yr1 (yellow rust-resistant) and Avocet-S (yellow rust-susceptible). An increase in PR-protein expression, in particular peroxidase activity, occurred 96h post-inoculation (hpi). Fluorescence microscopy of fungal development showed that infection structure differentiation and colonisation of leaf tissue were visible from 48hpi onwards. The characterisation of host-pathogen interaction at the biochemical level forms part of a more comprehensive understanding of defence mechanisms in plants and how these can be manipulated in disease control.

### Regeneration failure and the *Acacia karroo* successional pathway in coastal dune forests in KwaZulu-Natal, South Africa

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Forest Biodiversity Research Unit, School of Biological and Conservation Sciences, University of KwaZulu-Natal, Pietermaritzburg, South Africa

The *Acacia karroo* successional pathway has been used to rehabilitate mined dune sites near Richards Bay, South Africa. We investigated the relatively low species diversity and slow regeneration of coastal dune forests in areas dominated by *A. karroo* at Cape Vidal. Seed germination trials demonstrated that there are similar species and numbers of propagules in *A. karroo* corridors and adjacent climax dune forest, indicating that no dispersal barrier exists between these habitats. Surveys of naturally-occurring seedlings in *A. karroo* corridor and forest habitats found that community composition differed significantly, suggesting that a post-establishment factor influ-

ences seedling survival in the corridors. As an anthropogenically disturbed habitat and an area dominated by a nitrogen-fixing species, the *A. karroo* corridors were expected to have high levels of nitrogen that might limit growth of forest seedling species. No differences between soil nutrient levels in the two habitats were found, however. This indicates that excessive nitrogen enrichment does not occur and that growth and survival of forest species in the corridors are not affected by nitrogen levels or other soil nutrients. A high level of herbivory by large vertebrates was the most important constraint on the growth and survival of forest species in the *A. karroo* corridors. The impaired growth of forest seedlings and a lack of community convergence with climax forest composition suggest that the successional process in *A. karroo* corridors has been arrested by the actions of herbivores. This has resulted in regeneration failure in *A. karroo* corridors up to 50 years post-disturbance. The *A. karroo* successional pathway may successfully restore mined dune sites provided there are low densities of herbivores, but is less suitable for rehabilitating disturbed dune forest when herbivore densities are high.

### The systematic value of the leaf indumentum in *Lobostemon* (Boraginaceae)

MH Buys

AP Goossens Herbarium, School for Environmental Sciences and Development, North-West University, Potchefstroom Campus, South Africa

★ Awarded Local Organising Committee prize for best poster in Taxonomy

The character states pertinent to the indumentum of *Lobostemon* leaves are illustrated, tabulated and discussed. *Lobostemon* possesses similar trichome and indumentum types as described for the sister genus *Echium*, with the exception of the 'Alpine' indumentum type. Due to the apparent environmentally induced variation, the leaf indumentum characters of *Lobostemon* are viewed to be of limited phylogenetic value, although they do diagnose a number of taxa. Results from a phenetic analysis correspond in part with previous phylogenetic schemes.

### Genetic identification of kenaf (*Hibiscus cannabinus*) accessions using AFLP analysis

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Kenaf (*Hibiscus cannabinus* L.) is one of the world's most economically important fibre crops. Kenaf is classified in the genus *Hibiscus* in the Malvaceae family. The crop has great potential for its multipurpose utilisation (pulp and papermaking, board and filtration media making, and animal feed), in addition to its traditional usage (making ropes, sacs, and canvases). Kenaf was introduced to South Africa in the last few years as a potential commercial crop for fibre production. Since kenaf is a new and unknown crop to South Africa, it is necessary to determine the genetic relationships of accessions for possible breeding programmes. Varietal identification of kenaf is problematic and knowledge on genetic diversity of kenaf varieties is limited. In order to identify different varieties, and investigate their diversity and genetic relationships, ten commercial and nine wild type kenaf accessions were analysed using amplified fragment length polymorphism (AFLP) fingerprinting. More than 350 data points were generated using six AFLP primer combinations. Statistical analysis was done using NTSYS-pc version 2.02i. The AFLP fingerprinting technique was effective in uniquely identifying all nineteen kenaf accessions and determining their genetic relationships.

### Photoinhibition compared in $C_3$ and $C_4$ *Alloteroopsis* photosynthetic subtypes

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★ Awarded Local Organising Committee prize for best poster in Physiology

Due to fundamental differences in photosynthetic physiology,  $C_3$  and  $C_4$  plants are differentially affected by photoinhibitory conditions. In  $C_3$  plants photorespiration is an important sink for excess reductant, potentially reducing the requirement of other energy dissipating mechanisms.  $C_4$  plants lack photorespiration and are likely to require other mechanisms for energy dissipation including increased antioxidant activity, xanthophyll cycling and PSII inactivation. This study assesses the responses of  $C_3$  and  $C_4$  *Alloteroopsis semialata* photosynthetic subtypes to photoinhibitory conditions measuring PSII efficiency, antioxidant activity and xanthophyll cycling. Photoinhibitory conditions were induced with combinations of high light, drought and limited  $CO_2$  or  $O_2$  availability.

### Mysterious circles in the Namib Desert: An overview

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The secret of the origin of the so-called 'fairy circles' catch the attention of many scientists and tourists visiting Namibia. These fascinating phenomena occur in a broken belt in the pro Namib along the west coast of Southern Africa, extending from southern Angola to just south of the Orange River. These circles are concave depressions that occur roughly between 60 and 120 km inland from the coast. The altitude varies between 500 to 1 000 m. The rainfall varies from 50 to 100 mm per year; it may however be of importance that this area falls within the west coast fog belt as an additional source of moisture. It appears that the sizes of the circles are related to the amount of rainfall/moisture because they are generally larger in the north (15 m+) to an average of approximately 2 m in the south, while the average size is between 6–8 m in the central parts of the country. Densities can be as high as 20 circles/ha. A specific pattern in their distribution and size must still be determined. Grass species associated with the circles in Namibia are *Stipagrostis giessii* and *S. hochstetteriana* in the north, *S. uniplumis* in the central parts and *S. ciliata* and *S. obtusa* in the south. Several studies were conducted during the last 30 years in an effort to explain the origin of these circles. A brief overview of the current research will be covered in the poster.

### A comparative study of the concentration of naphthoquinones in South African *Euclea* species

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Four naphthoquinones (NQs) namely 7-methyljuglone, diospyrin, neodiospyrin and isodiospyrin, present in the tree, *Euclea natalensis* (Ebenaceae), have been found to have potent antituberculosis activity. A comparative study of these NQs was conducted on ten species of *Euclea* in order to select a species which has the highest concentration of NQs. These NQs are required in substantial amount for pre-clinical studies. Roots of the selected *Euclea* species were collected from all over South Africa, ground to a fine powder and extracted with chloroform. High Performance Liquid Chromatography (HPLC) was used to quantify the concentration of each naphthoquinone. The most potent anti-TB compound, 7-methyljuglone, was found in almost all species studied and its concentration ranged between 20–1 300 mg/kg. It was found that *E. pseudobenensis* had the highest concentration of 7-methyljuglone. Diospyrin was found in only 5 species of *Euclea*. Seven species of *Euclea* showed the presence of isodiospyrin whereas only one tree, belonging to *E. pseudobenensis* species was found to have 1 800 mg/kg of neodiospyrin. Of all the plants studied, *E. pseudobenensis* showed the presence of all four compounds, therefore, harvesting of *E. pseudobenensis* is recommended for the extraction of all anti-TB NQs. The concentration of the compounds varied to a great extent, both interspecifically and intraspecifically. It can be speculated that these variations may be due to different environmental factors, differences in the age of the roots and/or wound and other stress factors.



## ***Helichrysum* (Asteraceae) interactive**

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*Helichrysum* is a large, important genus in the family Asteraceae. It was revised in 1983 by Hilliard as part of the *Flora of southern Africa*. For most people the identification of species is a daunting task as there are 260 Asteraceae species in southern Africa. The text of Hilliard's treatment has been converted into a LUCID interactive key. Although this key is far from complete, it is already possible to demonstrate a workable aid to identification. In the final product each species will be illustrated with, photographs and a distribution map. Some additional information will be added to the original text.

## **Chlorophyll fluorescence-derived chill factor index (CFI) as a screening criterion for dark chilling tolerance in soybean**

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Due to chilling sensitivity, minimum night temperature represents the main constraint in soybean production in South Africa. Production can only be improved by directed breeding and genetic transformation. We describe and illustrate a quick, sensitive, non-destructive screening procedure for dark chilling tolerance in large numbers of soybean *Glycine max*, based on quantification of the fast chlorophyll a fluorescence transient O-J-I-P. Thirty South African soybean genotypes of unknown chilling tolerance, and two foreign genotypes of well known, but contrasting chilling tolerance, were grown in growth chambers under rigorously controlled environmental conditions. Plants were dark chilled (6°C) for seven consecutive nights but kept at normal day temperatures (26°C). Root zone temperatures were kept at 20°C to minimise the occurrence of chill-induced drought stress. Before the end of the respective dark periods, chlorophyll a fluorescence transients were recorded in fully dark-adapted attached leaves of control, and dark chilled, plants. The stress-induced changes in the fluorescence transients were translated to biophysical parameters quantifying the energy fluxes through photosystem II (PSII) using the JIP-test software. The performance index (PI<sub>ABS</sub>), a multi-parametric expression taking into account the three key steps of PSII function, namely light energy absorption, excitation energy trapping, and conversion of excitation energy to electron transport, was used as measure of dark chilling tolerance. Elaboration of the PI<sub>ABS</sub> resulted in the formulation of a novel parameter, the so-called chill factor index (CFI), capable of revealing large differences in dark chilling response among the genotypes. The CFI values of the two reference genotypes, Maple Arrow (chilling tolerant) and Java 29 (chilling sensitive), correlated with their known difference in chilling tolerance. We used the CFI to rank the thirty South African genotypes according to their dark chilling tolerance and could identify five genotypes with a dark chilling response comparable to that of the tolerant Maple Arrow.

## **Effect of emissions from Selibe-Phikwe copper/nickel mine on the soil and surrounding vegetation**

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The aim of this study was to investigate the effect of airborne smelter waste from BCL mine on the surrounding vegetation and soil. An Experimental Site, 4.0km west from the smelter, in the prevailing wind direction and two Control Sites, one 4.0km east of the smelter and the other 60km southwest of the smelter were demarcated. The results indicate that the BCL smelter emissions have had an adverse impact on the soil and vegetation on the Experimental Site. The Experimental Sites' soil was significantly

more acidic than that of the Control Sites (control site 1 and control site 2). *C. mopane* leaves in the experimental site exhibited lower leaf area and the maximum photochemical efficiency of PSII (i.e. dark adapted  $F_v/F_m$  ratio, compared to that of the other sites). *B. albitrunca* displayed higher leaf area in the Experimental Site compared to the two Control Sites. However, the lower maximum photochemical efficiency of PSII exhibited by *B. albitrunca* in the Experimental Site showed that it was stressed. These results clearly demonstrated that the plume from the BCI mine has an adverse effect on vegetation and soil, particularly, on the western side of the mine.

## **Methods to improve seed germination in *Bowiea volubilis* – a highly utilised bulbous medicinal plant**

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*Bowiea volubilis* Harv. ex Hook. f. is one of the top fifteen medicinal plants identified by herbal sellers in South Africa as likely to become scarce. The bulbs are commonly sold for traditional medicine and are used to treat a number of illnesses. Extracts prepared from roasted bulbs are administered as purgatives, while fresh bulbs are used to treat oedema (dropsy) and infertility in women. Lotions prepared from the plant are used in treating sore eyes and skin diseases. *B. volubilis* occurs naturally in the eastern parts of South Africa in grasslands and thickets, where it has now become vulnerable and in some regions is locally extinct. To date, methods to improve seed germination have not been investigated. The percentage germination under control condition (25°C) was only 15% with a mean germination time of 18 days. The seeds incubated at 20°C resulted in higher germination (90%) than with other temperatures tested, but had a mean germination time of 25 days. Seeds germinated in continuous dark at room temperature resulted in improved germination (88%) with a mean germination time of 24 days. In this study, mechanical scarification with sand paper was the best treatment to improve germination (100%) with a mean germination time of only 6 days. This technique could be used for large-scale production of *B. volubilis* seedlings.

## **Isolation of the antibacterial flavonol quercetin from *Melanthus villosus***

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Leaf and root extracts of *M. villosus* were screened for antibacterial activity using the dilution method. *Bacillus cereus*, *B. pumilis*, *B. subtilis*, *Staphylococcus aureus*, *S. pyogenes* and *Escherichia coli* were inhibited by ethyl acetate extracts. The ethyl acetate extract was subjected to chromatographic isolation and purification techniques, which resulted in a compound whose activity was investigated by direct bioassay with thin layer chromatography. Ultraviolet/visible spectral identification of the antibacterial active flavonoid compound indicated that it was a flavonol, quercetin.

## **Biflavones from *Rhus* species with affinity to the GABA<sub>A</sub>-benzodiazepine receptor**

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Plant material from three *Rhus* species were collected in South



Africa, one of these species, *Rhus pyroides*, is traditionally used in the treatment of epilepsy. From the leaves of *Rhus pyroides* Burch., *Rhus dentata* Thunb. and *Rhus pentheri* A. Zahlbr., six different extracts were made which were tested for activity in the [<sup>3</sup>H]-flumazenil binding assay. The ethanol extract of *R. pyroides* and the ethyl acetate extracts of *R. dentata* and *R. pentheri* had the highest activity. From the extract of *R. pyroides* two active compounds were isolated by HPLC. By structure elucidation (<sup>1</sup>H-NMR and <sup>13</sup>C-NMR) they were determined to be amentoflavone and agathisflavone. The IC<sub>50</sub> values of these compounds were 53nM and 77nM respectively. The K<sub>i</sub> values were calculated to be 57nM and 82nM respectively. The active fraction collected from *R. dentata* contained agathisflavone, apigenin, a triterpene and at least one other unknown compound. The fraction collected from *R. pentheri* contained agathisflavone, apigenin and more than one unknown compound. Apigenin, agathisflavone and amentoflavone were fitted into the pharmacophore model of ligands binding to the GABA<sub>A</sub> receptor benzodiazepine site. The fitting into the pharmacophore model reflected the affinity of the compounds in the [<sup>3</sup>H]-flumazenil binding assay. Apigenin, agathisflavone and amentoflavone were evaluated to be unfit as CNS active drug candidates due to their unspecific pharmacological profiles and their low penetration of the blood-brain barrier.

### Rust development and associated defence related responses in sunflower

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Sunflower (*Helianthus annuus* L.) cultivars that are either susceptible (S37-388) or resistant (PhRR3) to sunflower rust were inoculated with rust (*Puccinia helianthi*) spores and the fungal development monitored until disease symptoms were evident. Changes in the activities of pathogenesis related proteins were also monitored. In the susceptible plants infection structures were more abundant whereas in the resistant cultivar more infection structures were aborted and necrotic lesions developed around the haustorium mother cells. Infection selectively induced the activities of apoplastic  $\beta$ -1,3-glucanase (EC 3.2.1.39, PR 2) and chitinase (EC 3.2.1.14, PR 3) in the resistant plants. The induced enzyme activities occurred concomitant to an increase in the frequency of aborted infection structures as well as host cell necrosis in resistant plants. Furthermore, treatment of susceptible plants with benzothiadiazole, a functional analogue of salicylic acid, induced intercellular  $\beta$ -1,3-glucanase and peroxidase (EC 1.11.1.7) activities and reduced the degree of susceptibility to rust.

### Vegetation and habitat types of the OR Tambo Node, Eastern Cape, South Africa

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The OR Tambo Municipal district, previously known as Transkei, has been identified as a development node for poverty relief as part of the government's Integrated Sustainable Rural Development Strategy. To ensure a proper development a holistic audit, considering all natural resources and impacts, needs to be undertaken. The OR Tambo Node is situated in the Eastern Cape along the Indian Ocean coastline. This study presents the results of the regional vegetation audit to describe the rangelands and factors influencing rangelands. A total of 158 surveys were conducted during March–July 2002, where after the data were analysed using multivariate analysis techniques. Results obtained from a Minimum Variance and TWINSPAN classification were found to be complementary to each other. Therefore both these methods were used to describe the vegetation communities in the rangelands for potential development. Although the grasslands

are highly diverse (559 species), results showed a high dominance of specific grass species, such as *Aristida junciformis*, *Eragrostis plana* and *Sporobolus africanus*. Vegetation types were influenced mostly by elevation, precipitation, terrain position, soil depth and agricultural practices. Associations were also found between specific communities and certain broad soil groups for example leached, humic and shallow soil types. Three broad grassland vegetation types were primarily distinguished, namely the *Aristida junciformis* Vegetation type, *Sporobolus africanus* – *Eragrostis plana* Vegetation type and *Themeda triandra* – *Heteropogon contortus* Vegetation type.

### The influence of temperature, photoperiod and irradiance on flowering of *Watsonia* species

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*Watsonias* are members of the Iridaceae, which are native to South Africa. The genus exhibits potential as container subjects, but little is known about their flowering physiology. Experiments were designed to identify the environmental cues (temperature, photoperiod and irradiance) that induce flowering in three species; namely *Watsonia tabularis*, *W. borbonica* and *W. pillansii*. Representative species from winter- and summer-rainfall areas were included for comparison. Leaf number and elongation were promoted by high temperature regimes (21/15°C and 29/21°C), whilst low temperatures (12/7°C) produced shorter, less foliose plants. These data were independent of photoperiod. Compromising leaf number is critical, as control plants required a minimum of 5 leaves for successful spike emergence. However, higher temperatures resulted in an increased incidence of corm rot. Long days (16h light/8h dark), in combination with low irradiance (150  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>), nullified the negative effects of low temperature on flowering, and resulted in earlier flowering in warm-treated plants. Conversely, short days in combination with low irradiance (150  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>) delayed or completely inhibited flowering. Variation among species and the precise dynamic between temperature, light and flowering, are proposed consequences of ecological differences in species habitat.

### Turnover of Norway spruce genotypes in relation to their embryogenic competence during the transition from embryonic to vegetative growth

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In Norway spruce, the transition from embryonic to vegetative growth was accompanied by a transfer of the site of somatic embryo initiation from the hypocotyl to the cotyledons. This property was used to determine the embryogenic competence of individual genotypes either at embryonic or postembryonic stages. We demonstrated that, the competence of genotypes for somatic embryogenesis could be maintained, lost or recovered at different stages of development. This result suggests that all genotypes could be propagated via somatic embryogenesis, following the right definition of the developmental stage. If such an hypothesis is verified, there would be no selection in favor of genotypes that produce embryogenic cultures.

### Conspectus of the Free State flora

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The objective of the envisaged publication, **Conspectus of the Free State flora** is to contribute towards our knowledge of southern African plant diversity. The aim is to assist professional botanists, nature conservationists, students, amateur botanists

and informed laymen in their work on the plants of the region. The publication will include information on the vegetation, based on a map of Rutherford et al. (in prep.), a key to the relevant families and keys to genera occurring in the Free State. The species descriptions will contain information on habit, leaf, flower and fruit morphology, distribution and habitat, uses and common names. A glossary will be presented in three different languages: Afrikaans, English and Southern Sotho. It is hoped that the *Conspectus of the Free State flora* will stimulate and promote an interest in the flora of the region.

### An SEM examination of arbuscular mycorrhizal fungi distribution and localisation on the root structure of *Eragrostis curvula* (Umgeni)

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Frozen and critically-point dried root material of *Eragrostis curvula* (Schröd.) Nees (Umgeni) was examined using SEM for the distribution of hyphae from symbiotic mycorrhizal associations. Hyphae apparently enter the root epidermis via the cell wall making use of small cracks between cells, to gain entry into the underlying cortical cells. Our examination demonstrates hyphal entry and subsequent spread into the host cortical cells toward the endodermis. As in all roots, the endodermis plays a significant role in regulating movement of ions, water and nutrients across this cell layer. All roots examined demonstrated the presence of well developed pericyclic and endodermal layers, which even in young roots, appear to be barriers to hyphal invasion. This study therefore confirms that endodermis serves as a barrier to further inward progression of fungal hyphae, as arbusculature was never observed in pericyclic or vascular tissue. The mycorrhizal arbuscules appear to be optimally located near endodermal passage cells for nutrient exchange between the root and mycorrhizal fungi.

### Water utilisation by *Opuntia ficus-indica* and *O. robusta* over a soil-water gradient

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Cactus pear (*Opuntia* species) has great potential to improve productivity in arid areas, the main reason being its Crassulacean Acid Metabolism pathway, which is very efficient in terms of water requirements. This study is aimed at determining the influence of different water applications on the root/cladode ratio and water-use efficiency of *Opuntia robusta* Wendl. (cultivar: Monterey, blue cladode) and *O. ficus-indica* (L.) Miller (cultivar: Morado, green cladode). The cladodes were planted in pots (210mm diameter and 550mm deep soil) and kept in a greenhouse at day/night temperatures of 25–30/15–18°C. The water treatments applied were 0–25%, 25–50%, 50–75% and 75–100% depletion of total plant available water. After four weeks of keeping the soil at field water capacity, the water stress treatments started. The planted pots were washed out when reaching the different water levels, after which root lengths, root mass, cladode mass and water-use efficiency (dry matter produced per mm water used) were determined. The measurements were done on one-year-old plants. Water-use efficiency decreased ( $P < 0.01$ ) with an increase in water stress for *O. ficus-indica* while, in contrast, increased for *O. robusta*. The finer root system of *O. robusta* could be responsible for the more efficient water uptake with water stress. In both species root mass decreased and root length increased with water stress. The main reason could be the formation of more smaller side roots, while main roots die-off with water stress. The root/cladode ratios for *O. ficus-indica* and *O. robusta* decreased with water stress from 0.1364 to 0.0924 and 0.1443 to 0.0749 respectively. The amount of water needed to fill up a cladode, increased with water stress from 0.58 to 11.40mm and 1.15 to

10.97mm for *O. ficus-indica* and *O. robusta* respectively. The water percentage in the cladodes decreased with water stress from 90.31 to 86.86% and 91.29 to 86.44% for *O. ficus-indica* and *O. robusta* respectively. *Opuntia* species can use water more efficient than most other fodder plants and can therefore utilise arid areas to its full potential.

### Root dynamics of *Opuntia ficus-indica* and *O. robusta* as affected by water stress

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In contrast to the cladode system, the roots of *Cactaceae* have received little attention, however, they certainly differ from other plants, as they develop xeromorphic characteristics. The aim with this study was therefore to quantify some survival characteristics of the root system of *Opuntia robusta* Wendl. (cultivar: Monterey – blue cladode) and *O. ficus-indica* (L.) Miller (cultivar: Morado green cladode). Cladodes of each species were planted in root boxes (650 x 100mm, with 900mm deep soil), which were kept in the greenhouse at day/night temperatures of 25–30/15–18°C. The boxes were placed at a 15° angle, so that the roots growing downwards could easily be seen and measured through the glass side of the box. Growth of main roots, side roots and rain roots were measured on an hourly and daily basis (day and night) over a period of two months. The measurements took place over a soil-water gradient. The average daily increase in length of the main roots, growing in soil between field water capacity and permanent wilting point, were 39mm and 26mm for *Opuntia ficus-indica* and *O. robusta* respectively. With water stress the growth of the main roots, regardless the species, daily decreased from 12:00 to 18:00, but increased from 06:00 to 12:00 and from 18:00 to 06:00. The decrease in growth during the afternoon could be due to higher soil temperatures building up. The thinner side-roots, that develop during water-stress conditions, enable the plant to absorb water more efficiently under lower soil-water conditions. After rewetting the soil, rain roots developed and grew as much as 1mm after only an hour. These roots totally stopped growing after only three days for both species at an average length of 21mm and 12mm for *O. ficus-indica* and *O. robusta* respectively. The quick reaction of the rain roots to water, makes the plant adaptable under very low rainfall conditions. *Opuntia* species is a multifunctional crop (fodder and human use), which can be of great value in both developed and underdeveloped countries.

### Activity of traditional South African sedative and potentially CNS-acting plants in the GABA-benzodiazepine receptor assay

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Numerous plants are traditionally used to treat mental diseases in South Africa. Fifty six ethanol extracts from 39 plants, both indigenous and exotic, that are traditionally used predominantly as sedatives or to treat various CNS-related ailments, were tested in the GABA<sub>A</sub>-benzodiazepine receptor binding assay, where the binding of <sup>3</sup>H-Ro 15-1788 (flumazenil) to the benzodiazepine site is measured. The GABA<sub>A</sub>-benzodiazepine receptor complex is involved in sedation, epilepsy and convulsions. Of the 56 extracts tested, 11 extracts showed activity. The most active extracts were the ethanolic leaf extracts of *Arctopus echinatus* (Apiaceae), *Helichrysum ruderale* (Asteraceae) and *H. umbraculigerum* (Asteraceae) which all showed good dose-dependent activity. None of the active compounds have been isolated.

## QSAR studies on acetylcholinesterase enzyme inhibitory effects of Amaryllidaceae alkaloids

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Twenty-three Amaryllidaceae alkaloids having several different ring types were evaluated for their acetylcholinesterase enzyme (AChE) inhibitory activity. The alkaloid 1-O-acetyllycorine (IC<sub>50</sub>: 0.96 ± 0.04M) showed significant AChE inhibitory activity, two-fold more than the currently used drug, galanthamine (IC<sub>50</sub>: 1.9 ± 0.16M). QSAR studies of these Amaryllidaceae alkaloids as acetylcholinesterase inhibitors were carried out using physico-chemical properties as descriptors. Multiple linear regression analysis of the data has shown that strain energy, molecular weight, heat of formation and substitution on both the aromatic ring and ring C play an important role in the development of the QSAR model. The contribution of substituents in ring C to the model was further supported when energy was omitted from the model and ring-type based QSAR analysis for crinine- and lycorine-type alkaloids were performed.

## Influence of cytokinins on the *in vitro* flowering of *Kniphofia leucocephala*

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The role of cytokinins in the promotion of flowering in the endangered species, *Kniphofia leucocephala* Baijnath, was investigated using shoots maintained in culture for three years. The highest percentage flowering (65%) was obtained on media containing 20:M benzyladenine (BA). The inclusion of isopentenyladenine and zeatin in the media also resulted in flowering, but these treatments were less effective than BA in inducing flowering. The effect of cytokinins on flowering was dose-dependent, with high concentrations of BA inhibiting flower formation slightly. Treatments that resulted in rooting of explants produced no flowers. The resulting inflorescences in all treatments did not mature and senesced prematurely.

## Sustainability of *Elaeodendron transvaalensis* harvesting in Venda region, Limpopo province

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*Elaeodendron transvaalensis* is one of the medicinal plants used very often by people in the Venda region. It is known to treat a variety of diseases. Due to its wide usage by traditional healers it has found its way into the multi markets and is amongst the seven most commonly-traded plants in the Venda region. This study investigates the impact of bark harvesting on this species.

## Bush encroachment on Owen Sitole College (OSCA) campus, KwaZulu-Natal, South Africa

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Aerial photographs (1937–2001) indicated that woody plant densities on OSCA have changed significantly. The lowland areas

near drainage lines have been wooded for the longest period compared to higher-lying areas further away from the drainage lines. Woody species increased rapidly since the area was fenced during the 1960s. Woody classification through TWINSpan identified two vegetation types with four plant communities and four sub-communities. Ordination with CANOCO separated the drier areas with high disturbance factors like fire, overgrazing and previously cultivated lands from the areas closer to the drainage lines and riverine areas.

## An investigation of callose formation due to aphid feeding in resistant and non-resistant cultivars of wheat – evidence from phloem tracer studies

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Wheat plants, *Triticum aestivum* var 'Betta DN', containing the Dn-1 gene for resistance to the Russian wheat aphid (RWA) as well as the controls, the near-isogenic susceptible cultivar Betta and old SST variety, were examined for callose deposition in the absence and presence of feeding aphids were examined specifically with respect to the formation of wound callose. Our results indicate that aphid feeding does indeed stimulate the formation of wound callose in the non resistant lines, suggesting that the plant's response to aphid feeding, is a long-term, wounding response in which callose deposition plays a central role. Application of 5,6-carboxyfluorescein diacetate (5,6-CFDA) to attached leaves, examined some 5h after application, showed little evidence of phloem transport of the cleaved and fluorescent 5,6-carboxyfluorescein (5,6-CF), below known aphid probed sieve tubes. Low levels, or absence of 5,6-CF suggests that, either the aphids have successfully redirected sap to themselves, or that the phloem is no longer functional. In Betta DN, 5,6-CF transport was still evident below sites of aphid probing, suggesting that the phloem was still capable of long-distance transport. The reduced callose formation in the DN variety suggests that transport through the phloem is not as affected as is the case in non resistant varieties.

## Vegetation mapping with the aid of satellite imagery – lessons learnt from a transect in central Namibia

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★ Awarded Local Organising Committee prize for best poster in Ecology

This study was a first attempt to produce a highly accurate remote sensing derived vegetation map based on LANDSAT-ETM data and detailed phytosociological vegetation sampling and descriptions from surveys during the pilot phase of the BIOTA-Africa project ([www.biota-africa.org](http://www.biota-africa.org)). A supervised maximum-likelihood-classification was performed on subsets of satellite scenes 178/75 and 178/74, covering the study area in the Thornbush Savanna and Karstveld in central Namibia. These scenes are representative of the late growing season (17 May 2000), dated one year before the actual vegetation survey started. Accuracy assessments of the resulting maps showed that we were not successful in delineating all of the desired vegetation classes, which was especially conspicuous where the satellite scenes overlapped. The main factors we identified for this low mapping accuracy were (a) several contrasting vegetation classes which have a similar structure and also near-identical spectral properties, (b) the number of relevés did not cover the full phytosociological and spectral variation of the identified vegetation classes along the transect, and (c) the vegetation along the investigated transect changes drastically. Hence differences in the overall vegetation

composition on both satellite images influences the classification result, in spite of using the same ground data input. As a result, the classification in the overlapping area of both scenes are not identical. We expect that using additional multi-temporal data will compensate for enormous inter-annual phenological differences of this overall heterogeneous, semi-arid vegetation. Likewise, non-parametric classification procedures as well as textural indices may

further enhance the discrimination between vegetation types of identical spectral properties and so strengthen the accuracy of the resulting vegetation map. These results highlight the need for the continued critical evaluation of methodologies employed in vegetation mapping, especially in regions where the delineation and description of vegetation units itself is extremely limited, as is the case in Namibia.