

Table of Contents

Organising Committee.....	2
SAAB Council.....	3
Paper and Poster Organisation.....	3
Maps.....	4
Parking and Access to UP	5
Internet Access.....	5
Students.....	5
Notice Board.....	6
Meetings.....	6
Lunches and Social Functions.....	7
Exhibitions.....	7
Contact Numbers.....	8
Transport, Accomodation & Travel.....	8
Sponsors.....	9
Scientific Programme.....	10
Plenary Speakers.....	26
Abstracts of Plenary Speakers.....	28
Abstracts of Oral Papers.....	32
Abstracts of Posters.....	126
Author Index.....	158

Organising Committee

The Organising Committee would like to welcome all delegates to the 38th conference of the South African Association of Botanists hosted by the Department of Plant Science, University of Pretoria.

Chairperson	Prof M Meyer
Secretary and registration	Ms R Swart, Ms E Pienaar
Treasurers	Prof M Meyer, Dr Q Kritzinger
Accommodation, refreshments & lunches	Dr M Deutschlander, Ms R Swart, Ms J Bapela, Ms E v Wyk, Ms M Nel
Social evening functions	Ms N Uys, Dr M Deutschlander, Dr K Kiviet, Ms R Swart, Ms E van Wyk
Circulars, final programme, abstracts, conference bags, name tags	Dr B Crampton, Prof N Lall, Mr H Heyman, Ms P Degashu
Scientific programme	Prof M Meyer, Prof B v Wyk, Prof D Berger, Prof N Lall, Prof M Rouget, Prof G v Rooyen, Dr E Tshikalange, Dr B Crampton, Mr H Heyman, Ms P Degashu
Venues, equipment, projectionists	Mr G More, Mr T Nembudani, Mr N Olivier, Dr E Tshikalange, postgraduate students
Transport	Mr T Nembudani, Ms R Swart
Sponsorships and publicity	Dr M Deutschlander
Information technology	Mr N Olivier, Ms N Uys, Mr G More
SAAB and Van Staden awards	Prof M Rouget, Prof G v Rooyen
Poster organisation	Ms P Degashu, Ms J Bapela, Dr Q Kritzinger

SAAB Council

Dr N Makunga (President)

Prof J Farrant (Vice-President)

Prof GF Smith (SANBI Nominee)

Prof K Balkwill

Dr C Bredenkamp

Dr G Prinsloo

Dr M Muasya

Dr SM Jacobs

Ex Officio Members

Dr Paul Hills - Honorary Treasurer

Permanent Secretary

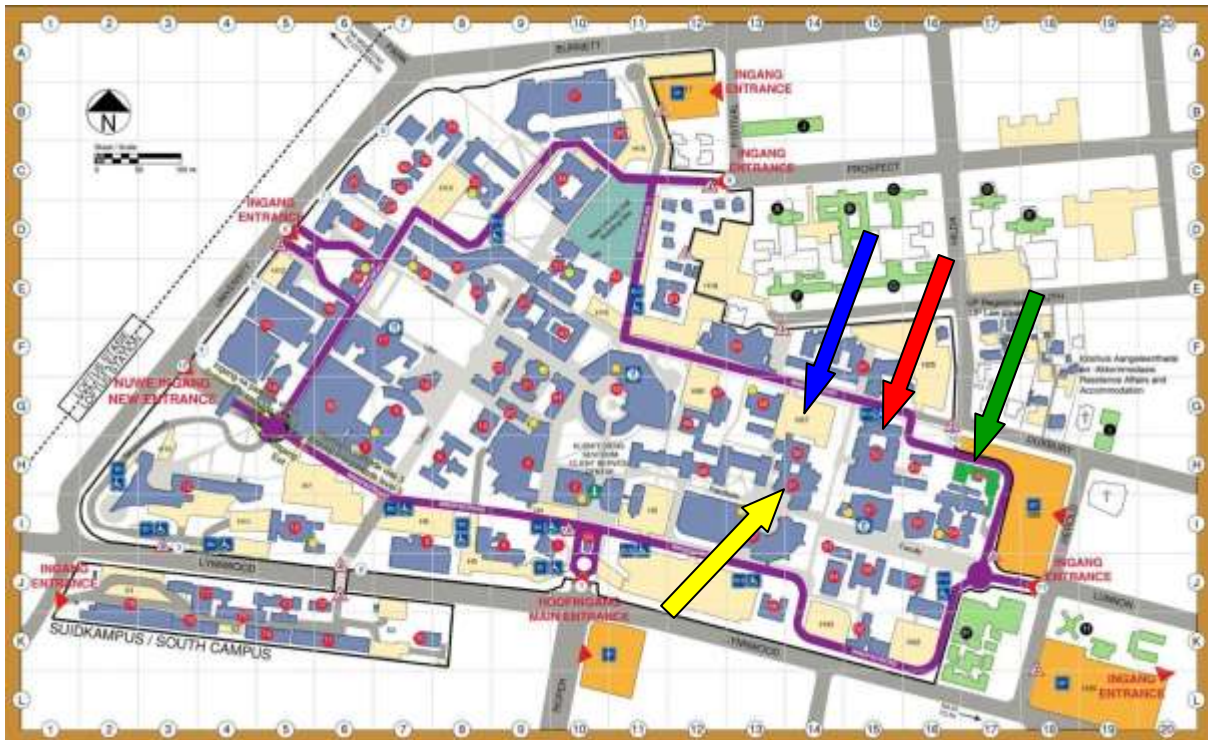
Mr Myke Scott (forsaab@telkomsa.net)

Paper and Poster Organisation

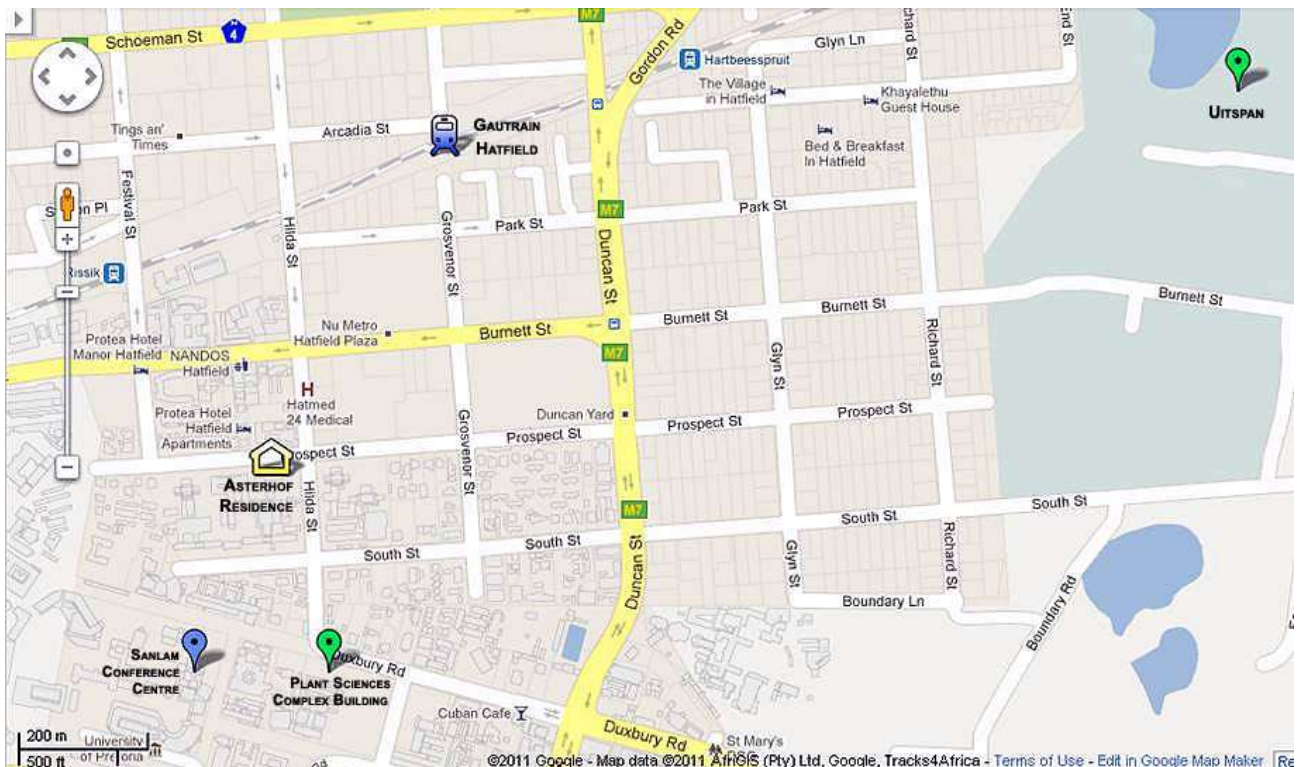
Presenters: Please submit your Powerpoint files as soon as possible, or at the latest, on 08:00 on the morning of your presentation to the assistant in Room D for distribution to the relevant lecture hall. Internet and computer facilities will also be available in Room D.

Posters will be displayed for the full duration of the conference. Delegates can put up their posters from 16:00 on Sunday, 15 January 2012. **PLEASE NOTE THAT PINS AND NAILS MAY NOT BE USED, ONLY VELCRO IS PERMITTED.** The poster session is scheduled for Monday, 16 January at 13:30.

Maps



Arrows: green = Plant Sciences Complex; blue = parking area for the conference; yellow = Sanlam Conference Centre; red = Klooster Dining hall



Parking and Access to UP

Parking for all delegates is available behind the Conference Centre (blue arrow on the map). The main entrance onto campus (c/o Lynwood Road/Roper Street) and the Prospect Road entrance are open 24 hours a day. The Lunnon Road entrance is open from 6 am until midnight only on weekdays.

Internet Access

Access to WiFi internet will be available in the conference venue and in the foyer of the Asterhof Hostel. The password can be requested at the registration desk.

Students

SAAB membership

All students registered at South African universities and attending the conference, will automatically be registered as SAAB members for 2012.

SAAB Awards

1. SAAB Young Scientist awards

Students of 30 years and younger (who are registered as SAAB members) and registered at a South African university are welcome to participate. One award is made to the best oral presentation by a young scientist.

2. Best MSc oral presentation award

One award is made to the best oral presentation by a MSc student.

3. Best PhD oral presentation award

One award is made to the best oral presentation by a PhD student.

4. Best poster presentation award

One award is made to the best poster presentation overall.

Notice Board

Please check the general notice board near Lecture Hall A for messages and announcements regarding changes to the programme etc. All personal messages received by the Secretariat will also be put on the notice board.

Meetings

SAAB Council Meeting

Date: Sunday 15 January

Time: 14:30

Venue: Plant Sciences Complex room 3-44

SAAB Annual General Meeting

Date: Wednesday 18 January

Time: 16:30

Venue: Lecture hall A

SAAB Council Special Meeting

Date: Wednesday 18 January

Time: Directly after the AGM

Venue: Lecture hall A

Lunches and Social Functions

Lunches will be served at the Klooster Hall's dining room to all persons who booked in advance. Delegates and registered companions are requested to wear their name tags at all times. The name tags and tickets found in conference bags will ensure admission to the lecture halls, tea, lunches and social functions.

Sunday 15 January

Meet-and-Greet

Time: 18:00

Venue: UP Conference Centre Foyer

Monday 16 January

Student Rumble@Outspan

Time: 19:00

Venue: OUTSPAN (LC de Villiers Experimental Farm)

Roof Top Braai for other delegates (non-students)

Time: 19:00

Venue: Tree House Tea Garden on the roof of the Plant Sciences Complex

Wednesday 18 January

SAAB Conference Dinner

Time: 19:00

Venue: UP Conference Centre Foyer

Exhibitions

Delegates are invited to visit the exhibitions of:

Separations

Umdauss Press (Pty)

Inqaba Biotechnical Industries

Labotec

Vividair

Lasec

Briza Publications

Contact Numbers

Police

Tel: 10111

Fire Brigade/Ambulance

Tel: (012) 310 6400

Tel: (012) 427 7111

Univeristy of Pretoria Campus Security

Tel: (012) 420 2310

Medical Services

HATMED Tel: (012) 362 7180

Transport, Accomodation & Travel

Agricultural Tours & Conferences Worldwide

Hestie Crous-Beckett

Tel: (012) 447-7718

Email: hestiecb@mweb.co.za

Roxburg Travel

Tel: 083 261 0455

Tel: 084 556 2304

Avis Rent-a-car

Tel: (012) 362 7073

Imperial Car Rental

Tel: (012) 342 6371

Europcar

Tel: (012) 342 3912

Sponsors

The kind support of the following sponsors is greatly acknowledged:



Scientific Programme

Papers will be presented in three different lecture halls (A, B and C). All the plenary lectures will be presented in Lecture Hall A (Sanlam Auditorium).

Presenters: Please submit your Powerpoint files as soon as possible or at the latest on 08:00 on the morning of your presentation to the assistant in Room D for distribution to the relevant lecture hall. Internet and computer facilities will also be available in Room D.

SUNDAY, 15 JANUARY 2012	
14:30	SAAB Council Meeting (<i>Plant Sciences Complex, room 3-44</i>)
17:00-19:00	Registration (<i>Conference Centre Foyer</i>)
18:00	Meet and Greet (<i>Conference Centre Foyer</i>)

MONDAY, 16 JANUARY 2012

07:45	Registration (<i>Conference Centre Foyer</i>)		
08:30	OPENING CEREMONY <i>Lecture Hall A (Sanlam Auditorium)</i> Prof Cheryl de la Rey – Vice Chancellor University of Pretoria Prof Nox Makunga – President of SAAB Prof Marion Meyer – Organising Committee Chairperson		
08:45			
08:55			
09:00	Plenary lecture: Prof Robert Verpoorte Metabolomics: A gateway to discoveries. Chairperson: Prof Marion Meyer		
10:00	TEA		
	<i>Lecture Hall A</i> Mon A1: Phytochemistry and Bioactivity Chairperson: Prof Kobus Eloff	<i>Lecture Hall B</i> Mon B1: Molecular Systematics and Barcoding Chairperson: Prof Ben-Erik van Wyk	<i>Lecture Hall C</i> Mon C1: Plant Biotechnology Chairperson: Dr Bridget Crampton
10:30	L.J. McGaw, T.J. Makhafola, O.O. Udom, K.T.V. Mayekiso and J.N. Eloff: Antimycobacterial activity, cytotoxicity and genotoxicity studies of <i>Ptaeroxylon obliquum</i> and <i>Sideroxylon inerme</i> leaf extracts.	K. Naidoo, M.P.A. Coetzee, E. Steenkamp, B. Slippers, M.J. Wingfield and B.D. Wingfield: Gene conversion of the Ribosomal RNA cistron.	B.J. Vorster, M.C. Goulet and D. Michaud: Plant cystatins and insect cysteine proteases: weapons in a molecular arms race.
10:45	H.M. Heyman, V. Maharaj, C. Kenyon and J.J.M. Meyer: Determination of anti-HIV active constituents of <i>Helichrysum</i> species by means of metabolomics.	A.R. Magee, A.N. Nicolas, P.M. Tilney, B.-E. van Wyk and G.M. Plunkett: Systematics of the early diverging Apiaceae tribe Annesorhizeae with emphasis on the South African genera.	I. Gazendam, D. Oelofse and D.K. Berger: Functional evaluation of a drought-induced “late embryogenesis abundant” protein gene from cowpea.
11:00	C.B. Oosthuizen, C. Hamilton and N. Lall: Potential of synthesized naphthoquinones for anti-tuberculosis activity.	P. Rousseau, P. Vorster, D. P. Little and M. van der Bank: A molecular phylogeny of <i>Encephalartos</i> Lehm.	N.M. Creux, M. Ranik, M.H. De Castro, A. Spokevicius, G. Bossinger, C. Maritz-Olivier and A.A. Myburg: Understanding the transcriptional regulation of the <i>Eucalyptus Cellulose synthase1</i> gene.

11:15	<u>A. Samie</u> , T. Nefefe, T. Mulaudzi and P.O. Bessong: Immunomodulatory effect of <i>Combretum molle</i> leaves water extracts on the production and expression of cytokines and toll like receptors.	<u>J. Gere</u> , O. Maurin and M. van der Bank: Phylogenetic relationships within subtribe Terminaliinae (Combretaceae).	<u>J. Bredekamp</u> , S. Naidoo and D.K. Berger: Challenging the dogma: Gene-for-gene tolerance, not resistance, to bacterial wilt in <i>Arabidopsis</i> .
11:30	<u>K.G. Mawela</u> , E. Chakauya and R. Chikwamba: Heterologous expression of RANTES analogues in <i>Lycopersicon esculentum</i> Mill.	<u>B.H. Daru</u> , O. Maurin, J.C. Manning, J.S. Boatwright and M. van der Bank: Phylogeny of the subfamily Alooideae (Asphodelaceae): Paraphyly of <i>Aloe</i> and <i>Haworthia</i> and consequences for classification.	<u>Maize eQTL project consortium</u> : Update on the maize eQTL project.
11:45	<u>S. Singh</u> and N. Lall: Evaluation of the mutagenicity and stability of plant extracts with potential use in the cosmetic field from the Myrsinaceae family.	<u>C.L. Taylor</u> and N.P. Barker: DNA fingerprinting of <i>Acacia karroo</i> : Attempting to resolve taxonomic uncertainties and inform ecological observations.	<u>M.M. van Dyk</u> , A.R.K. Kullan, E. Mizrachi, C.A. Hefer, L. Jansen van Rensburg, D. Newman, N. Coetzer, T.J. Tschaplinski, K.C. Cushman, N.E. Engle, G.A. Tuskan, N. Jones, A. Kanzler, A. Bayley and A.A. Myburg: Identifying genomic regions involved in growth, wood property, transcript and metabolite variation in an F ₂ pseudo-backcross pedigree of <i>Eucalyptus grandis</i> x <i>Eucalyptus urophylla</i> .
12:00	<u>R. Sharma</u> , A. Hussein and N. Lall: Evaluation of antibacterial, antioxidant and mechanistic activities of a plant from Myrtaceae family addressing acne vulgaris.	<u>P. Rousseau</u> , P. Vorster, D. P. Little and M. van der Bank: DNA Barcoding of Africa's endemic cycads: <i>Encephalartos</i> Lehm. and <i>Stangeria</i> T.Moore.	<u>I. van Jaarsveld</u> , E. Mizrachi, F. Joubert, Y. van de Peer and A.A. Myburg: Ensemble optimisation of <i>cis</i> -regulatory element discovery: <i>in planta</i> benchmark and discovery in <i>Eucalyptus</i> .
12:15	CONFERENCE PHOTO (Conference Centre Foyer)		
12:30	LUNCH		
13:30	POSTER SESSION		
14:30	TEA		

	Mon A2: Phytochemistry and Bioactivity Chairperson: Ms Johanna Bapela	Mon B2: Ecology - Pollination Biology Chairperson: Prof Sue Nicolson	Mon C2: Plant Physiology Chairperson: Dr Juan Vorster
15:00	<u>E.A. Adewusi</u> and V. Steenkamp: <i>In vitro</i> screening for acetylcholinesterase inhibition and antioxidant activity of medicinal plants from southern Africa.	<u>S.D. Johnson</u> : Using model and manipulated flowers to study pollinator-mediated selection.	<u>J. van Staden</u> and M.G. Kulkarni: Effects of smoke-water and smoke-derived stimulants on rooting.
15:15	J.S. Shelembe, D. Cromarty, M.J. Bester, A. Minnaar and <u>K.G. Duodu</u> : Characterisation of phenolic compounds in aqueous extracts from seed coats of marama bean (<i>Tylosema esculentum</i>) and their bioactive properties.	<u>G. Arena</u> , C. Symes and E.T.F. Witkowski: Pollination of <i>Aloe peglerae</i> - an endangered endemic of the Magaliesberg Mountains, South Africa.	B. Leboutteiller, C.W. King, J. Kossmann and <u>P.N. Hills</u> : Regulation of moss development by strigolactones.
15:30	<u>W. Cordier</u> , M. Gulumian, A.D. Cromarty and V. Steenkamp: Attenuation of <i>in vitro</i> oxidative stress by polyphenolic-rich fractions of <i>Burkea africana</i> and <i>Syzygium cordatum</i> .	<u>A. Heystek</u> and A. Pauw: The role of pollinators in the assembly of Erica communities.	<u>S. Figlan</u> , J.M. Kossmann and N.P. Makunga: Seed germination behaviour, micropropagation and <i>Agrobacterium</i> -mediated transformation of <i>Salvia runcinata</i> (L.f.): implications for conservation and cultivation.
15:45	<u>N. A. Masevhe</u> , A. Aroke, L.J. McGaw and J.N. Eloff: Evaluating antioxidant activity and cytotoxicity of the selected South African medicinal plant species.	<u>F. Martos</u> , T. Pailler, B. Bytebier and S. D. Johnson: Fly pollination in the achlorophyllous orchid <i>Gastrodia similis</i> : does <i>Gastrodia</i> provide a breeding site for its insect pollinator?	<u>M. Nakhooda</u> , M. P. Watt and D. Mycock: Phytohormone choice and the <i>in vitro</i> development of eucalypt roots.
16:00	<u>E. Muleya</u> , A.S. Ahmed, A.M. Sipamla, F.A. Mtunzi and J.N. Eloff: Antioxidant activities of nine medicinal plants used in treating inflammatory ailments in Zulu traditional medicine of South Africa.	<u>N. Hobbhahn</u> , B. Bytebier, S.D. Johnson and L. D. Harder: The role of resource limitation in the occurrence and persistence of rewardless pollination systems in <i>Disa</i> (Orchidaceae).	<u>A.O. Aremu</u> , M.W. Bairu, J.F. Finnie and J. van Staden: Influence of six aromatic cytokinins on the growth, phenolic and pigment contents of micropropagated banana (<i>Musa</i> spp. AAA cultivar 'Williams').
16:15	<u>M. Razwinani</u> , S.C.K.M. Motaung, T.E. Tshikalange, H.S. Abdillahi and J. van Staden: Anti-microbial and anti-inflammatory activities of <i>Pleurostyliya capensis</i> Turcz. (Loes) [Celastraceae].	<u>R.C. Turner</u> , J.J. Midgley and S.D. Johnson: Some Cape <i>Erica</i> species can self-pollinate: evidence for autogamy and geitonogamy, with a facultative and out-crossing seed-set advantage, in <i>Erica chloroloma</i> (Ericaceae).	<u>T. Mahlanza</u> , S.J. Snyman, M.P. Watt and R.S. Rutherford: Towards the <i>in vitro</i> generation of somaclonal variant plants of sugarcane (<i>Saccharum</i> spp. hybrids) for tolerance to <i>Fusarium sacchari</i>

			toxins.
16:30	<u>A.R. Ndhkala</u> , R. Anthonissen, G.I. Stafford, L. Verschaeve and J. van Staden: Toxicity studies on four <i>Hypoxis</i> species and a commercial herbal mixture.	<u>S. Lerch-Henning</u> and S.W. Nicolson: How do bird pollinators respond to nectar containing secondary metabolites?	<u>S.L. Beck-Pay</u> and K. Koen: The effect of temperature and relative humidity on <i>Acacia mearnsii</i> pollen viability.
16:45	Mon A3: Global Change Chairperson: Prof Mathieu Rouget <u>M. Masubelele</u> , M.T. Hoffman and W.J. Bond: Long-term vegetation change (1900-2010) with climate & land use as drivers change.	<u>R.J. Cozien</u> and S.D. Johnson: Better late than never? The high cost of self-incompatibility in <i>Aloe maculata</i> (Asphodelaceae).	<u>R. Sutherland</u> , A. Viljoen, A.A. Myburg and N. van den Berg: The effect of cold stress on infection of Cavendish bananas with <i>Fusarium oxysporum</i> f. sp. <i>ubense</i> .
17:00	<u>J. Puttick</u> , M.T. Hoffman and T. O'Connor: An analysis of woody cover change in the mesic eastern region of South Africa using repeat photography.	<u>I. Kiepiel</u> and S.D. Johnson: Breeding and pollination systems in <i>Clivia</i> (Amaryllidaceae).	<u>R. Janse van Rensburg</u> , G. Kemp, J.C. Pretorius and B. Visser: Proteomic analysis of wheat treated with a novel plant activator.
17:15			<u>B. Visser</u> , L.J. Szabo, L. Herselman and Z.A. Pretorius: Ug99 variants of <i>Puccinia graminis</i> f. sp. <i>tritici</i> in South Africa.
19:00	STUDENT RUMBLE@OUTSPAN (LC de Villiers Experimental Farm) ROOFTOP BRAAI FOR OTHER DELEGATES (Plant Sciences Complex)		

TUESDAY, 17 JANUARY 2012

08:30	<p><i>Lecture Hall A</i> Plenary Lecture: Prof Joseph Hirschberg Regulation of carotenoid biosynthesis and its connection to the biogenesis and function of plastids. Chairperson: Prof Dave Berger</p>		
	<p><i>Lecture Hall A</i> Tue A1: Plant Biotechnology Chairperson: Ms Nicky Creux</p>	<p><i>Lecture Hall B</i> Tue B1: Ecology - Pollination Biology Chairperson: Prof Steven Johnson</p>	<p><i>Lecture Hall C</i> Tue C1: Ethnobotany Chairperson: Prof Namrita Lall</p>
09:45	<p>P. Young, E. Alexandersson, D. Jacobson, J. Lashbrooke, Z. Coetzee, A. Deloire and <u>M. Vivier</u>: The molecular and metabolite profiling of grapevine berries in a model vineyard where the microclimate of the developing bunches has been altered.</p>	<p><u>S.W. Nicolson</u>: Bee food: the chemistry and nutritional value of nectar and pollen.</p>	<p><u>B.-E. van Wyk</u>: Ethnobotany of the Cederberg.</p>
10:00	<p>J.G. Lashbrooke, S.J. Dockrall, <u>P.R. Young</u> and M.A. Vivier: The Carotenoid Cleavage Dioxygenase (CCD) gene family in <i>Vitis vinifera</i> L.</p>	<p><u>C.W.W. Pirk</u> and H. Human: How healthy are our honeybees – Health check of a major pollinator.</p>	<p><u>M. Chinsamy</u>, J.F. Finnie and J. van Staden: Medicinal orchid research – a South African perspective.</p>
10:15	<p><u>M.C. Solofoharivelo</u>, A. van der Walt, J. McBride, D. Stephan, M.J. Freeborough, J.T. Burger and S. Murray: High throughput expression analysis of miRNAs and their targets involved in the water deficit response in <i>Vitis vinifera</i>.</p>	<p><u>R.J. Daniels</u> and C.I. Peter: On the wings of butterflies: unique floral morphology and flower orientation promotes cross-pollination in <i>Gloriosa superba</i>.</p>	<p><u>L.T. Mankga</u>, A.N. Moteetee, O. Maurin and M. van der Bank: DNA barcoding medicinal plants of South Africa.</p>
10:30	TEA		
	<p>Tue A2: Plant Biotechnology Chairperson: Dr Botma Visser</p>	<p>Tue B2: Ecology - Plant Invasions Chairperson: Dr Mark Robertson</p>	<p>Tue C2: Ethnobotany and Bioactivity Chairperson: Prof Namrita Lall</p>
11:00	<p><u>R. Naidoo</u>, S. Naidoo, D.K. Berger and A.A. Myburg: Elucidation and characterization of markers for the salicylic acid and methyl jasmonate pathways in <i>Eucalyptus grandis</i>.</p>	<p><u>J.R. Wilson</u>, P. Ivey, I. Nänni and P. Manyama: Biological invasion assessment and eradication-a national programme to reduce South Africa's invasion debt.</p>	<p><u>E.J. Mavundza</u>, R. Maharaj, J.F. Finnie, G. Kabera and J. van Staden: An ethnobotanical survey of mosquito repellent plants in uMkhanyakude district, KwaZulu-Natal province, South Africa.</p>

11:15	<u>H.D. Castelyn</u> , B. Visser and Z.A. Pretorius: Volatile emissions of <i>Puccinia triticina</i> infected wheat and its effect on uninfected wheat seedlings.	<u>R.F. Motlounq</u> , M.P. Robertson, M. Rouget and J.R. Wilson: The use of bioclimatic modelling in predicting the relative invasion success of introduced acacias in southern Africa.	<u>K.M. Mathibela</u> , <u>S.S. Semanya</u> , M.J. Potgieter, B.A. Egan, H.J. du Plessis and L.J.C. Erasmus: Bapedi traditional healers in the Limpopo Province, South Africa.
11:30	<u>T.T. Mhora</u> , R.S. Rutherford, D.L. Sweby, R.M. Jacob, L. Makome and J.W. Danson: Analysis of differential gene expression during the early stages of rust infection in a sugarcane breeding population.	<u>H.J.N. Sithole</u> : Dealing with newly detected invader: <i>Sagittaria platyphylla</i> .	<u>T.A. Netshivhulana</u> and M.P. Tshisikhawe: Perception and concept on diarrhea treatment amongst traditional healers of Vhembe District Municipality, Limpopo (RSA).
11:45	<u>C.N. Oates</u> , A.A. Myburg, B. Slippers and S. Naidoo: A hypothetical <i>Eucalyptus grandis</i> defence model against <i>Leptocybe invasa</i> based on transcriptome sequencing.	<u>T.P. Jaca</u> and H.M. Venter: Invasiveness of <i>Opuntia salmiana</i> – experimental study on its vegetative and sexual propagation.	<u>M. Adamu</u> , V. Naidoo and J.N. Eloff: Efficacy and toxicity of thirteen plants leaf acetone extracts used in ethnoveterinary medicine in South Africa on the egg and larva of <i>Haemonchus contortus</i> .
12:00	<u>R. Viljoen</u> , E.T. Steenkamp and G. Pietersen: Alternative indigenous Rutaceous hosts of ‘ <i>Candidatus Liberibacter africanus</i> ’ and ‘ <i>Candidatus Liberibacter africanus</i> subsp. <i>capensis</i> ’.	<u>O.K. Kambaj</u> , S.M. Jacobs and K.B. Mantlana: <i>In situ</i> and <i>ex situ</i> soil respiration in natural, invaded and cleared toposequences in the Fynbos Biome.	<u>K. Magwede</u> , M.P. Tshisikhawe and R.B. Bhat: Anti-microbial activities of indigenous medicinal plants used in treatment of tick infected cattle wounds in Vhembe District Municipality, Limpopo (RSA).
12:15	<u>K. Fitza</u> , A.A. Myburg, E.T. Steenkamp, K. Payn and S. Naidoo: Elucidation of defence responses associated with inducer application in <i>Pinus patula</i> .	<u>B.S. Bezeng</u> , O. Maurin, K. Yessoufou and M. van der Bank: What drives invasion success on Robben Island, South Africa?	<u>R. NemaKanga</u> and S.R. Sasa: The use of a locally developed innovation on biopesticide for pest management in dry beans in Limpopo Province.
12:30	LUNCH		

13:30	<p><i>Lecture Hall A</i> Plenary Lecture: Prof Petr Pysek From botanical data to macroecological patterns in invasion ecology. Chairperson: Prof Mathieu Rouget</p>		
	<p>Tue A3: Ecology - Plant Invasions Chairperson: Prof Mathieu Rouget</p>	<p>Tue B3: Plant Systematics and Comparative Morphology Chairperson: Mr Pieter Winter</p>	<p>Tue C3: Phytochemistry and Bioactivity Chairperson: Prof Alvaro Viljoen</p>
14:30	<p><u>D. Moodley</u>, J.R. Wilson, S. Geerts, D.M. Richardson and A.G. Rebelo: In the right place at the right time: why some introduced Proteaceae spread at some sites but fail at many others.</p>	<p><u>M.M. le Roux</u> and B.-E. van Wyk: The systematic value of flower structure in <i>Crotalaria</i> and related genera of the tribe Crotalarieae (Fabaceae).</p>	<p><u>O.O. Olaokun</u>, L.J. McGaw, J.N. Eloff and V. Naidoo: Antioxidant activity, total polyphenolic content and <i>in vitro</i> inhibition of activity of digestive enzymes significant to diabetes by leaf extracts of ten South African <i>Ficus</i> species.</p>
14:45	<p><u>M. Gaertner</u>, H. Nottebrock, H. Fourie, S.D.J. Privett and D.M. Richardson: Plant invasions, resilience, economics and restoration: Can fynbos pay for alien management?</p>	<p>A.R. Magee, <u>J.S. Boatwright</u> and L. Mucina: Taxonomic studies of <i>Ursinia</i>, parachute daisies, (Asteraceae, Anthemideae) in Namaqualand, South Africa.</p>	<p><u>A. Okem</u>, J.F. Finnie and J. van Staden: Anti-inflammatory, genotoxic and phytochemical properties of selected South African medicinal plants used in treating stomach-related ailments.</p>
15:00	<p><u>J.M. Kalwij</u>, M.P. Robertson and B.J. van Rensburg: Rapid colonisation by annual alien plants of a high montane roadside in the Drakensberg, South Africa.</p>	<p><u>P.P.J. Herman</u>: The taxonomic significance of the cypsela morphology in the genus <i>Nolletia</i> Cass. (Asteraceae) in southern Africa.</p>	<p><u>D. Berrington</u> and N. Lall: Anticancer activity of certain herbs and spices on the cervical epithelial carcinoma (HeLa) cell line.</p>
15:15		<p><u>R.R. Klopper</u>, O.M. Grace, A.E. van Wyk and G.F. Smith: <i>Aloe</i> section <i>Purpurascens</i>: the need for a new classification.</p>	<p><u>I. Vermaak</u> and A.M. Viljoen: Natural products in anti-obesity therapy – the good, the bad and the ugly.</p>
15:30	TEA		

	Tue A4: Ecology - Plant Invasions Chairperson: Dr John Wilson	Tue B4: Biodiversity Information and Classification of Mosses Chairperson: Mr Paul Herman	Tue C4: Phytochemistry and Biological Activity – Essential Oils Chairperson: Dr Gerhard Prinsloo
16:00	<u>G.D. Thompson</u> , D.U. Bellstedt, J.J. Le Roux, D.M. Richardson and J.R. Wilson: <i>Acacia saligna</i> invasions: genetics and species distribution modeling.	<u>N. Phephu</u> , A. Magee, J. van Rooy, D. Garcia-Avila and A.E. van Wyk: Evidence for a re-circumscription of the Thuidiaceae (Bryophyta) in Africa and the East African Islands.	<u>A.M. Viljoen</u> , M. Sandasi, J. Maree and N.P. Mncwangi: The application of vibrational spectroscopy and chemometrics in plant taxonomy.
16:15	<u>J.S. Boatwright</u> , M.T. Sethusa, O. Maurin, P. Ivey, M. Hamer and M. van der Bank: Progress towards DNA barcoding of invasive species in South Africa.	<u>F. Parker-Allie</u> : The South African Biodiversity Information Facility: Data mobilisation in South Africa.	<u>S. de Rapper</u> , S.F. van Vuuren, G.P.P. Kamatou and A.M. Viljoen: The additive and synergistic antimicrobial effects of Frankincense and Myrrh-Essential oils from the predynastic period.
16:30	<u>S. Geerts</u> , J.R. Wilson, D.M. Richardson, M. McGeoch, M. Gaertner, J.J. Le Roux, S. Kritzing-Klopper and C. Muofhe: <i>Banksia ericifolia</i> invading South Africa as predicted - a major threat or just symptom of a peculiar fire regime?	<u>R.H. Archer</u> : PRECIS, 35 years on.	<u>G.P.P. Kamatou</u> , M. Sandasi, S.F. van Vuuren, R.L. van Zyl and A.M. Viljoen: Quality control of commercially important essential oils and biological activities of selected oils from South African flora.
16:45	<u>W. Vardien</u> , D.M. Richardson, L.C. Foxcroft, J.R. Wilson and J.J. le Roux: Effective invasive species management around protected areas: Understanding the spatial dynamics of <i>Lantana camara</i> invasions in South Africa's Kruger National Park.	<u>P.J.D. Winter</u> : The transition of SANBI's herbaria from PRECIS to a BRAHMS database management system.	<u>K.M. Swanepoel</u> , P. Soundy and W.G. Alberts: Fragrances and flavours of some South African plants.
17:00	<u>S.N. Nzama</u> and H.J.N. Sithole: Early detection of the new potential invader; <i>Epipremnum aureum</i> (L.) Engl.	<u>M. Treurnicht</u> and I. Ebrahim: The CREW Programme: Making significant contributions to taxonomy and conservation.	<u>O.S. Soyngbe</u> , A.O. Oyedeji, M. Singh and A.R. Opoku: The chemical composition, antimicrobial and antioxidant properties of the essential oils of <i>Tulbaghia violacea</i> and <i>Eucalyptus grandis</i> .
FREE EVENING			

WEDNESDAY, 18 JANUARY 2012

08:30	<p><i>Lecture Hall A</i> Plenary Lecture: Prof Richard Cowling Desertification, carbon sequestration and job creation: the science behind the Subtropical Thicket Restoration Project. Chairperson: Prof Mathieu Rouget</p>		
	<p><i>Lecture Hall A</i> Wed A1: Ecology – Pollination Biology Chairperson: Prof Steven Johnson</p>	<p><i>Lecture Hall B</i> Wed B1: Bioactivity Chairperson: Dr Lyndy McGaw</p>	<p><i>Lecture Hall C</i> Wed C1: Floristics and Plant Anatomy Chairperson: Dr Glynis Cron</p>
09:45	<p><u>T. van der Niet</u>, R. Turner and J. Midgley: Pollinator ecotypes in the shrub <i>Erica plukenetii</i>.</p>	<p><u>J.N. Eloff</u> and G. Würger: Variation in biological activities of leaf extracts of 42 <i>Combretum molle</i> plants collected from different areas in Gauteng and Mpumalanga.</p>	<p><u>K.J. Duffy</u>: Probabilistic approaches to inferring plant extinctions from herbarium records.</p>
10:00	<p><u>A. Köhler</u>, C.W.W. Pirk and S.W. Nicolson: ‘Toxic nectar’: harmful and beneficial effects of nicotine on an important pollinator, the African honeybee.</p>	<p><u>S. Zonyane</u>, S. F. van Vuuren and N.P. Makunga: Pharmacological and phytochemical analysis of a medicinal plant mixture that is used as traditional medicine in Western Cape.</p>	<p><u>P. Cingo</u> and G.V. Cron: Floristic comparison of the northern and southern KwaZulu-Natal Drakensberg.</p>
10:15	<p><u>S. Steenhuisen</u> and S.D. Johnson: Breeding systems in <i>Protea</i>.</p>	<p><u>T.J. Makhafola</u>, L.J. McGaw and J.N. Eloff: Antibacterial, cytotoxic and mutagenic effects of five <i>Ochna</i> species.</p>	<p><u>P.M. Tilney</u>, A.E. van Wyk and C.F. van der Merwe: Do leaves play a more active role in the symbiotic relationship between domatia and mites?</p>
10:30	TEA		
	<p>Wed A2: Conservation Biology Chairperson: Prof Richard Cowling</p>	<p>Wed B2: Phytochemistry and Bioactivity Chairperson: Mr Heino Heyman</p>	<p>Wed C2: Plant Diversity Chairperson: Dr Glynis Cron</p>
11:00	<p><u>W.A. Haddad</u>, M. Rouget, A.E. van Wyk and J.E. Burrows: Towards an understanding of indigenous Afromontane forest regeneration and expansion in Buffelskloof Private Nature Reserve, Mpumalanga.</p>	<p><u>S.S. Semanya</u>, M.J. Potgieter and L.J.C. Erasmus: Species used by Bapedi Traditional Healers for the treatment of sexually transmitted diseases, Limpopo Province.</p>	<p><u>S.G. Mayonde</u>, G. Cron and M. Byrne: Genotypic and phylogeographic investigation of indigenous and alien <i>Tamarix</i> species in southern Africa.</p>
11:15	<p><u>M.L. van der Vyver</u>, R.M. Cowling, A. J. Mills, M. Difford and E.E. Campbell: Biodiversity and Restoration of Spekboom-</p>	<p><u>A.K. Ruiters</u>, B.-E. van Wyk, S.F. van Vuuren and P.M. Tilney: The ethnobotany, essential oil composition and</p>	<p><u>S.R. Dutta</u>: Phytogeography of Maharashtra: A taxonomic approach with reference to some</p>

	dominated thicket.	antibacterial activity of southern African <i>Teucrium</i> species (Lamiaceae).	selected species from India.
11:30	<u>L. Fourie</u> , M. Rouget and M.C. Lötter: An analysis of landscape connectivity of the Grassland Biome of Mpumalanga using graph theory.	<u>B.M. Sakong</u> , A.S. Ahmed, L.J. McGaw and J.N. Eloff: Isolation and characterization of compounds from <i>Calodendrum capense</i> and <i>Lydenburgia cassinoides</i> with antimicrobial potential against opportunistic pathogens.	<u>A.O. Bello</u> , O.T. Oladipo and S.A. Saheed: Numerical taxonomic study of some species of <i>Solanum</i> L. in Nigeria.
11:45	<u>K. Yessoufou</u> , O. Maurin, T.J. Davies, M. Kuzmina, H. Schafer, M. van der Bank and V. Savolainen: The role of megaherbivores in shaping the structure of subtropical plant communities.	<u>F.S. Botha</u> , C. van Wyk, V. Bagla and J.N. Eloff: The <i>in vitro</i> inhibitory effect of <i>Ptaeroxylon obliquum</i> (Thunb.) Radlk. on adhesion of <i>Candida albicans</i> to human buccal epithelial cells (HBEC).	<u>O.B. Ayeni</u> and S.A. Saheed: Morphological and leaf anatomical characters in relation to C ₃ and C ₄ photosynthetic pathways in some <i>Cyperus</i> species in South-Western Nigeria.
12:00	<u>S. Chen</u> , M.J. Wingfield, F. Roets and J. Roux: Identification, pathogenicity and population diversity of a new stem canker pathogen in the Cryphonectriaceae on <i>Rapanea melanophloeos</i> in South Africa.	<u>S.L. Stanton</u> , C.F. van der Merwe, A. Hussein, S.N. Venter, T.A. Coutinho and J.J.M. Meyer: A phytochemical and morphological comparison between <i>Pachystigma macrocalyx</i> , <i>P. pygmaeum</i> and <i>P. thamnus</i> .	<u>A.E. Folorunso</u> and O.S. Awosika: Comparative wood anatomy of six genera of the family Bignoniaceae in Nigeria.
12:15	<u>A. I. Odiwe</u> , A.A. Alimi, O. Ogunsanwo and R.O. Adewumi: Assessing biodiversity and nutrient status 10 years after reforesting a destroyed rain forest with <i>Tectona grandis</i> in Ile-Ife, Southwestern Nigeria.	<u>C.B.S. Hendriks</u> , J.J.M. Meyer and C.F. van der Merwe: Endophytes and their association with Dichapetalaceae.	<u>W.S.M. Gruezo</u> : Plant diversity of a lowland dipterocarp rainforest in Samar Island, Philippines.
12:30	LUNCH		
13:30	<i>Lecture Hall A</i> Plenary Lecture: Prof William Bond CO ₂ as a driver of global change in African ecosystems. Chairperson: Prof Mathieu Rouget		
	Wed A3: Global Change Chairperson: Prof Timm Hoffman	Wed B3: Plant Ecology Chairperson: Dr Miranda Deutschländer	Wed C3: Plant Physiology Chairperson: Dr Boney Kuriakose
14:30	<u>SAAB 2011 Best Young Botanist presentation:</u> <u>R.F. Powell</u> , M.T. Hoffman, L. Gillson and M. McGeoch:	<u>G. Prinsloo</u> : Allelopathic effects of <i>Amaranthus</i> .	<u>B. Ncube</u> , J.F. Finnie and J. van Staden: <i>In vitro</i> propagation of <i>Cyrtanthus</i> species.

	Long-term changes in the vegetation of Cape Point in response to climate and fire.		
14:45	<u>F.A. Engelbrecht</u> : Projections of regional climate change over southern Africa - the water balance in a warmer climate.	<u>O. Baloyi</u> , R.B. Bhat and M.P. Tshisikhawe: The germination and propagation potential of <i>Securidaca longepedunculata</i> Fresen.	<u>S.O. Amoo</u> , A.O. Aremu and J. van Staden: <i>In vitro</i> propagation and secondary metabolite production in <i>Aloe arborescens</i> : The role of aromatic cytokinins.
15:00	<u>B.S. Ripley</u> , R. Strauss and C.P. Osborne: Photosynthetic and anatomical acclimation by the C3 and C4 subspecies of <i>Alloteropsis semialata</i> in low CO2 atmospheres.	<u>S. Shaik</u> : In vitro germination of the threatened assegai tree, <i>Curtisia dentata</i> .	<u>S.G. van Wyk</u> , K.J. Kunert and B.J. Vorster: Characterisation of the cysteine protease and cysteine protease inhibitor family in soybean nodules.
15:15	<u>N. Stevens</u> , B.F.N. Erasmus, S. Archibald and W.J. Bond: Woody cover change in the north-east arid savannas of South Africa from 1940-present.	<u>M.P.A. Coetzee</u> , V.G. Muthelo, M.J. Wingfield and B.D. Wingfield: Ganoderma root rot: The scourge of <i>Jacaranda mimosifolia</i> in the “City of Jacarandas”.	<u>J.D. Hakizimana</u> , M. Gryzenhout, T.A. Coutinho and N. van den Berg: Root endophytes from <i>Persea americana</i> and their role against <i>Phytophthora cinnamomi</i> infection.
15:30	<u>J.A. van der Linde</u> , D.L. Six, M.J. Wingfield and J. Roux: Climate change-driven mortality of <i>Euphorbia ingens</i> in the Limpopo Province: Causal abiotic and biotic factors.	<u>T.P. Jaca</u> : <i>Iris pseudacorus</i> L. (yellow flag) has escaped cultivation in South Africa.	<u>R.O. Anyasi</u> and H.I. Atagana: Phytoremediation of polychlorinated biphenyls (PCBs) contaminated soil by <i>Chromolaena odorata</i> (L.) King & H.E. Robins.
15:45	<u>M. Whitecross</u> , S. Archibald and E.T.F. Witkowski: Is frost a demographic bottleneck for savanna trees? Testing the effect of freeze events on <i>Colophospermum mopane</i> population structure in Limpopo Province, South Africa.	<u>L. Jimu</u> , S. Chen, M.J. Wingfield, E. Mwenje and J. Roux: First report of <i>Coniothyrium</i> stem canker of eucalypts in Zimbabwe.	<u>M.M. From</u> and M. Landry: Employing biotechnology and cryobiological research to conserve threatened plant species.
16:00	<u>J.R. Pool</u> , B.S. Ripley and M. Powell: The carbon content of <i>Portulacaria afra</i> (L.) Jacq.	<u>J. Roux</u> and M.J. Wingfield: The rust pathogen <i>Puccinia psidii</i> , an eminent threat to South Africa.	<u>A.M. Al-Zaidi</u> and J. Khan: Molecular characterization of chilli leaf curl virus and satellite DNA associated with pepper in Oman.
16:15	TEA		
16:30	ANNUAL GENERAL SAAB MEETING (Lecture Hall A)		
	SPECIAL COUNCIL MEETING (directly after AGM)		
19:00	SAAB DINNER (Conference Centre Foyer)		

Poster Programme

POSTER NO.	PLANT ECOLOGY
E01	P.J. du Preez, <u>L.R. Brown</u> and L. Seipethlo: The peatlands of the Highlands of Lesotho – a unique aquatic system under threat.
E02	<u>C.H. Becker</u> , C. Coetsee and R.M. Cowling: Soil factors influencing the distribution of <i>Portulacaria afra</i> in subtropical thicket.
E03	<u>C-J. Thorne</u> , C. Gornall and C.I. Peter: How generalized is the pollination system of <i>Hypoestes aristata</i> ?
E04	<u>E. Davoren</u> , S.S Cilliers and S.J. Siebert: Socioeconomic gradients, garden design and plant diversity patterns in urban areas in the North-West Province and Gauteng.
E05	<u>T.P. Jaca</u> : Invasive <i>Crotalaria agatiflora</i> Schweinf. – ecology and distribution in South Africa.
E06	<u>R. Lalla</u> : Be warned! Invasive succulent, <i>Furcraea foetida</i> (Mauritius hemp) on the loose!
E07	L.E. Makwarela and <u>J.E. Victor</u> : South Africa's succulents under threat.
E08	<u>H.C. Mihiretu</u> , E. du Toit, J.M. Steyn and S.M. Laurie: Grafting method to improve vine production of orange fleshed sweet potato cultivar 'Resisto' under water stressed conditions.
E09	<u>B.T. Mthembu</u> and T.P. Jaca: A survey of invasive alien plants and their current distribution at Wonderboom Nature Reserve.
E10	<u>P. Masiagwala</u> , R.B. Bhat and M.H. Ligavha-Mbelengwa: Ecological, morphological and ethnobotanical assessment of <i>Acacia karroo</i> and <i>Acacia tortilis</i> at Nylsvley Nature Reserve, Limpopo Province South Africa.
E11	<u>I. Nänni</u> : <i>Hypericum pseudohenryi</i> – an invasive alien plant in the KZN Drakensberg and KZN Midlands.
E12	<u>M.M. Nxumalo</u> , H.J.N. Sithole and E. van Wyk: Surveillance of potential emerging wetland alien invaders.
E13	<u>L. Pretorius</u> , L. Brown, G. Bredenkamp, and T. Mostert: Characterization of wetland systems and –zones based on vegetation composition on the Maputaland Coastal Plain.
E14	<u>L. van der Walt</u> , S.S. Cilliers, K. Kellner, and M.J. du Toit: Landscape functionality and plant diversity in urban and rural grassland fragments in the Tlokwe Municipal area.

POSTER NO.	PLANT DIVERSITY
D01	<u>C.W. Beukes</u> , L. Mthombeni, E. van Zyl, S.N. Venter and E.T. Steenkamp: Description of novel <i>Burkholderia</i> species associated with the root nodules of indigenous Hypocalyppteae and Podalyriaceae.
D02	<u>G.V. Cron</u> : <i>Emilia</i> (Cass.) Cass. (Asteraceae, Senecioneae) in southern Africa.
D03	E.H. Biye, K. Balkwill and <u>G.V. Cron</u> : Sustaining <i>Gnetum</i> L. in Africa through improved taxonomy and domestication.
D04	<u>J. Grové</u> , M.P.A.Coetzee and E.T. Steenkamp: Identification of transposable elements in the pitch canker fungus, <i>Fusarium circinatum</i> .
D05	<u>R.R. Castillo</u> and W.SM. Gruèzo: Plant diversity of vegetation-over-ultramafic soil in Mount Bulanjao, Palawan Island, Philippines.
D06	<u>R.R. Castillo</u> and W.SM. Gruèzo: Plant Composition and Diversity of Forests-over-ultramafic soil in Mt.Victoria Range, Palawan Island, Philippines.
D07	<u>M. Koekemoer</u> , H.M. Steyn and S.P. Bester: A guide to plant families of southern Africa.
D08	<u>M. Mbenoun</u> , M.J. Wingfield, B.A.D. Begoude, B.D. Wingfield and J. Roux: Biodiversity of <i>Ceratocystis</i> species infecting wounds on elephant-damaged trees in the Kruger National Park.
D09	P.S. Mashile, <u>R.B. Bhat</u> , and M.P. Tshisikhawe: Variations in the epidermal structure of <i>Acacia tortilis</i> Hayne. grown in different climatic conditions.
D10	<u>T.E. Mphephu</u> and L.I. Ramovha: Comparative anatomy of the galls and equivalent healthy plant organs in <i>Terminalia sericea</i> (Combretaceae).
D11	<u>S. Situngu</u> and R.A. Lubke: A study of a population of a <i>Thamnochortus</i> species at the Sundays River, Eastern Cape.
POSTER NO.	PLANT BIOTECHNOLOGY AND PHYSIOLOGY
B01	M.W. Bairu, <u>A.O. Aremu</u> and J. van Staden: Stimulatory role of karrikinolide (KAR ₁) and smoke-water on photosynthetic pigment and phenolic contents of micropropagated 'Williams' bananas.
B02	P. Baskaran and <u>J. van Staden</u> : Somatic embryogenesis and plant regeneration from leaf explants of <i>Merwillia plumbea</i> (Lindl.) Speta.
B03	<u>J. Botha</u> , D. Pinard, N. Creux, S. Hussey, C. Maritz-Olivier, E. Mizrachi and A.A. Myburg: Promoter analysis of the <i>Eucalyptus grandis</i> secondary cell wall related <i>SND2</i> transcription factor gene.
B04	<u>N. Coetzer</u> , A.A. Myburg and D.K. Berger: Maize Microarray Annotation Database.
B05	<u>B.G. Crampton</u> and S.L. Murray: Micro RNA expression in maize leaves and flowers.

B06	<u>A. dos Santos</u> , E. Mizrachi, C.A. Hefer and A.A. Myburg: <i>De novo</i> assembly of the expressed gene catalog of <i>Equisetum ramosissimum</i> .
B07	O.O. Fajinmi, M.G. Kulkarni, J.F. Finnie and <u>J. van Staden</u> : Optimizing germination of <i>Coleonema album</i> seeds.
B08	<u>L. Ferreira</u> , S. Naidoo and A.A. Myburg: The application of salicylic acid to induce tolerance against <i>Chrysoporthe austroafricana</i> in <i>Eucalyptus grandis</i> clone ZG14.
B09	<u>A.T. Gill</u> , J.M. Farrant and M.S. Rafudeen: The identification of desiccation-responsive heat-stable proteins from the fronds of the desiccation tolerant form of the resurrection fern <i>Mohria caffrorum</i> .
B10	<u>J.N. Korsman</u> , T.G. Schmidt, B. Meisel, F.J. Kloppers, B.G. Crampton and D.K. Berger: Comparison between grey leaf spot lesion area and <i>Cercospora zeina</i> DNA content within maize leaves.
B11	<u>B. Kuriakose</u> , E. Viljoen, A. Killian, F. Carriero, S. Minoia, A. Bendahmane, and D.K. Berger: Diversity array analysis of <i>methyltransferase</i> (MET1) mutants of tomato.
B12	<u>B. Lombard</u> , C. Hefer, A. van der Merwe, E. Mizrachi and A.A. Myburg: Single nucleotide polymorphism (SNP) diversity in xylem and leaf expressed cellulose synthase genes of <i>Eucalyptus grandis</i> .
B13	<u>S.M. Reynolds</u> , M.M. van Dyk, M. Luo, E. Okogbenin, L.A. Becerra Lopez-Lavalle, M. Ferguson, A.A. Myburg and P. Rabinowicz: SNP-based genetic linkage maps of cassava (<i>Manihot esculenta</i>).
B14	<u>J.J. Scholtz</u> and B. Visser: Expression analysis of two genes during three different wheat- <i>Puccinia</i> interactions using Qpcr.
B15	<u>P. Singh</u> , S.G. Hussey, C. Maritz-Olivier, E. Mizrachi and A.A. Myburg: Expression and characterisation of <i>EgrSND2</i> , a <i>Eucalyptus grandis</i> transcription factor potentially involved in secondary cell wall regulation.
B16	<u>R. Smit</u> , E.S. du Toit and B.J. Vorster: Analysis of genetic variation in <i>Moringa oleifera</i> (lam.) using rapd and ssr's.
POSTER NO.	MEDICINAL PLANT SCIENCE
M01	<u>S.O. Amoo</u> , A.O. Aremu, M. Moyo and J. van Staden: Effects of long-term storage on antimicrobial and cyclooxygenase-1 inhibitory activities of South African medicinal plants.
M02	<u>M. de Canha</u> and N. Lall: Potential of two South African plants belonging to the <i>Greyiaceae</i> family for the treatment of skin hyper-pigmentation.
M03	<u>S. Emamzadeh-Yazdi</u> , A. Hussein, G. Prinsloo and Q. Kritzinger: Isolation and identification of compounds from an extract of <i>Strophanthus speciosus</i> (N. B. Ward. & Harv.) Reber., with emphasis on cardiac glycosides.
M04	<u>K.M. Mathibela</u> , B.A. Egan, H.J. du Plessis and M.J. Potgieter: Medicinal plants of the Blouberg Mountain, Limpopo Province, South Africa.
M05	<u>K.M. Mathibela</u> , B.A. Egan, H.J. du Plessis and M.J. Potgieter: The use of GIS in documenting medicinal plants of Blouberg mountain,

	Limpopo Province, South Africa.
M06	<u>T.M. Mulaudzi</u> and N.A. Masevhe: An inventory of indigenous medicinal plants used in the treatment of sexually transmitted infections in Vhembe district municipality, Limpopo Province. South Africa.
M07	<u>K.R. Phalandwa</u> and M.P. Tshisikhawe: Indigenous medicinal plants used by traditional healers in the stimulation of appetite of HIV/AIDS related patients in Vhembe District Municipality, Limpopo Province, South Africa.
M08	<u>T.E. Ramadwa</u> , E.E. Elgorashi and J.N. Eloff: Biological activity of methyl ursolate from <i>Funtumia africana</i> (Apocynaceae) leaf extracts.
M09	<u>S. Ramulifho</u> and N.A. Masevhe: An inventory of indigenous medicinal plants used in the treatment of thrush and related infections in Vhembe district municipality, Limpopo Province. South Africa.
M10	<u>S.S. Semanya</u> , L.J.C. Erasmus and M.J. Potgieter: <i>Catharanthus roseus</i> : Extraordinary Bapedi medicinal herb for gonorrhoea.
M11	<u>T. Tshilande</u> and M.P. Tshisikhawe: The treatment of persistent headache in HIV/AIDS related patients by the traditional healers of Vhembe district municipality, Limpopo Province (RSA).
M12	<u>C. van Wyk</u> , F.S. Botha, V. Steenkamp and J.N. Eloff: The effect of four medicinal plants on oral <i>Candida albicans</i> isolates.

Plenary Speakers

Professor William Bond

William Bond is a Professor in Botany at the University of Cape Town and holder of UCT's Harry Bolus Chair of Botany. He is an international expert on landcover change in Southern Africa and the ecology of sub-tropical grasslands, savannas and winter rainfall shrublands. His current research involves processes most strongly influencing vegetation change in the past and present, including fire, vertebrate herbivory, climate extremes, atmospheric CO₂ concentration and habitat fragmentation. He currently serves on the Editorial Boards of many scientific journals, including *Global Change Biology*, and *Global Ecology and Biogeography*.

Professor Richard Cowling

Richard Cowling is a professor in the Botany Department at Nelson Mandela Metropolitan University in the Eastern Cape. He has published extensively on the ecology and conservation of the fynbos, succulent Karoo and subtropical thicket biomes, and is widely acclaimed for his contribution to the theory and application of conservation science. He has published over 250 peer-reviewed items. Several awards have been conferred on him, including the National Research Foundation's President's Award (1987) and A- rating (1998 & 2003), Pew Fellows Program Award (1994), and the Society for Conservation Biology's Distinguished Service Award (2004).

Professor Joseph Hirschberg

Joseph Hirschberg is professor of the Department of Genetics at The Hebrew University of Jerusalem, Israel. Prof. Hirschberg's group pioneered the molecular analysis of the carotenoid biosynthesis pathway in plants, being the first to clone genes for carotenoid biosynthetic enzymes in cyanobacteria, algae and plants. Further studies elucidated the regulation of carotenoid accumulation in fruits and flowers and were imperative for genetically engineering plants to have altered carotenoid synthesis, such as in Golden Rice. Currently his laboratory uses a range of approaches including "classical" and molecular genetics, genomics and metabolomics. His research provides a paradigm of how natural diversity can be used to decipher complex biochemical pathways in plants. His research has been published in the top journals, including *PNAS*, *Plant Journal*, *Plant Physiology*, *Plant*

Cell etc. Prof Hirschberg is a lifetime honorary member of the American Society of Plant Biologists, and the most recent of his many awards was the The Trevor Goodwin Award for a Lifetime of Achievement in Research on Carotenoid Biochemistry and Biology from the International Carotenoid Society.

Professor Petr Pysek

Petr Pysek is the Deputy Director of the Institute of Botany of Academy of Sciences of the Czech Republic and the Head of the Department of Invasion Ecology. He has published extensively on the flora of Europe and the ecology of invasive plants, and he is a leading expert in invasion biology. He has received several awards, including the creativity award from the Charles University in 2010.

Professor Robert Verpoorte

Robert Verpoorte is the Head of Pharmacognosy, section Metabolomics at Leiden University in the Netherlands. He is author/co-author of 650+ scientific papers, 4 books and 4 patent applications and is Editor-in-chief of Journal of Ethnopharmacology and Phytochemical Reviews, Executive Editor of Biotechnology Letters and he serves on the editorial boards of 22 journals. His current research interests are mainly plant cell biotechnology, in particular the production of fine chemicals (including drugs) by means of plant cell cultures. He also works on the biosynthesis of secondary metabolites in plants and NMR-based metabolomics. He received an Honorary Doctorate from University of Amiens, France (2004) and the Phytochemical Society of Europe Medal in 2007.

Abstracts of Plenary Speakers

CO₂ as a driver of global change in African ecosystems

W. Bond

Botany Department, University of Cape Town, South Africa

Savannas evolved under low atmospheric CO₂ which favoured C4 grasses over their C3 predecessors. Over the last century, CO₂ has increased to levels exceeding those recorded in palaeo-atmospheres over at least the last million years. In the coming century they are expected to increase to levels last seen in the Eocene, more than 30 million years ago. These anthropogenic increases in CO₂ are likely to have profound effects on African ecosystems and especially C4 grassy ecosystems. Here I review studies of the contributions of increasing CO₂ to vegetation change in African ecosystems. Simulation studies, glasshouse experiments, and long term field experiments point to significant CO₂ effects on woody plant expansion, especially in higher rainfall savannas. The future of the C4 grassy biomes in Africa looks very uncertain in a high CO₂ world.

Desertification, carbon sequestration and job creation: the science behind the Subtropical Thicket Restoration Project

R.M. Cowling^a, A. Mills^b, A. Sigwela^a, S. Pierce^a, M. van der Vyver^a and C. Marais^c

^aRestoration Research Group, Department of Botany, P.O. Box 77000, Nelson Mandela Metropolitan University, Port Elizabeth, South Africa 6031

^bRestoration Research Group, Department of Soil Science, Stellenbosch University, Private Bag X1, Matieland 7602, South Africa

^cNatural Resource Management, Department of Environment Affairs, Parliament Towers, 103 - 107 Plein Street, Cape Town 8000, South Africa

Subtropical Thicket ecosystems dominated by *Portulacaria afra* (spekboom) store amounts of carbon comparable to forest ecosystems that receive up to three times their annual rainfall. This remarkable finding – an outcome of SANBI's Conservation Farming project – led to the initiation of research on the potential to finance the restoration of degraded spekboom thicket via the emerging carbon economy. The research, which is funded the Natural Resources Management Programme of the Department of Environment Affairs, was conceptualised as a monitoring and evaluation programme, in order to facilitate social learning and adaptive management for catalysing large-scale, biome-wide restoration in the private sector. This programme was formalised as the Subtropical Thicket Restoration Project (STRP), a learning organization comprising managers and researchers. Here I describe a decade of biophysical research that has culminated in the validation of the world's first carbon sequestration project involving the restoration of a desertified ecosystem. The STRP has successfully catalysed an initiative that is consistent with the South African government's commitment to create employment via a green economy.

Regulation of carotenoid biosynthesis and its connection to the biogenesis and function of plastids

J. Hirschberg

Department of Genetics, Alexander Silberman Institute of Life Sciences, The Hebrew University of Jerusalem, Jerusalem, 91904 Israel

Carotenoid pigments are essential components of the photosynthetic apparatus and thus are present in all green tissues of plants. These molecules are synthesized within plastids from the central isoprenoid pathway by enzymes that are nuclear encoded. In addition to their primary functions in photosynthesis, carotenoids play essential roles in plant reproduction by furnishing flowers and fruits with distinct pigmentation and as precursors for volatiles and aroma compounds that attract animals. Two phytohormones, abscisic acid (ABA) and strigolactones, are produced from carotenoids.

We are studying carotenoid biosynthesis and its regulation in flowers and fruits of tomato (*S. lycopersicum*), which has become a model system for chromoplast-containing plants. Over the years we have developed various genetic tools to decipher carotenogenesis in plants by cloning and analyzing genes that encode enzymes of the pathway. To this end, we have isolated novel mutations in tomato that alter pigmentation of flowers and fruit. Through characterization of these mutations we have identified new enzymes in the carotenoid biosynthesis pathway. Recent results from characterization of mutations demonstrated the importance of RedOx to the biosynthesis of carotenoids and revealed a link between carotenoid biosynthesis and plastid biogenesis.

From botanical data to macroecological patterns in invasion ecology

P. Pyšek

Institute of Botany, Academy of Sciences of the Czech Republic, Průhonice & Department of Ecology, Faculty of Science, Charles University in Prague, Czech Republic

Invasion ecology, as a field studying historical processes and their current outcomes, crucially depends on centuries of regional botanical tradition. The talk will explore how primary botanical data can be used to infer about macroecological patterns and processes of plant invasions at various scales, hence contribute to the invasion theory and our understanding of habitat invasibility, species invasiveness and spread, as well as factors determining these characteristics. Experiences with building national and continental databases of alien species in Europe, by using botanical literature and regional checklists will be also discussed.

Metabolomics: a gateway to discoveries

R. Verpoorte, N. Yuliana, H.K. Kim, and Y.H. Choi

*Natural Products Laboratory, Institute of Biology Leiden,
Leiden University, PO Box 9502, 2300RA Leiden, The Netherlands*

Metabolomics has developed in a major tool in all types of research in the past decade. Functional genomics, plant resistance, quality control of food and botanicals, you name it. The basis of metabolomics is making unbiased observations with highly reproducible analytical tools, followed by a biostatistical analysis to find correlations between all the available data. That is a systems biology approach, which may lead to new discoveries. This can be illustrated by the identification of the active compounds in medicinal plants. By measuring the metabolome of different extracts, accessions or fractions of a medicinal plant and combining these data with those of biological activity, signals related to the compounds related to activity can be found. That may include prodrugs and synergy in case of in-vivo experiments. To be able to measure all metabolites present in e.g. a plant, we developed comprehensive extraction as a way to rapidly identify the active compounds in a plant. NMR-based metabolomics of the fractions obtained with this method and combining these data with adenosine receptor binding activity data allowed the identification of flavonoids as the active compounds in *Orthosiphon stamineus* leaves.

A very different but quite exciting discovery we made through the NMR-based metabolomics was the Natural Deep Eutectic Solvents (NADES). The data we collected in NMR-based metabolomics made us to ask the question “why are a few very simple molecules always present in considerable and similar amounts in the spectra of any organism?” They must have a basic function in living cells. They include sugars, amino acids (e.g. proline, alanine, glutamine, asparagine), choline, and organic acids (e.g. malic, lactic, succinic acid). Sugars serve for storage and energy, the other compounds are in amounts that does not make sense to consider them only as metabolic intermediates. We found that mixtures of organic acids with bases form ionic liquids, whereas neutral solids may form deep eutectic solvents, e.g. sugars with choline or malic acid. NADES have a polarity like ethanol, and are excellent solvents for natural products, including DNA and proteins, often with orders of magnitude higher solubility than in water. In our hypothesis many cellular and physiological functions are connected with the occurrence of NADES in nature.

Abstracts of Oral Papers

Efficacy and toxicity of thirteen plants leaf acetone extracts used in ethnoveterinary medicine in South Africa on the egg and larva of *Haemonchus contortus*

M. Adamu^a, V. Naidoo^b and J.N. Eloff^a (Tue C2)

^a*Phytomedicine Programme, Department of Paraclinical Sciences, Faculty of Veterinary Science University of Pretoria*

^b*University of Pretoria Biomedical Research Centre*

In this study thirteen plants leaf acetone extracts used traditionally in ethnoveterinary medicine in South Africa were evaluated using the egg hatch assay and the larval development test. Cytotoxicity of these plants was also evaluated using the MTT cellular assay. Extracts of three plant species i.e. *Heteromorpha trifoliata*, *Maesa lanceolata* and *Leucosidea sericea* had EC₅₀ values of 0.62 mg/ml, 0.72 mg/ml and 1.08 mg/ml respectively for the egg hatch assay. Other plants with good anthelmintic activity include *Clausena anisata*; 1.08 mg/ml and *Clerodendrum glabrum*; 1.48 mg/ml. In the larval development test *H. trifoliata* extract was the best with EC₅₀ of 0.64 mg/ml followed by *L. sericea* 1.27 mg/ml. The activities in the larval development test were generally lower in most plant species compared to the egg hatch assay. Based on the cytotoxicity results *C. anisata* was the least toxic with an LC₅₀ of 171.86 µg/ml, while *Cyathea dregei* was the most toxic plant with an LC₅₀ of 3.32 µg/ml. The selectivity index of the thirteen plants shows *C. anisata* as the best with a value of 95.48 and 83.02 for both assays, this was followed by *H. trifoliata* and *L. sericea* with values of 68.58, 66.44 and 47.69, 40.55 for both assays and plant species respectively. The plant species with the worst S.I was *C. dregei* with a value of 0.19 for both assays. The result of this study confirms a scientific basis for the folkloric claim of the natives of South Africa in the use of these plants for the treatment of helminthiasis in livestock. The possibility of finding a compound with good anthelmintic activity from the most efficacious and less toxic plants will be evaluated. Further study will involve the fractionation of selected plants and evaluating their anthelmintic activity.

***In vitro* screening for acetylcholinesterase inhibition and antioxidant activity of medicinal plants from southern Africa**

E.A. Adewusi and V. Steenkamp (Mon A2)

Department of Pharmacology, School of Medicine, Faculty of Health Sciences, University of Pretoria, Private Bag X323, Arcadia 0007, South Africa

Neurodegenerative disorders primarily affect the elderly population. Alzheimer's disease, the most common neurodegenerative disorder, is associated with a deficiency in levels of the neurotransmitter acetylcholine as well as increased generation of reactive oxygen species (ROS). The aim of the study was to determine the acetylcholinesterase inhibitory (AChEI) and antioxidant activity of the ethyl acetate and methanol extracts of 12 traditional medicinal plants used in the treatment of neurological disorders. AChEI activity was determined spectrophotometrically using the Ellman's colorimetric method. Antioxidant activity was carried out by determining the ability of the extracts to scavenge the 2,2-diphenyl-1-picryl hydrazyl (DPPH) and 2,2'-azinobis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS) radicals. The levels of total phenols, flavonoids and flavonols were determined quantitatively using spectrophotometric methods. AChEI was observed to be dose-dependent.

Lannea schweinfurthii (Engl.) Engl. and *Scadoxus puniceus* (L.) Friis & I. Nordal. root extracts showed the lowest IC₅₀ value of 0.0003 mg/ml for the ethyl acetate extracts while *Zanthoxylum davyi* (I. Verd.) P.G. Watermann had the lowest IC₅₀ value of 0.01 mg/ml for the methanol extracts in the AChEI assay. The roots of *Piper capense* L.f., *L. schweinfurthii*, *Ziziphus mucronata* Willd., *Z. davyi* and *Crinum bulbispermum* (Burm.f.) Milne-Redh. & Schweick. showed noteworthy radical scavenging activity and good AChEI activity. Five plants showed good antioxidant and AChEI activity. These findings support the traditional use of the plants for treating neurological disorders especially where a cholinesterase mechanism and ROS are involved.

Molecular characterization of chilli leaf curl virus and satellite DNA associated with pepper in Oman

A.M. Al-Zaidi and J. Khan (Wed C3)

Department of Crop Sciences, College of Agricultural & Marine Sciences, Sultan Qaboos University, P.O. Box-34, Al-Khod, Sultanate of Oman

Pepper (*Capsicum annum*) is cultivated in the coastal region of Al-Batinah, A'Sharqiya and Dhofar regions in the Sultanate of Oman during the winter season to meet the high demand for fresh produce in the domestic market. To identify the causal agent of a widespread disease associated with infestations of the whitefly *Bemisia tabaci* (Genn.), leaves were collected from sweet pepper plants showing symptoms characteristic of the begomovirus disease in Al-Batinah and Dhofar during 2010 and 2011. Total nucleic acids were isolated from the pepper leaves and used as the template for Φ 29 DNA polymerase amplification of begomoviral circular DNA. Putative full unit length begomoviral DNA multimers were digested with *Pst*I and cloned into the plasmid vector pUC19. The complete nucleotide (nt) sequence was determined as 2758 base pairs (bp), indicative of a monopartite begomoviral genome. A comparison of the genome sequence for the ten field isolates examined indicated that they shared 98 – 99% nt identity. The virus from Oman was most closely related to ChLCV-Multan at 96% nt identity, a monopartite begomoviral isolate described previously from Pakistan. Based on the guidelines of the ICTV the Oman isolate has been designated ChLCV-Om and is considered a strain of ChLCV-Multan. A satellite DNA (DNA β), was amplified by polymerase chain reaction using degenerate primers and cloned, and the DNA sequence was determined. Analysis of the complete nt sequence of 1327 bp indicated that the DNA β shared 96% similarity with its closest relatives, which are TYLCV Al-Batinah DNA β molecules isolated from tomato in Oman. This is the first report of ChLCV from Oman and DNA β associated with the ChLCV-Om isolate. The ChLCV-Om and associated TYLCV Al-Batinah DNA β thus represent a begomovirus-complex at the Asian-Middle East crossroads that uniquely share geographical and genetic hallmarks of both.

***In vitro* propagation and secondary metabolite production in *Aloe arborescens*: The role of aromatic cytokinins**

S.O. Amoo, A.O. Aremu and J. van Staden (Wed C3)

Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa

Aloe arborescens is well known for its medicinal and horticultural values. Plant biotechnology offers an efficient tool for the production and modification of important plant secondary metabolites. A basic requirement for the transformation process is the development of a simple yet efficient propagation protocol. The choice of plant growth regulators (PGR), especially cytokinins, remains a critical factor in developing an efficient micropropagation system. In the present study, we investigated the effects of five different concentrations each of seven aromatic cytokinins (6-benzyladenine, *meta*-topolin and their derivatives) on *in vitro* shoot regeneration and secondary metabolite production in *Aloe arborescens*. All the treatments produced adventitious shoots with high frequency ranging from 86.7 to 100%. The highest number of adventitious shoots produced per shoot-tip explant (7.3 ± 0.71) was observed in Murashige and Skoog (MS) medium supplemented with 5 μM benzyladenine riboside after six weeks of culture. Variable amounts of iridoids, phenolics and flavonoids were present in all the treatments. Unlike with most of the cytokinin treatments, no proanthocyanidin was detected in regenerated plants cultured on PGR-free MS medium. In general, higher antioxidant and radical scavenging activities were observed in regenerated shoots produced in cytokinin-containing media when compared to the PGR-free medium. Our findings indicate that exogenous supply of different types and concentrations of aromatic cytokinins during micropropagation markedly influences *in vitro* production of bioactive secondary metabolites.

Phytoremediation of polychlorinated biphenyls (PCBs) contaminated soil by *Chromolaena odorata* (L) King and Robinson

R.O. Anyasi^a and H.I. Atagana^b (Wed C3)

^a*Department of Environmental Sciences University of South Africa, Pretoria, South Africa, 0003*

^b*Institute for Science and Technology Education, University of South Africa, Pretoria, South Africa, 0003*

The ability of *Chromolaena odorata* propagated by stem cuttings and grown for six weeks in the greenhouse to thrive in soil containing different concentrations of PCB congeners found in Aroclor and transformer oil (TO), and to possibly remediate such soil was studied under greenhouse conditions. *Chromolaena odorata* plants were transplanted into soil containing 100, 200, and 500 $\mu\text{g g}^{-1}$ of Aroclor 1254 and 1260, and soil containing 100, 200, and 500 ml kg^{-1} transformer oil (TO) in 1L pots. The experiments were watered daily to maintain 70% moisture at field capacity. Parameters such as fully expanded leaves per plant, shoot length, leaf colour as well as the root length at harvest were measured. *C. odorata* growth was differently affected by the different concentrations of transformer oil. The level of inhibition to plant growth increased with concentration. However, the Aroclor amended soil did not affect the plant. At the end of six weeks of growth, plants showed a diminished effect in TO amended soil to the parameters tested. Plants size was increased by 1.4, 0.46 and

-1.0% in 100, 200 and 500mg/kg respectively. In Aroclor amended samples, 45.9, 39.4 and 40.0% were plants sizes at different concentrations. Such trend was observed in the leaf numbers and root length. Leaf colour was pale green in TO samples but, middle green in Aroclor amended soils. The control sample has 43.3% increase in plant size which was not significant among the values in Aroclor treated soils, an indication that *C. Odorata* could survive PCB contamination as to remediate it. The result of the on-going GC-analysis would be presented in the main paper and would be used to establish the extent of remediation.

PRECIS, 35 years on

R.H. Archer (Tue B4)

National Herbarium, South African National Biodiversity Institute, Pretoria, Private Bag X101, South Africa 0001

Under the directorship of Bernard de Winter at the Botanical Research Institute in the 1970's, the then innovative National Herbarium, Pretoria (PRE) Computerised Information System (PRECIS) was planned and become operational. Encoding of 470 000 mainly southern African and type specimens began in June 1975 and was completed in December 1976. The system was maintained on a Burroughs 7800 mainframe at the Department of Agriculture. This first version was soon completely restructured and in April 1982 PRECIS II was implemented. Today, PRECIS is integral to most activities at PRE and NBG where it has expanded to more than 1 110 000 specimen records and more than 60 000 taxon name records. A pc based version, PRECIS.pc is used at NH but remains separate from PRECIS itself. Additionally, PRECIS.pc has been implemented at many smaller herbaria both within South Africa and 10 southern African countries as part of the 10 year SABONET project (1996 to 2005). We present a historical and critical review of intervening implementations of PRECIS on four different computer and software systems. At present we are preparing for the fifth migration on to the internationally acclaimed BRAHMS database (Botanical Research and Herbarium Management System) based at Oxford, England. It is time to ask the following questions: Who were the main role-players behind PRECIS? Did PRECIS achieve its initial expectations? What was the impact of PRECIS on herbarium management and research at PRE and NBG and within SANBI, and on botany in South Africa in general?

Influence of six aromatic cytokinins on the growth, phenolic and pigment contents of micropropagated banana (*Musa* spp. AAA cultivar 'Williams')

A.O. Aremu, M.W. Bairu, J.F. Finnie and J. van Staden (Mon C2)

Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa

The effect of benzyladenine (BA) and five topolins (*meta*-Topolin = *mT*; *meta*-Topolin riboside = *mTR*; *meta*-Methoxy topolin = *MemT*; *meta*-Methoxy topolin riboside = *MemTR* and *meta*-Topolin 9-tetrahydropyran-2-ylpurine = *mTTHP*) on the growth, phenolic and photosynthetic pigment contents of 'Williams' bananas were evaluated. Explants were cultured for 42 days on modified Murashige and Skoog media containing 10, 20 and 30 μM of the above aromatic cytokinins (CK). Using spectrophotometric methods, phenolic and pigment contents were quantified from the 50% methanol and acetone extracts, respectively. Analysis of growth parameters indicate that the use of 30 μM *mT* resulted in the highest

shoot multiplication rate (7 shoots/explant). Shoot length and fresh weight per plantlet were significantly lower in all the treatments compared to the control. However, at 30 μM CK treatments shoot dry weights were significantly higher than the control, with the highest being treatment with MemT. The lowest abnormality frequency was observed in 10 μM mTR regenerants. Treatments with 10 μM mT (25.3 ± 4.98) and mTTHP (27.0 ± 3.30) had the highest number of roots/explant. These treatments stimulated significantly higher total phenolic content in aerial parts of the plantlets. Conversely, 30 μM mTTHP favoured accumulation of the same secondary metabolites in the underground parts. Accumulation of these metabolites would probably enhance plant acclimatization. In terms of photosynthetic pigments (chlorophyll a, b and carotenoids), CK treatments brought no improvement in the regenerated plantlets. Studies on endogenous CK profiles and effect of the aromatic CKs on genetic stability are currently underway.

Pollination of *Aloe peglerae* - an endangered endemic of the Magaliesberg Mountains, South Africa

G. Arena, C. Symes and E.T.F. Witkowski (Mon B2)

School of Animal, Plant and Environmental Sciences, University of Witwatersrand, Private Bag 3, Johannesburg, 2050, South Africa

Aloe peglerae is a winter flowering succulent endemic to the Magaliesberg Mountains. It is listed as Endangered and is vulnerable to illegal collecting from the wild. The attractive inflorescence and the height of the raceme make it conspicuous to pollinators. Flowers produce nectar of low concentration (9.9%) and relatively high volumes (17.5 μl), hence we hypothesised that effective pollination is facilitated through avian-nectar feeding only. Pollinator exclusion experiments were used to determine contributions to fruit and seed set by different pollinator guilds. Three treatments were applied to sixty 'focal' aloes; 1) control - open to all floral visitors, 2) bird exclusion – mesh cage allowing only insect visitors, and 3) total exclusion – fine mesh netting excluding all floral visitors. Average % fruit set with bird and total exclusions was low (12.1 and 5.3%, respectively), while control plants had by far the highest fruit set (40.4%, $P > 0.05$), suggesting that birds are the major pollinators for *A. peglerae*. Similarly, seed set per fruit under bird and total exclusion were also much lower (16.9 and 15.7%, respectively) than in control plants (38.3%; $P < 0.0001$). Finally, total seed production per plant was much lower in bird and total exclusions (497 and 322, respectively), compared with the control (3868; $P < 0.0001$). Nectar production increased from morning to midday when feeding rates of birds was higher, resulting in nectar standing crop volume decreasing during the afternoon (28.8 μl). There was a slight increase in standing crop from the morning (29 μl) to midday (38.1 μl). However, nectar concentration remained constant through the day (9.7-10.2% w/w). Understanding the pollination biology of *A. peglerae*, together with information on its population demography, will contribute significantly to conserving this species.

Morphological and leaf anatomical characters in relation to C₃ and C₄ photosynthetic pathways in some *Cyperus* species in South-Western Nigeria

O.B. Ayeni and S.A. Saheed (Wed C2)

Department of Botany, Obafemi Awolowo University, Ile-Ife, BC220005 Nigeria

The genus *Cyperus* (Cyperaceae) is a taxonomically complex one containing species involved in both C₃ and C₄ photosynthetic pathways. The current study is a structure – function investigations where whole plant morphology and leaf anatomical characters are used to explain basic physiological and ecological functions among the species of *Cyperus* considered in addition to their delimitations along the different photosynthetic lines. Morphological characters used in the delimitation includes: type and arrangement of inflorescence as well as the length of spikelets. Anatomical characters examined includes: the presence or absence of kranz tissue since a species is classified as C₄ when kranz tissue is present and C₃ when kranz anatomy is absent. Other characters includes interveinal distances, maximum cell distant count using the “one cell distant criterion”, maximum lateral cell count, leaf thickness and mesophyll thickness. Our results clearly demonstrate the usefulness of these anatomical characters in elucidating the infrageneric affiliations of these species along the different photosynthetic pathways as well as the mechanism behind the physico-ecological expressions of these species.

The germination and propagation potential of *Securidaca longepedunculata* Fresen

O. Baloyi, R.B. Bhat, and M.P. Tshisikhawe (Wed B3)

Department of Botany, University of Venda, Thohoyandou 0950

Securidaca longepedunculata Fresen. is commonly known as the violet tree of the polygalaceae family. *S. longepedunculata* as a medicinal plant, is used in both Western and Southern Africa to treat a wide range of ailments. Because of its many uses, the plant is now faced with unsustainable harvesting pressure which in the long run may lead to the plant being extinct. Therefore, there is a need to implement cultivation tools to save this valuable, medicinal plant. In this study, we studied the germination potential of *S. longepedunculata* under three germination factors; temperature, light and soil depth. Seeds were subjected to three pre-treatment; Mechanical scarification, soaking in distilled water overnight and soaking in 3.5% m/v domesticated bleach for 60 minutes. Optimal temperatures for seed germination were 20°C and 30°C. With regard to soil depth, high germination percentage was obtained at soil depth of 4 cm, with seedling emergence of 30%. Both depth 2cm and 8 cm had seedling emergence of 10% respectively and only 3% at 6 cm. No seedlings emerged at 10cm level. Based on the findings, it is concluded that soil depth has an effect on seedling emergence and *S. longepedunculata* responds well to soil depth of 4 cm.

The effect of temperature and relative humidity on *Acacia mearnsii* pollen viability

S.L. Beck-Pay and K. Koen (Mon C2)

Institute for Commercial Forestry Research, P.O. Box 100281, Scottsville, Pietermaritzburg 3209, South Africa

Acacia mearnsii (black wattle) is a commercially important forestry species in South Africa, grown for its timber as well as its bark. It is, however, also considered to be an alien invader of indigenous vegetation and for this reason the production of a sterile variety would be highly desirable for commercial forestry in South Africa. Previous research on crosses between diploid and tetraploid parent plants to produce triploid progeny has resulted in poor seed set. One possible barrier preventing seed set could be the viability of the pollen used in the cross pollination operations. Thus a study was conducted to test the pollen viability. *In vitro* agar media germination tests (ACIAR and Brewbaker and Kwack media) together with vital stain tests (Sigma[®] DAB peroxidase and p-phenylendiamine) were used to test pollen germination and viability of *A. mearnsii* pollen. These were then compared to *in vivo* pollen germination on the stigma. Results showed that the vital stain tests gave significantly ($p < 0.05$) higher pollen viability (59.53 and 60.67 %) than the agar germination tests (11.92 and 24.50 %) and were more in agreement with the results from the pollen germination rate on the stigma (94.59 %). In 2011 pollen was subjected to various temperature and relative humidity combinations, to simulate conditions recorded within the isolation bags being used in the 2010 flowering season. The results from this study together with a comparison of pollen viability results across three seasons (2009, 2010 and 2010) will be discussed.

Numerical taxonomic study of some species of *Solanum* L. in Nigeria

A.O. Bello, O.T. Oladipo and S.A. Saheed (Wed C2)

Department of Botany, Faculty of Science, Obafemi Awolowo University, Ile-Ife, BC 220005 Nigeria

Numerical taxonomic techniques were used to evaluate the taxonomic status of the genus *Solanum* in the family Solanaceae in order to resolve the conflicting taxonomic issues within the genus *Solanum*. Fifty-eight characters were studied and observations were made from ten species. Principal Component Analysis (PCA) and Single Linkage Cluster Analysis (SLCA) were employed to elucidate the relationship among the taxa of the genus. Similarity matrix and dendrogram were constructed. The distinct anatomical and morphological characters which indicate close inter-relationship among the species include; presence of anisocytic to anomocytic stomata, amphistomatic leaf surfaces with lower number of stomata on the adaxial surface, presence of glabrous or pubescent epidermal surfaces, fruit type, anther colour, leaf arrangement as well as shape of the leaf. Erect stem is peculiar to members of the genus except *S. nigrum* with weak stem. Principal Component Analysis factor loading of the characters showed that foliar morphological and anatomical characters such as leaf base, leaf margin, stomata types and anticlinal wall patterns are important in separating and delimiting the taxa studied. In conclusion, that data obtained from this work taken along with data from other sources, can be used to enhance proper taxonomic characterisation of members of the genus *Solanum*.

Anticancer activity of certain herbs and spices on the cervical epithelial carcinoma (HeLa) cell line

D. Berrington and N. Lall (Tue C3)

Department of Plant Sciences, University of Pretoria, Pretoria 0002, South Africa

Acetone extracts, of nine herbs and one spice, were prepared and tested for their cytotoxic ability, *in vitro*, against a noncancerous African green monkey kidney (Vero) cell line and an adenocarcinoma cervical cancer (HeLa) cell line. The plants studied were: *Origanum vulgare* (Oregano), *Rosmarinus officinalis* (Upright rosemary and groundcove rosemary), *Lavendula spica* (Lavender), *Laurus nobilis* (Bay leaf), *Thymus vulgaris* (Thyme), *Lavendula-x-intermedia* (Margaret Roberts Lavender), *Petroselinum crispum* (Curly leaved parsley), *Foeniculum vulgare* (Fennel) and *Capsicum annuum* (Paprika). Of these plants only *L. nobilis* and *O. vulgare* exhibited pronounced cytotoxic effects on the HeLa cell line. Dose-dependent studies revealed fifty percent inhibitory concentrations (IC₅₀) values of 34.46 µg/ml and 126.3 µg/ml on the HeLa cell line and IC₅₀ of 124.1 µg/ml and 163.8 µg/ml on the Vero cell line for *L. nobilis* and *O. vulgare* respectively. The cytotoxic ability was measured using XTT (Sodium 3'-[1-(phenyl amino-carbonyl)-3,4-tetrazolium]-bis-[4-methoxy-6-nitro] benzene sulfonic acid hydrate) colorimetric assay. Antioxidant activity was determined using DPPH (1,1-Diphenyl-2-picryl hydrazyl). Both *L. nobilis* and *O. vulgare* showed good free radical scavenging capability with IC₅₀ values of 30.85 µg/ml and 26.43 µg/ml respectively. Light microscopy (eosin and haematoxylin staining) and confocal microscopy (Hoechst 33342, acridine orange and propidium iodide staining) were used to evaluate the mechanism of action of the acetone leaf extracts of *L. nobilis* and *O. vulgare*.

What drives invasion success on Robben Island, South Africa?

B.S. Bezeng^{a,b}, O. Maurin^{a,b}, K. Yessoufou^{a,b}, M. van der Bank^{a,b} (Tue B2)

^a*African Centre for DNA Barcoding, University of Johannesburg, P. O. Box 524 Auckland Park, 2006 Johannesburg, South Africa*

^b*Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa*

Robben Island was declared a world heritage site in 1999. It lays 11 km from the port of Cape Town with a surface area of about 500 ha. Natural ecosystems on the Island have been severely degraded and altered by the introduction of various fauna and alien vegetation. The woody shrub component of the original West Coast Strandveld vegetation has been eradicated and the diversity of the herbaceous component has been reduced, with major invasion by alien plant species occurring since 1933 with apparent little invasion prior to this period. Currently a new management plan for the natural environment of Robben Island has been developed. The main goal of this study is to determine what drives invasion success on Robben Island. Our main results will be discussed.

Progress towards DNA barcoding of invasive species in South Africa

J.S. Boatwright^{a,b}, M.T. Sethusa^c, O. Maurin^c, P. Ivey^d, M. Hamer^e and M. van der Bank^c (Tue A4)

^a*Compton Herbarium, South African National Biodiversity Institute, Private Bag X7, Claremont, 7735, Cape Town, South Africa*

^b*Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa*

^c*African Centre for DNA Barcoding, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa*

^d*Early Detection and Rapid Response Unit, South African National Biodiversity Institute, Private Bag X7, Claremont, 7735, Cape Town, South Africa*

^e*Biosystematics Division, South African National Biodiversity Institute, Private Bag X101, Pretoria 0001, South Africa*

South Africa's rich biodiversity, with ca. 20 456 species, is under constant threat mainly through agriculture, urbanization, habitat loss and encroachment of alien invasive species. More than ca. 660 naturalized plant species are currently known to be contributing to the widespread transformation of once pristine habitats in the country. Furthermore, an array of invasive animal species have also established feral populations. The early detection of invasive species and rapid response for eradication relies on accurate species level identification. This is often difficult as, outside their native range, there is a lack of herbarium and museum records, regional specific literature and expertise to aid in the identification of these species. In light of the challenges opposing accurate identification, alternative solutions need to be explored. DNA barcoding is one such tool that may aid in the identification of unknown material (especially when only incomplete material is available). This requires the use of short, highly informative DNA regions to discriminate between species. This lecture reports on a collaborative project between the Early Detection and Rapid Response Programme of the Working for Water Programme and South African National Biodiversity Institute (SANBI) and the African Centre for DNA Barcoding (ACDB) at the University of Johannesburg, funded by the Natural Resource Management Programme (Department of Environmental Affairs) which was started in August 2011. This project will form part of the International Barcode of Life Project (iBOL) and aims to collect invasive plant and animal samples in South Africa as well as South African species known to be invasive elsewhere in the world over an eight month period for DNA barcoding. The team consists of, apart from the co-ordinators, seventeen collaborators from 10 universities and research institutions in the country. Twenty-four recent graduates have been appointed at many of these institutions to undertake the collection and processing of samples for DNA barcoding.

The *in vitro* inhibitory effect of *Ptaeroxylon obliquum* (Thunb.) Radlk. on adhesion of *Candida albicans* to human buccal epithelial cells (HBEC)

F.S. Botha, C. Van Wyk, V. Bagla and J.N. Eloff (Wed B2)

Phytomedicine Programme, Department of Paraclinical Sciences, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, ONDERSTEPOORT, 0110, South Africa

Candidal adherence to human buccal epithelial cells (HBEC) is the critical initial step in the pathogenesis of oral candidiasis, which may eventually lead to a systemic infection. Since the ability of *C. albicans* to form a germ tube and hyphae is an indication of replication and growth, its adherence to HBEC can be seen as a step in the colonisation of buccal epithelial cells in oral candidiasis, because the organisms cannot be removed by natural cleaning mechanisms' in the oral cavity. The ability of *C. albicans* standard strain (ATCC 10231) and two clinical isolates to adhere to healthy buccal epithelial cells (HBEC) in the presence of *Ptaeroxylon obliquum* (Thunb.)Radlk., a medicinal plant used for several pharmaceutical purposes, were examined. Leaves of *P. obliquum* were harvested, dried and ground to powder. Extract of powdered leaves of *P. obliquum* were extracted with acetone at room temperature and 250 mg/ml *P. obliquum* acetone leaf extract was used as the starting concentration for the subsequent study. *P. obliquum* acetone leaf extract possessed antifungal activity towards *C. albicans* standard strain (ATCC 10231) and two clinical isolates. Adhesion of *P. obliquum* acetone leaf extract to HBEC was concentration dependent and exhibited marked inhibitory effect on the ability of *C. albicans* strains to adhere to healthy buccal epithelial cells which were evident at higher concentrations. Light microscopy images showed that *P. obliquum* acetone leaf extract had an effect on the adhesion of *C. albicans* to HBEC. Where no inhibitory effect was observed, hyphae formation or germ tube formation (budding) was evident. In some cases it was observed that the *C. albicans* cells were destroying the ultra structure of the epithelia cells. This study show the presence of substances in *P. obliquum* acetone leaf extracts with potential inhibitory effect on the adhesion of *C. albicans* to HBEC.

Challenging the dogma: Gene-for-gene tolerance, not resistance, to bacterial wilt in *Arabidopsis*

J. Bredenkamp^a, S. Naidoo^b and D.K. Berger^a (Mon C1)

^a*Department of Plant Science, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa*

^b*Department of Genetics, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa*

Ralstonia solanacearum, the causal agent of bacterial wilt, has been found to infect the host plant *Arabidopsis thaliana*. Our current understanding of this pathosystem is that *A. thaliana* ecotype Kil-0 shows gene-for-gene "resistance" to an African *Eucalyptus* isolate of *R. solanacearum*, BCCF402. However, a recent preliminary study indicated that Kil-0 may exhibit "tolerance" which is defined as supporting high pathogen load without symptoms or a reduction in fitness. Here, we report results from a more in depth study. As expected high bacterial numbers (1×10^{13} colony forming units/gram fresh weight) and severe disease symptoms were observed in Be-0 plants. Surprisingly, high concentrations (1×10^{12} colony forming units/gram fresh weight) of BCCF402 were found in Kil-0 plants displaying little or

no disease symptoms. The bacterial load of *R. solanacearum* was quantified in Kil-0 and Be-0 using quantitative PCR. The *cytC* gene region was used to quantify *R. solanacearum* in *A. thaliana* plants and the amount of bacterial DNA was normalized to “alien” DNA that was spiked into each sample. A small difference in the relative quantification of bacterial DNA to “alien” DNA was observed between the Kil-0 and Be-0 ecotypes, supporting the tolerance hypothesis. Kil-0 plants inoculated with BCCF402 were not significantly affected in terms of their seed fitness and biomass yield compared to control Kil-0. These results illustrate that Kil-0 is, in fact, tolerant to *R. solanacearum* isolate BCCF402.

Volatile emissions of *Puccinia triticina* infected wheat and its effect on uninfected wheat seedlings

H.D. Castelyn, B. Visser and Z.A. Pretorius (Tue A2)

Dept Plant Sciences, UFS, PO Box 339, Bloemfontein, 9300

Plants emit a vast array of different volatile organic compounds to which surrounding plants can respond. Recent research indicated a putative volatile signalling event between leaf rust (*Puccinia triticina*) infected and uninfected wheat (*Triticum aestivum*). In the current project uninfected wheat was exposed to volatiles emitted by leaf rust infected wheat in a continual air flow system. Volatile exposure was done for different combinations of susceptible (Thatcher) and resistant (Thatcher+*Lr9*) wheat lines. The induction of a plant defence response in the uninfected wheat was confirmed on phenotypical, biochemical (β -1,3-glucanase activity) and gene expression (*PR2* gene) levels. This defence activation in the exposed uninfected plants could be attributed to the released volatiles. Volatile profiles emitted by the infected wheat were determined and the results will be discussed.

Identification, pathogenicity and population diversity of a new stem canker pathogen in the Cryphonectriaceae on *Rapanea melanophloeos* in South Africa

S. Chen^a, M.J. Wingfield^a, F. Roets^a and J. Roux^a (Wed A2)

^a*DST/NRF Centre of Excellence in Tree Health Biotechnology (CTHB), Forestry and Agricultural Biotechnology Institute (FABI), Department of Microbiology and Plant Pathology, Private Bag X20, University of Pretoria, Hatfield, Pretoria, 0028, South Africa*

^b*Department of Conservation Ecology and Entomology, Stellenbosch University, Stellenbosch, 7600, South Africa*

Rapanea melanophloeos (Myrsinaceae), commonly known as Cape Beech, is native to Africa and forms an important component of afro-montane forests. During disease surveys in the Western Cape Province of South Africa, a serious stem canker disease on *R. melanophloeos* trees was observed in the Harold Porter National Botanic Garden. Infection on the trees often results in the death of branches and entire stems above the cankers. Fruiting structures typical of fungi in the Cryphonectriaceae were observed on the surfaces of cankers. The aim of this study was to identify the causal agent of this disease and obtain information on its possible origin. Multiple gene analyses of the partial *LSU* and *ITS* nuclear ribosomal DNA, and two regions of the β -*tubulin* gene, showed that the fungus represents a previously undescribed genus and species in the Cryphonectriaceae. Phylogenetic analyses, as well as

morphological comparisons suggest that it is most closely related to the genus *Microthia*. Stem inoculations indicated that the fungus is an aggressive pathogen of *R. melanophloeos* trees, with the ability to kill inoculated stems within six weeks. Based on vegetative compatibility groups (VCGs), the diversity of the pathogen in the Harold Porter National Botanic Garden is low, suggesting that it may have been introduced from outside the area where it is currently found.

Medicinal orchid research – a South African perspective

M. Chinsamy, J.F. Finnie and J. van Staden (Tue C1)

Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa

Orchidaceae, the largest Angiosperm family, has a worldwide distribution of an estimated 35 000 species and 796 genera. Approximately 75% of the ±1500 species occurring in southern Africa are endemic to the region and occupy very restricted distribution ranges. The relatively small number of species used in African traditional practices (±49 species) may represent a more refined list of the most used species and/or genera. Generally, orchids are fiercely protected on the basis that they are rare and under threat from excessive collection. However, earlier threats such as collection for cultivation and horticulture and more recent threats such as loss of habitat and urbanization pose more of a risk to orchid populations. The conservation status of most medicinally used orchid species are Least Concern and could either reflect a negligible impact by collection for medicinal uses or it could indicate a lack of knowledge, or both, on such orchids. One of the gaps in our knowledge of South African orchids is the ethnobotanical, pharmacological and phytochemical usage of medicinal orchids. Understanding the usefulness of, and possible threats to, traditionally used orchid species would enable increased protection of these orchids and their habitats. Factors influencing use, distribution and the conservation status of medicinally used orchids will be addressed, together with a summary of the pharmacological and phytochemical assessment of some South African exploited orchid species.

Floristic comparison of the northern and southern KwaZulu-Natal Drakensberg

P. Cingo and G.V. Cron (Wed C1)

School of Animal, Plant and Environmental Sciences, University of Witwatersrand, P Bag 3, WITS, 2050

The KwaZulu-Natal Drakensberg forms part of the Drakensberg Alpine Centre above 1800 m, which is renowned for its species richness and high levels of endemism. Within the KZN Drakensberg two regions have been recognized: the south-east facing southern region and northern region with a north-east aspect and their floras are expected to be different. The aim of the study was to compare the northern KZN Drakensberg to the southern KZN Drakensberg floristically and to add to the knowledge of the flora of the northern KZN Drakensberg by collecting in a relatively under collected region. The PRECIS data set was used (supplemented with Ezemvelo KwaZulu-Natal Wildlife data) and a floristic comparison was done on family, genera and species composition. The composition of the top ten families

was found to be very similar, but they differ in ranking except for the two largest families Asteraceae and Poaceae and three largest genera: *Helichrysum*, *Senecio* and *Erica*. The northern KZN Drakensberg appears to be more diverse with 1692 species compared to the southern Drakensberg which supports 1284 species above 1800 m. The southern KZN Drakensberg has, however been more thoroughly studied and has a greater number of collections compared to the northern KZN Drakensberg. Results from the chi-squared test show no significant difference between northern and southern KZN Drakensberg flora. 557 specimens, including 321 species in 170 genera and 59 families were collected from the Injisuthi region (northern KZN Drakensberg); with majority of collections in the altitudinal zone(s) 1800 – 1900 m and 1900 – 2025 m. Two new species were added to the known flora: *Aspilia natalensis* (Asteraceae) and *Bothriochloa insculpta* (Poaceae). Most plants were noted to flower in late-spring to early /mid-summer.

Ganoderma root rot: The scourge of *Jacaranda mimosifolia* in the “City of Jacarandas”

M.P.A. Coetzee^a, V.G. Muthelo^b, M.J. Wingfield^b and B.D. Wingfield^b (Wed B3)

^a*Department of Genetics, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa*

^b*Department of Microbiology, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa*

Pretoria is adorned by thousands of *Jacaranda mimosifolia* trees. The city is rightly referred to as the “City of Jacarandas”, and covered during spring in a cloud of purple. Large numbers of these trees are dying in the suburb of Brooklyn due to a root and butt rot disease apparently caused by a species of *Ganoderma*. Fruiting bodies of *Ganoderma* resembling those in the “*G. lucidum*” complex are commonly found at the bases of dying trees every year after the onset of rain in spring and early summer. The “*G. lucidum*” complex includes several paraphyletic groups that are considered to represent different species with similar morphology; the true identity of the fungus causing the disease on Jacaranda trees is therefore unknown. The aim of this study was to characterise the species of *Ganoderma* that is found on Jacaranda trees dying from this root rot disease based on morphological characteristics and DNA sequence comparisons. Basidiocarps were collected from infected trees and their morphology was examined. Fungal isolations were made and cultural characteristics including growth habit, colour and presence of chlamydospores were determined. DNA was extracted from the isolates and sequences obtained for the ITS regions (ITS-1, 5.8S gene and ITS-2), the IGS-1 region as well as the mitochondrial small subunit (mtSSU) gene. Morphological observations revealed that all of the isolates represented a species that was similar to those in the *G. lucidum* complex. DNA-based phylogeny confirmed that these isolates are closely related to *G. lucidum*. The gene phylogenies were, however, not congruent. Although a specific identification could not be made, the results of this study show that *G. lucidum* is a phylogenetically variable species complex and that the *Ganoderma* sp. commonly found associated with root rot disease of Jacaranda trees in Brooklyn resides in *G. lucidum sensu lato*.

Attenuation of *in vitro* oxidative stress by polyphenolic-rich fractions of *Burkea africana* and *Syzygium cordatum*

W. Cordier^a, M. Gulumian^b, A.D. Cromarty^a and V. Steenkamp^a (Mon A2)

^a *Department of Pharmacology, Faculty of Health Sciences, School of Medicine, University of Pretoria, P.O. Box X323, Arcadia, 0007, Pretoria, South Africa*

^b *Department of Toxicology, National Institute for Occupational Health, P.O. Box 4788, 2000, Johannesburg, South Africa*

Excessive production of ROS and free radicals in cells results in oxidative stress, a state which is involved in the aetiology of diseases such as diabetes, neurodegeneration and cardiomyopathy. Antioxidant supplementation has been suggested as a prophylactic measure to control disease progression. Due to potential toxicity with synthetic antioxidant usage natural alternatives are becoming ever-popular. *Burkea africana* Hook.F (Fabaceae) and *Syzygium cordatum* Hochst. ex C.Krauss (Myrtaceae) are both used ethnomedically for oxidative stress-related diseases. Plant extracts were fractionated through liquid-liquid extraction and assessed for polyphenolic content, antioxidant activity, inherent cytotoxicity and potential to reverse oxidative stress-induced parameters. Polyphenolic content was higher in *B. africana* (90.88 and 105.71 mg/g GAE and RE, respectively) than *S. cordatum* (43.36 and 49.80 mg/g). Cytotoxicity assessed through neutral red uptake was apparent for *B. africana* and *S. cordatum* in 3T3-L1 pre-adipocyte (IC₅₀ = 24.3 µg/ml and 25.0 µg/ml, respectively) and C2C12 myoblast (13.9 µg/ml and 20.5 µg/ml) cell lines, but not in normal human dermal fibroblasts and 48 h PMA-stimulated U937 monocytic cells (>100 µg/ml). Antioxidant activity was found to be greater for *S. cordatum* when using the TEAC and DPPH assays (2.03 and 2.18 TE, respectively) than for *B. africana* (2.46 and 2.98). Both plants decreased the oxidative stress-parameters induced by AAPH in terms of cytotoxicity, ROS formation, apoptosis and lipid peroxidation. *S. cordatum* elicited the most potent decrease in ROS formation (>80% at 2.5 µg/ml). *B. africana* had the greatest anti-apoptotic effect, reducing caspase-3 activity by 40% at 20 µg/ml. *B. africana* and *S. cordatum* had similar activity in attenuating lipid peroxidation with 206.7% and 195.9% reduction at 20 µg/ml, respectively. The usage of these polyphenolic-rich fractions may be beneficial, in the treatment or prevention of oxidative stress-related disorders.

Better late than never? The high cost of self-incompatibility in *Aloe maculata* (Asphodelaceae)

R.J. Cozien and S.D. Johnson (Mon B2)

School of Biological and Conservation Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa

Close to half of all flowering plant species show reduced reproductive success following self-pollination. This was long assumed to be achieved primarily through self-incompatibility mechanisms acting in the stigma or style to prevent germination of self pollen or successful growth of pollen tubes to the ovary. A few studies have shown, however, that even after self-pollen tubes penetrate the ovules, fecundity may be similarly reduced. Because of the high reproductive cost of disabled or aborted ovules, such late-acting self-incompatibility mechanisms should occur less frequently. To establish the relative

importance of the different mechanisms, we investigated the breeding system of *Aloe maculata*, a South African succulent from a family in which self compatibility known but largely uncharacterized. Hand pollinations with self and cross pollen revealed very high levels of self incompatibility. Pollen chase experiments following cross pollination 24 hours after self pollination indicated that self pollen usurps ovules. This was supported by a significant reduction in fecundity after hand pollination with mixtures of self and cross pollen relative to pollination with only cross-pollen. Hand pollinations with mixtures of cross and self, and cross and dead pollen showed that reduced fecundity does not result from stigma clogging by self pollen. Examination of pollen germination and tube growth using fluorescence and light microscopy confirmed that most ovules are fertilized within 24 hours of pollination with either self or cross pollen. Pollinator exclusion completely prevented fruit set. Open-pollinated flowers had similar fruit and seed set to intact flowers supplemented with cross pollen, but both were less successful than flowers which received only cross-pollen, indicating that pollen quality limits reproductive success in natural populations. Further investigations are required to establish whether reduced fecundity following self pollination results from late-acting self-incompatibility or early inbreeding depression.

Understanding the transcriptional regulation of the *Eucalyptus Cellulose synthase1* gene

N.M. Creux^{a,b}, M. Ranik^{a,b}, M.H. De Castro^{a,b}, A. Spokevicius^c, G. Bossinger^c, C. Maritz-Olivier^a and A.A. Myburg^{a,b} (Mon C1)

^aDepartment of Genetics, University of Pretoria, Pretoria, 0002, South Africa

^bForestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, 0002, South Africa

^cDepartment of Forest and Ecosystem Science, Melbourne School of Land and Environment, University of Melbourne, Water Street, Creswick, Victoria, 3363, Australia

Cellulose is a highly abundant biopolymer found in plant cell walls and has received considerable attention from various industries due to its unique molecular characteristics. Cellulose is deposited into plant cell walls by a large cellulose synthase (*CesA*) protein complex embedded in the cell membrane. The *CesA* proteins within this complex differ depending on whether primary or secondary cell walls are being deposited. While many studies have focused on the characterization of this enzyme and its biosynthetic pathway, there are few studies on the spatio-temporal regulation of the corresponding genes. We aimed to identify the promoter regions that interact with the transcription factors and modulate the expression of the *Eucalyptus grandis CesA1* gene. We performed an *in silico* analysis on the promoters of six *Eucalyptus CesA* gene family members and identified a number of highly conserved cis-elements. The positions of these conserved elements allowed for targeted truncation of the *EgCesA1* promoter. β -glucuronidase reporter-gene analysis in *Arabidopsis* and *Eucalyptus* enabled us to assign putative functions to some of the conserved regions. We also identified regions in the promoter that may contain novel elements necessary for *EgCesA1* expression as they influenced β -glucuronidase expression but contained no known cis-elements. Several previously identified transcription factors are known to regulate *CesA* genes and we have cloned these to test which regions of the promoter these proteins bind. We are also screening the functional promoter regions against a cDNA expression library to identify novel proteins which may bind to the *CesA1* promoter. Pairing regions of promoter sequence with transcription factors that bind them will aid in completing the regulatory model for this gene's promoter. This information will be useful on a fundamental level to

understand the transcriptional control of cellulose biosynthesis in trees, as well as being applicable to future biotechnology approaches to enhance cellulose production.

On the wings of butterflies: unique floral morphology and flower orientation promotes cross-pollination in *Gloriosa superba*

R.J. Daniels and C.I. Peter (Tue B1)

Department of Botany, Rhodes University, Grahamstown, 6140

Gloriosa superba L. (Colchicaceae) has conspicuous, brightly coloured crimson and yellow flowers with reflexed and inverted tepals. Butterflies and sunbirds have been reported to visit the flowers but little work has been done to investigate the pollination biology of this species. In this paper, the identity of pollinators was investigated. In addition, measurements of stigma and pedicel orientation, nectar concentration and volume measurements and pollinator observations were used to test a number of hypotheses regarding the direction from which pollinators approach the flowers and the orientation of the stigma. It was hypothesised that (a) arriving pollinators would approach from clearings, (b) flowers would project toward clearings by pedicel orientation (c) stigmas would be orientated toward clearings to increase the likelihood of cross-pollination and (d) there is stigma-anther spatial separation to limit selfing. Results indicated that *G. superba* is specialised for butterfly pollination. *Eronia cleodora* is by far the most common visitor, although sunbirds occasionally visit in gardens but they are unlikely to be important pollinators. *G. superba* is one of the few known angiosperms that loads its pollen onto the wings of its pollinator. Anthers are separated from stigmas horizontally but overlap vertically. There was support for pedicel orientation away from vegetation and stigmas orientation toward clearings to intercept incoming pollinators. Pollinator observations of tepal position corresponding to the sequence in which tepals were visited also support this hypothesis. Few butterflies visited >1 tepal/flower or >1 flower/patch indicating potentially high outcrossing rates.

Phylogeny of the subfamily Alooideae (Asphodelaceae): Paraphyly of *Aloe* and *Haworthia* and consequences for classification

B. H. Daru^{a,b}, O. Maurin^{a,b}, J.C. Manning^c, J.S. Boatwright^{a,b,c} and M. van der Bank^{a,b} (Mon B1)

^a*African Centre for DNA Barcoding, University of Johannesburg, P. O. Box 524 Auckland Park, 2006 Johannesburg, South Africa*

^b*Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa*

^c*Compton Herbarium, South African National Biodiversity Institute, Private Bag X7, Claremont 7735, South Africa*

Alooideae (Asphodelaceae: Asparagales) comprises five genera, four of which are endemic to southern Africa. Despite their importance in commercial horticulture, little is known about their evolutionary history and their genetic diversity, and the taxonomy remains unresolved. This study addresses generic delimitation of the subfamily Alooideae using an expanded molecular sequenced datasets from three plastid regions (*rbcLa*, *matK* and *trnH-psbA*) and the first subunit of the nuclear ribosomal internal transcribed spacer (ITS1) as well

as using morphological trait data. Representatives of other genera within the order Asparagales were also included to infer the placement of the five Alooideae genera within Asphodelaceae. The plastid and nuclear DNA were analysed using Maximum Parsimony and Bayesian statistics as well as the morphological matrix data that was mapped onto the molecular phylogeny. Both Parsimony and Bayesian analyses of combined cpDNA and ITS1 regions yielded two major clades in Alooideae. The findings will be discussed.

The additive and synergistic antimicrobial effects of Frankincense and Myrrh – Essential oils from the predynastic period

S. de Rapper^a, S.F. van Vuuren^a, G.P.P. Kamatou^b and A.M. Viljoen^b (Tue C4)

^a*Department of Pharmacy and Pharmacology, Faculty of Health Sciences, University of the Witwatersrand, 7 York Road, Parktown 2193, South Africa*

^b*Department of Pharmaceutical Sciences, Faculty of Science, Tshwane University of Technology, Pretoria, 0001, South Africa*

The historical use of frankincense and myrrh essential oils date back to biblical times, however, very little is known of their antimicrobial effects when used in combination. This study investigated the *in vitro* antimicrobial activity between three essential oil samples of frankincense (*Boswellia rivae*, *B. neglecta*, *B. papyrifera*) and two essential oil samples of myrrh (*Commiphora guidotti* and *C. myrrha*) independently and in combination against various micro-organisms. When examined independently, it was noted that generally *Cryptococcus neoformans* and *Pseudomonas aeruginosa* often appeared to be the most susceptible micro-organisms against oils both of *Boswellia* and *Commiphora* spp, with noteworthy minimum inhibitory concentration (MIC) values varying between 0.50-1.50 mg/mL. When assayed in various combinations, the frankincense and myrrh oils displayed synergistic (11.11%), additive (41.67%) and non-interactive properties (45.95%), with no antagonism noted. When investigating different ratio combinations against the pathogen *Bacillus cereus*, the most favourable combination observed was between *B. papyrifera* and *C. myrrha*. The GC-MS showed that the oils are chemically diverse. Major compounds for *C. myrrha* were furanogermacrene (15.90%) and furanoeudesma-1.3-diene (44.30%), present only as minor components in *C. guidotti*. Major compounds for *C. guidotti* were (*E*)- β -ocimene (52.60%) and α -santalene (11.10%) and (*E*)- α -bisabolene (16.00%). α -Pinene (36.10-67.70%) was present in high concentrations in both *B. rivae* and *B. neglecta*. Other major compounds for *B. rivae* include σ -3-carene (12.20%) and limonene (12.00). One other major compound (terpinen-4-ol at 11.30%) was detected for *B. neglecta* and only one major compound (octyl acetate at 64.80%) was predominant for *B. papyrifera*. The historical and antimicrobial importances of these oils prove to be extremely promising when examined both independently and in combination.

Probabilistic approaches to inferring plant extinctions from herbarium records

K.J. Duffy (Wed C1)

School of Biological and Conservation Sciences, University of KwaZulu-Natal, Pietermaritzburg

The probability and time of extinction of taxa is often inferred from statistical analyses of herbarium records. Such spatially explicit, temporally aggregated data may be useful for identifying historical sighting clusters of endangered taxa in space and time. Identification of such sighting clusters can help highlight changes in the historical recording of endangered taxa. I used two methods to identify sighting clusters in historical records: the Ederer-Myers-Mantel (EMM) test and the space-time permutation scan (STPS). I applied these methods to the spatially explicit sighting records of three nationally endangered orchids from the Republic of Ireland: *Cephalanthera longifolia*, *Hammarbya paludosa*, and *Pseudorchis albida*. Results show that the STPS is a more flexible statistical tool with sparse historical record data. This is because the STPS can identify regions that contain sighting clusters because it uses a flexible scanning window (defined by cylinders of varying size that move over the study area and evaluate the likelihood of clustering), and it identifies regions with high and regions with low rates of historical sightings. I show that, coupled with other probabilistic methods to infer extinction, the STPS analyses can be used to detect sighting clusters of endangered species that may be related to regions of extirpation and under-recording and may assist in the categorization of threat status.

Phytogeography of Maharashtra: A taxonomic approach with reference to some selected species from India

S. R. Dutta (Wed C2)

Department of Botany, R. D. National College, Bandra (W), Mumbai -400 050

Maharashtra is the third largest state in India and includes an area of 308,000 km² (119,000 mi²). It lies between 18.96°N and 72.82°E. The Western Ghats, are a hilly range running parallel to the coast, at an average elevation of 1,200 metres (4,000 ft). Total area under forest cover in Maharashtra is only 17%. Maharashtra has five national parks, three game reserves, twenty four wildlife and bird sanctuaries which have been created with the aim of conserving the rich bio-diversity of the region. The present paper deals with the geographic origin of about 5100 species of angiosperms belonging to 1600 genera and 215 family in the state of Maharashtra (India); A quantitative report of continentwise analysis of exotic, endemic species(about 319 endemic species for which no record is found beyond the state) is given here . Taxonomic identity of some taxa and typification of some Linnaean species like *Crateva tapia* Linn., *Ixora alba* Linn., *Lawsonia inermis* Linn., *Phyllanthus emblica* Linn., etc. are also discussed in this paper.

Variation in biological activities of leaf extracts of 42 *Combretum molle* plants collected from different areas in Gauteng and Mpumalanga

J.N. Eloff and G. Würger (Wed B1)

Phytomedicine Programme, Faculty of Veterinary Science, University of Pretoria

Combretum molle is an important medicinal plant that is used widely for many indications in several countries in Africa. *Combretum* species are also included in the 50 most important medicinal plants in the African Herbal Pharmacopoeia. One of the major problems in using medicinal plants that are collected in nature is uncertainty on the variation in biological activity. We investigated the antibacterial activity of acetone leaf extracts of 42 *C. molle* plants collected in different areas in the northern parts of South Africa against *Escherichia coli* and *Staphylococcus aureus* using a twofold serial dilution microplate method. Because other parts of the Combretaceae contain high tannin concentrations we also determined the tannin content of plants to determine if there is a correlation. In general although there were some differences in the chemical composition of different extracts and the tannin content, there were little differences in antibacterial activity of plants collected in different areas. The average minimum inhibitory concentration against *E. coli* was 220 µg/ml (SD 70) µg/ml and against *S. aureus* was 399 (SD 162) µg/ml. The inverse correlation between tannin content and antibacterial activity was low with R^2 values of 0.183 (*E. coli*) and 0.286 (*S. aureus*). When cultures were incubated for 120 minutes to determine if the activity was mainly bacteriostatic or bactericidal there was very little decrease in the activity. The results indicate that at least in the case of this species environmental factors do not play a major role in the antibacterial activity and that information obtained for a plant can be extrapolated to other plants. There were some cases where much higher activities were obtained. If this higher activity is maintained it indicates that it may be feasible to search for chemotypes to propagate for the delivery of high activity plant extracts.

Projections of regional climate change over southern Africa - the water balance in a warmer climate

F.A. Engelbrecht^{a,b} (Wed A3)

^a*CSIR Natural Resources and the Environment - Climate Studies, Modelling and Environmental Health, Pretoria, 0001, South Africa*

^b*Climatology Research Group, GAES, University of the Witwatersrand, South Africa*

A variable-resolution global atmospheric circulation model is used to downscale the projections of six different coupled climate models that contributed to Assessment Report Four (AR4) of the Intergovernmental Panel on Climate Change (IPCC) to high resolution (about 0.5° in latitude and longitude) over southern Africa. All the simulations are for the A2 (business as usual) emission scenario and for the period 1961-2100. The variable-resolution model used is the conformal-cubic atmospheric model (CCAM) of the Commonwealth Scientific and Industrial Research Organisation (CSIRO) in Australia. In these simulations, CCAM was integrated using a static description of the land-surface (e.g. fields such as the vegetation type and surface albedo did not vary at an inter-annual time-scale). Additionally, simulations are shown where the land-surface responded dynamically to the changing climate. For these simulations, CCAM was applied coupled to the CABLE (CSIRO

Atmosphere-Biosphere Land-Exchange) dynamic land-surface model. The southern African region is projected to warm rapidly during the 21st century, at about twice the global rate of temperature increase. For large parts of the interior regions of southern Africa, the climate for the period 2071-2100 is projected to be 4 °C to 6 °C warmer than the climate of the baseline period 1961-1990. This drastic rise in surface temperature is shown to occur in association with a large increase in potential evaporation. The southern African region is simultaneously projected to become generally drier in response to enhanced anthropogenic forcing, with only slight rainfall increases projected to occur over parts of the central and eastern interior of South Africa. That is, the water balance over the region is projected to become increasingly negative under conditions of enhanced anthropogenic forcing.

Seed germination behaviour, micropropagation and *Agrobacterium*-mediated transformation of *Salvia runcinata* (L.f.): implications for conservation and cultivation

S. Figlan^a, J.M. Kossmann^a and N.P. Makunga^{a,b} (Mon C2)

^a*Institute for Plant Biotechnology, Department of Genetics, Faculty of Natural Sciences, Stellenbosch University, Private Bag XI, Matieland, South Africa*

^b*Department of Botany and Zoology, Faculty of Natural Sciences, Stellenbosch University, Private Bag XI, Matieland, South Africa*

The seed germination behaviour, optimal conditions for micropropagation and transformation efficiency of *Salvia runcinata* was determined. *S. runcinata* is particularly rich in rosmarinic acid and has also been identified as an alternative source of natural α -bisabolol; a compound which is an important component of cosmetic products, making it highly valued in the cosmetic industry. Nevertheless medicinal plants of the mint family are under pressure owing from overexploitation from their natural environment. The methods of extraction employed are almost invariably crude and unsystematic. As a result, the roles of exploitation may exceed those of local natural regeneration. There is thus an urgent need to develop and implement regeneration/conservation strategies for this sage species. In this study, the common means of regeneration and propagation of *Salvia runcinata* included *in vitro* seed germination and micropropagation methods. The efficacy of smoke and scarification treatments for germination improvement was tested. Hairy roots were also established using three different strains of *Agrobacterium rhizogenes* (A4T, LBA9402 and C58C1) as an attempt to enhance secondary metabolite production of the plant. The most effective germination treatment proved to be a 3 minute exposure of seeds to 25% H₂SO₄ combined with a concentration of 10⁻⁵ M smoke solution resulting to more than 80% germination. Shoot proliferation was significantly higher using nodal explants with addition of 4.43 μ M BA generally promoting the average number of shoots per nodal explant. The LBA9402 strain was more efficient in the establishment of putative hairy root lines. However, only two hairy root clones survived subculturing and polymerase chain reaction with primers for the genes *rolA*, *rolB*, *rolC* and *ags* confirmed the integration of T-DNA fragment from the Ri plasmid of *A. rhizogenes* to the genome of these clones obtained after transformation by A4T strain. Southern blot analysis also confirmed the presence of *rolA* gene in the two A4T clones.

Elucidation of defence responses associated with inducer application in *Pinus patula*

K. Fitza^a, A.A. Myburg^a, E.T. Steenkamp^b, K. Payn^c and S. Naidoo^a (Tue A2)

^a*Department of Genetics, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria 0002, South Africa*

^b*Department of Microbiology and Plant Pathology, FABI, University of Pretoria, Pretoria 0002, South Africa*

^c*Mondi South Africa, P.O. Box 12, Hilton 3245, South Africa*

A major challenge for the pine species in South Africa is the necrotrophic fungus *Fusarium circinatum*, which causes the disease pitch canker. Commercial forestry incurs large economic losses from the pathogen *F. circinatum*, mostly occurring due to post planting stress, where 20-30% of the planting stock of *Pinus patula* is lost due to infection annually. One means of enhancing defence is through applying chemicals or biologically derived treatments known to activate induced resistance, throughout the entire plant. Induced resistance may involve chemical defences, e.g. production of terpenoids, phenolic metabolites, protein defences such as pathogenesis related proteins and anatomical defences such as cell wall lignification. A panel of 10 different inducers were chosen to determine their efficiency in activating induced resistance in *P. patula*. The application of chitosan at a concentration of 10mg/ml resulted in significant reduction in lesion length over a 6 week period after challenge with *F. circinatum*. Subsequent characterization of the molecular basis of induced resistance was achieved by analysing the expression profiles of selected putative defence genes. These *P. patula* putative orthologs were identified based on Expressed Sequence Tag (EST) sequences available from *Pinus taeda* using a phylogenetic approach. Reverse transcription-quantitative PCR assays revealed that *phenylalanine ammonia lyase (PAL)* showed a four-fold up-regulation in *P. patula* seedlings treated with chitosan compared to un-treated plants, suggesting the onset of induced resistance. Chitosan application may hold promise in inducing resistance in *P. patula* against *F. circinatum* and could form part of an integrated management strategy to control the disease.

Comparative wood anatomy of six genera of the family Bignoniaceae in Nigeria

A.E. Folorunso and O.S. Awosika (Wed C2)

Botany Department, Obafemi Awolowo University, Ile-Ife, Nigeria

Comparative wood anatomy of six genera of Bignoniaceae in Nigeria were carried out to assess the taxonomic and diagnostic characters from the wood samples of the selected genera, identify the primitive characters and the species possessing them, investigate the phylogenetic relationship among members of the family and use the wood characters in producing a key for easy identification of the species of Bignoniaceae. Wood specimens were collected from different latitudes and ecological zones such as the wet forest, dry forest and derived savanna areas of Nigeria and assigned accession numbers. Small pieces of the species were macerated in Schutz's fluid; a mixture of equal 10% solution of Chromium trioxide and 10% solution of concentrated nitric acid, stained in 1% Safranin O and mounted on a slide using dilute glycerol as a mountant. The dimension and anatomical characteristics of the vessel elements and fibres were described respectively. The length and diameters of twenty vessel

elements and fibres were measured from the macerated materials; fibre-length and vessel-length ratio (F/V ratio) was also calculated. Small block of about 1cm x 1cm x 1cm were removed from the wood samples; transverse, tangential and radial longitudinal sections were made on Reichert Sledge Microtome at varying thickness between 8 – 15 µm. The sections were stained in 1% solution of Safranin O for 15 minutes, washed in three changes of water, counter-stained in 1% alcian blue for 3-5 minutes, dehydrated through series of ethylalcohol: 30, 50, 70, 90 and 95%, cleared in xylene and mounted in DPX mountant. Photomicrographs of the wood sections were taken with Leitz camera mounted on Dialux research microscope. The wood is diffuse porous and show the presence of growth ring in all the six genera. Diagnostic wood characters are mainly confined to perforation plates, vessel length, fibre types, ray types and number of adjoining vessel members. Major primitive characters shown include presence of more tracheids, elongated and narrow vessel elements, highly oblique end wall, vessel angular in shape and more solitary vessels in transverse section distribution. Phylogenetically, *Tecoma stans* and *Crescentia cujete* are the highly primitive members, *Stereospermum acuminatissimum* and *Markhamia tomentosa* are highly advanced species while the position of *Newbouldia laevis* and *Spathodea campanulata* is intermediate.

An analysis of landscape connectivity of the Grassland Biome of Mpumalanga using graph theory

L. Fourie^a, M. Rouget^a and M.C. Lötter^b (Wed A2)

^a*Department of Plant Science, University of Pretoria, South Africa 0002*

^b*Mpumalanga Touris and Parks Agency, Private Bag X1088, Lydenburg 1120*

The South African Grassland biome is one of the most threatened biomes in South Africa. According to National Land Cover data, 45% of the Grassland biome area is transformed, degraded or severely invaded by alien plants and the remaining natural areas are highly fragmented. The connectivity between habitat patches is very important for populations to survive in this fragmented landscape and the analysis and quantification of this connectivity can be used for reserve design and to direct conservation efforts. The aim of this study is to analyse connectivity in the grassland biome of Mpumalanga using graph theory. Graph theory-based connectivity indices have the ability to combine spatially explicit habitat data with species specific dispersal data and can quantify structural and functional connectivity over large landscapes. Natural areas were identified using 2008 land cover data for Mpumalanga. This study shows the level of connectivity of the Grassland Biome of Mpumalanga for different grassland species with dispersal distances ranging from small to large. The habitat patches most important for maintaining connectivity in this area are identified. Additionally, the importance of protected areas as well as the importance of abandoned croplands for maintaining overall connectivity is demonstrated. These results can be used to inform management decisions and reserve design in this biome.

Employing biotechnology and cryobiological research to conserve threatened plant species

M.M. From and M. Landry (Wed C3)

Laboratory for Endangered Plants, Centre for Conservation & Research, Omaha's Henry Doorly Zoo, Omaha, Nebraska, USA 68107

Plant biotechnology developed in the 21st century can serve as a means to forestall the final extinction of many plant species. The vast majority of plant species on earth today have not yet been researched for their possible benefits to mankind and all other life forms. Omaha's Henry Doorly Zoo established an *ex situ* plant laboratory to research plant species from threatened habitats and reintroduce propagules of those species in their original habitats. Sustainable populations are thus created by increasing the total number of specimens at a given site. As an example, Over 840 endangered native orchid species, representing 5 genera: *Aeranthus spp.*, *Aerangis, spp.*, *Bulbophyllum spp.*, *Calanthe sp.*, and *Cryptopus sp.* (13 species) have been reintroduced into Madagascar in collaboration with Malagasy scientists from CITES and graduate students who have received biotechnology training in the zoo's laboratory. The head of the Madagascar forestry department and local residents were collaborators for plant reintroductions made in the rainforest. Seedling survival, based on the number of plants displaying active shoot and root growth five years after reintroduction, ranged an average of 76%. Samples of seed and spore accessions from threatened plant species are also preserved in the zoo's frozen seed bank. Currently, seeds and spores from more than 150 species are in the cryobank, some preserved using novel techniques developed in the zoo's laboratory, creating a backstop to extinction by saving germplasm for future research projects and long term conservation plans that can eventually lead to species recovery on a broader scale.

Plant invasions, resilience, economics, and restoration: Can fynbos pay for alien management?

M. Gaertner^a, H. Nottebrock^b, H. Fourie^c, S. D.J. Privett^d and D.M. Richardson^a
(Tue A3)

^a*Centre for Invasion Biology, Department of Botany and Zoology, Stellenbosch University, Private Bag x1, Matieland 7602, South Africa*

^b*Potsdam University, Plant Ecology and Conservation Biology, Maulbeerallee 2, 14469 Potsdam, Germany*

^c*Western Cape Department of Agriculture, Private Bag X1, Elsenburg, 7607, South Africa*

^d*Fynbos Ecoscapes, Witkrans, Gansbaai, 7220*

We explored the influence of different invasive plant species on the resilience of three different invaded sites in the Cape Floristic Region by testing a variety of management strategies through field trials and experimental manipulations. To investigate the financial feasibility of introducing native species which can be used for sustainable flower harvesting as an incentive for restoration we linked ecosystem services to the beneficiaries of ecosystem restoration conducting a cost-benefit analysis. Our results show that the ecosystem was sufficiently resilient to allow autogenic recovery to occur following removal of the invasive species. Native cover, species richness, diversity or evenness increased significantly after

restoration at all three sites, whereas alien cover decreased significantly. Soil restoration treatments resulted in a decrease of available nitrogen, but this had no effect on plant cover (alien or native), species richness, diversity or evenness. We therefore argue that in this case soil restoration treatments were futile and added unjustifiable expense to the restoration program. We learned that an adaptive approach involving revising management objectives throughout the management process would have been more appropriate. The financial analysis shows that the density of invasion is the key determinant influencing whether clearing of invasive plants and restoration results in a net gain or loss when land is restored to a state conducive to sustainable harvesting.

Functional evaluation of a drought-induced “late embryogenesis abundant” protein gene from cowpea

I. Gazendam^a, D. Oelofse^a and D.K. Berger^b (Mon C1)

^a*Agricultural Research Council-Roodeplaat Vegetable and Ornamental Plant Institute, Private Bag X293, Pretoria, 0001 South Africa*

^b*Department of Plant Science, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, 0002 South Africa*

Drought tolerance of plants is governed by multiple genes. Biotechnology can be applied to improve a crop's drought tolerance through candidate gene identification, followed by transformation or molecular breeding strategies. Cowpea (*Vigna unguiculata* (L.) Walp) was used as a source of candidate drought responsive genes, since it is a highly drought tolerant legume crop. A cDNA library, enriched for drought responsive cowpea genes, was generated following suppression subtractive hybridisation of two cultivars differing in their drought tolerance. DNA microarrays were employed to identify genes responding to drought stress. A gene encoding an atypical group 5 late embryogenesis abundant protein (LEA5) was selected for further characterisation. It was shown to be highly upregulated by drought, application of the plant stress hormone abscisic acid and other abiotic stress conditions (salt and cold) in cowpea leaves. LEA proteins play an important protective role during drought and other abiotic stresses of plants, but group 5 proteins are not as well studied as the classical LEA groups. This gene was therefore over-expressed in the model plant *Arabidopsis thaliana* to study its role in tolerance to drought. Seedling stress tests were performed *in vitro* in the presence of osmotic agents (high molecular weight polyethylene glycol) that simulate drought conditions in the soil. Quantitative growth parameters (hypocotyl and primary root elongation) of homozygous transgenic T4 seedlings were measured and compared to non-transgenic controls. No statistically significant improvement in growth and recovery after stress could be attributed to the cowpea LEA5. This LEA protein could, therefore, not improve the drought tolerance of transgenic *A. thaliana* lines. Its possible role in cold stress protection is suggested as a follow-up study.

***Banksia ericifolia* invading South Africa as predicted – a major threat or just symptom of a peculiar fire regime?**

S. Geerts^{a,b}, J.R. Wilson^{a,b}, D.M. Richardson^a, M. McGeoch^a, M. Gaertner^a, J.J. le Roux^a, S. Kritzinger-Klopper^a and C. Muofhe^a (Tue A4)

^a Department of Botany and Zoology, Centre for Invasion Biology, Stellenbosch University, Private Bag XI, Matieland 7602, South Africa

^b South African National Biodiversity Institute, Kirstenbosch National Botanical Gardens, Claremont, South Africa

In the Cape Floristic Region (CFR) of South Africa there is a long history of alien invasive Australian plant species. Some of these species, for example those in the genus *Hakea* (Proteaceae family), are well adapted and thrive under Fynbos fire regimes and have become widespread invaders. However, many potentially invasive species are only small infestations and assessing and controlling these at an early stage is the most cost effective. Recently, another Australian Proteaceae genus, *Banksia*, was listed as invasive in South Africa. With similar life history traits than *Hakeas* (serotinous, with large canopy-stored seed banks dispersed by wind in the post fire invasion window) *Banksias*, and in particular *Banksia ericifolia* have been predicted to be high risk introductions in fynbos. We assess the status of all known *B. ericifolia* populations in the CFR. At the only site where *B. ericifolia* has become invasive, we determine the cause of its spread. The invasive population arose from an ornamental farm hedge planted 30 years ago with sporadic recruitment into the surrounding natural vegetation. It now covers an area of about 150 hectares with approximately 10 000 individuals. The population was mapped and levels of recruitment and plant size determined. From these data, size distribution and time to first flowering and post fire seed dispersal distances were calculated. The ability to attract local pollinators in this preferential outcrossing species is discussed. We present preliminary analyses linking a particular human-induced fire regime in the immediate area of the invasion to the observed episodic recruitment. We contrast this to other sites where *B. ericifolia* has been grown under different fire regimes, and discuss our results in the light of risk analysis of the species. Lastly we assess the effectiveness of clearing operations on *B. ericifolia*.

Phylogenetic relationships within subtribe Terminaliinae (Combretaceae)

J. Gere^{a,b}, O. Maurin^{a,b}, M. van der Bank^{a,b} (Mon B1)

^aAfrican Center for DNA Barcoding, University of Johannesburg, Department of Botany and Biotechnology, PO Box 524, Auckland Park 2006, South Africa

^bDepartment of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa

Subtribe Terminaliinae is a taxonomically and phylogenetic complex group within Combretaceae. Previous morphological studies on the family recognized five genera, *Terminalia*, *Buchenavia*, *Conocarpus*, *Pteleopsis* and *Anogeissus* with *Bucida* transferred to *Terminalia*. Recent molecular studies indicated that the largest genus, *Terminalia* (approximately 200 species) is polyphyletic with the proposal that all genera, except for *Conocarpus*, should be transferred to *Terminalia*. In the current study an extra plastid gene, *ndhF*, as well as an increased number of taxa, was added to the existing data set (ITS, *rbcL* and spacers *trnH-psbA* and *psaA-ycf3*). Within Terminaliinae three major clades are

retrieved: clade I comprises mainly Asian *Terminalia* and *Anogeissus* species; clade II includes *Buchenavia*, *Bucida*, *Pteleopsis* and American *Terminalia* species, and clade III comprises mainly African *Terminalia* with a few Asian and Australian *Terminalia* species embedded in it. *Conocarpus* is supported as sister to the rest of the subtribe.

Plant diversity of a lowland dipterocarp rainforest in Samar Island, Philippines

W.S.M. Gruezo (Wed C2)

Professor 12, Plant Biology Division, Institute of Biological Sciences, College of Arts and Sciences, University of the Philippines Los Banos, College 4031, Laguna, Philippines

The plant diversity of a lowland dipterocarp rainforest in Samar Island, Philippines (*i.e.* Borongan, Eastern Samar Province) is composed of 485 species in 276 genera and 99 families. Of the 485 species, 9 species are epiphytes, 24 ferns, 17 grasses, 83 herbs, 43 lianas, 15 vines, 17 shrubs, 91 small trees, 120 medium-sized trees, 53 large trees and 29 tree-like plants. In terms of economic or functional importance, these 485 species serve the following uses: 207 landscape plants, 117 timber sources, 58 ornamentals, 42 for cottage industry, 33 weeds, 25 medicinals, 17 fruit crops, 9 each as fiber and poison sources, 6 pasture and forages, 3 spices, 2 each with edible fruits, for masticatory purposes, ecological functions and soil erosion control, 1 each for beverage, dye source, perfume and animal food. In addition to a number of new species records for Samar Island, one important taxonomic and biogeographic discovery was made, particularly in the vicinity of Mt. Apoy. This involves the discovery of *Falcatifolium gruezoi* de Laubenfels, (Podocarpaceae) known locally as *kari-karis* (Waray), *binaton* (Tagalog) and Gruèzo sickle-leaf (in English). Its discovery represents a new genus record for Samar Island and also made possible the description of a new forest formation type termed as mixed *Falcatifolium* forest, which can only be found in a specific habitat type, here described also as new under the name “pseudo-peat swamp”. The discovery of another species of conifer, *Podocarpus nerifolius* D.Don [*malaadelfa* (Tagalog), Podocarpaceae] brings the total number of conifer species in Samar Island to five, thus this particular species is a new species record for the island. In terms of plant endemism, the study area which is a part of the Suribao Watershed has 256 endemic species representing 53% of the total species record for this particular watershed, which largely belongs to a lowland dipterocarp rainforest type. This watershed has a very high plant diversity *i.e.* its calculated Shannon-Weiner diversity index (H') is 4.56 (based on measurements of 25 circular sample plots, each with an area of 1/8 ha).

Towards an understanding of indigenous Afromontane forest regeneration and expansion in Buffelskloof Private Nature Reserve, Mpumalanga

W.A. Haddad^a, M. Rouget^a, A.E. van Wyk^a and J.E. Burrows^b (Wed A2)

^a*Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002*

^b*Buffelskloof Herbarium, PO Box 710, Lydenburg 1120*

Several recent studies show a marked increase of woody vegetation in many different biomes across the globe. The aim of this honours project was to understand forest regeneration under different land uses and to quantify the spatial extent of indigenous

Afromontane forest over time in the Buffelskloof Private Nature Reserve, Mpumalanga, South Africa. A time series of aerial photographs from 1956, 1981 and 2004 was constructed in a GIS and used to locate and quantify the areas converted from grassland and pine plantations to indigenous woody communities. A marked increase in woody cover was found in some areas, while the forest/grassland interface remained stable in other areas. A total of four 10m x 10m plots were investigated, two located in former grassland and two in areas that were under pine plantations up to twenty years ago. Further, two 100m transects were recorded through an area of former grassland and an area of former plantation. None of these areas have been burnt in the past two decades. Investigation was made into the reproductive and dispersive biology of some of the most commonly encountered species in an effort to see whether this has an effect on which woody species colonizes an area first. From the results it seems that there is not a major difference between the species and structural composition of plots located on areas with different vegetation histories (grassland vs. plantation), but that other factors such as distance to established forest (seed source), soil moisture and aspect has a greater influence. This study and further work along these lines can be of great value in the rehabilitation of old commercial plantations to indigenous afromontane forests along the Escarpment.

Root endophytes from *Persea americana* and their role against *Phytophthora cinnamomi* infection

J.D. Hakizimana^{a,b}, M. Gryzenhout^e, T.A. Coutinho^{a,d} and N. van den Berg^{a,c} (Wed C3)

^aForestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, 0002, South Africa

^bDepartment of Plant Science, ^{3c}Genetics, and ^dMicrobiology and Plant Pathology, University of Pretoria, 0002, South Africa. ^eDepartment of Plant Sciences, University of the Free State

Endophytes are microorganisms that, during a viable period of their life, symptomlessly colonise the living internal tissues of plants. Root endophytes have been investigated in plants and used as biocontrol agents against plant pathogens. However, little is known about the diversity of endophytes in *Persea americana* (avocado) roots and their potential role in biocontrol of *Phytophthora cinnamomi* (*Pc*). This Oomycete is the causal agent of Phytophthora root rot, the most important disease in avocado producing countries worldwide. The aim of this study was to identify bacterial and fungal endophytes from avocado roots and to use selected endophytes with potential biological activity against *P. cinnamomi* in planta. The identification was based on morphological characteristics of the isolates as well as using ITS, β -tubulin, EF-1 α and 16S ribosomal RNA gene sequencing. Twenty four different fungal species and 8 bacterial species were identified as endophytes from the avocado roots from various locations in South Africa. Two bacterial strains and 2 fungal strains were selected on their antagonistic effects against *P. cinnamomi*. Clonal as well as endophyte-free tissue-cultured avocado plants were inoculated with each of the selected endophytes at 10^8 CFU ml⁻¹ for bacteria and 10^6 spores ml⁻¹ for fungi. Four weeks after endophyte inoculation, each plant received 10^5 *Pc* zoospores ml⁻¹ directly sprayed onto roots, except for negative control plants. Positive control plants received no endophytes. Phytophthora symptoms were assessed 21 days post infection and disease incidence was calculated. Avocado plants that received endophytes prior to *Pc*-infection showed a significant decrease in disease incidence with ratings from 2-40% compared to 94-100% for the positive control plants that received no endophytes.

Endophytes and their association with Dichapetalaceae

C.B.S. Hendriks^a, J.J.M. Meyer^a and C.F. van der Merwe^b (Wed B2)

^a*Department of Plant Science, University of Pretoria, Pretoria, 0002, South Africa*

^b*Laboratory for Microscopy and Microanalysis, University of Pretoria, Pretoria, 0002, South Africa*

Dichapetalum cymosum (Poison leaf) is a very common problem plant in southern Africa. Fluoroacetic acid, is believed to be the poisonous entity in the plant, might be produced by the plant, but the micro-organisms associated with this plant may also play a role in the production thereof. By isolating the bacteria the capabilities of the isolated endophytes to synthesise fluorinated compounds were studied. It is suggested from preliminary results that symbionts might play a role in the synthesis of the poisonous entities in the Dichapetalaceae family, but further investigation is required. The detection of glandular lesions on the abaxial side of the leaf led to closer examination and the cross sections revealed unusually deformed epidermis cells with adjacent cells containing vacuoles filled with phenolic-like crystals. Transmission electron microscopy (TEM) of the spongy parenchyma cells directly above the glandular lesions indicated the presence of clusters of small, virus-like particles (VLPs) in the chloroplasts. Observations by TEM show that these VLPs have unusual lattice formation, differing from the typical viral-capsid protein arrays and, hence, the question is posed whether this might be of virus origin. *Tapura fischeri* (Leafberry tree) is a tree member of the same family, and it was found to contain a fluorinated compound. Endophytes were also found in the plant and similar glandular lesions with analogous VLPs were observed at these sites. This might implicate that endophytes have a share in the biosynthesis of the fluorinated compounds found in Dichapetalaceae. Numerous factors ought to be considered to establish the chemical ecology of the intricate system regarding the endophytes and the toxicity of the family Dichapetalaceae.

The taxonomic significance of the cypselas morphology in the genus *Nolletia* Cass. (Asteraceae) in southern Africa

P.P.J. Herman (Tue B3)

National Herbarium, South African National Biodiversity Institute, Private Bag X101, Pretoria, South Africa 0001

The genus *Nolletia* Cass. belongs to the tribe Astereae of the family Asteraceae. Currently eight species are recognised in southern Africa. The genus has never been revised and there is no key available dealing with all of the species. Full descriptions of the species are also scanty or non-existing in the available taxonomic literature. A species was identified as being toxic to cattle and another species from the Pretoria area recognised as a new species. A taxonomic revision of the genus is currently under way. Based on the cypselas morphology, the genus can be divided into two groups. The one group has plain, flat epidermal cells and longish twin hairs. The other group has globose epidermal cells each with very small twin hairs covering the entire surface of the cypselas. In this group a taxon with a slight variation was observed: it has globose epidermal cells arranged in pairs, each with one twin hair between them. The paired globose cells are scattered over the cypselas surface.

Determination of anti-HIV active constituents of *Helichrysum* species by means of metabolomics

H.M. Heyman^a, V. Maharaj^b, C. Kenyon^b and J.J.M. Meyer^a (Mon A1)

^aDepartment of Plant Science, University of Pretoria, Pretoria 0002

^bBiosciences, CSIR, P.O Box 395, Pretoria, 0001, South Africa

Being the home to more than 35% of the world *Helichrysum* species (c.a. 244 species) of which many are used as traditional medicine it is only fitting that research into these species should be conducted in South Africa. The most commonly used *Helichrysum* species for their medicinal properties are *H. cymosum*, *H. odoratissimum*, *H. petiolare* and *H. nudifolium*. The medicinal uses include the treatment of coughs, colds, fever, infection, headaches, menstrual pain and they are very popular for wound dressing. In previous studies conducted on 12 *Helichrysum* species, it was clear that some species have significant anti-HIV activity (5 - 12 µg/ml). In this current study another 35 randomly selected *Helichrysum* species were collected and tested for anti-HIV properties. Using an in-house DeciphR method on a full virus model (University of Basel), it was established that six species had significant anti-HIV activity ranging between 12 – 21 µg/ml (IC₅₀). Subsequent toxicity tests also revealed little or no toxicity for these extracts. With the use of NMR-based metabolomic analysis the search for active constituents was conducted and areas of interest were identified. Regions 2.55ppm – 3.16ppm, 6.20ppm – 6.40ppm, 5.72ppm - 6.28ppm, 6.60ppm – 7.00ppm, 7.24ppm – 8.04ppm and 8.24ppm – 8.98ppm were identified as being the main contributors to bioactivity. The isolation of active constituents will be guided by focusing on compounds having chemical shifts in these regions to identify possible lead compounds for further studies.

The role of pollinators in the assembly of *Erica* communities

A. Heystek and A. Pauw (Mon B2)

Department of Botany and Zoology, Stellenbosch University, Private Bag X1, Matieland, 7602, South Africa

Plant-pollinator interactions are proposed to be one of the main factors responsible for the diversification and maintenance of the exceptional plant species richness of the Cape Floristic Region. We investigated the role that pollinators play in the assembly of *Erica* communities since 426 members of this genus are crammed into a small area in the southwestern Cape and they display a range of pollination syndromes (80% entomophilous, 15% ornithophilous, 5% anemophilous). Vegetation plot data from ten regions in the fynbos biome was used to test whether the frequency of co-occurrence of species with the same pollination syndrome differs from a null model of random community assembly. We found that *Erica* communities were assembled randomly with respect to pollination syndrome in six regions, but were significantly overdispersed in four regions. Overdispersion is consistent with competition for pollinators between co-flowering species. To better understand these ecological processes, we conducted field experiments with six *Erica* species that are all primarily pollinated by the Orange-breasted Sunbird *Anthobaphes violacea*. We transplanted inflorescences of a focal *Erica* species into communities containing another ornithophilous *Erica* species and recorded pollination rates in the transplanted and resident species in relation to the floral density of the resident species. In five cases pollination rate in the

transplanted species was not related to the floral density of the resident species. However, there was one case of inter- and intraspecific competition for pollinators and one case of intraspecific pollination facilitation. Our investigation of patterns and processes of *Erica* community assembly concur that pollination can structure communities in non-random ways with potential for both facilitation and competition.

The role of resource limitation in the occurrence and persistence of rewardless pollination systems in *Disa* (Orchidaceae)

N. Hobbhahn^{a,b}, B. Bytebier, S.D. Johnson^a and L.D. Harder^b (Mon B2)

^a*School of Biological and Conservation Sciences, University of KwaZulu-Natal, Private Bag X01, Scottsville, Pietermaritzburg, 3209*

^b*Department of Biological Sciences, University of Calgary, 2500 University Drive NW, Calgary, Alberta, T2N0N5, Canada*

Pollination by deceit, whereby flowers do not offer any rewards to their pollinators, is frequently explained as a means of avoiding the resource costs of reward production, especially when resources are limited. To mitigate fitness losses due to reduced attractiveness, rewardless species may allocate more resources to other floral functions that promote reproduction, such as display size, floral longevity, or gamete production. Given the lack of estimates of the resource costs of reward production in lineages with deceitful species, the role of resource limitation in the occurrence and persistence of rewardlessness remains unclear. We assessed this resource-limitation hypothesis of rewardlessness with a phylogenetically controlled comparison of the energy costs of flower production and maintenance, and nectar production in the African orchid genus *Disa*, in which nectar production has evolved at least seven times from rewardless ancestors. The costs of 24-h nectar sugar production and maintenance are minute compared to those of flower production, but can accrue considerably due to floral longevity. However, pollination-induced wilting following earlier and more frequent pollination of rewarding than of rewardless flowers limits cost accrual in a higher proportion of rewarding than rewardless flowers. We detected no trade-offs between nectar production and other floral traits in rewarding species, which invested more energy overall than rewardless species in reproduction. Overall greater resource investment in reproduction in rewarding than in rewardless species suggests that the resource costs of reward production may not be limited to the costs of the actual rewards. Instead, reward production may be associated with a life history that trades off present reproduction against long-term survival, whereas rewardless plants may live longer due to lower reproductive investment per season. Transitions from rewardlessness to reward production may be hampered by lower efficiency of resource acquisition in rewardless than in rewarding species.

***Iris pseudacorus* L. (yellow flag) has escaped cultivation in South Africa**

T.P. Jaca (Wed B3)

Early Detection and Rapid Response programme, South African National Biodiversity Institute, Private bag X101, Pretoria 0001, South Africa

The majority of plants that have been introduced to South Africa perform the purpose for which they are intended and therefore benefit humans in multiple ways. A small portion of introduced plants, however escape from cultivation. While most introduced plants will not establish populations outside of cultivation, some probably will become invasive. *Iris pseudacorus* native to Eurasia was introduced sparingly to South Africa as an ornamental plant. The aim of the study was to examine if *Iris pseudacorus* has escaped cultivation or if it is still contained under cultivation. Surveys were conducted in three provinces of South Africa, Gauteng, Limpopo and KwaZulu Natal. The results indicated that *Iris pseudacorus* has escaped from cultivation to wetlands, rivers and other moist habitats, where it makes brilliant yellow displays during spring to early summer. In most of the areas surveyed, the first plants probably have arisen from rhizomes discarded or else removed from nearby gardens and nurseries. Subsequent spread has resulted from the breaking up of rhizomes or from seeds, which are produced in abundance. In some places his species has stated to spread to such an extent and forms such a scattered pattern that it has the appearance of a native plant.

Invasiveness of *Opuntia salmiana* – experimental study on its vegetative and sexual propagation

T.P. Jaca^a and H.M. Venter^b (Tue B2)

Early Detection and Rapid Response programme, South African National Biodiversity Institute, Private bag X101, Pretoria 0001, South Africa

^b*Plant Propagation and Rehabilitation Technologies cc, P.O. Box 99273, Garsfontein East, 0060, Pretoria, South Africa*

Opuntia salmiana (bur cactus) resembles the well known jointed cactus (*Opuntia aurantiaca*) is less spiny with long round cladodes that do not detach that easily. It is native to Chaco of northern Argentina, South-Eastern Bolivia and Paraguay and was probably introduced to South Africa as an ornamental. Plants are shrub like, ±1 m high, often sprawling with long cylindrical stems 1-2 cm in diameter. During summer the plants are recognized by small bright red club-shaped fruits which bear several small 1-2 cm long joints on their tips. Several species of the genus *Opuntia* e.g. *O. imbricata*, *O. kleiniae*, *O. phaeacantha* and *O. rufida*, have fruits that proliferate joints which develop into new plants. These joints are clonal propagules also known as bulbils and are easily detached by passing animals. They are armed with many small bristle-like barbed spines which attach easily to fur, clothes or skin for potential long distance dispersal. Production of attractive red fleshy fruits increases the chances of the bulbils being spread by frugivores. Plant material collected at Wagpos in the Brits district of North West province was studied to determine if the species propagates both through sexual and vegetative regeneration. A stereo microscope study showed that possible viable seeds are produced within the fruits. Vegetative and seed propagation trials were studied in controlled environments. The regeneration ability of bulbils, cladodes stems, timing of shoot emergence and root emergence were studied. Seeds were sown in two treatments: in filter paper in petri-dishes treated with distilled water and

sand in the laboratory. Bulbils and Cladodes regenerated well and seeds did not germinate at all in both treatments. It is concluded that bulbils and cladodes are more crucial than seeds for the spread of *O. salmiana*, contributing to an aggressive increase in population size and the importance of frugivores.

Proteomic analysis of wheat treated with a novel plant activator

R. Janse van Rensburg^a, G. Kemp^b, J.C. Pretorius^c and B. Visser^a (Mon C2)

^a*Dept. Plant Sciences, UFS, PO Box 339, Bloemfontein, 9300*

^b*Dept. Microbial, Biochemical and Food Biotechnology, UFS, PO Box 339, Bloemfontein, 9300*

^c*Dept. Soil- and Crop- and Climate Sciences, UFS, PO Box 339, Bloemfontein, 9300*

Plant activators present a new and exciting approach to improve crop production by increasing overall plant health and yield. The aim of this study was to determine the effect of a novel plant activator called SS on wheat (*Triticum aestivum*) after foliar treatment using shot-gun proteomics. Total protein extracted from SS treated wheat was separated on SDS-PAGE. Polypeptides present in different sections of each gel lane was digested with Trypsin and subjected to LC-MS/MS analysis. Expression levels of identified polypeptides were calculated using spectral counting. A total of 365 polypeptides were identified with 95 showing increased levels following SS treatment. Preliminary results indicated that SS application improved the plant's photosynthetic ability with three different aspects being affected. The proteomic results for selected polypeptide encoding genes were confirmed using qPCR analysis. The positive influence of SS application on photosynthesis in wheat was confirmed using chlorophyll fluorescence data.

First report of Coniothyrium stem canker of eucalypts in Zimbabwe

L. Jimu^{a,c}, S. Chen^b, M.J. Wingfield^b, E. Mwenje^c and J. Roux^b (Wed B3)

^a*Forestry Department, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Private Bag X20, Hatfield, Pretoria 0028, South Africa*

^b*Department of Microbiology and Plant Pathology, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Private Bag X20, Hatfield, Pretoria 0028, South Africa*

^c*Bindura University of Science Education (BUSE), P. Bag 1020, Bindura, Zimbabwe*

Coniothyrium stem canker, caused by species of *Raederiella* (previously *Coniothyrium zuluense*) is one of the most important eucalypt stem diseases in the southern hemisphere. During a disease survey conducted in Zimbabwe, symptoms of the disease, characterised by discrete dark brown sunken lesions, kino exudation and pockets in the wood, were observed in four sites on *E. grandis*, *E. camaldulensis*, *E. grandis*-*X*-*camaldulensis* hybrid clone and *E. propingua*. The ITS region, exons 3 through 6 of the β -tubulin gene and the intron of the translation elongation factor 1- α gene were sequenced to identify the causal pathogen. Representative isolates from Zimbabwe were compared with authenticated isolates of *T. zuluensis* and *T. gauchensis*. Sequence comparisons identified the pathogens as *T. gauchensis*. This study is the first to confirm the presence of Coniothyrium stem canker on eucalypts in Zimbabwe. Furthermore, it is the first to report *T. gauchensis* in Southern Africa.

Using model and manipulated flowers to study pollinator-mediated selection

S.D. Johnson (Mon B2)

School of Life Sciences, University of KwaZulu-Natal, P. Bag X01, Scottsville, Pietermaritzburg 3209, South Africa

Strong selection in the wild has the effect over time of reducing phenotypic variation. To understand the historical influence of selection, it is therefore often necessary to reintroduce phenotypic variation into populations. This can be done expensively through genetic engineering or much less expensively through the use of natural hybridization, reciprocal translocations, physical manipulation of the phenotype or even construction of model flowers. We have used simple morphological manipulations and reciprocal translocations to detect the potential for selection to act on flower colour, nectar guides, shape, scent, protandry, and plant height, and used model flowers to detect the potential for selection to act on flower colour, shape and scent. Measurement of the male component of fitness remains a challenge and we have refined the Peakall method of pollinia staining so that we can now track the fate of individual pollen massulae and thereby estimate male pollination success in orchids with greater precision.

Rapid colonisation by annual alien plants of a high montane roadside in the Drakensberg, South Africa

J.M. Kalwij^{a,c}, M.P. Robertson^b and B.J. van Rensburg^{b,c} (Tue A3)

^a *Institute of Ecology and Earth Sciences, University of Tartu, Tartu, 51005, Estonia*

^b *Department of Zoology and Entomology, University of Pretoria, Pretoria, 0002, South Africa*

^c *DST-NRF Centre for Invasion Biology, Stellenbosch University, Private Bag XI, Matieland, 7602, South Africa*

Invasive alien plants pose a major threat to biodiversity in general and to that of montane ecosystems in particular. The elevational gradient of montane ecosystems offers ample opportunities to study the colonisation rate of alien plant species. Yet to date few such studies exist mostly due to limitations in research funding and scarcity of long-term monitoring programs. This study aimed at filling that gap by annually recording the upper altitudinal limits of alien plant species in the Drakensberg mountain range. From 2008 to 2011 we sampled a 20-km long stretch of the Sani Pass road covering an elevational gradient of 1500–2874 meter above sea level. The average alien species richness did not change over time, with 29.5 annual and 42.0 perennial species. With each subsequent observation we recorded ~7 new alien species to the study area. However, only 19 annuals and 31 perennials were observed annually. A repeated-measures ANOVA showed a significant change in the upper altitudinal limits of annuals that was equivalent to an ascend of ~70 m per year (ANOVA, $F = 5.502$, $df_{\text{year(adj)}} = 2.340$, $df_{\text{error}} = 54$, $P = 0.005$), but not so for the perennials. This rapid ascend could not be attributed to changes in local climatic conditions, density of vehicle movements, or minimum residence time of these alien species in South Africa. Nearly all of our observations were spatially clustered around potential introduction sources

such as the South African border post or residential dwellings. Therefore, the large number of new and disappearing species in combination with rapidly ascending upper altitudinal limits around potential introduction sources suggests that propagule pressure is high and that the process of colonisation and distribution is still ongoing. These results also suggest that an observation period of five years is already sufficient to detect trends in alien plant species.

Quality control of commercially important essential oils and biological activities of selected oils from South African flora

G.P.P. Kamatou^a, M. Sandasi^a, S.F. van Vuuren^b, R.L. van Zyl^b and A.M. Viljoen^a
(Tue C4)

^a*Department of Pharmaceutical Sciences, Faculty of Science, Tshwane University of Technology, Pretoria, South Africa*

^b*Department of Pharmacy and Pharmacology, Faculty of Health Sciences, University of the Witwatersrand, Parktown, South Africa*

Essential oils are volatile compounds isolated from aromatic plants. Like any other product intended for commercial purpose, essential oils sold on the market should meet prescribe standards for safety and efficacy in order to give the desired effect to consumers. The quality control of five essential oils including frankincense (*Boswellia rivae*), buchu (*Agathosma betulina*), cape chamomile (*Eriocephalus tenuifolius*), geranium (*Pelargonium graveolens*) and tea tree (*Melaleuca alternifolia*), will be discussed with regard to the major constituents used to determine “good oil”. The oil is usually evaluated based on the relative percentage calculated from the FID detector. Limitations of the use of relative percentage area, rather than quantification based on calibration curves, will be highlighted. In addition, the biological activities (antimicrobial, antimalarial and anticancer) of the oils isolated from selected South African aromatic plants (e.g. *Vitex* spp, *Salvia* spp) and the problems encountered when investigating their biological properties will be discussed.

***In situ* and *ex situ* soil respiration in natural, invaded, and cleared toposequences in the Fynbos Biome**

O.K. Kambaj^a, S.M. Jacobs^a and K.B. Mantlana^b (Tue B2)

^a*Department of Conservation Ecology and Entomology, Stellenbosch University, Private Bag X1, Maitieland, 7602*

^b*Climate Change and Bio-adaptation Division, South African National Biodiversity Institute, Kirstenbosch Research Centre, Private Bag X7, Claremont 7735, Cape Town, South Africa*

Acacia mearnsii, *A. longifolia*, and *A. dealbata* are among the most widespread alien invasive plants in South Africa and are often located within riparian catchments with extensive woody stands that dominated areas. These invasions may be significant to the overall catchment carbon cycling as soils are often high in soil organic matter and elevated biological productivity due to the availability of nutrients and water in riparian environments. As such seasonal soil respiration measurements were conducted in selected riparian ecotones of the Western Cape, South Africa, with differing invasion status: natural, invaded, and cleared riparian zones. Soil respiration at all sites and within all landscape positions showed significant seasonality, with highest and lowest values observed during the warm and dry

season and the wet season, respectively. Of all environmental factors, R_s were most closely related to soil temperature, while high soil water content during the wet season seemed to inhibit soil respiration. In general soil water content alone was poorly correlated to R_s while a combination of both factors provided a strong relationship with soil respiration. Overall, R_s rates were significantly greater ($P < 0.05$) in the invaded sites compared to the natural and cleared sites with dry banks showing highest soil respiration rates, followed by wet banks and the terrestrial area. Within invasion status, mean daily soil respiration ranged from 0.70 to 3.50 g C m⁻² day⁻¹ in the natural, 1.58 to 6.35 g C m⁻² day⁻¹ in the invaded, and 0.16 to 3.32 g C m⁻² day⁻¹ in the cleared sites. Potential soil respiration, determined in soil microcosms incubated in the laboratory, showed significantly higher R_s in the soils from invaded sites on day 7 and day 14. This study contributes significantly to the understanding of soil respiration and factors driving soil respiration in fynbos biome, and across landscapes. The findings in the field suggest that soil microbial activity increases in riparian areas invaded by *Acacia* species, but upon removal of these woody plants a semblance of the pre-invasion state may return over time, though the work on soil microcosm reveals that some differences may remain.

Breeding and pollination systems in *Clivia* (Amaryllidaceae)

I. Kiepiel and S.D. Johnson (Mon B2)

School of Life Sciences, University of KwaZulu-Natal, P. Bag X01, Scottsville, Pietermaritzburg 3209, South Africa

Clivia (Amaryllidaceae), a small genus of evergreen perennials endemic to southern Africa, has received considerable horticultural and taxonomic attention. Most of the species possess pendulous tubular flowers and are thought to be pollinated by birds. *Clivia miniata* is recently diverged and has upright-trumpet shaped flowers consistent with butterfly pollination. This study investigates the reproductive biology of *C. miniata* and the robust form of *C. gardenii*, also known as *C. robusta*. We document the self-incompatibility systems, floral traits, pollinators, and natural levels of fecundity in these species, and interpret the findings in the context of the phylogeny of *Clivia* and its close relatives.

***Aloe* section *Purpurascentes*: the need for a new classification**

R.R. Klopper^{a,c}, O.M. Grace^b, A.E. van Wyk^c and G.F. Smith^{a,c} (Tue B3)

^a *Biosystematics Research & Biodiversity Collections Division, South African National Biodiversity Institute, Private Bag X101, Pretoria, South Africa 0001*

^b *Jodrell Laboratory, Royal Botanic Gardens, Kew, TW9 3AB, Surrey, United Kingdom / Marie Curie Fellow, Department of Medicinal Chemistry, University of Copenhagen, Universitetsparken 2, Copenhagen, Denmark 2100*

^c *H.G.W.J. Schweickerdt Herbarium, Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002*

Southern Africa is a centre of diversity for the genus *Aloe* L. and the region harbours ± 150 members of this genus that has ± 600 species in total. Many societies utilise aloes for a multitude of biocultural uses, and these succulent plants are immensely popular among plant collectors and horticulturalists. *Aloe* section *Purpurascentes* Salm-Dyck is restricted to South Africa and Namibia and is characterized by acaulescent or caulescent plants with densely

rosulate ensiform leaves and cylindrical flowers with free segments in cylindrical to conical racemes on simple or up to 8-branched inflorescences. Currently this section is usually treated as comprising six species, namely *Aloe framesii* L.Bolus, *A. gariensis* Pillans, *A. khamiensis* Pillans, *A. knersvlakensis* S.J.Marais, *A. microstigma* Salm-Dyck and *A. succotrina* Weston. A taxonomic revision of *Aloe* section *Purpurascetes* is underway as differing views regarding the demarcation of species exist. Matters in need of clarification include: populations of *A. microstigma* in southern Namibian were described under the name *A. juttae* Dinter in 1923, but were alleged to be conspecific with *A. microstigma* in 1974; *Aloe khamiensis* is sometimes included as a synonym of *A. microstigma* subsp. *microstigma* while *A. framesii* is reduced to subspecific rank [*A. microstigma* subsp. *framesii* (L.Bolus) Glen & Hardy]. Furthermore, several morphologically distinct populations of aloes belonging to this section, which warrant possible description as new taxa, are known. New comparative data will be presented based on field studies, evidence from morphology, ecology and biogeography, as well as DNA-based phylogenetic analyses.

‘Toxic nectar’: harmful and beneficial effects of nicotine on an important pollinator, the African honeybee

A. Köhler, C.W.W. Pirk and S.W. Nicolson (Wed A1)

Department of Zoology and Entomology, University of Pretoria, Pretoria 0002, South Africa

Nicotine, best known from the Solanaceae, is a plant defence metabolite and highly toxic to most herbivores. The presence of nicotine in floral nectar and pollen seems paradoxical, and the use of nicotine and neonicotinoid insecticides on agricultural crops has been suggested to contribute to the pollinator declines that are currently observed internationally. We examined the effects of nectar nicotine on honeybee (*Apis mellifera scutellata*) foraging choices and longevity. Using preference tests on free-flying honeybees, we showed that the dose-dependent deterrent effect of nicotine is stronger in more dilute nectars. Secondary metabolites (SM) may act as partial repellents that keep pollinators moving between plants and ensure outcrossing. Plants with nectar SM may be more attractive to pollinators when flowering in isolation or secreting concentrated nectar, and pollinators can avoid intoxication by foraging on multiple nectar sources. We found that caged honeybee worker groups tolerate naturally occurring nectar nicotine concentrations ($\leq 30 \mu\text{M}$) in 0.63 M sucrose, but survival decreased on high dietary nicotine (300 μM). Honeybees injected with *E. coli* lipopolysaccharides showed even lower survival when fed nicotine, suggesting that the combined effect of immune challenges and dietary toxins can be detrimental to colony fitness. However, antimicrobial properties of nicotine may also be beneficial to honeybees: weak workers (possibly affected by a bacterial brood disease) and workers infested with parasitic *Varroa* mites (known to transmit viral diseases) demonstrated prolonged survival on nicotine diets. A possible exploitation of nectar SM for therapeutic purposes may help to explain the role of these compounds in mediating interactions between plants and their pollinators.

The Carotenoid Cleavage Dioxygenase (CCD) gene family in *Vitis vinifera* L.

J.G. Lashbrooke, S.J. Dockrall, P.R. Young and M.A. Vivier (Tue A1)

Institute for Wine Biotechnology, Department of Viticulture and Oenology, Stellenbosch University, Matieland, South Africa, 7602

In plants carotenoids perform crucial roles in photosynthesis, and also serve as the precursors for apocarotenoids: a group of compounds with diverse biological functions. Enzymatic cleavage of carotenoids by the carotenoid cleavage dioxygenase (CCD) family has been shown to produce volatile flavour and aroma apocarotenoid compounds (e.g. β -ionone, geranylacetone, pseudoionone, α -ionone and 3-hydroxy- β -ionone), as well as the phytohormone, strigolactone. Here we identify and characterise the grapevine CCDs involved in apocarotenoid production: CCD1, CCD4a and CCD4b. VvCCD1 has previously been observed to catalyse the *in vitro* cleavage of zeaxanthin and lutein while we show that it is additionally able to cleave β -carotene. *Vitis vinifera* lines were transgenically altered for *CCD1* expression through an RNAi-mediated gene silencing strategy and overexpression of the *CCD1* gene. A transgenic grapevine population showing a 12-fold range of VvCCD1 expression was generated and metabolically characterised. Photosynthetic tissue (leaf) from these lines was analysed for carotenoid and apocarotenoid content via RP-HPLC and HS-SPME-GC/MS, respectively. Results indicate that a large amount of control is exerted on VvCCD1 activity, both post-transcriptionally and possibly by means of subcellular compartmentalisation. Since weak correlation between VvCCD1 gene expression and apocarotenoid production was observed, it appears that the *in planta* action of VvCCD1, certainly in photosynthetic tissue, is distinct from the demonstrated *in vitro* activity. Identification, isolation, expression and functional characterisation of VvCCD4a and -4b provide alternative mechanisms for carotenoid cleavage in *V. vinifera*, L. and an explanation for the divergence between the reported *in vitro* and the *in vivo* activities of VvCCD1.

The systematic value of flower structure in *Crotalaria* and related genera of the tribe Crotalarieae (Fabaceae)

M.M. le Roux and B.-E. van Wyk (Tue B3)

Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park, 2006, Johannesburg, South Africa

Molecular systematics have recently provided a major step towards a profound insight into generic relationships, thereby creating the opportunity to re-evaluate the taxonomic and functional significance of flower structure in the tribe, with emphasis on the large genus *Crotalaria*. Flowers in the tribe Crotalarieae are generally adapted to bee pollination mechanisms. A representative sample of flowers from 211 species was dissected to record morphological character states. Six structural-functional flower types were identified: (1) pump; (2) gullet; (3) hugging; (4) saddle; (5) tunnel and (6) brush (saddle and tunnel types described here for the first time). *Crotalaria* uniquely has the brush type; specialized flowers characterized by a rostrate keel, highly dimorphic anthers, stylar trichomes and four types of elaborate callosities on the standard petal: (1) ridge callosities – vertical swellings on blade and claw; (2) disc callosities – sub-circular swellings on the blade; (3) columnar callosities – cylindrical protruding appendages on the blade; and (4) lamelliform callosities – plate-like

protruding appendages on the blade. Remarkably, *Crotalaria* and *Bolusia* are the only genera of the tribe Crotalariaeae with callosities present in all of the species. In other genera, callosities are generally absent or infrequent. Trends toward specialization are apparent as assemblages of specialized flower characters; individual characters are labile and diagnostically less valuable. Specialized characters often appear to be non-homologous and a result of convergence. Unique combinations of flower characters are often useful to support current generic concepts in Crotalariaeae. The shape of the callosities in *Crotalaria* is diagnostically important to distinguish between unspecialized and specialized infrageneric groups and is congruent with the molecular phylogeny.

Regulation of moss development by strigolactones

B. Lebouteiller, C.W. King, J. Kossmann and P. N. Hills (Mon C2)

Institute of Plant Biotechnology, Department of Genetics, University of Stellenbosch, Private Bag X1, Matieland, 7602, South Africa

Physcomitrella patens, an ephemeral moss widely distributed in temperate zones, is an emergent model for non-vascular plant biology. Because of its simple structure and the intermediate place of mosses in the plant phylogeny, it can be used as a model system for plant growth studies. In the gametophyte stage, the moss colony is formed by two types of filaments, the chloronemata and the caulonemata, with respectively assimilatory and adventitious roles. Plant hormones, particularly auxin, act as internal signals to regulate cell differentiation. The predominance of one filament type over the other is also influenced by external factors including nutrition status, such as the presence of sugars or the source of nitrogen, and light intensity. Analysis of a *P. patens* mutant deficient in strigolactone biosynthesis suggested that strigolactones, like auxin, inhibit protonemal branching. Moreover, the synthetic strigolactone analogue GR24 restores the wild-type phenotype when applied to the mutant, as in *Arabidopsis* or rice strigolactone-deficient mutants. In higher plants, strigolactones participate in the hormonal network which regulates shoot branching and root architecture. Moreover, their effects on the plant development seem to be highly dependent on the environmental conditions, especially nutrient availability in soils. The place of strigolactones in this complex signal network might be conserved in non-vascular plants. Our study aims to better understand the roles of strigolactones in conjunction with other factors regulating the differentiation of moss filaments. For this, we compared moss development on different media, with or without GR24. We have found that some, but not all, hormone effects depend on the medium composition. This new finding suggests that this model system is suitable to investigate the integration of external and internal signals regulating plant development.

How do bird pollinators respond to nectar containing secondary metabolites?

S. Lerch-Henning and S.W. Nicolson (Mon B2)

Department of Zoology and Entomology, University of Pretoria, Pretoria, South Africa 0002

The function of nectar is to reward pollinators. Plants defend themselves against herbivores by producing secondary metabolites (SM) and these are also present in nectar of at least 21 plant families. We need to better understand the effects of SM in nectar on pollinating animals. We investigated how specialist (white-bellied sunbirds, *Cinnyris talatala*) and generalist bird pollinators (dark-capped bulbuls, *Pycnonotus tricolor*) respond to fresh and artificial nectar containing SM. *Erythrina* is an important bird-pollinated genus in Africa and the Americas: leaves and seeds are rich in alkaloids and nectar in amino acids. When offered pure nectar of *E. caffra* and *E. lysistemon* in choice tests, sunbirds rejected the nectar of *Erythrina* species but bulbuls did not. Our experiments with artificial nectar used nicotine, an alkaloid of the Solanaceae and present in nectar of *Nicotiana* spp (3-30 μ M). Firstly, the preference for different nicotine concentrations (0.1-300 μ M) in different sucrose concentrations (0.25 and 1 M) was tested. Birds were deterred by high nicotine concentrations but higher sucrose concentration led to higher nicotine intake, more so in sunbirds than in bulbuls. Secondly, we tested if nicotine influences the feeding behaviour and patterns of white-bellied sunbirds. In the presence of nicotine, sunbirds showed better compensatory feeding at higher sucrose concentrations. Feeding frequency, total time spent feeding and mean duration of feeding events decreased significantly at a high nicotine concentration (50 μ M). If plants have nectar containing SM, generalist pollinators will be less deterred than specialist pollinators but the sugar concentration of nectar can influence this response. High levels of SM had a negative effect on the feeding behaviour of a specialist pollinator, and we did not see the increase in feeding frequency which might translate to more efficient pollination.

Systematics of the early diverging Apiaceae tribe Annesorhizeae with emphasis on the South African genera

A.R. Magee^{a,b}, A.N. Nicolas^c, P.M. Tilney^b, B.-E. van Wyk^b and G.M. Plunkett^c
(Mon B1)

^a*Compton Herbarium, South African National Biodiversity Institute, Private Bag X7, Claremont 7735, Cape Town, South Africa*

^b*Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa*

^c*Cullman Program for Molecular Systematics, The New York Botanical Garden, 2900 Southern Blvd., Bronx, NY 10458-5126, USA*

Annesorhizeae is an early diverging tribe belonging to the family Apiaceae. The tribe forms part of a paraphyletic assemblage referred to as the protoapioids, defined by the presence of scattered druse crystals of calcium oxalate in the mesocarp of the fruit. The tribe currently comprises six genera, four of which are endemic to South Africa. Within the tribe *Annesorhiza*, *Chamarea*, and *Itasina* have been shown to form a strongly supported clade.

The three genera are herbaceous perennials with hysteranthous, summer-deciduous leaves and periodically-replaced fleshy roots. The hysteranthous leaves (withering or absent at anthesis), coupled with the poor collection record and the need to either return to the population at least twice or to cultivate them in order to obtain complete vegetative and reproductive material, has resulted in the number of species within these genera being greatly underestimated. As part of a re-assessment of generic boundaries within the *Annesorhiza* clade, extensive field excursions over the last few years have been undertaken in order to obtain complete material for morphological, anatomical and molecular phylogenetic studies. This has resulted in the clarification of species concepts and affinities and several new species have been identified. Within *Annesorhiza*, the number of species has almost doubled (12 to 21) and includes two species previously assigned to the genus *Peucedanum*. The number of species within *Chamarea* has also doubled with at least five new species awaiting formal description. Generic delimitations of the three genera are explored through phylogenetic analyses for 51 accessions representing 18 spp. of *Annesorhiza*, 9 spp. of *Chamarea*, 1 sp. of *Itasina* and two outgroup species (*Astydamia latifolia*, *Molopospermum peloponnesiacum*). Resolution within *Annesorhiza* was poor, but better resolution within *Chamarea* was obtained. While the monophyly of *Chamarea* is well supported, *Annesorhiza* is shown to be paraphyletic with the monotypic genus *Itasina* embedded within.

Taxonomic studies of *Ursinia*, parachute daisies, (Asteraceae, Anthemideae) in Namaqualand, South Africa

A.R. Magee^{a,b}, J.S. Boatwright^{a,b} and L. Mucina^c (Tue B3)

^aCompton Herbarium, South African National Biodiversity Institute, Private Bag X7, Claremont 7735, Cape Town, South Africa

^bDepartment of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa

^cDepartment of Environment & Agriculture, School of Science, Curtin University of Technology, G.P.O. Box U1987, Perth, WA 6845, Australia

Ursinia is a mainly southern African genus (1 species extending to Ethiopia) comprising ca. 39 species. They are easily recognized by their often large showy ray florets, paleate involucre, scarious involucre bracts and fruit crowned by a pappus of large, white spreading scales. The latter character gives rise to the common name, parachute daisy. The species are currently divided into two subgenera, *Ursinia* and *Sphenogyne*, distinguished most readily by the presence or absence of appendages on the paleae. While preparing the treatment of *Ursinia* for *Greater Cape Plants II: Namaqualand-southern Namib and western Karoo*, a high percentage of misidentified specimens, particularly from Namaqualand, were noted in the herbarium record as well as an undescribed species uncovered from the Kamiesberg. In particular, the delimitation of *U. anthemoides* and *U. calenduliflora* was seen to be problematic. This necessitated a study of the species from this region, the results of which are presented herein. Eight species are here recognised, two from subgenus *Sphenogyne* and six from subgenus *Ursinia*. The species can be distinguished by a combination of their life history, presence or absence of appendages on the paleae, and shape of the involucre bracts and their scarious apices.

Anti-microbial activities of indigenous medicinal plants used in treatment of tick infected cattle wounds in Vhembe District Municipality, Limpopo (RSA)

K. Magwede, M.P. Tshisikhawe and R.B. Bhat (Tue C2)

Department of Botany, University of Venda, Thohoyandou 0950

Plant extracts used for cattle wound treatment in Vhembe District were tested for anti-microbial activity using acetone, ethanol and water as extraction mediums using micro dilution method. Information on plant uses for animal health care administration by the local substantial livestock or cattle breeders was gathered through individual interviews as well as observation. Out of more than 10 plant species mentioned only the most frequently used species were tested. Among plant species mentioned *Terminalia sericea* was one of the most commonly used plant followed by *Cissus quadrangularis* and *Targetes minuta*. All plants species tested were mainly used for the treatment of cattle wounds as the main indication.

Towards the *in vitro* generation of somaclonal variant plants of sugarcane (*Saccharum spp.* hybrids) for tolerance to *Fusarium saccharitoxins*

T. Mahlanza^{a,b}, S.J. Snyman^{a,b}, M.P. Watt^b and R.S. Rutherford^a (Mon C2)

^a*South African Sugarcane Research Institute, Private Bag X02, Mount Edgecombe, 4300, KwaZulu-Natal, South Africa*

^b*School of Biological and Conservation Sciences, P. bag X54001, University of KwaZulu-Natal, Durban, South Africa, 4000*

The fungus *Fusarium sacchari* causes stem rot in sugarcane and the toxicity of the isolate PNG40 to the stalk borer *Eldana saccharina* renders it potentially useful as an endophytic biocontrol agent of this lepidopteran pest. However, its usefulness is restricted due to its pathogenicity on sugarcane. To produce sugarcane lines tolerant to toxins produced by *F. sacchari*, chemical mutagenesis and *in vitro* selection using *F. sacchari* culture filtrates (CF) were utilised. Various selection treatments were established. The first determined the effect of 0-100 ppm CF and at which of the *in vitro* culture developmental stages (embryo maturation, embryo germination and plantlets) they were most effective. Incorporation of CF in the media at either embryo maturation or germination stages resulted in significant callus necrosis, and consequent decreased plantlet yield; percentage callus necrosis was highest with the latter treatment. Another selection treatment established that root re-growth in plantlets with trimmed roots was inhibited by 750-1500 ppm CF. The third selection treatment involved immersing *in vitro* plantlets in varying concentrations of *F. sacchari* conidial suspensions, resulting in 33.3% and 100 % mortality from the 10³ and 10⁵ conidia/ml treatments, respectively. To obtain somaclonal variants, chemical mutagenesis was carried out using ethyl methanesulphonate (EMS). Calli and potential tolerant plants were selected using the established CF and inoculation treatments. In order to confirm tolerance to PNG40, plants that survived all the *in vitro* CF selection pressures were inoculated with *F. sacchari* conidia and are currently being observed for disease symptom development and endophytic colonisation. Future work includes verifying of fungal colonisation by staining and re-isolation of PNG40 from plant tissue and molecular analysis of isolates using inter-simple sequence repeat (ISSR) polymorphisms.

Update on the maize eQTL project

Maize eQTL project consortium (Mon C1)

Department of Plant Science, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria; PANNAR SEED Pty (Ltd), Greytown; African Centre for Crop Improvement, University of KwaZulu-Natal, Pietermaritzburg; Centre for Proteomic and Genomic Research, Cape Town

The maize eQTL project addresses the broad hypothesis that phenotypes are controlled to some extent by gene expression differences between genotypes. The agricultural problem that the project is focusing on is grey leaf spot (GLS) disease of maize, caused by the fungus *Cercospora zeina*. GLS is an economically important constraint for commercial as well as subsistence maize farmers in southern Africa. The project's main aims are to develop improved maize varieties with quantitative resistance to GLS as well as gain an understanding of the molecular basis of this type of resistance, particularly the role of gene regulation. The development of hybrid maize suitable for agro-ecological zones in southern Africa will be discussed as well as an update on the characterization of gene expression differences identified using an eQTL pipeline developed in-house.

Antibacterial, cytotoxic and mutagenic effects of five *Ochna* species

T.J. Makhafola, L.J. McGaw and J.N. Eloff (Wed B1)

Phytomedicine Programme, Department of Paraclinical Sciences, University of Pretoria, Private Bag x04, Onderstepoort, 0110, South Africa

The antibacterial activity and number of antibacterial compounds present in the acetone leaf extracts of *O. natalitia*, *O. pretoriensis*, *O. pulchra*, *O. gamostigmata*, and *O. serullata*, against *Staphylococcus aureus*, *Escherichia coli*, *Enterococcus faecalis* and *Pseudomonas aeruginosa* were determined using the serial dilution microplate assay and bioautography respectively. Some of the *Ochna* species are traditionally used by the Zulu tribe of South Africa for the treatment of dysmenorrhoea, fertility problems, diarrhoea, haemorrhoids, stomach pains and gangrenous rectitis. The cytotoxic effects of the extracts and selective index values were also determined in monkey kidney cells (Vero), human hepatocellular carcinoma cells (C3A) and bovine dermis cells using the MTT assay. Furthermore, their potential mutagenic effects were determined using the Ames test (*Salmonella typhimurium* TA98 and TA100). The MIC values of the five extracts ranged from 39 to 1250 µg/ml. Bioautography showed that each extract had at least 1 antibacterial compound and *O. gamostigmata* had at least 4 active compounds. The IC₅₀ values of the extracts ranged from 26 to 99 µg/ml. Overall all the plant extracts had low selective index values (SI) with SI ≤ 1.3. This is a clear indication of non-selective toxicity i.e. extracts are toxic to bacteria as well as mammalian cell lines. None of the extracts was mutagenic (mutagenic index values ≤ 1.59 for TA98 and ≤ 0.92 for TA100). As a result the extracts may have limited application as ingestible/intravenous therapeutic agents. It may be useful to evaluate acute toxicity in animal studies because cellular toxicity does not always equate to *in vivo* toxicity. However, the extracts may be useful for the treatment of topical bacterial infections.

DNA barcoding medicinal plants of South Africa

L.T. Mankga^{a,b}, A.N. Moteetee^{a,b}, O. Maurin^{a,b} and M. van der Bank^{a,b} (Tue C1)

^a*African Centre for DNA Barcoding, University of Johannesburg, P. O. Box 524 Auckland Park, 2006 Johannesburg, South Africa*

^b*Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa*

The market and public demand for medicinal plants over the past few decades has increased dramatically with more than 1000 plant species actively traded for medicinal purposes throughout South Africa. Intensive harvesting of wild materials is now acknowledged as a serious threat to biodiversity in this country. Also the substitution of a valuable commodity (medicinal plant) by a cheaper alternative (other plant species), either inadvertently due to misidentification, or deliberately to cheat consumers, raises some serious concerns as these adulterants may not be as effective medicinally or may even be toxic and cause harm to consumers. To add to the problem many species are either traded as dried leaf, root, bark products, or extracts and their identification becomes problematic. In the current study we targeted the most commonly used medicinal plants in South Africa and produced a set of herbarium identified vouchers, a set of barcodes for fast and easy DNA-based species identification (*rbcLa*, *matK*, ITS, *trnH-psbA*), and illustrations on how customs officers could use DNA barcoding to identify plant fragments from species in which trade is controlled by the Convention on International Trade of Endangered Species (CITES).

Fly pollination in the achlorophyllous orchid *Gastrodia similis*: does *Gastrodia* provide a breeding site for its insect pollinator?

F. Martos^a, T. Pailler^b, B. Bytebier^a and S.D. Johnson^a (Mon B2)

^a*School of Biological and Conservation Sciences, University of KwaZulu-Natal, P/Bag X01, Scottsville, 3209, South Africa;*

^b*UMR 53 Peuplements Végétaux et Bioagresseurs en Milieu Tropical, Université de La Réunion, 15 av. René Cassin, 97715 Saint-Denis cedex 9*

Among the vast diversity of angiosperms, only a few taxa are known to have engaged in a mutualistic interaction with insect pollinators by providing them a breeding site, such as in the case of some *Ficus* (Moraceae), *Yucca* (Agavaceae) and *Aristolochia* (Aristolochiaceae) species. Orchidaceae are known for a high diversity and specialisation of pollination syndromes. We specifically report a recent pollination study on the tropical, achlorophyllous orchid *Gastrodia similis*, which is endemic to La Réunion Island (Mascarene Archipelago). This orchid attracts drosophilid flies for pollination by producing volatile compounds that mimic fruit odour. We suggest that this nectarless orchid rewards its insect pollinators by providing breeding site.

Evaluating antioxidant activity and cytotoxicity of the selected South African medicinal plant species

N. A. Masevhe^{a,b}, A. Aroke^a, L.J. McGaw^a and J.N. Eloff^a (Mon A2)

^a*Phytomedicine Programme, Department of Paraclinical Sciences, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, Onderstepoort 0110, South Africa.*

^b*Department of Botany, School of Mathematics and Natural Sciences, University of Venda, Thohoyandou 0950, South Africa*

Fourteen different plant species were evaluated for their antioxidant potential using three methods, namely DPPH (1,1-diphenyl-2-picrylhydrazyl radical), ABTS+ (2,2'-azinobis-(3-ethylbenzthiazoline-6-sulfonic acid)), and ferric reducing antioxidant power (FRAP). They were also evaluated for their in vitro toxicity against Vero cell lines using MTT assay ((3-(4,5-dimethylthiazolyl-2)-2,5-diphenyltetrazolium bromide). Four plant species (29 %): *S. acmella*, *S. pinnata*, *O. lanceolata* and *S. brachypetala* exhibited high cytotoxic activity against Vero cell lines and had IC₅₀ values of 13.7±0, 19.88±0.001, 21.62±0.007 and 28.34±0.001 respectively. However, the IC₅₀ values of these plant extracts were higher than that of the positive control, Doxorubicin which was 9.87±0 µg/ml. Plant species that was relatively non-toxic was *F. saligna* with IC₅₀ value of 201.2±0.004 µg/ml. The rest of the plant species (64%) had moderate cytotoxicity activity (30 µg/ml < IC₅₀ < 100 µg/ml. Plant species that had a high level of antioxidant activity in both DPPH, ABTS and FRAP were *O. lanceolata*, *F. saligna*, *A. caffra* and *R. brasiliensis*; and *H. longipetiolata* had potent antioxidant activity in DPPH and ABTS methods while *S. brachypetala* exhibited radical scavenging efficiency in DPPH assay only and *E. transvaalensis* had a reducing power activity in FRAP assay only. These plant species had less antioxidant activity than the controls used. The rest of the plant species had weak antioxidant activity. The results obtained in this study revealed that a high proportion of plant species (93%) contained cytotoxic compounds in their crude extract. On the other hand, medicinal plant species tested in this study showed some free radical scavenging activities using three different methods and can be considered as promising sources of natural antioxidants for medicinal uses.

Long-term vegetation change (1900-2010) in the Karoo Midlands biomes with climate & land use as drivers change

M.L. Masubelele^{ab}, M.T. Hoffman^b and W.J. Bond^c (Mon A3)

^a*South African National Parks, Cape Research Centre, Scientific Services: Cape Node, End of Tokai Road (West) Tokai, 7945, email: mmoto.masubelele@sanparks.org*

^b*University of Cape Town, Plant Conservation Unit, Botany Department, Rondebosch, Cape Town, 7701,*

^c*University of Cape Town, Botany Department, Rondebosch, Cape Town, 7701*

Global climate predictions suggest aridification due to drought in the central interior of South Africa. Biomes in this region are expected to undergo dramatic decline in cover of grasses as shrubs spread from the Nama-karoo into the Grassland biome. Long-term changes in the vegetation within biomes of the Eastern Karoo Midlands Region were studied using historically repeated photography. The aim of the study was to understand the direction of change and also quantify the nature of change for the past 100 years using. Firstly, historical changes in climate variables such as rainfall, drought, temperature and drought were

analyzed. Second, changes in stocking rate between 1910 and 1996 were assessed. Third, 65 repeat photographs were taken within the region and analyzed in terms of the major changes that have occurred within key landform units of the area (e.g. slopes, plains and rivers). We calculated the decadal change in the cover of grasses, dwarf shrubs, tall shrubs and total cover within each landform and biome. Results showed no long-term change in annual rainfall and drought incidence while temperature had increased and evaporation rate were variable. Nama-karoo biome sites showed an increase in the incidence of wet periods and an increase in early seasonal rainfall while the Grassland and the Albany Thicket biomes showed no significant changes. Stocking rates within all three biomes declined in the 20th century. Grass cover increased substantially across the region while dwarf shrubs declined in the Nama-karoo and Grassland biomes. The cover of Tall shrubs increased in Grassland and Albany Thicket biomes. Historical accounts of the region's climate and land use are important since they help to determine the direction and trajectory of change. Trajectories of change are opposite in sign to what is anticipated in the latter part of the 21st century. A long-term monitoring approach, incorporating historical assessment is needed in order to understand how landscape level processes affect human and ecosystem wellbeing.

Bapedi traditional healers in the Limpopo Province, South Africa

K.M. Mathibela^a, S.S. Semanya^a, M.J. Potgieter^a, B.A. Egan^a, H.J. du Plessis^a and L.J.C. Erasmus^b (Tue C2)

Departments of ^aBiodiversity and ^bPhysiology and Environmental Health, School of Molecular and Life Sciences, University of Limpopo, Private Bag X1106, Sovenga 0727, South Africa

This survey documented aspects related to Bapedi traditional healers' practice such as their social characteristics and knowledge of relevant legislation and conservation. Sixty four traditional healers were surveyed via a semi-structured questionnaire across the whole of the Limpopo Province. Results showed that the vast majority of traditional healers are male, aged between 41 and 50, with more than 30 years of experience in traditional healing. The majority have no formal education, with a minority that managed to reach secondary school. Because of this low level of literacy they tend to shy away from sources of written information, with the result that none of the questioned healers had any knowledge of the various national or provincial environmental legislations. This had a knock on effect in their view of anything formal, with traditional healers tending to view conservation efforts that involve procedures such as permits as obstacles to their practice. Rituals play an integral part of traditional healing, especially prior to plant harvesting. Ancestral guidance stands central in traditional healing. For example the ancestors are consulted when information is shared; to determine the efficacy of their medicine and to obtain healing knowledge.

An ethnobotanical survey of mosquito repellent plants in uMkhanyakude district, KwaZulu-Natal province, South Africa

E.J. Mavundza^{a,b}, R. Maharaj^a, J.F. Finnie^b, G. Kabera^c and J. van Staden^b (Tue C2)

^a*Malaria Research Unit, Medical Research Council, 491 Ridge Road, Overport, Durban 4001, South Africa*

^b*Research Centre for Plant Growth and Development, School of Biological and Conservation Science, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa*

^c*Biostatistics Unit, Medical Research Council, 491 Ridge Road, Overport, Durban 4001, South Africa*

An ethnobotanical survey of plants used for repelling mosquitoes was carried out in uMkhanyakude district, KwaZulu-Natal province, South Africa. The specific objectives of this survey were to (1) identify plant species and their parts being used; (2) determine the condition of plant material used and the method of application. Data was collected from 60 respondents in five villages in the district using standardised and pre-tested questionnaires. The study revealed that 13 plant species are used in the study area to repel mosquitoes; 2 of the plants remain to be identified. The identified plant species were belonging to 11 genera from 9 families. Meliaceae and Anacardiaceae were the most represented families with two species each. The most frequently recorded species were *Lippia javanica* (91.67%), followed by *Aloe ferox* (11.67%), *Sclerocarya birrea* (5%), *Melia azedarach* (3%), *Balanite maughamii* (3%) and *Mangifera indica* (3%). Leaves were the most (38%) common plant part used. The majority (82%) of the plant parts were used in a dry state. Burning of plant materials to make smoke was the most (92%) common method of application. Nine plant species, namely: *A. ferox*, *Calausena anista*, *Croton menyharthii*, *S. birrea*, *B. maughamii*, *Oxalys dissitiflora*, *Trichilia emetic*, *M. indica*, and *Atalaya alata* are documented for the first time as mosquito repellents. This documentation provides the basis for further studies in developing new, effective, safe and affordable plant-derived mosquito repellents especially for Africa where malaria is highly prevalent. The study also plays a part in documenting and conserving traditional knowledge of mosquito repellent plants for future use.

Heterologous expression of RANTES analogues in *Lycopersicon esculentum* Mill

K.G. Mawela^{a,b}, E. Chakauya^b and R. Chikwamba^b (Mon A1)

^a*Phytomedicine Programme, University of Pretoria; Private Bag X04, Onderstepoort 0110 South Africa*

^b*CSIR Biosciences, PO Box 395, Pretoria, 0001*

AIDS is one of the most severe, deadly diseases that claimed lives of many people across the globe. An estimate of 42 million people are found to be living with HIV/AIDS worldwide, and more than 3 million die every year from AIDS-related sicknesses. Women are the most susceptible people of the pandemic. Hence there is an urgent need for microbicides research intervention. RANTES analogues are natural ligands for the CC chemokine receptors such as CCR5 which block HIV infection and have been found to be effective ingredients for microbicides. The current study investigated the expression of

RANTES analogues in plants as an alternative production system for the microbicides. Two RANTES analogues were transiently expressed in *Lycopersicon esculentum* Mill via agrobacterium-mediated transfection. Magnifection system was used to deliver the RANTES constructs in *L. esculentum* species. The RANTES genes were successfully transfected into four developmental stages of mature tomato fruits for protein expression. Protein expression was evaluated by ELISA and was detectable from 3-9 day post infiltration (dpi). The highest expression level was obtained in apoplast at 9 dpi. A gradual decrease in the expression level was observed from green to red tomato fruits per construct. 5P12-RANTES construct gave the highest expression level of about 25ug/kg. Western blot was conducted to confirm the size of recombinant proteins in *L. esculentum* species which corresponded with the RANTES positive controls. Our results highlight the potential usefulness of plants for the production of RANTES in tomato fruits, a food/feed crop, for preventive intervention of HIV in humans.

Genotypic and phylogeographic investigation of indigenous and alien *Tamarix* species in southern Africa

S.G. Mayonde, G. Cron and M. Byrne (Wed C2)

Department of Animal, Plant and Environmental Science, University of Witwatersrand, Private BagX3, WITS 2050, Johannesburg, South Africa

Tamarix (Tamaricaceae) is from the Old World, but has become naturalized and invaded other parts of the world. *Tamarix usneoides* is the only species native to southern Africa, but *T. aphylla*, *T. chinensis*, *T. parviflora* and *T. ramosissima* have been reported as exotic in South Africa. *Tamarix* species are hypothesized to be hybridizing among themselves, and *T. ramosissima* has been declared invasive. *Tamarix usneoides* is used for phytoremediation in the mines in southern Africa as it hyper-accumulates sulphate and metals from Acid Mine Drainage from Tailing Storage Facilities and excretes gypsum (CaSO₄). *Tamarix* species are morphologically and ecologically similar, making them very difficult to distinguish and hybridization adds to the taxonomic confusion. Identification of *Tamarix* species in South Africa is of great importance because of the invasive potential of *T. ramosissima* and also the potential usefulness of *T. usneoides*. This investigation aims to identify populations of pure *Tamarix usneoides* that can be cloned for cultivation for phytoremediation on the mines, and to reveal the geographic origin of the invasive species (e.g. *Tamarix ramosissima*) to facilitate a biocontrol programme. In this study, nuclear and plastid DNA sequence markers are used to characterise southern African *Tamarix* species and their putative hybrids genetically and to assist in recognition of hybrids. Phylogenetic analyses and analysis of polymorphisms indicate that many of the populations sampled are of hybrid origin, most notably those currently being used on the mines for phytoremediation. Morphological characters are not always reliable detectors of hybridization.

Antimycobacterial activity, cytotoxicity and genotoxicity studies of *Ptaeroxylon obliquum* and *Sideroxylon inerme* leaf extracts

L.J. McGaw, T.J. Makhafola, O.O. Udom, K.T.V. Mayekiso and J.N. Eloff (Mon A1)
Phytomedicine Programme, Department of Paraclinical Sciences, University of Pretoria, Private Bag X04, Onderstepoort 0110, Pretoria, South Africa

Two plant species, namely *Ptaeroxylon obliquum* and *Sideroxylon inerme*, were investigated for antimycobacterial, cytotoxic and genotoxic activity following good activity detected in an initial random screening procedure of a large number of extracts. Acetone extracts of the leaves were prepared and tested *in vitro* for activity against *Mycobacterium smegmatis* and *M. bovis* BCG. MIC values ranged from 156 to 313 µg/ml. *P. obliquum* was relatively cytotoxic to human liver (C3A) cells, with an LC₅₀ value of 86 µg/ml. The selectivity index (SI) values for this extract with regard to *M. smegmatis* and *M. bovis* BCG were 0.552 and 0.275 respectively. *S. inerme* was less cytotoxic, with an LC₅₀ value of 1 039 µg/ml, resulting in promising SI values of 6.66 for both *M. smegmatis* and *M. bovis* BCG. Neither of the two extracts was mutagenic in the Ames test against *Salmonella typhimurium* strains TA98 and TA100, without metabolic activation. However, in the comet assay, the two extracts were moderately genotoxic when using tail moment (a measure of both amount of DNA in the tail and distribution of DNA in the tail) as an assessment parameter, with *P. obliquum* showing a more pronounced effect. This extract may be responsible for the induction of double stranded DNA breaks and may thus be a potential mutagen (with a higher % DNA in the comet tail and short tail length). Isolation of antimycobacterial compounds from these species is currently being undertaken.

Analysis of differential gene expression during the early stages of rust infection in a sugarcane breeding population

T.T. Mhora^{a,b}, R.S. Rutherford^a, D.L. Sweby^a, R.M. Jacob^a, L. Makome^a and J.W. Danson^b (Tue A2)

^a*South African Sugarcane Research Institute, Private Bag X02, Mount Edgecombe, 4300, South Africa*

^b*School of Agricultural Sciences and Agribusiness, University of KwaZulu-Natal, Pietermaritzburg, Private Bag X01, Scottsville, 3209, South Africa*

Resistant cultivars are the most effective way of controlling brown rust of sugarcane caused by *Puccinia melanocephala*. Molecular techniques can be used to identify undocumented rust resistance mechanisms, which can then be utilised in breeding strategies to develop cultivars with increased resistance and durability. Suppression Subtractive Hybridization (SSH) was used to identify genes differentially expressed between sugarcane genotypes that are resistant (two groups with different modes of resistance, each group having three genotypes), intermediately resistant and susceptible to rust. Leaves were sampled 48h after inoculation with *P. melanocephala*. mRNA for each genotype was purified, pooled into its respective groups and converted into cDNA using the Clontech SMARTer™ cDNA synthesis kit. Two subtracted cDNA libraries were constructed using a PCR-Select™ cDNA subtraction kit. For Library 1 pooled samples of Susceptible and Intermediate resistant genotypes were subtracted from Resistant Group One. For Library 2, Resistant Group Two was subtracted from Resistant Group One. Subtracted cDNA libraries

were transformed into *Escherichia coli*. The cDNA inserted using the pGEM®-T easy vector in individual bacterial colonies was amplified by PCR and showed successfully transformed clones with cDNA inserts ranging from 200-1300bp. DNA Sequence analysis of inserts and BLAST searches for both libraries revealed that over 70% of the sequences were identified as various disease and drought stress related gene sequences. Library 1 revealed enrichment for serine/ threonine kinases, reticulon-like proteins and RNA recognition motifs, domains found in proteins involved in post-transcriptional gene expression processes. The *Mob1* gene was also identified in Library 1. *Mob1* is known to play a role in hypersensitive responses and in cytokinesis, critical processes in host resistance to pathogens. Processing of Library 2 results is still in progress. These results show the usefulness of SSH in providing information on gene expression in response to brown rust infection. This information could be used to develop markers for resistance, having potential application in the breeding and selection programme.

In the right place at the right time: why some introduced Proteaceae spread at some sites but fail at many others

D. Moodley^a, J.R. Wilson^{a,b}, S. Geerts^{a,b}, D.M. Richardson^a and A.G. Rebelo^b (Tue A3)

^a*Centre for Invasion Biology, Department of Botany and Zoology, Stellenbosch University, Private Bag X1, Matieland 7602, South Africa*

^b*South African National Biodiversity Institute, Kirstenbosch National Botanical Gardens, Claremont, South Africa*

The success of invasive alien plants is attributed to them overcoming a series of barriers during the introduction-naturalization-invasion process. We explore why certain species are invasive in some locations, but not in others using Proteaceae as a test case. Many species of Proteaceae are used as cut flowers, for hedges and ornamental plants, in landscaping and for food. Consequently, many species of Proteaceae have had a long history of introductions to regions outside their native ranges. Some Australian Proteaceae species are notable invaders, but others are notoriously difficult to cultivate. However, although some species are rather difficult to cultivate, a few of the Proteaceae introduced to South Africa, particularly *Hakea salicifolia* and *Banksia integrifolia*, have naturalised or are spreading at some sites, but have failed to naturalise at many other sites. The Protea Atlas and the Southern Africa Plant Invasion Atlas were used as initial sources for obtaining locality information for alien Proteaceae in South Africa. Populations in unmanaged sites were selected, since plants are more likely to spread at these sites. The extent and abundance of planted and self-sown plants was determined. Sampling involved mapping the source population(s), and mapping and categorizing the number of naturalised plants into different size classes, namely seedlings, juveniles and young adults. Data on the presence of herbivory, time since last fire, presence of phytophthora, land use, topography, reason for planting, management, seed viability, rainfall, temperature, elevation and soil type were collected. The status of populations, whether naturalised or non-naturalised, was related to the predictors by means of generalized linear models in order to determine which factors facilitate spread. Preliminary results will be discussed in terms of the importance of site specific factors in predicting the likelihood of a species naturalizing and these factors include climate, disturbance, introduction history and propagule pressure.

The use of bioclimatic modelling in predicting the relative invasion success of introduced acacias in southern Africa

R.F. Motloung^a, M.P. Robertson^a, M. Rouget^b and J.R. Wilson^{c,d} (Tue B2)

^a*Department of Zoology and Entomology, University of Pretoria, Pretoria, South Africa 0002*

^b*Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002*

^c*Centre for Invasion Biology, Department of Botany and Zoology, Stellenbosch University, Matieland, South Africa 7602*

^d*South African National Biodiversity Institute, Private Bag x7, Claremont, South Africa 7735*

The likelihood of successful establishment of introduced plant species has been predicted using various modelling techniques. Although these models have considerable potential in risk assessment, this has been rarely tested. We aim to determine how useful bioclimatic models are at predicting the establishment or invasion success of introduced plant species and its implications for risk assessment. About 69 Australian *Acacia* species have been introduced into southern Africa for various purposes, of which 38 species were introduced for government forestry trials. Firstly, we compare the introduction success of each species with respect to climatic suitability. And secondly, for 15 Australian acacias reported to be invasive in Southern African Plant Invaders Atlas (SAPIA), we determine whether bioclimatic models could have successfully predicted their spread. Bioclimatic models were calibrated using occurrence datasets from Australia as the native range, and evaluated in the introduced range using government forestry trials and the Southern African Plant Invaders Atlas. Models were tested against independent records by calculating true skill statistic which quantifies omission and commission errors, sensitivity and specificity. Bioclimatic models suggest that a large portion of southern Africa is climatically suitable for Australian acacias. Overall there was a fair agreement between the models and government forestry trials dataset (specificity of 0.83, sensitivity of 0.68, and TSS score of 0.51). However, the models poorly predicted the distribution of invasive acacias (specificity of 0.23).

Antioxidant activities of nine medicinal plants used in treating inflammatory ailments in Zulu traditional medicine of South Africa

E. Muleya^{a,b}, A.S. Ahmed^b, A.M. Sipamla^a, F.A. Mtunzi^a and J.N. Eloff^b (Mon A2)

^a*Vaal University of Technology, Chemistry Department, Private Bag X021, Vanderbijlpark, 1900, South Africa*

^b*Phytomedicine Programme, Department of Paraclinical Sciences, Faculty of Veterinary Sciences, University of Pretoria, Private Bag X04, Onderstepoort, 0110, South Africa*

Inflammation is a complex interaction between pro-inflammatory and anti-inflammatory mediators in response to induced negative stimuli in which the former predominates. Many allopathic medications currently used in alleviating inflammation are associated with some major side effects such as intestinal and cardiac toxicity. However, medicinal plants are also used in many traditional practices against inflammatory complications. If the active components present in medicinal plant extract affect the same molecular targets as mainstream interventions with no side effects, such extracts could provide attractive and cost-effective alternatives to the conventional anti-inflammatory agents. Considering the importance of oxidation in inflammatory mechanisms, the free

radical scavenging activities of *Pentanisia prunelloides*, *Pomaria sandersonii*, *Alepidea amatymbica*, *Gunnera perpensa*, *Carissa bispinosa*, *Artemisia afra*, *Eucomis autumnalis*, *Ledebouria revoluta* and *Berkheya setifera* used to remedy inflammation in Mabandla Village, Kwa-Zulu Natal, South Africa was carried out against ABTS^{•+} and DPPH[•] radicals. Results from both assays indicated that some of the plants had good antiradical properties. For DPPH[•] radical assay, EC₅₀ values ranged between 1.9 mg/L from *L. revoluta* crude to 467 mg/L from hexane fraction of *C. bispinosa*. The trend of activity in ABTS^{•+} radical scavenging was similar to the DPPH trend. However, *P. sandersonii* extracts was the most active in this case inhibitory with EC₅₀ values of 1.27 mg/L for crude extract, 1.66 mg/L for DCM and 1.27 mg/L for acetone fraction. *Carissa bispinosa* crude extract had the lowest activity against the ABTS^{•+} and DPPH[•] radicals (190.6 mg/L and 25.45 mg/L respectively). The good antiradical results obtained for some of the plants indicate that antioxidant activities may contribute to their anti-inflammatory potential, therefore validating their traditional use as therapeutic in inflammatory disorders.

Elucidation and characterization of markers for the salicylic acid and methyl jasmonate pathways in *Eucalyptus grandis*

R. Naidoo^a, S. Naidoo^a, D.K. Berger^b and A.A. Myburg^a (Tue A2)

¹*Department of Genetics, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria 0028*

²*Department of Plant Science, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria 0028*

The South African forestry industry relies significantly on the efficient production of various *Eucalyptus* species primarily for their use in manufacturing wood and paper products. Although these trees are relatively sturdy, they can succumb to diseases caused various pathogens including *Chrysosporthe austroafricana*. Much of the information pertaining to plant defence stems from model organisms such as *Arabidopsis thaliana* and therefore there is a need to investigate this niche in *Eucalyptus*. Among the plethora of defences, signalling pathways such as salicylic acid (SA) and methyl jasmonate (MeJA) are triggered and assist in curbing the spread of the pathogen. Each of these signalling pathways initiate the expression of a suite of pathogenesis related (*PR*) genes which have been shown to be markers for the induction of the signalling pathway. Based on this premise, the aim of this study was to elucidate and characterize the *PR* genes associated with SA and MeJA in *Eucalyptus grandis*. Using the genome of *E. grandis* and a phylogenetic approach, orthologous markers for the SA (*PR1*, *PR2*, *PR5*) and MeJA (*PR3*, *PR4*, *LOX*) pathway were identified based on *Arabidopsis* sequences. Expression profiling using Reverse Transcription quantitative PCR (RT-qPCR) was performed with these markers in a dose response and time dependent experiment. The dose response experiment indicated that 5 mM and 100 μ M induced the most significant change in transcript expression for the putative marker genes associated with SA and MeJA respectively. Additionally the marker genes supported an antagonistic relationship between SA and MeJA in *Eucalyptus* as previously described in *Arabidopsis*. These putative markers were furthermore investigated in *Eucalyptus* plants challenged with *Chrysosporthe austroafricana*, which revealed a possible role for SA in the tolerant interaction. This study adds to the current knowledge of defence responses in *E. grandis* and provides a stepping stone for understanding plant resistance mechanisms to curb future tree diseases.

Gene conversion of the ribosomal RNA cistron

K. Naidoo^a, M.P.A. Coetzee^a, E. Steenkamp^b, B. Slippers^a, M.J. Wingfield^a and B.D Wingfield^a (Mon B1)

^a*Department of Genetics, Forestry and Agricultural Biotechnology Institute (FABI), Faculty of Natural and Agricultural Sciences, University of Pretoria, South Africa, 0002*

^b*Department of Microbiology, Forestry and Agricultural Biotechnology Institute (FABI), Faculty of Natural and Agricultural Sciences, University of Pretoria, South Africa, 0002*

The nuclear ribosomal RNA cistron, comprising of the 18S-5.8S-26S gene regions, is most frequently targeted for phylogenetic reconstruction and DNA based identification in the eukaryotic kingdom. Identity among the numerous tandem repeats of this element are assumed to be maintained through gene conversion. During this process, concerted evolutionary forces are thought to prevent genes within individual repeat elements from accumulating mutations. It is however, not yet known when during the cell cycle this process occurs. By making use of a fungal model, our overarching aim was to determine if gene conversion in the ribosomal RNA cistron was as a result of meiotic events, mitotic events or a combination of both. *Ceratocystis manginecans*, a haploid homothallic ascomycete, was found, through routine population studies, to have two distinct sequences within the internal transcribed spacer region of the cistron. These two sequence types were profiled in mitotically and meiotically derived populations using PCR and restriction fragment length polymorphism. The relative ratios of these two sequence types changed during mitosis when the fungal isolates were cultured as well as during meiosis when spores are produced. These changes were shown to occur more frequently during meiosis than mitosis. These results thus provide empirical evidence of gene conversion occurring during both meiosis and mitosis, and not only as previously thought during meiosis. This is the first time that gene conversion of ribosomal RNA repeat elements has been shown experimentally.

Phytohormone choice and the *in vitro* development of eucalypt roots

M. Nakhooda^a, M. P. Watt^a and D. Mycock^b (Mon C2)

^a*School of Life Sciences, University of KwaZulu-Natal, Durban, South Africa*

^b*School of Animal, Plant and Environmental Sciences, University of the Witwatersrand, Johannesburg, South Africa*

Eucalyptus micropropagation has great potential in supplying large numbers of superior but otherwise difficult-to-root clones for forestry deployment. Studies indicate, however, that micropropagated plants possess an inferior root system once acclimatised, compared with those of seedlings and mini-cuttings. Since adventitious rooting *in vitro* is often under the influence of empirically-selected exogenous auxins, the specific actions of these auxins on root induction and development needed investigation. The roles of two natural auxins - indole-3-acetic acid (IAA) and indole-3-butyric acid (IBA) – were presently investigated, using two eucalypt clones of differing rooting ability (clone 1- roots to 87% and clone 2 roots to 45%, when using a standard protocol), in conjunction with the IAA-specific transport inhibitor 2,3,5-triiodobenzoic acid (TIBA). It was found that while IBA was able to induce roots *in vitro*, subsequent root development and graviperception was dependent on shoot IAA content, as established using gas chromatography-mass spectrometry (GC-MS). However, owing to its greater stability over IAA, IBA remains the preferred auxin for adventitious rooting, particularly for difficult-to-root clones. The apparent inability of poor-rooters to

respond to IAA was therefore investigated using clone 2. It was found that this clone was not deficient in auxin perception (using the inhibitor of auxin signal transduction ρ -chlorophenoxyisobutyric acid - PCIB), nor was its poor rooting properties attributed to auxin conjugation (using the auxin conjugation inhibitor dihydroxyacetophenone (DHAP)). However, using GC-MS, a strong relationship was realised between shoot cytokinin:auxin and rootability. Reducing cytokinins from the pre-rooting culture stages, or using the less stable cytokinin, *trans*-zeatin, significantly improved the rooting ability of clone 2 (100% with IAA). This indicated that the phytohormones chosen for eucalypt micropropagation should be based on phytohormone properties and interactions, with regard to developmental requirements of shoots and roots, rather than in isolation, based on potency.

***In vitro* propagation of *Cyrtanthus* species**

B. Ncube, J.F. Finnie and J. van Staden (Wed C3)

Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa

Cyrtanthus (Amaryllidaceae) is a genus of perennial geophytes, endemic to the southern African region. In South Africa, the genus represents an important group of monocotyledonous bulbous plants of which the bulbs and flowers are highly sought after for both medicinal and ornamental purposes. Due to their valuable use in traditional medicine and potential for the production of novel alkaloids, the three *Cyrtanthus* species (*C. contractus*, *C. guthrieae*, *C. obliquus*) have been reported to be declining rapidly in their natural habitats in South Africa. In recognition of this fact, increasing the population of these plant species through cultivation would be an important strategy for meeting a growing demand as well as maintaining an ecological diversity. Twin scales from the three species were cultured on solid Murashige and Skoog (MS) media with concentrations of 6-benzyladenine (BA) (0 - 17.7 μ M) and naphthalene acetic acid (NAA) (0 - 10.7 μ M) under continuous darkness and 16/8 h light/dark conditions. The best shoot initiation responses were obtained on MS medium containing 8.9 μ M BA and 5.4 μ M NAA under continuous darkness at 25 °C (3.1 shoots/explant) for *C. contractus*, 4.4 μ M BA and 1.1 μ M NAA at 25 °C under 16 h light /8 h dark regime (6.3 shoots/explant) for *C. guthrieae* and 6.7 μ M BA and 2.7 μ M NAA at 25 °C under 16 h light /8 h dark (1.9 shoots/explant) for *C. obliquus*. The effects of different concentrations of cytokinins (BA, kinetin and zeatin) on shoot multiplication were evaluated. The developed micropropagation protocols provide rapid and cost effective ways for the conservation and domestication of *Cyrtanthus* species.

Toxicity studies on four *Hypoxis* species and a commercial herbal mixture

A.R. Ndhkala^a, R. Anthonissen^b, G.I. Stafford^c, L. Verschaeve^b and J. van Staden^a (Mon A2)

^aResearch Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3201 South Africa

^bToxicology, Scientific Institute of Public Health, Juliette Wytsmanstreet 14, B-1050 Brussels, Belgium

^cBotanical Garden and Museum, Natural History Museum, University of Copenhagen, Sølvgade 83, DK-1307 Copenhagen, Denmark

Several claims have been made for extracts emanating from *Hypoxis* species. These include efficacy as stimulant of the immune system, which has led to the species being given credit for its health benefits to HIV/AIDS sufferers. Another claim made for *Hypoxis* spp. includes their use as agents for slowing down the growth of certain types of cancer, having a positive effect in combating colds and being highly effective in alleviating arthritis. These widespread claims have led to the appearance of commercial preparations made of *Hypoxis* sp. Extracts. Amongst them the *African potato extract*TM. Within the genus, two species, *H. hemerocallidea* and *H. colchicifolia* are particularly popular both as African traditional remedies and for the preparation of herbal teas and tinctures. Using the comet and micronucleus/cytome and neutral red uptake assays, four *Hypoxis* species commonly used in traditional medicine in South Africa and an unregistered commercial *Hypoxis* product were evaluated for their potential toxicity. The results reflected lack of toxicity in all four *Hypoxis* species analyzed as well as in the commercial herbal mixture. It is, however, important to carry out confirmatory tests using other tests such as the VITOTOX® test for toxicity and/or *in vivo* assays.

The use of a locally developed innovation on biopesticide for pest management in dry beans in Limpopo Province

R. NemaKanga and S.R. Sasa (Tue C2)

Limpopo Department of Agriculture, Private Bag X9487, Polokwane 0700

The production of dry beans in Mbahela has been decreasing rapidly in recent year's even though there were lots of flowers during the flowering stage of the crop. However farmers started to observe the Cape Mounted Rifle (CMR) beetles during flowering stage and realized that the beetles were feeding on the flowers of the dry beans hence a decrease in production. At first the farmers have been using commercial pesticides but it soon came out that they could not continue with this practice due to the high costs of the input. In order for the small holder farmers to reduce the costs of expensive pesticides and also eliminate the health hazards caused by these inputs, farmers came up with their own innovation to control the pests in a biological way. The objectives of the study are to determine the effectiveness of the biopesticide mixture on the CMR beetles affecting dry beans and to determine the effect of the mixture on the dry beans yields. Special attention should be given to the dry beans plant during flowering stage because it determines and affect the yield of the crop. During reproductive stages, there are lots of insects that linger around the beans because they are attracted by the flowers. Categories of these insects are: 1) Pollinators such as bees and 2) Pests that feed on these flowers resulting to reduced production of the dry beans. The CMR

beetles are often very abundant in South Africa and do a great deal of harm to crops. Dry bean seeds were planted on deep well drained sandy loamy soil of at least 90cm with no nutrient deficiencies with the planting in between space of 900m and spacing within the rows of 75mm and the population seeds ha⁻¹ of 150 000 on 2x3 factorial experiment in a RCRD replicated three times with two red speckled dry beans cultivars Kranskop and OPS-RS1 and three treatments namely Control, Mercaptothion and Biopesticide mixture (100%). Each plot will consist of three 5m rows. The furrow irrigation was used in the trial.

Perception and concept on diarrhea treatment amongst traditional healers of Vhembe District Municipality, Limpopo (RSA)

T.A. Netshivhulana and M.P. Tshisikhawe (Tue C2)

Department of Botany, University of Venda, Thohoyandou 0950

The study focused on the perception and the concept of traditional healers on diarrhoea in Vhembe District Municipality, Limpopo Province, South Africa. The study was dealt with the understanding and belief system towards diarrhoea as per traditional healers' understanding. Data was collected through using questionnaire establishing patterns towards description of diarrhoea and categories of diarrhoea. Dietary requirements for diarrhoeal patients were also recorded. The study revealed that traditional healers in Vhembe district have a lot of information on the causes as well as categories of diarrhoea and dietary requirements of diarrhoeal patients. The study showed that traditional healers of Vhembe District have a better understanding when it comes to the perception and concept of diarrhoeal disease.

Bee food: the chemistry and nutritional value of nectar and pollen

S.W. Nicolson (Tue B1)

Department of Zoology and Entomology, University of Pretoria, Pretoria 0002, South Africa

Bees are herbivorous insects, consuming nectar and pollen throughout their life cycles, and are excellent pollinators as a result. One of the factors contributing to current declines in bee populations may be poor nutrition, due to forage loss and dependence on monocultures, and this may interact with the adverse effects of pesticides and pathogens. I present a brief overview of the chemistry of floral resources for bees and the implications for their nutrition. Nectar is primarily an energy source, but in addition to sugars contains various minor constituents that may, directly or indirectly, have nutritional significance. Pollen provides bees with the protein, lipids, vitamins and minerals that are essential for larval rearing. Chemical analyses of pollen have tended to focus on the protein component of bee-collected pollens as an index of nutritional quality. However, the substantial nectar content of such samples (~ 50% dry mass) should not be ignored, especially in view of current interest in assessing the nutritional quality of floral resources for bees.

Early detection of the new potential invader; *Epipremnum aureum* (L.) Engl.

S.N. Nzama and H.J.N. Sithole (Tue A4)

Early Detection and Rapid Response to emerging invasive alien plants programme (EDRR – funded by Working for Water) South African National Biodiversity Institute, P.O Box 52099, Berea Road, 4007, Durban, South Africa

Epipremnum aureum (L.) Engl. (Silver vine) is a potential invader recently detected by the Early Detection and Rapid Response (EDRR) programme in KwaZulu-Natal. It is an epiphytic climber from Araceae family and is characterized by remarkable huge leathery leaves with yellow or white marks. This plant is native to Southeast Asia and has spread to tropical areas where it is highly invasive hence poses an invasion threat to South Africa's subtropical coastal areas. *E. aureum* was introduced for ornamental purposes/as a garden plant where it has escaped to the wild and now is realizing the potential of becoming invasive. Forest margins and disturbed sites are prone to its invasion. In South Africa this species is not yet included in the draft list of declared invaders. In preparation to get it listed, a preliminary survey is conducted to map it in the wild and assess its invasiveness. Biology studies are also done to determine its life/growth form. Nurseries within the South coast in KwaZulu- Natal are surveyed to determine if horticultural trade have a role to play in distributing *E. aureum*. The purpose of this talk is to highlight our preliminary ecology results and present the nursery survey findings. This talk also serves to motivate for a need of long-term research on ecological studies and control methods for rapid response.

A hypothetical *Eucalyptus grandis* defence model against *Leptocybe invasa* based on transcriptome sequencing

C.N. Oates, A.A. Myburg, B. Slippers and S. Naidoo (Tue A2)

Faculty of Natural and Agricultural Sciences, Department of Genetics, Forestry and Agricultural Biotechnology Institute, University of Pretoria, 0002

Eucalyptus species are widely used in commercial plantations due to their superior growth, adaptability and wood properties. A number of industries rely on these trees to supply the fibre, pulp and sawn wood for their products. The *Eucalyptus* gall wasp, *Leptocybe invasa* (Hymenoptera: Eulophidae), was first described in Israel in 2000 following the extensive damage it caused in plantation nurseries. Adults oviposit in young tissue along the midribs, petioles and stems of numerous *Eucalyptus* species. The larvae are endophytic herbivores whose feeding leads to the induction of gall formation. Severely infested trees show a range of symptoms including a gnarled appearance, stunted growth and death. Variations in resistance and susceptibility have been noted across different *Eucalyptus* genotypes. There is currently no information available regarding the mechanism of defence in these plants. The aim of this study is to investigate the defence response of a resistant *E. grandis* clone against *L. invasa*. Year old *E. grandis* seedlings were coppiced and maintained in a *L. invasa*-free environment until sufficient young material had been produced. Three biological replicates of six plants each were naturally infested over seven days. Thereafter, leaf midribs were excised and total RNA was isolated from attacked and control plants for RNA sequencing. Initial analyses have identified 1381 significantly differentially expressed genes from the 44974 current *E. grandis* gene models. Over representation data appears to

support a defence model that includes a number of well described responses, such as the oxidative burst and the jasmonate-mediated signalling pathway. Some other defences, which are less well characterised in previous studies, include evidence for a gene-for-gene recognition system, the suppression of nutrient availability to the larvae and the induction of secondary metabolism. This research represents the first step towards understanding *Eucalyptus* resistance against this destructive plantation pest.

Assessing biodiversity and nutrient status 10 years after reforesting a destroyed rain forest with *Tectona grandis* in Ile-Ife, Southwestern Nigeria

A.I. Odiwe, A.A. Alimi, O. Ogunsanwo and R.O. Adewumi (Wed A2)

Department of Botany, Obafemi Awolowo University, Ile-Ife, BC220005

Plantation ecosystems are becoming dominant constituents of many tropical ecosystems. Yet we do not have enough understanding of the values of these habitats for understorey species diversity and nutrient status. Changes in soil properties, forest floor litter accumulation, litter nutrients concentration, and understorey species diversity were evaluated in four, 25 m x 25 m plots in *Tectona grandis* (re-forested area) plantation and a nearby undisturbed secondary rainforest (non-forested) in Ile-Ife, Southwestern, Nigeria. Five soil samples were randomly collected both in the plantation and the secondary forest at the depth of 0-20 cm. The amounts of forest floor litter accumulated and nutrients concentration were determined. Five line transect were systematically laid and a quadrat of 50 cm x 50 cm placed at every 1 m point to identify the understorey species (herbaceous, shrubs, tree saplings and climbers) present in each area. Species diversity using Shannon-Wiener, Simpson's index and Evenness were determined, while species similarity was determined using the Jaccard's similarity index. Results showed that the following soil properties, bulk density, pH, Ca²⁺, Mg²⁺ and Available P were significantly higher (P < 0.05) in the re-forested area than the secondary forest. Reforested area had more forest floor litter and nutrient concentrations (compared with the secondary forest). The dominant understorey species in the *Tectona grandis* plantation are *Asystacia gangentica*, *Combretum spp* and *Cyathula spp* and *Setaria barbata* while *Chromolaena odorata*, *Asystacia gangentica* and *Setaria barbata* are in the secondary. The implication of this result is that re-forestation with *Tectona grandis* can help to address some of the negative effects of forest destruction. However, these replacements do not provide a substitution for original primary forest destroyed.

Anti-inflammatory, genotoxic and phytochemical properties of selected South African medicinal plants used in treating stomach-related ailments

A. Okem, J.F. Finnie and J. van Staden (Tue C3)

Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa

Ethyl acetate (EtOAc), ethanol (EtOH) 70% and water extracts of South African medicinal plants used in treating stomach-related ailments were evaluated for their cyclooxygenase-inhibitory activity against COX-1 and -2 isoenzymes. The standard plate incorporation assay for Ames test was used to evaluate the genotoxic potential of the plant extracts. A spectrophotometric method was used to determine the total phenolics,

gallotannins, flavonoids and saponins in the evaluated plants. All the EtOAc extracts exhibited percentage inhibition in the range of 50.7 to 94.7% against COX-1 and -2 isoenzymes at 250 µg/mL. All the evaluated plant extracts were non-mutagenic towards *Salmonella typhimurium* tester strains TA98, TA100 and TA1537 without metabolic activation. Phytochemical analysis revealed relatively high amounts of total phenolics, gallotannins and flavonoids in the evaluated plant extracts. Total and steroidal saponins were detected in two plant samples, *Canthium spinosum* and *Cassinopsis ilicifolia* (bark). The general pharmacological activities exhibited by some of the plant extracts in this study warrant their traditional uses in treating stomach-related ailments. Further studies will be aimed at isolation and identification of the bioactive compounds in the evaluated plant species.

Antioxidant activity, total polyphenolic content and *in vitro* inhibition of activity of digestive enzymes significant to diabetes by leaf extracts of ten South African *Ficus* species

O.O. Olaokun^a, L.J. McGaw^a, J.N. Eloff^a and V. Naidoo^{a, b} (Tue C3)

^a*Phytomedicine Programme, Department of Paraclinical Sciences, University of Pretoria, Private Bag X04, Onderstepoort, 0110, South Africa*

^b*Biomedical Research Centre, Faculty of Veterinary Sciences, University of Pretoria, Onderstepoort 0110, South Africa.*

Hyperglycaemia is the hallmark of diabetes mellitus. Postprandial blood glucose levels may be high with normal fasting blood glucose in early diabetes. Continuous elevated blood glucose deteriorates β -cell function by glucose toxicity, which provokes oxidative stress. One way to manage hyperglycaemia is to inhibit the activity of α -amylase and α -glucosidase, the enzymes which breakdown carbohydrate. We investigated the potential of crude acetone extracts of ten South African *Ficus* species to inhibit the digestive enzyme activities, and the possible relationship between these activities, total polyphenolic content and antioxidant activity. A literature survey revealed that about 10 species are presently reported to have anti-diabetes activity through their glucoselowering activity in alloxan or streptozotocin induced diabetic laboratory animals. In no case were the mechanism(s) involved in the glucose lowering effects elucidated. All the *Ficus* species we studied had varying polyphenolic contents and antioxidant activity. The crude acetone extract of *F. lutea* had the highest phenolic content (56.85 ± 1.82 mg GAE/g of dry material) and the strongest antioxidant activity with a TEAC value of 4.80 ± 0.90 . Generally, no relationship was observed between the total polyphenolic content and the antioxidant activity. The *Ficus* species weakly inhibited α -glucosidase activity with *F. sycomorus* having the best EC₅₀ (217 ± 69 µg/ml) followed by *F. lutea* (289 ± 111 µg/ml), but strongly inhibited α -amylase activity with *F. lutea* having the best EC₅₀ (9 ± 2 µg/ml) followed by *F. craterostoma* (11 ± 5 µg/ml). The results of this study revealed that the crude acetone extract of *F. lutea* is high in total polyphenolic content, antioxidant activity and is a potent inhibitor of α -amylase activity. The polyphenolic compounds present in *F. lutea* may likely be responsible for the inhibitory activity against the carbohydrate hydrolyzing enzymes. Further work is continuing on this species.

Potential of synthesized naphthoquinones for anti-tuberculosis activity

C.B. Oosthuizen^a, C. Hamilton^b and N. Lall^a (Mon A1)

^a*Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002*

^b*School of Pharmacy, Medicinal Chemistry, University of East Anglia, Norwich, NR4 7TJ, United Kingdom*

Mycobacterium tuberculosis is one of the world's biggest killers, with an incident rate of 9.4 million with a death of 1.7 million in 2009. Tuberculosis (TB) is also a major problem in South Africa due to the high incidence of HIV/Aids patients. Even though relatively adequate treatments are in place, the emergence of multiple drug resistant (MDR) TB has become a major threat, and thus calls for an urgent search for new and effective treatments. Medicinal plants have been used traditionally to treat the symptoms of TB. Naphthoquinones have been isolated from plant extracts and have shown activity against *Mycobacterium* previously. Nineteen chemically synthesized naphthoquinone derivatives were used to screen their activity against *M. smegmatis* and *M. tuberculosis*, where five of these compounds showed high activity with Minimum Inhibitory Concentrations (MIC's) ranging between 7.8ug/ml to 200ug/ml. These five compounds were tested for possible enzymatic mechanism of action utilising glutathione disulfide reductase (Gtr - human analog) and mycothiol disulfide reductase (Mtr – *Mycobacterium* analog) assays. None of the samples showed inhibition of Mtr or Gtr but all five samples showed some subversive substrate activity, with Km-values ranging between 0.3mg/ml to 2mg/ml. The cytotoxicity of the compounds was evaluated on Vero cells as well as on differentiated U937 macrophages.

The South African Biodiversity Information Facility: data mobilisation in South Africa

F. Parker-Allie (Tue B4)

Biodiversity Information Management Directorate, Kirstenbosch Research Centre, SANBI, Cape Town

South Africa is one of the world's most mega diverse countries, with a rich biological heritage but, direct and indirect drivers of change, including habitat destruction and degradation, invasive alien species and climate change, threaten the existence of many species in the country. These changes and threats highlight the urgent need to record, monitor and report on biodiversity. The South African Biodiversity Information Facility was initiated to address the country's need for an enabling platform for researchers, policy makers, conservation scientists and practitioners to access, share and disseminate primary biodiversity data while at the same time having access to global information. Currently SABIF serves over 14 million biodiversity data records (including images), using the Darwin Core Standard, with a network of more than 15 organisations which contributes to the programme. Data sharing takes place through both a funded and non-funded mechanism, thereby ensuring that resources are available to initiate digitization activities. A comprehensive policy framework has also been put in place by SANBI, to enable data sharing which takes into account intellectual property rights, citations and sensitive data. The next phase of SABIF looks at strengthening the biodiversity information “strategic partnership projects” with organizations in the country to ensure that data is relevant for both science and policy uptake.

Evidence for a re-circumscription of the Thuidiaceae (Bryophyta) in Africa and the East African Islands

N. Phephu^{a,b}, A. Magee^{c,d}, J. van Rooy^b and A.E. van Wyk^a (Tue B4)

^a*Department of Plant Science, University of Pretoria, Pretoria, 0002, South Africa*

^b*South African National Biodiversity Institute, National Herbarium, Private Bag X101, Pretoria, 0001, South Africa*

^c*South African National Biodiversity Institute, Compton Herbarium, Private Bag X7, Claremont, 7735, Cape Town, South Africa*

^d*Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park, 2006, Johannesburg, South Africa*

The moss family Thuidiaceae consists of about 19 genera and more than 150 species worldwide. The group is characterized by attractive, regularly 2- or 3-pinnate branched stems; uniseriate paraphyllia on stem and branch surfaces; dimorphic stem and branch leaves, a single costa and ornate cells; perfect peristome; asymmetrical capsule; occasionally papillose seta and ciliate inner perchaetial leaf margins. Members of the family are morphologically very similar and exhibit tremendous infraspecific variability. As a result the delimitation of genera and species has been problematic. In an attempt to resolve uncertainties in classification of the group and re-circumscribe the African Thuidiaceae, morphological and anatomical characters of selected Thuidiaceae were studied and reconstructed onto the phylogenetic trees using parsimony. The relationship of the African species of *Haplocladium*, *Hylocomiopsis*, *Abietinella* and *Raiiella* with other members of the Thuidiaceae is investigated. The exclusion of these genera from Thuidiaceae has been suggested by previous studies and a relationship rather with Leskeaceae seems likely. Based on morphological and anatomical data we support their exclusion and recognize only three genera and 16 species of Thuidiaceae in Africa. Plant size, stem branching pattern, paraphyllia morphology and ornamentation of leaf cells is shown to be taxonomically useful, particularly in combination at the generic level.

How healthy are our honeybees – Health check of a major pollinator

C.W.W. Pirk and H. Human (Tue B1)

Social Insect research group, Department of Zoology & Entomology, University of Pretoria, Pretoria, South Africa 0002

South Africa seems to escape the worldwide observed colony losses. Despite that all the major pests and diseases have been reported for South Africa, the honeybee population seems to be unaffected. Moreover, the outbreak in 2009 of American Foulbrood (AFB) does not have the expected negative effect on the South African population. The vital natural population with a high genetic diversity, thus buffering the negative effects of pests and diseases on the population, could play a role or alternatively, there is a lack of data. We have started a survey in order to fill this potential gap of knowledge and also to evaluate the impact of common honeybee diseases in the country. As expected all the major diseases are present, but it also shows a fundamental lack of identifying skills by the beekeepers. However, local pests, e.g. *Capensis*, are catching the attention of the industry. This suggests that the other pests and diseases are below the economical threshold, resulting in beekeepers ignoring them. If this conclusion holds, it also suggests that the natural population of African honeybees has traits and features successfully dealing with the diseases compared to its European

counterparts. Therefore the African population is the ideal model for investigating the underlying mechanisms since and the overall population appears not to be diseased despite the presence of diseases.

The carbon content of *Portulacaria afra* (L.) Jacq

J.R. Pool^a, B.S. Ripley^a and M. Powell^b (Wed A3)

^a *Department of Botany, Rhodes University, P.O. Box 94, Grahamstown 6140, South Africa*

^b *Department of Environmental Science, Rhodes University, P.O. Box 94, Grahamstown 6140, South Africa*

The demand for carbon credits, to offset carbon emissions, has resulted in the evolution of the carbon market where rights to carbon are bought as a method of offsetting carbon emissions. The Albany Thicket biome of the Eastern Cape (South Africa) has been degraded by anthropogenic activities and its restoration has the potential to restore biodiversity, promote ecosystem services, provide sustained employment, and generate carbon credits which will have the potential to create a strong and lucrative South African based carbon economy. *Portulacaria afra*, commonly known as Spekboom, is a succulent facultative Crassulacean acid metabolism (CAM) plant from the family *Portulacaceae*. It is an endemic to the semi-arid Subtropical Thicket. The exact carbon content of *P. afra* is yet to be scientifically verified. Because of its widespread adoption by restoration specialists and people with an interest in carbon credits and the South African carbon economy it is important to identify the exact carbon content of *P. afra* as a percentage of total biomass. Currently carbon stock calculations are based on the estimate that 48 % of *P. afra* dry biomass is elemental carbon. Secondly, it is important to understand if and how carbon content varies with plant size. Allometric regression analyses found plant volume (cm³) to be the best predictor of total plant above ground biomass. Exact carbon content (as a percentage of total dry biomass) results have far reaching implications. The allometric regressions reported in this study could aid in future biomass and carbon studies in subtropical Thickets and may have value in rangeland condition assessments. Priority areas, in need of restoration, which will provide the most economic benefit (due to increased plant biomass accumulation) should be identified and restored preferentially.

Long-term changes in the vegetation of Cape Point in response to climate and fire

R.F. Powell^a, M.T. Hoffman^a, L. Gillson^a and M. McGeoch^b (Wed A3)

^a *Plant Conservation Unit, Botany Department, University of Cape Town, Private Bag x 3, Rondebosch, Cape Town, South Africa, 7701*

^b *Cape Research Centre, South African National Parks, Steenberg, South Africa, 7945*

Climate change is a known threat to biodiversity worldwide and is predicted to have a major impact in the winter rainfall zone (WRZ) of South Africa. Projections show that temperatures are expected to increase and rainfall to decrease in the South Western Cape. The Cape of Good Hope Section of Table Mountain National Park (CGH) lies within this crucial area. This study investigates the drivers of change (climate, fire and land use) and the subsequent response of key vegetative growth forms to these changes in the conservation area. To understand past changes in CGH, climate data was analysed over a 100 year period

and fire frequency was investigated using Geographic Information Systems (GIS). Aerial photographs were compared and analysed to get a further understanding of changes in land cover over a 63-year period. The response of key growth forms to these drivers was studied using repeat photography, with stratified points set out across the reserve. It was found that climate has not vary significantly over the time period, although mean annual maximum temperatures have increased (0.175°C/decade). The frequency of fire in the reserve has also not changed significantly over the last 50 years. Comparison of the repeat photographs from 1966 showed that vegetation response varied across the reserve. In many cases there was very little difference in cover and growth form composition while in others significant changes in large proteoid shrubs (especially *Protea lepidocarpodendron*) was evident. Time since the last fire, alien plant clearing and local disturbance effects (e.g. mole rat (*Bathyergus suillus*) activity) appear to have an important impact on vegetation cover and composition. Although not evident over the recording period, climate change impacts on fire regimes and vegetation composition might be more evident over a longer time scale.

Allelopathic effects of *Amaranthus*

G. Prinsloo^a and C.P. Du Plooy^b (Wed B3)

^a*Department of Agriculture and Animal health, University of South Africa (UNISA), Florida Campus, Private bag x 6, Florida, 1710*

^b*Agricultural Research Council, Roodeplaat Vegetable and Ornamental Plant Institute (VOPI), Private bag x 293, Pretoria, 0001, South Africa*

Amaranthus is highly valued by different communities in South Africa and it is preferred for its flavour and palatability by rural communities. It plays an important role in nutrition among the leafy vegetables grown worldwide. As these crops were growing as weeds, allelopathy has been known to exist for centuries as way of survival mechanisms by plants. In a crop rotation or intercropping production system, *Amaranthus* may pose serious problems for the follow-up crop. The project consisted of three different trials to confirm and evaluate the extent of the allelopathic effects of *Amaranthus* on other crops. Each trial investigated a different aspect of the mechanisms of the possible allelopathic effect. A field trial, pot trial and seed germination trial were conducted to determine the allelopathic effects exhibited by *Amaranthus*. The field trial aimed at determining the effect of *Amaranthus* plants on four different vegetable crops namely spinach, tomato, cabbage and green peppers. The pot trial aimed at determining the allelopathic effects of three different *Amaranthus* species on tomato seedlings and the seed germination trial investigated the effects of a crude extract of *Amaranthus* and the effect of extracts from different plant organs on the seed germination of tomato, cabbage and two weeds namely *Conyza bonariensis* and *Campuloclinium macrocephalum*. All the extracts inhibited the seed germination of *Conyza bonariensis*, tomatoes and cabbage seeds. A high Electrical conductivity (EC) value of the extract was an indication of a high concentration of ions which influenced seed germination and probably the inhibition of growth that were observed in the pot trials. The presence of a high concentration of oxalate in *Amaranthus* contributes to the effects observed as it is generally responsible for root inhibition of seed germination and protection of plants when accumulated in plant organs.

An analysis of woody cover change in the mesic eastern region of South Africa using repeat photography

J. Puttick^a, M.T. Hoffman^a and T. O'Connor^b (Mon A3)

^a*Plant Conservation Unit, University of Cape Town, Rondebosch, 7701*

^b*SAEON Grasslands-Wetlands-Forest Node, Ezemvelo KZN Wildlife, PO Box 13053, Pietermaritzburg, 3202*

Increases in woody cover in grasslands and savannas is a phenomenon observed worldwide over the last century and has social, economic, and conservation impacts due to associated losses in herbaceous productivity and biodiversity. Woody thickening is well documented in South Africa although most studies have been focused at local scales. We examined woody cover change over the last century for the mesic eastern region of South Africa using repeat photography. 200 Repeat photographs were spread over an altitudinal gradient from the coast to the Drakensberg, and a latitudinal gradient from the Tugela Valley in the north to the Border region in the south. Woody cover change was quantified within landform units defined for each repeat photograph pair based on slope, aspect and catenal position. Woody cover increased within 64% of the landform units (n=488), while 30% remained stable and 6% experienced a decrease in woody cover. Woody cover increases were greatest within the savanna, Albany thicket and Indian Ocean coastal belt biomes but also occurred within the grassland biome, particularly at the lower altitudes of the grassland range. Quantile regression of woody cover change versus altitude revealed a ceiling on woody cover increase, which declined with increasing altitude. Although a maximum ceiling on woody cover increase was apparent, observed woody cover change was highly variable ($r^2 = 0.07$) along the altitudinal gradient. We propose that the ceiling on woody cover increase is determined by global drivers such as climate and CO₂ concentration while local land use drivers and abiotic factors such as soils and landform type contribute to the observed variability between sites.

Anti-microbial and anti-inflammatory activities of *Pleurostylia capensis* Turcz. (Loes) [Celastraceae]

M. Razwinani^a, S.C.K.M. Motaung^a, T.E. Tshikalange^b, H.S. Abdillahi^c and J. van Staden^c (Mon A2)

^a*Department of Biomedical Science, Tshwane University of Technology, Private Bag X680, Pretoria, South Africa, 0001*

^b*Department of Plant Science, University of Pretoria, Pretoria, South Africa, 0002*

^c*Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal, Private Bag X01, Scottsville 3209, South Africa*

Pleurostylia capensis Turcz. (Loes.) is large tree that is used in traditional African medicine as a treatment of various diseases including epilepsy, mental illness and in combination with other plants for treatment of several ailments. It is also used to encourage sleep and bring good dreams. Extracts of three parts (roots, leaves and bark) of *Pleurostylia capensis* (Celestraceae) were investigated scientifically for antimicrobial and anti-inflammatory activities. Water, ethanol and dichloromethane (DCM) plant extract were prepared and screened for antimicrobial activity using the micro dilution method against nine microorganism namely *Staphylococcus aureus*,

Escherichia coli, *Klebsiella pneumonia*, *Klebsiella oxytoca*, *Streptococcus pyogenes*, *Bacillus cereus*, *Pseudomonas aeruginosa*, *Salmonella typhimurium*, *Mycobacterium smegmaris* and *Candida albicans*. Minimum inhibition concentration (MIC) and minimum bacteria concentration (MBC) of water, ethanol and DCM extracts were determined against these microorganisms. The anti-inflammatory activity of water, ethanol and dichloromethane extracts were evaluated against both cyclooxygenase enzymes (COX-1 and COX-2). Out of three part plant extract, the roots and bark exhibited the MIC values ranging from 25.0 mg/ml to 1.563 mg/ml. The leaf extracts were not active especially against Gram-negative bacteria. This may be due to more chlorophyll being present. Gram-negative bacteria were found to be more resistant than Gram-positive bacteria. For anti-inflammatory activity the ethanol and DCM extracts were the most potent. Ethanol (80%) barks and root extracts, gave the highest inhibitory activity against both COX-1 and COX-2. Inhibitory activity for COX-1 is 98.0% and 98.1% respectively. For COX-2, percentage inhibitions were 78.17% and 83.07% respectively. Water extracts, which is the most used solvent by traditional healers gave more than 50% inhibition against both COX-1 and COX-2.

Photosynthetic and anatomical acclimation by the C₃ and C₄ subspecies of *Alloteropsis semialata* in low CO₂ atmospheres

B..S. Ripley^a, R. Strauss^a and C.P. Osborne^b (Wed A3)

^a*Botany Department, Rhodes University, PO Box 94, Grahamstown 6140, South Africa*

^b*Department of Animal and Plant Sciences, University of Sheffield, Sheffield S10 2TN, UK*

The past 1-25 Myr have been characterised by sub-ambient partial pressures of atmospheric CO₂ (C_a) which impose a greater limitation on C₃ than C₄ photosynthesis. In response to low CO₂ it was hypothesized that C₃ plants alter leaf anatomy and photosynthetic responses more than C₄ plants in order to offset reductions in productivity. Here, we report the first phylogenetically controlled comparison of the effects of sub-ambient C_a on leaf anatomy and photosynthetic responses of the C₃ and C₄ subspecies of *Alloteropsis semialata*. Glacial CO₂ concentrations resulted in marked alterations to C₃ leaf anatomy that were associated with increased mesophyll conductance and even when combined with increased biochemical acclimation, was insufficient to maintain photosynthetic rates. In contrast, the C₄ subspecies maintained photosynthetic rates and displayed less altered leaf anatomy. Our findings highlight the importance of biochemical and anatomical acclimation to atmospheric CO₂ and add to the understanding of how historically low CO₂ gave C₄ plants a photosynthetic advantage.

A molecular phylogeny of *Encephalartos* Lehm.

P. Rousseau^{a,b}, P. Vorster^c, D.P. Little^d and M. van der Bank^{a,b} (Mon B1)

^a*African Center for DNA Barcoding, University of Johannesburg, Department of Botany and Biotechnology, PO Box 524, Auckland Park 2006, South Africa*

^b*Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa.*

^c*Department Botany and Zoology, University of Stellenbosch, Private Bag XI, 7502 Matieland, South Africa*

^d*Lewis B. and Dorothy Cullman Program for Molecular Systematics, The New York Botanical Garden, Bronx, New York 10458-5126, U.S.A.*

Encephalartos is an African endemic cycad genus of 65 species and 2 subspecies and is the most endangered group of cycads with 80% considered threatened (CR, EN, or VU) by the IUCN and all classed as CITES appendix 1. Currently relationships within the genus, including species delimitation, are uncertain and in need of taxonomic investigation. Most studies to date have used a combination of more traditional lines of evidence such as morphology, anatomy and geography. Also no all-inclusive phylogenetic framework currently exists for *Encephalartos*. In the current study, DNA sequence from three plastid regions (*rbcLa*, *psbA-trnH* and *matK*) along with the nuclear region ITS were used to produce a phylogeny using multiple accessions per species where possible. Results show an increase in resolution at both the species and higher level and the delimitation of several new groupings. Each species grouping is characterised by shared derived morphological, ecological, and geographic characters. This study provides the first steps towards a much-needed monograph of the entire genus.

DNA barcoding of Africa's endemic cycads: *Encephalartos* Lehm. and *Stangeria* T. Moore

P. Rousseau^{a,b}, P. Vorster^c, D. P. Little^d and M. van der Bank^{a,b} (Mon B1)

^a*African Center for DNA Barcoding, University of Johannesburg, Department of Botany and Biotechnology, PO Box 524, Auckland Park 2006, South Africa*

^b*Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa.*

^c*Department Botany and Zoology, University of Stellenbosch, Private Bag XI, 7502 Matieland, South Africa*

^d*Lewis B. and Dorothy Cullman Program for Molecular Systematics, The New York Botanical Garden, Bronx, New York 10458-5126, U.S.A.*

Africa's cycads (66 species and 2 subspecies in two endemic genera: *Encephalartos* and *Stangeria*) are extremely endangered with four species Extinct in the wild and 80% threatened (CR, EN, or VU) with all included in CITES appendix 1. Although South Africa has some of the world's strictest cycad legislation, cycads are still under threat from illegal collection for horticulture and medicine especially where plants seized in an unidentifiable condition. Currently developed legislation demands accurate identification for permit issuing. Ex situ conservation of genetic and locality based diversity is paramount. Furthermore, taxonomically many species of unknown origin are difficult to identify especially when diagnostic characters are absent. Species delimitation and numbers are

uncertain with field observations often contradicting current understanding. DNA barcoding can assist in all the above-mentioned scenarios and as such all proposed DNA barcoding regions (*matK* + *rbcLa* + *psbA-trnH*, nrITS) along with several additional regions were tested for all species encompassing ~350 samples. Results will focus on amplification success and discriminatory power of the different markers.

The rust pathogen *Puccinia psidii*, an eminent threat to South Africa

J. Roux and M.J. Wingfield (Wed B3)

Department of Microbiology & Plant Pathology, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa

Fungal pathogens represent one of the greatest threats to the survival of plants on earth, especially trees that live for long periods and adapt slowly. Several examples exist of the devastating impact that fungi have had on human existence via the destruction and contamination of food crops. The fact that trees suffer from diseases has only been recognised for about 130 years but there are many examples of fungal diseases changing entire ecosystems. The rust fungus *Puccinia psidii* was first described in 1884 and has been described as the “biggest threat to the ecosystem” in Australia. It has a host range of more than 100 known hosts in the sub-family Myrtoideae. Until the 1970s *P. psidii* was known only from South and Central America and the Caribbean Islands, but it has subsequently spread to the southern USA, Hawaii and Japan. Early in 2010 *P. psidii* was detected for the first time in Australia. In the 18 months since its first detection, it has spread throughout NSW and into Queensland, affecting various native Australian plants in the Myrtoideae. Current predictions, based on its impact on native *Melaleuca* species, are that it will eliminate several of these iconic tree species. The appearance of *P. psidii* in Australia should be of great concern to South Africa. Tests on South African *Heteropyxis natalensis* showed that this tree species is highly susceptible to this pathogen. South Africa has several tree genera in the Myrtaceae and the local forestry industry relies on susceptible Eucalypt species. We are, therefore, at risk of significant ecological and economic losses when this pathogen enters the country, as it undoubtedly will. This is particularly due to the ready exchange of people and products between these two countries. Significant effort should be placed on quarantine and breeding programmes to prepare for the appearance of *P. psidii* in South Africa and this should be done sooner rather than later.

The ethnobotany, essential oil composition and antibacterial activity of southern African *Teucrium* species (Lamiaceae)

A.K. Ruiters^a, B.-E. van Wyk^a, S.F. van Vuuren^b and P.M. Tilney^a (Wed B2)

^a*Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa*

^b*Department of Pharmacy and Pharmacology, Faculty of Health Sciences, University of the Witwatersrand, 7 York Road, Parktown 2193, Johannesburg, South Africa*

Three southern African endemic species of the genus *Teucrium*, *T. africanum*, *T. kraussii* and *T. trifidum* of the family Lamiaceae are poorly studied with regard to their anatomy, essential oil composition and the scientific validation of their traditional medicinal uses as well as their taxonomic position in the genus. All three are used in traditional

medicine to treat digestive and respiratory ailments. A summary of the published ethnobotanical information is presented. The essential oils were extracted from the three species, and water, methanol and dichloromethane: methanol extracts of the different plant parts made. These were used for the antimicrobial studies to determine the minimum inhibitory concentrations (MIC) against the bacteria, *Staphylococcus aureus*, *Escherichia coli*, *Bacillus cereus*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Moraxella catarrhalis* and *Streptococcus pyogenes*. The composition of the oils was also investigated with gas chromatography- mass spectroscopy (GC-MS). The essential oils of *T. africanum* and *T. trifidum* are very similar with α -Cubebene, β -Cubebene and β -Caryophyllene as main compounds. Antibacterial activity was recorded for *T. africanum* against *E. coli*, with an MIC value of 0.125 mg/ml (dichloromethane: methanol extract) and against *S. pyogenes*, with an MIC value of 0.16 mg/ml (essential oil). This data seems to at least partially validate the traditional uses of *Teucrium* species to treat digestive and respiratory ailments.

Isolation and characterization of compounds from *Calodendrum capense* and *Lydenburgia cassinoides* with antimicrobial potential against opportunistic pathogens

B.M. Sakong, A.S. Ahmed, L.J. McGaw and J.N. Eloff (Wed B2)

Phytomedicine Programme, Department of Paraclinical Sciences, University of Pretoria, Private Bag X04, Onderstepoort 0110, Pretoria, South Africa

Infectious diseases are a serious concern worldwide especially in immune-compromised patients. The problem is compounded by the emergence of microbial resistant pathogens. A wide range of microbes including bacteria, fungi, parasites, viruses and protozoas are implicated as causative agents of various diseases. Two plant species (*Calodendrum capense* and *Lydenburgia cassinoides*) used in South African traditional medicine for treating infectious diseases were screened for antimicrobial activities against *Candida albicans*, *Cryptococcus neoformans*, *Aspergillus fumigatus*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Enterococcus faecalis*. *L. cassinoides* acetone extract generally had good activity, with MIC values ranging from 0.04 to 0.15 mg/ml while *C. capense* MIC values ranged from 0.31 to 0.62 mg/ml against the fungi. The two plant extracts had good antibacterial activity against *S. aureus* and *P. aeruginosa*, with MIC values ranging from 0.16 to 0.32 mg/ml. Antibacterial activity against *Escherichia coli* and *Enterococcus faecalis* resulted in MIC = 0.63 mg/ml for both plants. Bioassay-guided evaluation of the antimicrobial active components led to the isolation and characterization of lupeol from *C. capense* and β -amyryn from *L. cassinoides*. The MIC of lupeol and β -amyryn ranged from 1.5 to 6.2 μ g/ml against all the tested organisms. Crude extracts and compounds were also tested for cytotoxicity against Vero (monkey kidney) cells. Both plant extracts had low toxicity with average IC₅₀ values of 205.8 \pm 8.38 μ g/ml, for *L. cassinoides* crude extracts had 83.07 \pm 44.66 μ g/ml for *C. capense* crude extracts while IC₅₀ values for the compounds were greater than 200 μ g/ml, the highest concentration tested. This gave an excellent selectivity index of >30 indicating the safety of the compounds. In conclusion both plant species showed broad antimicrobial activity against the standard strains of bacterial and laboratory isolates of fungal pathogens. The results validate the use of two plants as anti-infectious agents in traditional medicine.

Immunomodulatory effect of *Combretum mole* leaves water extracts on the production and expression of cytokines and toll like receptors

A. Samie, T. Nefefe, T. Mulaudzi and P.O. Bessong (Mon A1)

University of Venda, Department of Microbiology, Private Bag X5050, Thohoyandou 0950

Medicinal plants are known to have positive as well as negative effects on the immune system before and during infection. In the present study, peripheral blood mononuclear cells (PBMCs) were isolated from volunteers and maintained in culture in the presence of a stimulating agent (Phytohaemagglutinin: [PHA]) and the plant extracts. PBMCs isolated from HIV negative and HIV positive patients were tested. The effect of the plant extracts on the immune cells was assayed by measuring the production of interferon gamma in the cells supernatant in the first protocol and the measurement of mRNA production of several cytokines and Toll like receptors (TLRs) using a real time PCR procedure. Measurements of the effect of the extracts were conducted with and without activation by phytohemagglutinin. The measurement of INF γ in the cell supernatant indicated a slight increase in the production of the cytokine by cells that were exposed to the plant extracts following stimulation by PHA not before. There was a dose dependent variation of the effect of the extracts on the cytokine expression. PBMCs from HIV positive patients showed greater activation and higher production of INF γ compared to HIV negative patients. The mRNA expression of the cytokines and Toll like receptors was evaluated by real time PCR and showed high expression of IL6 by the cells. NFkappa B was also highly expressed by the cells as well as TLR2. The present study showed that *Combretum molle* has immunomodulatory capacities with special effect on improving the production of interferon gamma. The activation of the cells and consequently the improved expression of certain cytokines seem to use the NF kappa B pathway. Further studies are needed to identify the chemical constituents responsible for the observed effects.

Species used by Bapedi Traditional Healers for the treatment of sexually transmitted diseases, Limpopo Province

S.S. Semanya^a, M.J. Potgieter^a and L.J.C. Erasmus^b (Wed B2)

^a*Departments of Biodiversity and* ^b*Physiology and Environmental Health, School of Molecular and Life Sciences, University of Limpopo, Private Bag X1106, Sovenga 0727, South Africa*

Annually, millions of people are exposed to and affected by a variety of curable sexually transmitted diseases (STDs). These diseases can have a detrimental effect on the health status of the individual. This study found that 47 plant species from 44 genera belonging to 31 families are used to treat STDs, such as *drop* (gonorrhoea), HIV/AIDS, *khutlega*, *nta* and *tshofela* (syphilis). Six of the 47 species are exotics, and were of significant value to traditional healers. The majority (87.5%, n=41) of the species were used in the treatment of a single STD; only 12.5% (n=6) were used in the treatment of more than one STD. Fifteen of the 47 species were used more than once for the preparation of an extract. The remaining 32 species all had a single application, thus was used to treat a single STD. A number of species used in the treatment of STDs are protected by National and Provincial legislation. These include, amongst others rare species such as *Boscia albitrunca* and *Eleaodendron transvaalense* (used to manage HIV infections), which are protected under the

National Forest Act. Thirty five (76%) of these species appear in the Red Data List. These include *Dioscorea sylvatica* (vulnerable), *Drimia elata* (insufficient data), *Eucomis pallidiflora* (near threatened), and *Hypoxis hemerocallidea* (declining). These species, as noted by traditional healers are either declining or rare, and are increasingly being cultivated in home gardens. The large number of species employed in the treatment of STDs clearly reflect the diversity of treatment protocols used by Bapedi traditional healers. In the treatment of the more prominent STDs a number of alternative species can be used, which in itself will ensure that treatment options are always available.

In vitro* germination of the threatened assegai tree, *Curtisia dentata

S. Shaik (Wed B3)

School of Biological & Conservation Sciences, University of KwaZulu-Natal, Westville Campus, Private Bag X54001, Durban 4000, South Africa

Curtisia dentata (Cornaceae) is a medicinally important indigenous tree native to southern Africa. In South Africa the stem bark of this threatened species has been overharvested to treat many medical conditions in humans and animals. As a result of its threatened and protected status this study was performed to investigate whether *in vitro* germination of *C. dentata* seeds could be utilized as a possible mechanism to conserve the species. In the wild, propagation of *C. dentata* through seed may be difficult and could take many weeks provided the climate and rainfall is favourable. Seed yields can also fluctuate on a year to year basis and current diminishing populations can exacerbate seed shortage. In addition, beetle predation of seeds is a current and real problem. If seeds do survive to seedling stage they become vulnerable to damage from drought, high surface soil temperature, frost and damping off fungi. Therefore, the alternative cultivation and conservation technique of *in vitro* germination was considered. Experiments using physical, mechanical and chemical pre-sowing treatments were conducted in light and dark conditions to determine the germination response of this species. Among the various treatments, the highest germination percentage was achieved in seeds treated with concentrated H₂SO₄ for 10 minutes and incubated in the light. The quickest germination response was obtained in mechanically scarified treatments on day 3 in both light and dark conditions. The results indicate that *C. dentata* seeds possess exogenous dormancy due to the thick tough seed coat which is the main inhibitor of germination.

Evaluation of antibacterial, antioxidant and mechanistic activities of a plant from Myrtaceae family addressing acne vulgaris

R. Sharma, A. Hussein and N. Lall (Mon A1)

Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002

Fifty plant species grown in South Africa were selected for their antibacterial activity. Ethanol extracts were prepared and tested for their action on *Propionibacterium acnes* and the Minimum Inhibitory Concentration (MIC) were evaluated. The results using the broth dilution methods showed that many crude extracts could inhibit the growth of *P. acnes*. The leaf extract of a plant from a family Myrtaceae exhibited best MIC value of ≤ 62.5 $\mu\text{g/ml}$. The

cytotoxicity effects of this extract were determined by colorimetric XTT {2,3-bis (2-methoxy-4-nitro-5-sulfophenyl)-5-[(phenylamino) carbonyl]-2H-tetrazolium hydroxide} assays on human macrophages U937 cells. The extract showed low cytotoxicity exhibiting a fifty percent inhibitory concentration (IC₅₀) of 400 µg/ml. Phytochemical test revealed the presence of phenols, tannins, anthraquinones and flavonoids in the ethanol extract. Bioassay-guided isolation of ethanolic extract led to the isolation of two compounds. The first compound (triterpene), did not show activity against *P. acnes* at the highest concentration (500 µg/ml) tested; whereas the second (6-alkyl salicylic acid) compound exhibited MIC of 0.25 µg/ml. The extract was further investigated for its inhibitory activity on glutathione disulfide reductase and mycothiol disulfide reductase enzymes at concentrations ranging from 800 µg/ml to 1.56 µg/ml. The IC₅₀ was found to be between 6.25 to 3.125 µg/ml for glutathione disulfide reductase and 36.77 µg/ml for mycothiol disulfide reductase. The antioxidant activity was detected by DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging capacity and EC₅₀ (substrate concentration to produce 50% reduction) was found to be 0.89 µg/ml. The antibacterial activity of potent samples were further confirmed by means of Transmission Electron Microscopy.

Characterisation of phenolic compounds in aqueous extracts from seed coats of marama bean (*Tylosema esculentum*) and their bioactive

J.S. Shelembe^a, D. Cromarty^b, M.J. Bester^c, A. Minnaar^a and K.G. Duodu^a (Mon A2)

^aDepartment of Food Science, ^b Department of Pharmacology, ^c Department of Anatomy
University of Pretoria, Pretoria, South Africa 0002

The marama bean is an underutilised legume growing wild in the arid and semi arid regions of Southern Africa. The seed coats of marama beans contain antioxidant phenolic compounds with potential health benefits, but are not consumed. Aqueous extracts for use as antioxidant functional food ingredient from seed coats of marama beans were prepared by extracting with water (water extract) or with water acidified to pH 2 (acidified water extract). The extracts were analysed for phenolic content using spectrophotometric methods. Phenolic acids, flavonoids and proanthocyanidins were analysed by HPLC-MS. Free radical scavenging activities of the extracts and their protective effect against free radical induced human erythrocytes hemolysis, oxidative DNA damage and human LDL oxidation were determined. Extraction of marama bean seed coats under acidic conditions resulted in reduction in phenolic compound content, antioxidant activity and lower protective effect against erythrocyte hemolysis with no significant differences in their protective effect against oxidative DNA damage and LDL oxidation. Gallic acid and methyl (epi)afzelechin-3-O-gallate were the major phenolic acid and flavonoid compounds respectively in the marama bean seed coat aqueous extracts. Proanthocyanidins were predominantly highly galloylated prodelphinidins. The findings of this study show that the extracts have a potential to reduce oxidative stress which is implicated in many chronic diseases such as neurodegenerative diseases, cancer and cardiovascular disease.

Evaluation of the mutagenicity and stability of plant extracts with potential use in the cosmetic field from the Myrsinaceae family

S. Singh and N. Lall (Mon A1)

Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002

The leaves and stems of a plant from the *Myrsinaceae* family, which is indigenous to Southern Africa, have shown substantial anti-tyrosinase activity. The 10% propylene glycol plant extract, as well as its formulation, were tested under different temperatures in order to determine their stability at Stability Testing Services. The storage period extended over 3 months. The pH, specific gravity, appearance, odour and viscosity were tested at these intervals. Analysis of the results revealed that the sample stored at 25°C was most stable as it retained most of the original activity. Approximately 90% of tyrosinase inhibition was observed when the sample was tested at 1%. The physio-chemical properties such as pH, refractive index, relative density, appearance and odour were determined. In order to determine the mutagenicity of the extract, samples of the extract were tested on strains of *Salmonella typhimurium* (*S. typhimurium*) in accordance with the Ames test. The number of revertant colonies observed for the T98 and T100 strains at the highest concentration (5mg/ml) was 18.67 ± 1.15 and 103.33 ± 2.52 respectively. The number of revertant colonies in the blank plates was observed to be 112.40 ± 3.65 . According to the Ames test, a sample must have more than twice the number of revertant colonies produced in the blank plates in order to be mutagenic. The extract was classified as non-mutagenic according to these tests. The irritancy and the skin-depigmentation properties of the extract were tested at Future Cosmetics. The irritancy potential of the extract was calculated to be - 9.00% and overall the extract was observed to be a non-irritant by Future Cosmetics cc. This plant can be beneficial in the treatment of hyper-pigmentation, a common skin disorder.

Dealing with newly detected invader: *Sagittaria platyphylla*

H.J.N. Sithole (Tue B2)

Early Detection and Rapid Response to emerging invasive alien plants programme (EDRR – funded by Working for Water) South African National Biodiversity Institute, P.O Box 52099, Berea Road, 4007, Durban, South Africa

The Early Detection and Rapid Response to emerging invasive alien plants programme (EDRR) aim to reduce the incidence of plant invasions through identifying them at an early stage of invasion, plan rapid responses, explore and recommend effective control methods. *Sagittaria platyphylla* is one of the newly detected invaders from Tropical America. It has severe ecological impacts and poses a great threat to wetlands in South Africa. The coastal areas are under a high threat because the climatic conditions resemble that of the country of origin. *Sagittaria platyphylla* is not yet declared as an invader in South African legislative context therefore there are no recommended methods to control it, but the process to get it declared has been initiated. In the meantime EDRR has performed several activities and explored possible control methods. Achieved tasks are highlighted: species identity has been verified, awareness raising, mapping and herbicide field trials are in progress. Thus the talk

serves to encourage the like minded to support EDRR Programme with sightings and provide inputs.

High throughput expression analysis of miRNAs and their targets involved in the water deficit response in *Vitis vinifera*

M.C. Solofoharivelo^a, A. Van der Walt^{b*}, J. McBride^b, D. Stephan^a, M.J Freeborough^a, J.T. Burger^a and S. Murray^b (Tue A1)

^a*Vitis lab, department of Genetics, Stellenbosch University, Private Bag XI, Matieland 7602*

^b*Centre for Proteomic and Genomic Research, P.O. Box 81, Observatory, 7935, Cape Town*

^{*}*Current address: Central Analytical Facility, Stellenbosch University, Private Bag XI, Matieland 7602*

MicroRNAs (miRNAs) are small non-coding RNAs that regulate diverse biological processes across various organisms. In plants, miRNAs have been associated with plant development and in the response to biotic and abiotic stress. In this study, we investigated the role of miRNAs in *Vitis vinifera* (grapevine) under mild to severe water deficit. Grapevine is one of the most important fruit crops in the world. Abiotic stress caused by water deficit can severely limit plant development, growth and yield. Although grapevine has been described as moderately tolerant to drought, recent evidence has shown that gene expression is modulated by low to severe water deficit. Here, using miRNA microarray technology to analyze miRNA expression during drought treatment of grapevine plants, we identified several miRNAs that were differentially expressed. Putative targets of these miRNAs were also identified using a whole genome *Vitis* microarray. Both miRNAs and miRNA targets were validated by quantitative real-time qPCR. Several of these miRNAs have been implicated in drought responses in other plant species.

The chemical composition, antimicrobial and antioxidant properties of the essential oils of *Tulbaghia violacea* and *Eucalyptus grandis*

O.S. Soyngbe^a, A.O. Oyedeji^b, M. Singh^c and A.R. Opoku^a (Tue C4)

^a*Department of Biochemistry and Microbiology, University of Zululand, Private Bag X1001 KwaDlangezwa, 3886*

^b*Department of Chemistry, Walter Sisulu University, Private Bag XI Mthatha, 5117*

^c*Department of Biochemistry, University of Kwazulu-Natal (Westville Campus), Durban 4000*

Tulbaghia violacea (Harv.) L.f. and *Eucalyptus grandis* W.Hill ex Maiden are medicinal plants used by Zulu traditional healers to treat respiratory track diseases. Essential oils hydrodistilled from the rhizomes of *T violacea* and the leaves of *E grandis* were evaluated for the chemical composition, antioxidant and antibacterial activities. The GC and GC-MS analysis of the oils revealed the main constituents of the essential oils of *T violacea*, to be 2,4- Dithiapentane (51.04%), p- Xylene (20.59%), Chloromethylmethyl sulfide (8.69%), o- Xylene (7.38%), Thiodiglycol (6.43%), and p- Xylol (5.88%). The main constituents of the essential oils of *E grandis* are m- Xylene (49.25%), Ethylbenzene (16.86%), Eucalyptol (15.50%), o- Xylene (14.24%) and Limonene (5.14%). While the oils of *T violacea* showed weak antioxidant activity, the essential oils of *E grandis*, exhibited a high DPPH and ABTS scavenging activity. The antimicrobial activity of the essential oils

showed that the oils of *T violacea* were affective against 8 of the 16 microorganisms tested with MIC values ranging from 2.5µg/ml - 5.0µg/ml; the oils of *E grandis* were active against 13 of the 16 organisms tested with the MIC ranging from 0.625µg/ml – 5.0µg/ml, and the MBC values ranging from 2.5µg/ml - 10µg/ml. The essential oils of *E grandis* were also tested against 8 antibiotic resistant bacteria, and were seen to show activity against 7 of the 8 with MIC ranging from 5µg/ml - 10µg/ml. The Oils had low (1218µg/ml, and 1641 µg/ml) cytotoxicity levels against HEK293 and HepG2 cell lines respectively. It is apparent that the bioactivity of the essential oils of *T violacea* and *E grandis* contribute to the use of these plants in folk medicine.

A phytochemical and morphological comparison between *Pachystigma macrocalyx*, *P. pygmaeum* and *P. thamnus*

S.L. Stanton^a, C.F. van der Merwe^b, A. Hussein^a, S.N. Venter^c, T.A. Coutinho^c and J.J.M. Meyer^a (Wed B2)

^a*Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002*

^b*Laboratory for Microscopy and Microanalysis, University of Pretoria, Pretoria 0002*

^c*Department of Microbiology and Plant Pathology, University of Pretoria, Pretoria 0002*

The plants of the Rubiaceae family, specifically *Pachystigma macrocalyx*, *P. pygmaeum* and *P. thamnus* have been the focus of very little research. *P. pygmaeum* and *P. thamnus* have been assumed to contain the polyamine pavettamine and thus cause the disease “Gousiekte” in domestic ruminants; however extensive studies have not been done on *P. macrocalyx*. The aim of this study was to compare these plants on morphological and phytochemical aspects. The bacterial endophytes, which may cause the toxicity, were compared using electron microscopy and isolation techniques. The chemical compositions of the three plants were analysed using TLC, NMR and metabolomic procedures. In the TLC analysis the standard, pavettamine was spotted along with the crude extracts of the three plants; the TLC plates were run on both polar and non-polar mobile phases to reveal the compound composition. The compounds which reacted with ninhydrin, possible polyamines, were noted and also analysed by NMR to determine their chemical structures. Twenty samples of each species were used for the metabolomic study to determine the differences between different collection sites and the individual plants within the sites. These toxic compounds will be subjected to toxicity tests along with compounds isolated from the endophytes in future. Due to the hypothesis that extracts from *P. pygmaeum* and *P. thamnus* are responsible for the degradation of the ruminant’s myofibres, H9c2 cells which are derived from embryonic rat cardiac cells, will be used to determine toxicity.

Breeding systems in *Protea*

S-L. Steenhuisen and S.D. Johnson (Wed A1)

School of Biological and Conservation Sciences, University of KwaZulu-Natal, P/Bag X01, Scottsville, 3209, South Africa

It has been assumed that species of the large African genus *Protea* have strong self-incompatibility systems. However, this assumption was based largely on studies conducted on a clade of bird-pollinated species that occur in the shrubby fynbos vegetation of the Cape region of southern Africa. To test whether self-incompatibility occurs in a grassland/savanna *Protea* clade, which is largely insect-pollinated, we performed controlled pollination experiments on four species - *Protea caffra*, *P. simplex*, *P. dracomontana* and *P. welwitschii*. Although pollen-ovule ratios of all four species fall within the range for outcrossers, all four species are self-compatible and capable of autogamous seed production. In *P. caffra*, a small tree expected to carry substantial genetic load, selfed progeny had rates of germination and early seedling survivorship which were identical to those of crossed progeny. Selective exclusion experiments and allozyme analysis of eight polymorphic loci in progeny revealed that exclusion of vertebrate visitors to *P. caffra* did not reduce outcrossing rates, and we could infer that insects are effective agents of cross-pollination in *P. caffra*. However, high inbreeding depression ($\delta=1$) and a marked difference between maternal and progeny Wright's fixation indices suggest that selfed progeny may not reach reproductive maturity. The grassland *Protea* species studied are visited intensively by insects and are thus likely to be facultatively autogamous with mixed mating systems. If one assumes previous reports of self-incompatibility in *Protea* to be reliable, there have been at least five losses of SI and two gains of autonomous self pollination in this genus. However, earlier studies in the genus were often methodologically flawed and a thorough re-analysis of breeding systems in *Protea* is required.

Woody cover change in the north-east arid savannas of South Africa from 1940-present

N. Stevens^{a,c}, B.F.N Erasmus^b, S Archibald^c and W. J Bond^a (Wed A3)

^a*Botany Department, University of Cape Town, Private Bag, Rondebosch 7701, South Africa*

^b*School of Animal, Plant and Environmental Sciences, University of the Witwatersrand, Wits 2050, South Africa*

^c*Natural Resources and the Environment, CSIR, PO Box 395, Pretoria 0001, South Africa*

Increasing tree cover in “open” systems is occurring globally. Tree cover increases in arid savanna regions of South Africa have been noted to be extensive and widespread. Numerous theories have been proposed as to what is determining increases in these dry (<450mm) savannas. Local land-use management actions or global drivers particularly elevated CO₂ have been considered to be the primary drivers, however some authors suggest that the perceived changes are part of the inherent variability of the system. This debate remains unresolved as very few multi-site, multi land-use, large-scale evaluations of woody plant encroachment exist, with the majority of the studies documenting woody encroachment in an area less than 10km². We measured the magnitude of woody cover change in the north-east arid savannas of South Africa across a 160km² area consisting of three distinctly different land-use types; communal rangelands, commercial rangelands, conservation areas with elephants. Tree cover change was measured between 1940 and 2009 using the aerial photo

record. Detection of woody cover from each aerial photograph was automated using ECognitions' Object based image analysis (OBIA). Tree cover increased in commercial and communal areas and decreased in conservation areas with elephants, however tree cover changes were approximately 10% and therefore unlikely to be a significant alteration to the landscape. The overall composition of the canopy structure however has shifted towards smaller canopied plants. Our study indicates that overall land cover has remained constant but there may be a shift in the woody structure of these systems.

The effect of cold stress on infection of Cavendish bananas with *Fusarium oxysporum* f. sp. *cubense*

R. Sutherland^a, A. Viljoen^b, A.A. Myburg^c and N. van den Berg^c (Mon C2)

^a*Department of Microbiology and Plant Pathology, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa 0002*

^b*Department of Plant Pathology, University of Stellenbosch, Private Bag XI, Matieland, South Africa 7600*
^c*Department of Genetics, FABI, University of Pretoria, Pretoria, South Africa 0002*

The soil-borne fungus *Fusarium oxysporum* f. sp. *cubense* (*Foc*) causes Fusarium wilt of banana, one of the most destructive plant diseases known. Some strains of the fungus, designated *Foc* sub-tropical race 4 (STR4), cause disease to Cavendish bananas in cooler regions of the world, particularly after winter. The aim of this study was to determine why Cavendish banana plants become susceptible to *Foc* STR4 following predisposition by cold temperatures. Cavendish banana plantlets were subjected to three different treatments: (a) inoculation with *Foc* STR4 at 25°C, (b) incubation at of 10°C for 2 weeks followed by inoculation with *Foc* STR4 at 25°C, and (c) inoculation with *Foc* STR4 at 10°C. RNA was extracted from banana roots at 3 and 12 hours post inoculation and cDNA generated. cDNA libraries were constructed and sequenced on a 454 GS FLX pyrosequencer (15464 reads, 3.9 Mbp). An average of 417 contigs per library with an average contig length of 615 bp was obtained with 55.64% of the transcripts showed similarity to the rice genome. The putative functions of transcripts were determined by homology searches on the National Center for Biotechnology Information (NCBI) database. Genes identified include PR1, peroxidase, abscisic stress ripening protein and phenylalanine ammonia lyase. Gene expression analysis is currently underway to better understand the roles of these in defense and cold stress at different time intervals. Preliminary results suggest down-regulation of defense genes during cold stress. By reducing cold stress during the winter months, or by uncoupling cold stress from down-regulation of defense genes, *Foc* STR4 may be controlled in sub-tropical countries.

Fragrances and flavours of some South African plants

K.M. Swanepoel^a, P. Soundy^b and W.G. Alberts^c (Tue C4)

^a*Southern African Essential Oil Producers Association (SAEOPA), Post box 462, Newlands, Pretoria 0049*

^b*Department of Soil Science, University of Pretoria, Pretoria, South Africa 0002*

^c*KARWIL Consultancy, Coral str 111, Lynnwood Glen, Pretoria, 0082.*

According to the fragrance and flavour industry, there is a constant need for new flavours and aromas in the world. Latest trends show an interest in South African plants with known and unknown fragrance and flavours. Few indigenous plants have been investigated for potential in the perfumery and flavouring industry. South Africa with its variety of flora, has untapped potential in the flavour and fragrance industries. Many rural areas rich in bio diversity are also poverty stricken will need opportunities for development e.g. natural products in the fragrance and flavour industries. Some of the aromatic plants were identified with potential of commercial production for the fragrance and flavour industry. The indigenous plants that have been investigated for potential further production include: *Mondia whitei*, *Athrixia phylicoides*, *Warburgia salutaris*, *Siphonochilus aethiopicus*, *Osyris lanceolata*, *Cymbopogon validus*, *Spirostachys africana*, *Tarchonanthus camphoratus*, *Helichrysum oderatissimum*, *Acacia nilotica*, *Croton gratissimus*, *Heteropyxis natalensis*, *Lippia javanica* and *Dovyalis caffra*. *Pelargonium spp.*, *Agathosma spp.* (Buchu) and *Sclerocarya birrea* (Marula) and are already well known for valuable and distinctive properties in the flavour and fragrant applications. There are many more species in South Africa that should be researched for the unique South African flavour and fragrance properties. Sensory evaluations were done by the flavour and fragrance industry and chemical analysis need to be completed, before the agricultural potential could be investigated and explored for selection as future crops.

DNA fingerprinting of *Acacia karroo*: Attempting to resolve taxonomic uncertainties and inform ecological observations

C.L. Taylor and N.P. Barker (Mon B1)

Department of Botany, Rhodes University, PO Box 94, Grahamstown, 6140, South Africa

Acacia karroo Hayne is a very common woody species in South Africa. The species displays a large amount of variation, especially in terms of its morphology with enormous variation in the architecture of fully grown trees. Genetic variability of the species throughout South Africa was examined to test whether it is correlated to the different morphotypes or ecotypes. The species is also currently encroaching in some areas of its distribution and it is hypothesised that one ecotype/genotype is responsible for this encroachment. These findings could inform management practices in the encroached areas. The study uses Inter-Simple Sequence Repeat (ISSR) fingerprinting method to investigate the genetic diversity in this species. Additionally DNA extracts from fresh and silica dried samples of the same plant were tested to determine whether the treatment of silica drying compromises DNA extract and thus the quality of the Inter-Simple Sequence Repeat Polymerase Chain Reaction. The ISSR analysis found no specific genotypes within *Acacia karroo*. No difference was found in the quality of DNA extracted from fresh compared to dried specimens.

***Acacia saligna* invasions: genetics and species distribution modelling**

G.D. Thompson^a, D.U. Bellstedt^b, J.J. Le Roux^a, D.M. Richardson^a and J.R. Wilson^{a,c}
(Tue A4)

^a*Centre for Invasion Biology, Department of Botany and Zoology, Stellenbosch University, Matieland, 7602, South Africa*

^b*Department of Biochemistry, Stellenbosch University, Matieland, 7602, South Africa.*

^c*South African National Biodiversity Institute, Kirstenbosch National Botanical Gardens, Claremont, 7735, South Africa*

Acacia saligna, a species complex native to Western Australia, is highly invasive in 20 countries on four continents. Using genetic analyses, we determined which lineages of the species complex (putative subspecies) are represented in the invasive range in South Africa. In addition, we used species distribution models to explore whether high genetic structure influenced native and introduced distribution patterns. From population genetic (microsatellites) and phylogenetic (nDNA and cpDNA) analyses, we found deep phylogenetic divergences between the native and introduced South African populations, such that we were unable to associate South African entities with any native subspecies of *A. saligna*. We then assessed distribution patterns by constructing correlative species distribution models for the different subspecies of *A. saligna*. These models accurately distinguished between the native subspecies niches, but were unable to accurately predict introduced distributions in South Africa and the Mediterranean Basin. Both the ecological and genetic findings suggest that a novel genetic entity of *A. saligna* is present in South Africa. We speculate that this novel entity may have arisen due to cultivation practices in Western Australia, and that this was the entity that was introduced to South Africa. As a consequence, the invasive populations of *A. saligna* may have no historical analogue in the native range (genetically or in terms of a realised climatic niche). This highlights the need to consider that native and invasive plant populations are distinct entities (genetically or in terms of realised climatic niche) which is in contrast to current assumptions that such populations are one and the same.

Do leaves play a more active role in the symbiotic relationship between domatia and mites?

P.M. Tilney^a, A.E. van Wyk^b and C.F. van der Merwe^c (Wed C1)

^a*Department of Botany and Plant Biotechnology, University of Johannesburg, PO Box 524, Auckland Park, South Africa 0006*

^b*Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002*

^c*Laboratory for Microscopy and Microanalysis, University of Pretoria, Pretoria, South Africa 0002*

Domatia are small structures on the lower surface of a leaf, usually consisting of cavities or pouches covered by hairs or leaf tissue, located in the axils between the midrib and secondary veins. They are found in many dicotyledons including certain members of the Rubiaceae. In the tribe Vanguerieae of this family, they may or may not be present. As part of an ongoing study of selected southern African members of the Vanguerieae, their structure in transverse section was investigated. Light microscopic (LM) observations revealed the presence of a number of “channels” extending from the outer periclinal cell walls of the

epidermal cells across the cuticle towards the cavity of the domatia in some of the taxa. These structures in *Plectroniella armata* were also examined using transmission electron microscopy (TEM). Domatia are known to house mainly mites, many of which are predatory or fungivorous, in a symbiotic (mutualistic) relationship. To date, the role of domatia is thought to be confined to the provision of shelter for these organisms, their eggs and their young during development. However, the present study of the microscopic structure of domatia revealed not only that the cuticle of the epidermis of the domatia is thicker than in other parts of the leaf, but also the presence of pronounced and closely-spaced cuticular folds which are particularly conspicuous in domatia. What appear as transcuticular "channels" under LM turned out to be cuticular folds with electron dense inclusions under TEM. The functional significance of the folds requires further investigation. In plants such folding of walls and membranes at ultrastructural level is usually functionally associated with an increase in surface area to facilitate the active exchange of compounds/metabolites. This may indicate that translocation of substances is possible from the domatium to the inhabitants (or vice versa) and therefore suggests a far more active role for the leaf in the symbiotic relationship than was previously thought. More work is required to test such a possibility.

The CREW Programme: Making significant contributions to taxonomy and conservation

M. Treurnicht and I. Ebrahim (Tue B4)

South African National Biodiversity Institute, Kirstenbosch Research Centre, Private Bag X7, Claremont, 7735

The Custodians of Rare and Endangered Wildflowers (CREW) is a programme that involves civil society in monitoring and conserving threatened plants. Our CREW volunteers has been providing us with critical data required for updating Red List Assessments, contributing to identifying conservation priority areas, making significant new plant discoveries and submitting important distribution records and specimens to several South African herbaria. The focus of this paper is to illustrate the important contributions that citizen scientists make to Botany by showcasing their new discoveries, collaborative projects with taxonomists and building on valuable herbarium collections. Since the inception of the CREW programme in 2003 we have discovered 15 new species, rediscovered 13 species and collected population data on 825 species of conservation concern.

Some Cape *Erica* species can self-pollinate: evidence for autogamy and geitonogamy, with a facultative and out-crossing seed-set advantage, in *Erica chloroloma* (Ericaceae)

R.C. Turner^a, J.J. Midgley^b and S.D. Johnson^a (Mon B2)

^a*School of Biological and Conservation Sciences, University of KwaZulu-Natal, P.Bag X01, Scottsville, Pietermaritzburg 3209, South Africa*

^b*Department of Botany, University of Cape Town, Rondebosch 7701, South Africa*

High levels of observed fruit-set in wild populations of *Erica chloroloma*, Cape St. Francis, South Africa, led us to hypothesise that, despite frequent visitation by Malachite Sunbirds and Greater Double Collared Sunbirds, a certain amount of self-fertilisation was occurring. We employed breeding experiments to determine the relative amounts of seed-set in terms of autogamous, geitonogamous and allogamous pollination. Our results indicate highest seed set with outcrossing but that autogamous and geitonogamous fertilisation can contribute significantly towards seed-set. Furthermore, facultative autogamy yielded higher seed-set than unmanipulated autogamy. We also determined that insects, mostly *Apis mellifera capensis* bees, were responsible for up to 20% of seed-set through selfing but not necessarily pollen transfer. Observed floral robbery by Cape Weavers and White Eyes also contributes towards seed-set through facultative disturbance of anther rings and subsequent selfing. As a result of these factors, high fertilisation levels are achieved and the species thus has a negligible pollen limitation value of 0.005 by the Larsen-Barrett index. Despite protogyny, the stigmas of individual flowers are receptive to their own pollen as soon as the stigma emerges from the corolla mouth. Our results showing that *E. chloroloma* has hedged its reproductive bets through the ability to self-pollinate, and that it is thus likely, despite relatively large sub-populations and abundant pollinators, that genetic diversity within stands of the species is lower than in similar-sized populations of allogamous, bird-pollinated Cape *Erica* species.

Climate change-driven mortality of *Euphorbia ingens* in the Limpopo Province: Causal abiotic and biotic factors

J.A. van der Linde^a, D.L. Six^b, M.J. Wingfield^a and J. Roux^a (Wed A3)

^a*Department of Microbiology and Plant Pathology, DST/NRF Centre of Excellence in Tree Health Biotechnology (CTHB), Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Private Bag X20, Hatfield, Pretoria, 0028, South Africa*

^b*College of Forestry and Conservation, Department of Ecosystem and Conservation Sciences, The University of Montana, Missoula, MT 59812, United States of America*

The plant genus *Euphorbia* is one of the most diverse in the world including more than 2100 species of both succulent and woody plants. In South Africa, the largest of the succulent tree-like euphorbias is *Euphorbia ingens*. In the last 10 to 15 years, high levels of mortality of these trees have been observed in the Limpopo Province of South Africa. The main symptoms include rotting and browning of the succulent branches, grey discoloration of the foliar parts, various spots and lesions on succulent branches, blue stain of the main woody stems as well as insect damage. In the past five years, a number of studies have been undertaken to investigate the possible causes of disease and mortality in this tree, looking at both biotic (fungal and insect) and abiotic (temperature, rainfall, evapotranspiration) factors.

Diseased plant material and associated insects were collected from four sites in the Limpopo Province. Fungi were isolated from insect tunnels in the succulent branches, blue stained wood as well as from insects collected from diseased plant material. Insects included weevils (Scolytinae), bark beetles (Cossoninae) and a moth (Pylalidae). A wide diversity of fungi were isolated belonging to the Botryosphaeriaceae, Cordycipitaceae, Microascales, Nectriaceae, Ophiostomataceae and the Teratosphaeriaceae. Analyses of weather data revealed changes in annual temperature and precipitation over the 40 year period analyzed, with greater upward trends in temperature and downward trends in precipitation in the Limpopo Province compared with the North West Province. Estimates of potential evapotranspiration and water balance indicated an increasing water demand while precipitation has remained the same or is decreasing. The dramatic death of *E. ingens* observed, since about 2000, appears to be linked to increasing moisture deficits resulting in tree stress. This in turn results in increased damage by opportunistic pathogens and insects ultimately contributing to tree mortality.

Pollinator ecotypes in the shrub *Erica plukenetii*

T. van der Niet, R. Turner and J. Midgley (Wed A1)

Botany Department, University of Cape Town, University Private Bag, Rondebosch 7700, South Africa

Pollinator-driven speciation has been proposed as an important factor driving speciation in the mega-diverse Cape Floristic Region. Most evidence in favour of this hypothesis put forward to date has come from large geophytic irid and orchid lineages, while data from proteoid and legume lineages seem to suggest that pollinators were not important. Here we present a case of pollinator ecotypes, arguably a key element in demonstrating pollinator-driven speciation, for *Erica*, the largest genus of the Cape flora. The widespread *Erica plukenetii* is characterized by great variation in vegetative and floral traits. The most common form, with long-tubed red flowers, was visited by orange-breasted sunbirds that carried large amounts of *Erica* pollen at the base of their beaks. Beak length closely matched the length of the floral tube, and flowers were unscented, as is typical of bird-pollinated flowers. Another form was characterized by white, short-tubed flowers which emit a pleasant floral scent. We speculate that this form is visited by nocturnal moths. The two forms have been found to co-occur in nature, reinforcing their status as genuine pollinator ecotypes. This is the first evidence for a role for pollinators in driving diversification in the florally diverse heaths, which constitute almost 10% of the entire Cape flora, and it refutes the idea that this process is limited to geophytic herbs only.

Biodiversity and restoration of Spekboom-dominated thicket

M.L. van der Vyver^a, R.M. Cowling^a, A.J. Mills^b, M. Difford^a and E.E. Campbell^a (Wed A2)

^a Restoration Research Group, Department of Botany, P.O. Box 77000, Nelson Mandela Metropolitan University, Port Elizabeth, South Africa 6031

^b Restoration Research Group, Department of Soil Science, Stellenbosch University, Private Bag XI, Matieland 7602, South Africa

An accepted criterion for measuring the success of ecosystem restoration is the return of biodiversity relative to intact reference ecosystems. The emerging global carbon economy has made landscape-scale restoration of severely degraded *Portulacaria afra* (Spekboom)-dominated subtropical thicket, by planting multiple rows of Spekboom truncheons, a viable land-use option. Although large amounts of carbon are sequestered when planting a monoculture of Spekboom, it is unknown whether this is associated with the return of other thicket biodiversity components. We planted in degraded, intact and three differently aged post-restoration (*P.afra* truncheons) sites nursery-propagated individuals of two woody canopy dominants (*Pappea capensis* and *Searsia longispina*), and two inter-canopy shrubs (*Lycium ferocissimum* and *Rhigozum obovatum*) in September 2008 (spring). The experiment was repeated again in May 2009 (autumn) and a succulent canopy species (*P. afra*) was added. We assessed restoration success in terms of the survival of planted individuals after 24 months (spring planting) and 12 months (autumn planting). Contrary to expectations, survival was not related to a gradient of intactness encompassing degraded, restoration and intact treatments that are associated with increasing biomass and soil carbon. The costs of incorporating the four woody canopy species into the restoration programme's protocol were 2.4 times the costs of restoring with *P. afra* alone. We also used carbon stock data from a restored and intact sites, and sampled carbon stocks at restored stands at a second site in the same thicket plant community. Similarly, we sampled plant community composition at both sites. The total carbon stock of the oldest post-restoration stand ($250.8 \pm 14 \text{ t C ha}^{-1}$) approximated that of intact stands (245 t C ha^{-1}) and we observed a general increase in carbon content with restoration age ($71.4 \pm 24 \text{ t C ha}^{-1}$ after 35 and $167.9 \pm 20 \text{ t C ha}^{-1}$ after 50 years). A multiple correspondence analysis separated degraded stands from stands under restoration based on ground cover, floristic composition and total carbon stock. Older post-restoration and intact stands were clustered according to woody canopy recruit abundance. Our results suggest that Spekboom is an ecosystem engineer that promotes the spontaneous return of canopy species and other components of thicket biodiversity.

Identifying genomic regions involved in growth, wood property, transcript and metabolite variation in an F₂ pseudo-backcross pedigree of *Eucalyptus grandis* x *E. urophylla*

M.M. van Dyk^a, A.R.K. Kullana^a, E. Mizrachi^a, C.A. Hefer^a, L. Jansen van Rensburg^a, D. Newman^b, N. Coetzer^c, T.J. Tschaplinski^d, K.C. Cushman^d, N.E. Engle^d, G.A. Tuskan^d, N. Jones^e, A. Kanzler^e, A. Bayley^f and A.A. Myburg^a (Mon C1)

^aDepartment of Genetics, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, 0002, South Africa

^bDepartment of Plant Science, University of Pretoria, Pretoria, South Africa

^cBioinformatics and Computational Biology Unit, Department of Biochemistry, University of Pretoria, Pretoria, South Africa

^dBiosciences Division, Oak Ridge National Laboratory, Oak Ridge, TN 37831, USA

^eSappi Forests Research, Shaw Research Centre, PO Box 473, Howick, 3290, South Africa

^fSappi Technology Centre, PO Box 6, The Innovation Hub, Lynnwood Pretoria 0087, South Africa

Breeding of *Eucalyptus* hybrids for clonal forestry in subtropical regions of South Africa is aimed at combining the growth, form and rooting ability of *E. grandis* with the disease tolerance of *E. urophylla*. The underlying genetic basis of the observed hybrid superiority remains unclear. To identify genetic factors controlling growth and wood property traits in eucalypt hybrids, an F₂ pseudo-backcross mapping family (n = 314) derived from a cross between an F₁ hybrid (GUSAP1, *E. grandis* x *E. urophylla*, Sappi Forest Research) and an *E. urophylla* parent (USAP1) was used for genetic linkage map construction, using microsatellite (SSR) and Diversity Arrays Technology (DArT) markers. Phenotypic trait assessment for quantitative trait locus (QTL) analysis included physical measurements of tree diameter and wood density performed on three-year-old individuals. Klason (acid-soluble & -insoluble) lignin and cell wall sugar content were determined for a selection of 100 backcross progeny and used for near-infrared analysis (NIRA) calibration. NIRA predictions for glucose, xylose, arabinose, cellulose and total lignin content, as well as pulp yield were made for all individuals. Total lignin and S:G ratios were also separately measured for all individuals. Immature xylem tissues, collected from 192 backcross progeny, were used for metabolite profiling (ORNL, Oak Ridge, TN) and Illumina mRNA-Seq (15 million PE50, BGI Americas) quantification of transcript levels of more than 30,000 genes for which xylem expression was detected. To bridge the gap between fine mapping and QTL validation studies, transcript and metabolite levels were treated as quantitative traits and used for eQTL and mQTL mapping, respectively. Co-localization of wood property, expression and metabolite QTLs will facilitate the identification of positional candidate genes and other components of regulatory networks underlying phenotypic variation in this interspecific backcross pedigree.

Ensemble optimisation of *cis*-regulatory element discovery: *in planta* benchmark and discovery in *Eucalyptus*

I. van Jaarsveld^{a,b}, E. Mizrachi^b, F. Joubert^a, Y. van de Peer^c and A.A Myburg^b (Mon C1)

^a*Bioinformatics and Computational Biology Unit, Department of Biochemistry, University of Pretoria, Pretoria, 0002, South Africa*

^b*Department of Genetics, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, 0002, South Africa*

^c*Bioinformatics and Systems Biology, Plant Systems Biology Department, University of Ghent, Ghent, 9000, Belgium*

Cis-regulatory motif discovery is extensively applied in the *in silico* analysis of plant promoters. This is a pattern recognition endeavour which ascribes regulatory function to subtly conserved 5 – 20 nt words. Guiding this approach is the “guilt-by-association” assumption that regions in the promoters of co-expressed or orthologous genes bind the same or similar transcription factors, and so regulate gene expression. Numerous algorithms have been developed to detect regulatory motifs. Their performance is generally poor with sensitivity < 0.223 and precision < 0.308 in yeast, *Drosophila*, mouse and human. Despite this performance, the necessity for annotating transcription factor binding sites in hierarchical regulatory pathways, ensures that it remains a widely used practice. Spatial conservation of promoter motifs in plants is expected to be less than that of metazoans, as large-scale genome rearrangements and transposable element activity have engendered a loss of colinearity, expanded gene families, and resulted in poor conservation in proximal non-coding DNA. Before attempting *cis*-element discovery in *Eucalyptus grandis* promoters, we are comprehensively benchmarking existing motif discovery algorithms on plant-specific, simulated and generic promoters. We aim to decipher the abilities and limitations of motif discovery tools in plants, and employ a synergistic ensemble implementation for secondary cell wall and cellulose biosynthesis related promoters in *E. grandis* trees. This will ensure as accurate as possible detection of regulatory motifs which are associated with the economically important properties of secondary cell walls in woody tissue, in this newly sequenced, fast-growing and globally dominant plantation tree species.

Effects of smoke-water and smoke-derived stimulants on rooting

J. van Staden and M.G. Kulkarni (Mon C2)

Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa

It is now well documented that smoke and smoke-derived compounds stimulate germination and in the process significantly increase root growth. When considering the literature it is clear that the effect on root growth *per se* has never been dissected properly. The main objective of this study was to test the effect of smoke-water and smoke-isolated butenolide (3-methyl-2*H*-furo[2,3-*c*]pyran-2-one) on root system of a wide range of plant species including agricultural and horticultural crops. Findings of these studies have shown that smoke solutions have an ability to stimulate rooting of a number of plant species. It is interesting to note that smoke solutions also showed an improvement of rooting under temperature and water stress conditions. The results of these studies are of considerable

importance as root structure and developed forms the basis of successful plant establishment, nutrient mobilization and ultimate plant survival and crop production.

Ethnobotany of the Cederberg

B.-E. van Wyk (Tue C1)

Department of Botany and Plant Biotechnology, University of Johannesburg, PO Box 524, Auckland Park 2006, South Africa

The traditional uses of plants in the Cederberg region of the Western Cape Province, South Africa, are poorly documented. The Cederberg is well known as the source of commercial natural products such as rooibos tea (*Aspalathus linearis*) and round leaf buchu (*Agathosma betulina*); some others are now obsolete, including cedar wood (*Widdringtonia cederbergensis*) and tan bark from waboom (*Protea nitida*), suikerbos (*Protea neriifolia*), kliphout (*Heeria argentea*) and pruimbas (*Osyris compressa*). This report forms part of an ongoing study to document the useful plants of the Cederberg as a contribution towards a more complete understanding of the broader patterns of traditional plant use in the Cape region. A checklist of more than 106 ethnobotanically relevant plant species has been compiled, including plants that are important as medicine or for wound healing, food, beverages, timber, firewood and several other everyday uses. Local inhabitants of the Cederberg (in villages such as Wuppertal, Heuningvlei and Kleinvlei) still regularly use indigenous plants. Until recently, these villages were isolated from the main centres (with no easy access to clinics and pharmacies) but an improved road system will undoubtedly result in the people becoming less dependent on indigenous plants so that the knowledge may gradually be lost. There is a need for quantitative studies to allow for comparisons between different communities and also between different generations.

Characterisation of the cysteine protease and cysteine protease inhibitor family in soybean nodules.

S.G. van Wyk^a, K.J. Kunert^a and B.J. Vorster^b (Wed C3)

^a *Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002.*

^b *Department of Plant Production and Soil Science, University of Pretoria, Pretoria, South Africa 0002.*

Soybean (*Glycine max*) is one of the most important sources of vegetable protein for food and animal feed world-wide. The symbiotic relationship between soybean and the Rhizobia, *Bradyrhizobium japonicum*, enables soybean to assimilate fixed nitrogen in the root nodules. This symbiotic association allows soybean to be cultivated with reduced nitrogen supplementation, lowering production costs and maintaining high-protein yield. However, the nodule life is limited and sensitive to environmental stress. The plant cysteine protease-cystatin system plays an important role during plant development processes and has been shown to be involved in nodule senescence. The release of the complete genome data of soybean has allowed all putative cysteine proteases and cystatin sequences to be identified. However, it is not known whether all of these putative sequences are expressed, where and when they are expressed, or which cysteine proteases and cystatins are involved in nodule development and senescence. In soybean, only limited information is currently available

about the expression of cysteine proteases with no cystatin expression data, despite soybean being one of the major crop plants in the world and of significant importance to South Africa. The hypothesis of this study is that there is a balanced interplay between individual cysteine proteases and cystatins during nodule development, with changing expression profiles during nodule development and during senescence. The overall aim of the study will be to advance our knowledge regarding the cysteine protease-cystatin system in soybean and to establish a relationship between individual cysteine protease and cystatin expression during nodulation. Further characterization of the individual components of the protease–protease inhibitor system, might make it possible to silence a particular cysteine protease or recombinantly express a specific natural or engineered cystatin in soybean nodules that could possibly delay either natural or stress-induced nodule senescence.

Effective invasive species management around protected areas: Understanding the spatial dynamics of *Lantana camara* invasions in South Africa's Kruger National Park

W. Vardien^a, D.M. Richardson^a, L.C. Foxcroft^{a,b}, J.R. Wilson^{a,c} and J.J. Le Roux^a (Tue A4)

^a*Centre for Invasion Biology, Department of Botany & Zoology, Stellenbosch University, Matieland 7602, South Africa*

^b*Conservation Services, South African National Parks, Skukuza 1350, South Africa*

^c*South African National Biodiversity Institute, Kirstenbosch National Botanical Gardens, Claremont 7735, South Africa*

Invasive plants are one of the greatest threats to biodiversity in Kruger National Park (KNP), South Africa's flagship protected area, primarily because of their impact on ecosystem services and functioning. The globally invasive and highly variable species complex, *Lantana camara*, is particularly troublesome in this regard. *Lantana camara* reproduces sexually and vegetatively, and is associated with multiple dispersal vectors. Here we use molecular techniques to explore how *L. camara* disperses in the KNP's Sabie-Sand River catchment, with the aim of providing management-relevant insights. The history of the species' presence in the park suggests either: a) downstream dispersal of seeds during normal flow and flood events, and/or b) long distance dispersal by vertebrates and wind. Using inter simple sequence repeat (ISSR) and amplified fragment length polymorphism (AFLP) molecular markers, we assessed population genetic structure and used it as a proxy for patterns of dispersal and to identify the main contributing sources of propagules. Our results indicate that genetic variation is partitioned largely within populations as opposed to between them. Populations located at the confluence of the Sabie and its main tributary, the Sand River, show the highest genetic diversity. We also found that populations along the Sand River contribute substantially more propagules to the lower reaches of the Sabie than the upper Sabie itself. Spatial analysis revealed a positive correlation between genetic and geographic distance along the river. The most likely spread scenario for *L. camara* in this area is continuous, from the Sand River sub-catchment down into the lower Sabie River. However, only a small part of the Sand River falls under KNP management. We therefore recommend that control efforts should be focussed on the parks' neighbouring areas as well, if the spread and re-infestation of *L. camara* is to be minimized.

Natural products in anti-obesity therapy – the good, the bad and the ugly

I. Vermaak and A.M. Viljoen (Tue C3)

Department of Pharmaceutical Sciences, Tshwane University of Technology, Pretoria, South Africa 0001

Obesity is a major health concern of modern times and should be considered a global epidemic. In 2005 it was estimated that 1.6 billion adults were overweight globally with at least 400 million classified as obese. Obesity is associated with and can lead to many disease conditions including type-2 diabetes, cardiovascular disease, hypertension, sleep apnoea, and cerebrovascular accidents amongst many others. Conventional medicines used to treat obesity have high abuse potential and frequently exhibit side effects. The commercial market for anti-obesity preparations is enormous due to public awareness of the ill-effects of obesity and the general perception that being obese is less than attractive. This multibillion-dollar industry expands on a daily basis as new ‘miracle cure’ products become available. The continued search for new therapies has revealed multiple targets to combat obesity and highly complex plant extracts are ideally suited to fulfil a multi-targeted approach. Many herbal formulations are currently commercially available but most of them have been poorly researched in terms of efficacy and safety, if at all. Not surprisingly, associations between consumption of botanical products and instances of toxicity have been and are still being made. It is therefore imperative that these plants and derived herbal products be extensively investigated in terms of quality, efficacy, and especially safety in order to validate their widespread consumption. The phytochemistry, mechanism of action, *in vivo* efficacy or lack thereof and toxicity will be discussed for several of the most important plants to date used to treat obesity including: *Camellia sinensis*, *Citrus aurantium*, *Ephedra sinica*, *Hoodia gordonii*, *Ilex paraguariensis* and *Panax ginseng*.

The application of vibrational spectroscopy and chemometrics in taxonomy

A.M. Viljoen, M. Sandasi, J.E. Maree and N.P. Mncwangi (Tue C4)

Department of Pharmaceutical Sciences, Tshwane University of Technology, Private Bag X680, Pretoria, South Africa 0001

Vibrational spectroscopy has been identified as an important quality control technique in the pharmaceutical, food and beverages, agriculture and mining industries. The diverse applications include; the inspection and identification of raw materials and quantification of active constituents or contaminants. The technique has gained popularity as it is non-destructive, inexpensive and requires minimal sample preparation. This work reports on the use of vibrational spectroscopy in the quality assessment of several commercially important South African species (*Agathosma*, *Pelargonium*, *Salvia*, *Eriocephalus*, *Harpagophytum*). Chemometric processing of the spectral data using orthogonal projections to latent structures (OPLS) discriminant analysis, partial least squares (PLS) and multivariate calibration models has shown that vibrational spectroscopy is a powerful technique to distinguish between closely related species.

Alternative indigenous Rutaceous hosts of ‘*Candidatus Liberibacter africanus*’ and ‘*Candidatus Liberibacter africanus* subsp. *capensis*’

R. Viljoen^{a,b}, E.T. Steenkamp^{a,b}, and, G. Pietersen^{a,b,c} (Tue A2)

^aDepartment of Microbiology and Plant Pathology, University of Pretoria, Pretoria 0002

^bForest and Agricultural Biotechnology Institute, University of Pretoria, Pretoria 0002,

^cAgricultural Research Council - Plant Protection research institute, Pretoria 0002.

‘*Candidatus Liberibacter africanus*’ (Laf) is the causal agent of citrus greening disease in South Africa. This bacterium is of economical importance to the citrus industry as the disease it is associated with results in a considerable reduction in yield. A related bacterium, ‘*Candidatus Liberibacter africanus* subsp. *capensis* (Laf C), is commonly associated with an indigenous member of the Rutaceae, *Calodendrum capense*. It has been postulated that Laf C may represent an ancestral form of Laf. This study aims to determine whether other alternative indigenous Rutaceous host exist that harbours either Laf or Laf C. 269 *Vepris lanceolata*, 135 *Zanthoxylum capense* and 75 *Clausena anisata* trees were sampled from across South Africa. These Rutaceous members were selected as the psyllid vector of Laf, *Triozaerytrae*, is capable of completing its life cycle on these trees. Total DNA was extracted from these samples which were then subject to a generic *Liberibacter* species real-time PCR. Samples with a Ct value of below 35 were considered positive. A conventional PCR specific for *Liberibacter* were then performed on these samples and directly sequenced thereafter to identify the *Liberibacter* species detected. Thus far LafC has been identified from 11 *V. lanceolata* trees collected from Knysna. Other samples are in the process of being screened. This is the first report of LafC being present in an indigenous Rutaceae other than *C. capense*. Laf has only been associated with citrus trees from various African countries including South Africa, Mauritius and Reunion Island where citrus is not indigenous. Thus the possibility exists that Laf originated through a “host jump” from an indigenous bacterium onto citrus upon its introduction into the region. A thorough understanding into the host range of Laf will also aid in control of citrus greening disease.

Ug99 variants of *Puccinia graminis* f. sp. *tritici* in South Africa

B. Visser^a, L.J. Szabo^b, L. Herselman^a and Z.A. Pretorius^a (Mon C2)

^a Dept. Plant Sciences, University of the Free State, Bloemfontein, South Africa

^b USDA-ARS Cereal Disease Laboratory, University of Minnesota, St. Paul, USA

The discovery of the Ug99 wheat stem rust race in Uganda in 1999 heralded a new era for wheat rust research. Ug99 was the first race to possess virulence for *Sr31*, a durable and widely used stem rust resistance gene. Initial concerns about Ug99 in terms of food security were not completely unfounded, since not only has it spread to several African countries, Yemen and Iran, but seven variants in the Ug99 race group have also been described. Based on field tests in eastern Africa, 90% of the world’s wheat varieties are susceptible to Ug99 and its related races. Even though wheat cultivation in South Africa is small compared to other countries, three of the seven Ug99 variants were originally identified in South Africa. Microsatellite analysis of South African stem rust races divided the population in two distinct groups. The first consisted of historically detected races whereas the second grouping

contained the Ug99-related races, confirming the close relationship between Ug99 and its South African variants. Two of these races most probably represent exotic introductions into South Africa, while the third seems to be a single step mutation. In 2010, an eighth variant in the Ug99 lineage was identified in South Africa, when race TTKSF acquired virulence for an unknown resistance gene in a local cultivar. The application of new marker technology and implications of Ug99 diversity will be discussed.

Plant cystatins and insect cysteine proteases: weapons in a molecular arms race

B.J. Vorster^a, M-C. Goulet^b and D. Michaud^b (Mon C1)

^a*Department of Plant Production and Soil Science, University of Pretoria, Pretoria, South Africa 0002.*

^b*Département de Phytologie, Faculté des Sciences de l'Agriculture et de l'Alimentation, Université Laval, Québec (QC) Canada G1V 0A6*

On a molecular level the war between plants, and the insects that eat them, is akin to an arms race, with each organism continually striving to out match the other. One of the weapons in the arsenal of plants, targeted against the digestive proteases of herbivorous Coleoptera, such as the Colorado potato beetle, is cystatins or cysteine protease inhibitors. Cystatins are small proteins that act as natural inhibitors of cysteine proteases, thereby disrupting the digestion of plant material by insects such as the Colorado potato beetle. However the Colorado potato beetle is able to compensate towards the presence of dietary protease inhibitors in plant tissues through a multi-component defensive strategy involving the over-expression of inhibitor-sensitive digestive proteases, the expression of proteases insensitive to the inhibitors as well as through proteolytic inactivation of the inhibitory proteins using non-target proteases. More than 30 digestive cysteine proteases have been identified so far in the potato beetle, in line with the observed ability of this insect to elude the detrimental effects of cystatins induced in wounded potato leaves. Our goal, in this study, was to determine whether functional diversity of the potato beetle digestive cysteine protease complement was matched with similar functional variability of the potato host cystatin complement, using the wound-inducible eight-domain potato multi-cystatin (PMC), as a model. Computational modelling of the PMC domains interacting with cysteine proteases of the beetle suggested the onset of variable interaction strengths for the PMC domains, then confirmed empirically with protease inhibitory data showing complementary protease inhibitory spectra among the eight domains. This data suggest, overall, that functional variability among PMC inhibitory domains has evolved in response to predatory challenges by the Colorado beetle relying on cysteine proteases for potato leaf protein digestion.

Is frost a demographic bottleneck for savanna trees? Testing the effect of freeze events on *Colophospermum mopane* population structure in Limpopo Province, South Africa

M. Whitecross^a, S. Archibald^b and E.T.F. Witkowski^a (Wed A3)

^a*School of Animal, Plant and Environmental Sciences, University of the Witwatersrand, Johannesburg, South Africa 2050*

^b*Natural Resources and the Environment, CSIR, PO Box 395, Pretoria, South Africa 0001*

Frost disturbance is often mentioned in southern African savanna literature, but is seldom discussed in great detail. Nevertheless, it can represent an above-ground disturbance as effective as fire depending on the resistance capacity of the effected tree species. A severely freeze damaged stand of *Colophospermum mopane* in the Venetia Limpopo Nature Reserve provided an opportunity to investigate the mechanisms behind freeze damage impacts on *C. mopane*. Frost may therefore act as a possible demographic limitation of *C. mopane* in preventing its southwards spread. Freeze damage of individual trees was assessed according to tree height and landscape position -with lower elevations representing the most severe freeze zones and higher elevations the least severe freeze zones. A high freeze severity threshold (558.75 m.a.s.l.) and a low freeze severity threshold (564m.a.s.l) were found to distinguish between populations of smaller, coppicing trees at lower elevations, and taller, non-coppicing trees at higher elevations. Little freeze damage was observed on tree canopies above 4 m in height. Trees below 4 m that had experienced 100% freeze damage, failed to regrow to their original heights from the previous season. This is a possible driver of the pre-freeze height differences seen across the slope; with low elevation trees having to recover from freeze events more frequently, and subsequent topkill resulting in a decrease in height over time. Taller trees at higher elevations experience less freeze damage and can grow beyond the 4 m freeze zone which decreases their chance of severe topkill and stunted growth. Long term effects linked to heights and densities suggested fewer, smaller trees at lower elevations due to relatively higher freeze frequency impacts. It appears *C. mopane* has limited resistance to freeze events, and this may be linked to the absence of this species at colder latitudes in the Southern Hemisphere.

Biological invasion assessment and eradication—a national programme to reduce South Africa's invasion debt

J.R. Wilson^{a,b}, P. Ivey^a, I. Nänni^a and P. Manyama^a (Tue B2)

^a*South African National Biodiversity Institute, Kirstenbosch Research Centre, Claremont 7735, South Africa.*

^b*Centre for Invasion Biology, Department of Botany and Zoology, Stellenbosch University, Private Bag XI, Matieland 7602, South Africa*

The number of biological invasions in South Africa is increasing as introduced species naturalise, naturalised species become invasive, and species which are invasive in small areas become much more widespread. Therefore, even if there are no new introductions, this country has a massive invasion debt. However, to date, most management and research has focussed either at the border or after species have become widespread. What has been missing are general post-border assessments of the risks posed by introduced species, and the

consideration of eradication as a management goal where feasible. To address this, a national programme of Early Detection and Rapid Response was initiated in 2008. In this talk we discuss the rationale for such an approach, how it was achieved, and progress to date. We use Australian acacias as a case-system to explore some of the main issues.

The transition of SANBI's herbaria from PRECIS to a BRAHMS database management system

P.J.D. Winter (Tue B4)

Biodiversity Information Management Directorate, Kirstenbosch Research Centre, SANBI, Cape Town.

The PRECIS database was an international leader in its class for several decades (1974—1990), but with the field of databases moving more into the realm of PC's, Windows, Internet, and general connectivity, it failed to progress much beyond a mainframe database model where users have only limited access to the data holdings. Other databases were in a better position to make use of technological advances, and have overtaken it. Currently on an InterBase & Delphi platform, the expertise of developing PRECIS to user's needs is no longer available or cost-effective. There has been a steady increase in demand among our users (for at least the last seven years) for the basic type of relational database functionality from PRECIS that could no longer be supplied with the InterBase/Delphi system given the resources available for its operation and management. SANBI will provide a herbarium collections' data service that meets the current demand of users, and is built upon the sound management, staffing and operation of a core BRAHMS database. This will be the preferred national plant information system to support the capture & management of fundamental taxonomic, specimen & floristic information for SA. The project involves an integration of data from several sources in SANBI. Some of this data will be centralised, while others will be brought in as needed from external sources. The benefits and risks associated with centralisation are discussed.

The role of megaherbivores in shaping the structure of subtropical plant communities

**K. Yessoufou^{a,b}, O. Maurin^{a,b}, T.J. Davies^c, M. Kuzmina^d, H. Schafer^e,
M. van der Bank^{a,b} and V. Savolainen^f (Wed A2)**

^a*African Center for DNA Barcoding, University of Johannesburg, Department of Botany and Biotechnology, PO Box 524, Auckland Park 2006, South Africa*

^b*Department of Botany and Plant Biotechnology, University of Johannesburg, P.O. Box 524, Auckland Park 2006, Johannesburg, South Africa.*

^c*McGill University, 1205 Avenue Docteur Penfiel, Montreal, Quebec, Canada*

^d*Canadian Centre for DNA Barcoding, University of Guelph, 50 Stone Road East, Guelph, N1G 2W1, Canada*

^e*Harvard University, 22 Divinity Avenue, Cambridge, 02138, USA*

^f*Imperial College London, Silwood Park Campus, Ascot, SL5*

The assembly of organisms into communities is influenced by a variety of ecological and climatic factors, but evolutionary relationships also play a key role in shaping the structure of these assemblages. Examining phylogenetic trees of species within communities provides a framework in which processes such as competition or adaptation leave distinct signatures - an approach that has been successfully applied to describe the patterns of a few rainforests, microbial and animal communities. However, understanding the processes underlying phylogenetic community structure has been hampered by a lack of long-term experiments to test complex biotic interactions. We analysed plant communities of the subtropical savanna-woodland biome of southern Africa, renowned for its large mammalian fauna. Reconstructing the phylogeny of species of trees and shrubs based on DNA barcodes data and measuring multiple defense strategies, we then compared the phylogenetic structure of these communities under various pressures of herbivory, including 'exclosures' where megaherbivores have been denied access for decades. We demonstrate that heavy pressure from generalist megaherbivores shapes the structure of plant assemblages, favouring clusters of closely related species, while also increasing functional diversity and providing a buffer to disturbances. Our results provide the first empirical test of theoretical expectations for the effect of herbivory on community structure. These results help understand the evolution of savanna-woodlands under drivers of change, while shedding new light on how management of large game animals, such as elephants, impacts biodiversity conservation in these habitats.

The molecular and metabolite profiling of grapevine berries in a model vineyard where the microclimate of the developing bunches has been altered

P. Young^{a,b}, E. Alexandersson^c, D. Jacobson^{a,b}, J. Lashbrooke^{a,b}, Z. Coetzee^b, A. Deloire^b and M. Vivier^{a,b} (Tue A1)

¹*Institute for Wine Biotechnology,* ²*Department of Viticulture and Oenology, Stellenbosch University, Private Bag X1, Matieland 7602, South Africa*

³*Department of Plant Protection Biology, Swedish University of Agricultural Sciences, SE-230 53 Alnarp, Sweden*

An estimation of grape composition is considered to be essential in establishing grape quality. Grape composition is influenced by a number of factors and can to a certain extent be manipulated in the vineyard by viticultural management practices. This study investigated the molecular response of *Vitis vinifera* cv. Sauvignon blanc berries to treatments that led to an altered microclimate in the bunch zone of a highly characterised vineyard setting (i.e. a model vineyard). The carotenoid biosynthetic pathway was investigated due to its crucial role in photosynthesis (especially photoprotection); as well as its role in the formation of the norisoprenoids: compounds formed by the degradation of carotenoids and responsible for the characteristic flavour and aroma of certain cultivars. Pathway analysis was used to analyse the expression of the carotenoid biosynthetic genes at three time points during berry development using grape whole genome arrays. The photosynthetic pigments were analysed and the concentrations in the berries correlated to gene expression. The altered microclimate in the bunch zone resulted in significant changes in gene expression and carotenoid and chlorophyll concentrations. The expression of the genes involved in specifically the xanthophyll cycles, as well as the photoprotective xanthophylls was differentially affected in the treated grapevines, relative to control plants. This approach proved valuable to understand the molecular response of developing grape berries and the impact of changing environmental conditions.

Pharmacological and phytochemical analysis of a medicinal plant mixture that is used as traditional medicine in Western Cape

S. Zonyane^a, S.F. van Vuuren^b and N.P. Makunga^a (Wed B1)

^a*Department of Botany and Zoology, Stellenbosch University, Private Bag X1, Stellenbosch, 7600, South Africa*

^b*Department of Pharmacy and Pharmacology, Faculty of Health Sciences, University of the Witwatersrand, 7 York Road, Parktown 2193, South Africa*

In traditional medicine, different plant species which are often not related are combined as to enhance the efficacy of a medicinal preparation. As part of indigenous herbal knowledge, it is believed that better therapeutic effects are derived from synergistic interactions between different plant constituents. However, there are few studies which have confirmed this practice as it is mainly single plant extracts that are screened for bioactivity in South Africa. We thus investigated a plant mixture which consisted of three plant species; *Agathosma crenulata*, *Dodonaea viscosa* and *Eucalyptus globulus* for synergistic interactions. This plant combination was the most potent out of eight plant mixtures which were screened for antibacterial activity, with MIC value of 0.049 against *S. aureus* and *B.*

subtilis. Individual plant extracts were not as effective as the whole plant combination. An in-depth investigation of phyto-synergism in this plant combination using different combinations and ratios was conducted. Fractional inhibitory concentrations (FIC) for two-plant combinations revealed enhanced activity when plants are combined (FIC values ranged from 0.079 to 0.750). The combination of *D. viscosa* and *E. globulus* (0.079) exhibited the strongest synergistic interaction while the combination of *D. viscosa* and *A. crenulata* was mildly synergistic for all test bacteria. Combining plants into a mixed herbal preparation was beneficial for improving the action of plant mixtures as individual plants were less active on their own.

Abstracts of Posters

Effects of long-term storage on antimicrobial and cyclooxygenase-1 inhibitory activities of South African medicinal plants

S.O. Amoo, A.O. Aremu, M. Moyo, J. van Staden (M01)

Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa

The antimicrobial and cyclooxygenase-1 (COX-1) inhibitory activities of 50% methanolic extracts obtained from ten different medicinal plants stored for 16 years at room temperature were compared to those of fresh plant materials collected from the same locations. The plants evaluated are traditionally used for treating pain and infection-related ailments and they included *Acokanthera oppositifolia*, *Artemisia afra*, *Buddleja salvifolia*, *Clausena anisata*, *Dombeya rotundifolia*, *Ekebergia capensis*, *Ocotea bullata*, *Protorhus longifolia*, *Spirostachys africana* and *Tetradenia riparia*. Lower minimum inhibitory concentrations (MICs) against *Staphylococcus aureus* were observed in stored plant materials of all the species except *A. afra* and *P. longifolia* when compared to the respective fresh materials. With the exception of *D. rotundifolia* and *P. longifolia*, the MICs of the stored plant materials against both *Candida albicans* and *Pseudomonas aeruginosa* were either lower than or the same as those of fresh materials. The percentage COX-1 inhibition values of the stored plant materials were generally higher than their respective fresh materials. Current findings indicate that stored plant materials may retain their biological activities over a period of time. Better activities observed in majority of the stored plant materials are probably due to the production of more potent metabolites arising from the breakdown of some constituent chemicals.

Stimulatory role of karrikinolide (KAR₁) and smoke-water on photosynthetic pigment and phenolic contents of micropropagated 'Williams' bananas

M.W. Bairu, A.O. Aremu, J. van Staden (B01)

Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa

At low concentrations, smoke-water (SW) and smoke-derived karrikinolide (KAR₁) are compounds with potential cytokinin- and auxin-like activity. In comparison with *meta*-topolin (*mT*), their role on the growth, photosynthetic pigment and phenolic contents of micropropagated 'Williams' bananas was investigated. Explants were cultured in modified Murashige and Skoog (MS) basal media supplemented with either SW (1:125; 1:250; 1:500; 1:1000; 1:2000 dilutions) or KAR₁ concentrations ranging from 3.3×10^{-12} to 1.2×10^{-24} . After 42 d, growth parameters were measured while the photosynthetic pigments and phenolic contents were determined using spectrophotometric methods. The photosynthetic pigments chlorophyll a, b and total carotenoid contents were significantly improved by KAR₁ (4.8×10^{-22}) and SM (1:125 and 1:1000). These treatments were approximately two-fold and three-fold better than the control and *mT*, respectively. Total phenolic content was highest in KAR₁ at 1.0×10^{-19} and 7.8×10^{-17} for the leaves and roots, respectively. Furthermore, KAR₁ at 1.0×10^{-19} had a significantly higher amount of total flavonoids and proanthocyanidin in the root parts. At 1:500 dilutions SM stimulated the highest amount of total flavonoids in the leaves across the treatments. Since photosynthetic pigments and phenolic compounds play a

significant role in the growth, defense and survival strategy of plants, the findings of this study will have practical significance for the acclimatization of micropropagated plants. The results are also demonstration of the potential of smoke as an eliciting agent.

Somatic embryogenesis and plant regeneration from leaf explants of *Merwillia plumbea* (Lindl.) Speta.

P. Baskaran and J. van Staden (B02)

Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Scottsville 3209, South Africa

Merwillia plumbea, a popular but threatened medicinal plant in South Africa, was investigated for somatic embryogenesis by cell suspensions and plant regeneration. Embryogenic callus was initiated from *in vitro* grown *M. plumbea* leaf explants on agar-solidified MS medium containing 8.3 μM picloram, 2.3 μM thidiazuron (TDZ) and 20 μM glutamine. Callus was subsequently proliferated on solid medium containing 4.1 μM picloram, 2.3 μM TDZ and 20 μM glutamine before transfer to liquid MS medium supplemented with 0.4 μM picloram and 0.9 μM TDZ for somatic embryo development. The highest frequency of somatic embryogenesis (5.4 - 35.6, globular to cotyledonary somatic embryos/settled cell volume (SCV)) was induced on liquid medium containing 0.4 μM picloram and 0.9 μM TDZ and 150 mg l^{-1} haemoglobin. The embryos were regenerated (94.4%) into plantlets on solid media containing 1.4 μM gibberellic acid. The plantlets were grown further on half-strength MS medium and successfully acclimatized. This is the first report on induction of *M. plumbea* somatic embryogenesis. The protocol developed offers a viable means for mass propagation and germplasm conservation, and it provides a system for physiological, biochemical, molecular and cellular studies of embryo development.

Soil factors influencing the distribution of *Portulacaria afra* in subtropical thicket

C.H. Becker^{a,b}, C. Coetsee^b and R.M. Cowling^a (E02)

^a*Department of Botany, P.O. Box 77000, Nelson Mandela Metropolitan University, Port Elizabeth, South Africa 6031*

^b*School of Natural Resource Management, Private Bag X6531, Nelson Mandela Metropolitan University, George, South Africa 6530*

Subtropical thicket in South Africa has been heavily degraded and transformed to an open desert like state, with a major loss of the succulent *Portulacaria afra*. Restoration of thicket can be achieved by planting *P. afra* cuttings, and this study aims to investigate the relationship between selected soil properties and *P. afra*, over a range of soil types and landscape positions, to aid in the identification of the most appropriate restoration sites. It investigates *P. afra* occurring in both natural stands and in restoration sites; to determine factors that influence its distribution. Observations and research shows that *P. afra* dominates crests of hills and decreases with distance down slope; that it prefers growing on north facing slopes and barely exists on south facing

slopes; that it's tolerant of a wide range of soil conditions but thrives on rocky, dry soils and avoids soils that may be seasonally waterlogged. It appears to survive better in conglomerate soils than in alluvial soils and a possible clear indication is that sodium concentrations could be a controlling factor. The data collection is split into three parts, whereby 1) in experimental plots *P. afra* cuttings planted in different soil types are subjected to different levels of salinity; 2) on a landscape level ten sites between Grahamstown and Calitzdorp are analysed to determine which factors (aspect, gradient, soil depth, water infiltration and soil texture and sodium levels) could be important in driving distribution; and 3) intensive studies on north and south facing slopes at three of Working for Water's restoration sites whereby soil is analysed for macro-nutrients, particle size, texture, and pH. This will combine with the survival rates of *P. afra* determine if its distribution is related to soil nutrients or physical properties of the soil and if it's avoiding soils with higher concentrations of sodium.

Description of novel *Burkholderia* species associated with the root nodules of indigenous Hypocalypteae and Podalyrieae

C.W. Beukes, L. Mthombeni, E. van Zyl, S.N. Venter and E.T. Steenkamp (D01)

Department of Microbiology and Plant Pathology, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria 0002, South Africa

Most legumes can establish nitrogen-fixing symbioses with root-nodule bacteria. This group of bacteria can represent either Alpha- or Betaproteobacteria. Although much is known regarding root-nodule bacteria of agriculturally important legumes, the nitrogen-fixing symbionts of hosts indigenous to South Africa remains to be characterized. The research in this study focused on the rhizobial symbionts of the three indigenous *Hypocalyptus* species (subfamily Papilionoideae, tribe Hypocalypteae) as well as those associated with species in the genera *Virgilia*, *Podalyria* and *Cyclopia* (subfamily Papilionoideae, tribe Podalyrieae). A set of 69 rhizobial isolates were obtained from these hosts and shown to be capable of nodulating their original hosts, as well as the promiscuous legumes siratro (*Macroptilium atropurpureum*) and cowpea (*Vigna unguiculata*). To characterize and describe these bacteria, we utilized a polyphasic approach in which both the phenotypic and genotypic traits were studied. DNA sequence and phylogenetic analyses for four housekeeping genes (16S rRNA, *recA*, *rpoB* and *atpD*) placed all of the isolates into the genus *Burkholderia*. From these data we distinguished 25 distinct rhizobial lineages, which were also supported by a range of phenotypic characters (e.g., cultural growth properties and metabolic abilities). However, the majority of these lineages apparently lacked close relationships with known species of *Burkholderia*. The uniqueness of the isolates examined in this study was also reflected in the phylogenies for a nodulation gene, *nodA*, and a gene involved in nitrogen-fixation, *nifH*. The root-nodule bacteria examined in this study are thus unlike other *Burkholderia* species, not only in terms of their housekeeping genes and functions, but also with respect to the information located on their symbiotic loci. Taken together, our results suggest that the *Burkholderia* species associated with indigenous legumes have African origins and that both horizontal and vertical gene transfer determine the evolution of their symbiotic abilities.

Sustaining *Gnetum* L. in Africa through improved taxonomy and domestication

E.H. Biye, K. Balkwill and G.V. Cron (D03)

School of Animal, Plant and Environmental Sciences, University of the Witwatersrand, Private Bag 3, WITS, Johannesburg, South Africa, 2050

The African species of *Gnetum* L. (Gnetales) are lianas in the humid tropical forests of Central and West Africa. These gymnosperms are nutritionally and economically valuable and contribute greatly to poverty alleviation of the local people as they are widely used as a vegetable ('*eru*'). The African species of this genus are however difficult to distinguish from one another, and are threatened due to over-exploitation and face the risk of disappearing before they are sufficiently known. The names *Gnetum africanum* Welw. and *G. buchholzianum* Engl. have been interchangeably applied on specimens that apparently represent similar forms of *Gnetum* in Africa and this has resulted in taxonomic confusion. This study aimed to clarify the taxonomy of the African species of *Gnetum* and to promote sustainable utilization and conservation of the species. An evaluation of variation in taste among *Gnetum* populations was performed to identify the best tasting species for cultivation. Cluster analyses using morphological characters were carried out to investigate the variation among the African populations and to reveal additional characters to differentiate the species for both male and female plants. Molecular markers (AFLPS) were also used to clarify species boundaries. Reproductive structures were found to be most useful in distinguishing species and the applications of the names *Gnetum africanum* and *Gnetum buchholzianum* were clarified and two new African species have been described: *Gnetum interruptum* E.H. Biye *sp. nov.* and *Gnetum latispicum* E.H. Biye *sp. nov.* Taste analyses identified the leaves of *G. buchholzianum* and young leaves in general as best for an '*eru*' (*Gnetum*) meal. This has implications for cultivation and harvesting methods.

Promoter analysis of the *Eucalyptus grandis* secondary cell wall related *SND2* transcription factor gene

J. Botha^{a,b}, D. Pinard^{a,b}, N. Creux^{a,b}, S. Hussey^{a,b}, C. Maritz-Olivier^a, E. Mizrachi^{a,b} and A.A. Myburg^{a,b} (B03)

^a*Department of Genetics, ^bForestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, 0002, South Africa*

NAC and MYB transcription factors (TFs) are important components of the regulatory network controlling secondary cell wall (SCW) biosynthesis in plants. *Arabidopsis SND2*, *SND3* and *MYB103* are targets of the wood fibre SCW master regulators NST1 and SND1, and are thought to play a role in cellulose biosynthesis in fibre cell walls. Recent studies have shown that although much of this regulatory network is conserved between herbaceous and woody model species, there may be unique TFs or combinations of TFs that are important for regulating xylem deposition in wood fibres. It is hypothesised that there is an analogous regulatory network in the commercially important tree genus *Eucalyptus*, which remains to be characterised. In this study we aim to identify *cis*-regulatory sequences in the promoter regions of the *Eucalyptus grandis EgrSND2*, *EgrSND3* and *EgrMYB103* genes, to characterize the expression patterns of these promoters in *Arabidopsis* and to identify specific TF-promoter interactions of *EgrSND2* using a yeast-1-hybrid (Y1-H) approach. Using the

RSA tools suite (<http://rsat.ulb.ac.be>), several previously described and novel *cis*-elements were identified in all three *Eucalyptus* promoters. Promoter sequences (1.5 kb upstream of the translational start site) of the three transcription factors were amplified and fused to the β -glucuronidase (GUS) reporter gene for expression analysis in transgenic *Arabidopsis* plants. All three promoters were able to drive strong reporter gene expression, but not in a fibre-specific fashion as expected. This may be caused by a lack of a fibre-specific element or combination of elements in the promoter sequence investigated, or may be due to a functional or regulatory divergence of *Eucalyptus* and *Arabidopsis* promoters. Using a yeast one-hybrid analysis (Y1-H), we aim to detect direct interaction of several upstream SCW associated TFs with the promoter of *EgrSND2* in order to characterise its central role in the SCW transcriptional network.

Plant diversity of vegetation-over-ultramafic soil in Mount Bulanjao, Palawan Island, Philippines

R.R. Castillo^a and W.S.M. Gruèzo^b (D05)

^a*Center for Strategic Policy and Governance, Palawan State University, Puerto Princesa City 5300, Palawan Island, Philippines*

^b*Plant Biology Division, Institute of Biological Sciences, College of Arts & Sciences, University of the Philippines Los Baños, College 4031, Laguna, Philippines*

Mount Bulanjao is a low mountain (highest peak - 1036 masl) with extant natural vegetation-over-ultramafic soil in the Municipalities of Bataraza and Rizal, Palawan Island, Philippines. Results of the assessment and characterization of plant diversity and vegetation types of this mountain showed a relatively high diversity level of $H' = 4.17$. A total of 193 plant species belonging to 152 genera and 73 families were recorded from the study area based from a total of 33 (10 x 10 m) sample plots. The dominant plant forms are trees with 117 species followed by vines (30 species), herbs (25 species) and shrubs (21 species). In terms of importance value per habit type, *Alstonia macrophylla* (Apocynaceae), *Xanthostemon* sp. (Myrtaceae), *Dillenia monantha* (Dilleniaceae) ranked top for trees; *Dracaena angustifolia* (Agavaceae), *Pandanus luzoniensis* (Pandanaceae), *Melastoma* sp. (Melastomataceae) for shrubs; *Fimbristylis globulosa* (Cyperaceae) *Zingiber* sp. (Zingiberaceae), *Imperata cylindrica* (Poaceae) for herbs and *Dinochloa acutiflora* (Poaceae), *Calamus* sp. (Arecaceae) and *Lygodium circinatum* (Schizaeaceae) for climbers. Additional noteworthy plants of these vegetation-over-ultramafic soil are *Brackenridgea palustris* subsp. *foxworthyi* (Ochnaceae), *Eurycoma longifolia* (Simaroubaceae), *Gymnostoma rumphiana* (Casuarinaceae), *Jasminum aemulum* (Oleaceae), *Messersmidia argentea* (Boraginaceae), *Gymnostachyum pictum* (Acanthaceae), *Wrightia hanleyi* (Apocynaceae), *Scaevola micrantha* (Goodeniaceae), *Protium connarifolium* (Burseraceae) and *Phyllanthus balgoyii* (Phyllanthaceae). The results of the study showed that vegetation of Mt. Bulanjao is still intact, with types belonging to those of primary to secondary forests-over-ultramafic soil to a large extent, with grassland and brushland as well as riparian formation. All these vegetation types are still capable of providing excellent niches for the wildlife fauna of this endangered mountain ecosystem.

Plant composition and diversity of forests-over-ultramafic soil in Mt. Victoria Range, Palawan Island, Philippines

R.R. Castillo^a and W.S.M. Gruèzo^b (D06)

^{1a}Center for Strategic Policy and Governance, Palawan State University, Puerto Princesa City 5300, Palawan, Philippines ^bPlant Biology Division, Institute of Biological Sciences, College of Arts and Sciences, University of the Philippines Los Baños, College 4031, Laguna, Philippines

Mount Victoria is a medium-sized mountain range (with peaks ranging from 1500 to 1726 masl) straddling the Municipalities of Aborlan, Narra and Quezon, Palawan Island, Philippines. Generally, a greater portion of the mountain range harbors the largest nickel deposit in Palawan Island. A mixed broad leafed forest-over-ultramafic soil is present in this mountain range from sea level to 700 m elevation but with apparently two altitudinal zones of plant diversity. At lower elevations (0-300 masl) of this mountain, a forest zone is present which is composed of 141 tree, 32 herb, 27 vine and 22 shrub species, for a total of 222 species belonging to 174 genera and 75 families. The four most dominant species in this lower forest zone are *Xanthostemon speciosum*, *Tristaniopsis decorticata* (Myrtaceae); *Casuarina equisetifolia* (Casuarinaceae) and *Swintonia foxyworthyi* (Anacardiaceae). It had a relatively high diversity level of $H' = 4.25$. In the upper elevations (400 to 700 masl), a total of 81 species belonging to 67 genera and 46 families were recorded, composed of 50 tree, 13 vine, 11 shrub and 7 herb species. The dominant species in this zone are *Casuarina equisetifolia* (Casuarinaceae), *Tristaniopsis decorticata* (Myrtaceae), *Myristica guatteriaefolia* (Myristicaceae) and *Dinochloa acutiflora* (Poaceae). This upper forest zone had a diversity index value of $H' = 3.76$, which is slightly lower compared to the lower forest zone.

Maize Microarray Annotation Database

N. Coetzer^a, A.A. Myburg^b and D.K. Berger^c (B04)

^aACGT Bioinformatics and Computational Biology Unit, Department of Biochemistry, University of Pretoria, Private Bag X20, 0028, South Africa

^bDepartment of Genetics, ^c Department of Plant Science, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Private Bag X20, 0028, South Africa

Microarray technology is a cost-effective solution with established data analysis protocols for global gene expression profiling. The Agilent-016047 maize 44K microarray was custom-designed from EST sequences, but only reporter sequences with EST accession numbers are publicly available. The following information is lacking: (a) reporter - gene model match, (b) number of reporters per gene model, (c) potential for cross hybridization, (d) sense/antisense orientation of reporters, (e) position on B73 genome sequence (for eQTL studies), and (f) functional annotations of genes represented by reporters. To address this, we developed a strategy to annotate the Agilent-016047 maize microarray, and built a publicly accessible annotation database. Genomic annotation of the 42,034 reporters was based on BLASTN results of the 60-mer reporter sequences and their corresponding ESTs against the maize B73 RefGen v2 “Working Gene Set” (WGS) predicted transcripts and the genome sequence. The agreement between the EST, WGS transcript and gDNA BLASTN results were used to assign the reporters into six genomic annotation groups: (i) “annotation by sense

gene model” (23,668 reporters), (ii) “annotation by antisense gene model” (4,330); (iii) “annotation by gDNA” without a WGS transcript hit (1,549); (iv) “annotation by EST”, in which case the EST from which the reporter was designed, but not the reporter itself, has a WGS transcript hit (3,390); (v) “ambiguous annotation” (2,608); and (vi) “inconclusive annotation” (6,489). Functional annotations of reporters were obtained by BLASTX and Blast2GO analysis of corresponding WGS transcripts against GenBank. The annotations are available in the Maize Microarray Annotation Database (<http://MaizeArrayAnnot.bi.up.ac.za/>), as well as through a GBrowse annotation file that can be uploaded to the MaizeGDB genome browser as a custom track. This database will assist users of the Agilent-016047 maize microarray in (i) refining gene lists for global expression analysis, and (ii) confirming the annotation of candidate genes before functional studies.

Micro RNA expression in maize leaves and flowers

B.G. Crampton^a and S.L. Murray^{b,c} (B05)

^a*Department of Plant Science, Forestry and Agricultural Biotechnology Institute, University of Pretoria, Pretoria, South Africa, 0002*

^b*Centre for Proteomic and Genomic Research, P.O. Box 81 Observatory, Cape Town, 7945*

^c*Department of Molecular and Cell Biology, University of Cape Town, Rondebosch, 7700*

Micro RNAs are approximately 20-22 nucleotides in length, and act as master regulators of gene expression. Micro RNAs recognise complementary mRNAs and bind to them by base-pairing, thereby regulating expression of the mRNA, either by blocking translation or by cleavage and subsequent degradation of the mRNA. In order to investigate the role of microRNAs in maize development, we employed Exiqon miRCURY LNA arrays (www.exiqon.com) and profiled micro RNA expression in leaves and male and female flowers. These arrays contain probes for micro RNA genes listed in the database miRBase (www.miRBase.org), and are representative of micro RNAs identified in all eukaryotic species. Comparison of micro RNA profiles of leaves and male and female flowers indicated that nine micro RNA genes were up-regulated in both male and female flowers relative to leaves, whereas nine and 25 genes were up-regulated only in either in male or female flowers respectively. We obtained similar results for down-regulated micro RNA genes. Of particular interest is the up-regulation of zma-MIR166, and members of the zma-MIR171 family, both of which have previously been shown to play a role in flower development. This study emphasises the utility of microarrays in rapid identification of micro RNAs and in profiling micro RNA gene expression in plant tissues.

***Emilia* (Cass.) Cass. (Asteraceae, Senecioneae) in southern Africa**

G.V. Cron (D02)

School of Animal, Plant and Environmental Sciences, University of the Witwatersrand, Private Bag 3, WITS, Johannesburg, South Africa, 2050

Emilia (Cass.) Cass. (Asteraceae, Senecioneae) is a tropical herbaceous genus comprising about 100 species, most of them African. The mostly glabrous herbs are annual or perennial with variable leaf forms. Similarly, the capitula may be solitary or few to several in lax corymbs, radiate or discoid, and the florets range from white to pink, red, orange or rarely yellow. Chromosome numbers also vary considerably within the genus. Distinguishing

features include the ecalyculate capitula, elliptic-oblong cypselas with an obscure carpodium and a pappus of slender, persistent bristles. Twenty species of *Emilia* occur south of the DRC and Tanzania, including seven from southern Africa (Botswana, Namibia, South Africa and Swaziland). *Emilia hantamensis* from near Nieuwoudtville was thought to be the first record of the genus from the winter rainfall region of South Africa with a large disjunction between it and either *E. transvaalensis* (northern KwaZulu-Natal and Gauteng) or *E. ambifaria* (North West Province). It has large, yellow radiate capitula - quite distinct from the other discoid, purple or white-flowered *Emilia* species in South Africa. This study aimed to investigate whether *E. hantamensis* was more closely related to the other radiate members of the genus *Emilia* (the nearest being *E. discifolia* in Zimbabwe), or to the southern African discoid species. Phylogenies based on nuclear markers have revealed that *E. hantamensis* is not a member of *Emilia* at all, but a distinct genus closely related to *Bolandia*, *Mesogramma* and *Stilpnogyne*.

Socioeconomic gradients, garden design and plant diversity patterns in urban areas in the North-West Province and Gauteng

E. Davoren, S.S Cilliers and S.J Siebert (E04)

School of Environmental Sciences and Development, North-West University, Private Bag x6001, Potchefstroom, 2520, South Africa

In contrast to the main focus of studies in developed countries, home garden research over the last three decades in developing countries have been aimed primarily at either ethnobotanical documentation of the diversity and uses of garden species, or the promotion of home gardening for nutritional improvement of low-income groups. Similarly in South Africa the majority of studies have focused on the value of cultivated floras in homegardens of rural areas for household consumption and poverty alleviation, the cultivation of medicinal plants as a tool for biodiversity conservation and indigenous knowledge research. Between 2007 and 2010 several home garden projects have been and are still being conducted by the Urban Ecology Research Group at the North-West University. The overall objective of these studies was to develop a better understanding of the plant diversity of homegardens, their contribution to urban plant diversity, as well as their structure and function in a city, town, rural settlement and deep-rural area. The current study aims to compare garden design (traditional European approach versus a specific local ethnical approach, e.g. “tshimos”) and plant diversity with the mentioned home garden studies along a socioeconomic gradient. Socio-economic status is determined using variables such as monthly income, income generation, total number of residents per house, modes of transport and schooling status. The degree of urbanization surrounding the home gardens is also important and was quantified using fourteen demographic, physical and landscape metrics measures. The completed and ongoing studies provide a large dataset of home garden floras and the socioeconomic aspects of the rural, peri-urban and urban environments. Results have indicated correlations between species diversity (total, native, alien, medicinal, ornamental, vegetable, fruit trees), type of garden design, socioeconomic status and degree of urbanization.

De novo* assembly of the expressed gene catalog of *Equisetum ramosissimum

A. dos Santos^a, E. Mizrachi^b, C.A. Hefer^a and A.A. Myburg^b (B06)

^a*Bioinformatics and Computational Biology Unit, Department of Biochemistry, University of Pretoria, South Africa, 0002*^b*Department of Genetics, University of Pretoria, Pretoria, South Africa, 0002*

Horsetails (*Equisetum* spp.) represent some of the earliest and most dominant vascular plants of the early carboniferous. They diverged more than 360 mya from the rest of the ferns and have remained relatively unchanged since. In addition to their important phylogenetic position in the evolution of plant vasculature, horsetails possess unique cell wall properties that could provide insight into the evolution of cell walls in plants. High-throughput transcriptome sequencing using Illumina mRNA-Seq presents the opportunity to explore entire transcriptomes in a high-throughput and relatively low cost manner. However, the *de novo* assembly of transcript-derived short-read data presents new challenges, including variable coverage across assembled contigs, reads derived from alternatively spliced transcripts, as well as sampling bias of highly expressed genes. A gene catalog of *Equisetum ramosissimum* was assembled from ~220 million, 80 bp paired-end Illumina high quality reads (8.8 Gbp) generated from stem- and sporangiophore-derived cDNA. The depth of sequencing and high quality of the reads offered an opportunity to gain insight into the efficiency of some of the available software for the complex task of assembling short-read data, while simultaneously providing a resource to answer further biological questions. Trans-ABYSS, Trinity, Oases and Velvet assemblers were compared using standard sequence assembly metrics including N50, L50, median contig length, proportion of incorporated bases and total number of assembled contigs. Assembled gene catalogs were also evaluated according to annotation, such as predicted coding sequence and significant blast hits to other sequenced plant transcriptomes. Oases was found to produce the highest quality assembly in terms of contig contiguity and total amount of unique transcriptome assembled when compared to the other assemblers. The final assembly was annotated using BLAST2GO and InterProScan. The results include a comprehensive expressed gene catalog of *Equisetum ramosissimum* comprising of 26,318 non-redundant contigs with a median contig length of 1027 bp and total size of 34 Mbp, which could shed light on many aspects of vascular plant evolution, as well as the unique biology of horsetails.

Potential of two South African plants belonging to the *Greyiaceae* family for the treatment of skin hyper-pigmentation

M. de Canha and N. Lall (M02)

Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002

Melanin biosynthesis is regulated by the tyrosinase enzyme, which is responsible for the catalysis of the rate limiting step in the melanin biosynthetic pathway. In humans, melanin biosynthesis takes place within the skin melanocyte cells. Ten percent propane-1, 2-diol (PG) leaf extracts of two plants (S1 and S2) belonging to the *Greyiaceae* family were screened for anti-tyrosinase activity, mutagenicity, stability and irritancy. Ten percent PG: deionised water extracts for S1 and S2 showed approximately 98% inhibition of the tyrosinase enzyme. Samples were compared to that of known inhibitor Kojic Acid which exhibited an IC₅₀ of 2.841 µg/ml. Stability tests were performed at a corporate firm, Chemgrit

Cosmetics, over a 3 month period and results recorded at 1, 2, 4, 8, and 12 week intervals. Stability was compared to that of a control sample and both S1 and S2 were found to be stable at room temperature. The samples also showed no mutagenic effects when *Salmonella typhimurium* strains TA98 and TA100 were used during the bioassay. A clinical study done by private firm Future Cosmetics concluded that both S1 and S2 were non-irritants with an irritant potential of -34.21% and -37.90% for S1 and S2 respectively. S1 showed a large potential for the development of a skin even toner when incorporated into a formulation. The findings of the present study indicate that sample 1 has the potential to be used as effective and alternative treatments for the skin hyper-pigmentation disorder.

The peatlands of the Highlands of Lesotho – a unique aquatic system under threat

P.J. Du Preez^a, L.R. Brown^b and L. Seipethlo^a (E01)

^a*Department of Plant Sciences University of the Free State*

^b*Applied Behavioural Ecology & Ecosystem Research Unit, University of South Africa*

Numerous hill-slope seeps and valley bottom peatlands occur in the high-altitude headwaters of the Senqu River in Lesotho. These peatlands deliver important ecosystem services such as water retention, purification, and slow release of water as well as sustaining a unique plant and animal biodiversity. Due to the high altitudes (mostly above 2 000m) a slow process of peat formation is taking place. On these peatlands a very unique plant community exists which supports several endemic plant species. A communal grazing system exists in the highlands of Lesotho and these peatlands are one of the main sources of pasture for the domestic stock. In order to describe the vegetation and species composition of these peatlands, sampling plots were randomly placed within the peatlands. All species present were recorded and their cover abundance assessed using the Braun-Blanquet cover abundance scale. Environmental data included slope, aspect, grazing, and erosion while the average height of the plants was also estimated. The data analysis was done by using TWINSpan classification which was refined by using Braun-Blanquet procedures. An ordination of the data was also done by using the CANOCO ordination techniques. The results reveal that the thurfur (elevated humps of peat) on the peatland supports a different plant species composition. Environmental factors such as wetland hydrology, drainage, erosion, and biotic impacts play important roles in the distribution of the wetland associated species. Several impacts are threatening to destroy these peatlands and drastic measures to conserve these unique ecosystems need to be put in place.

Isolation and identification of compounds from an extract of *Strophanthus speciosus* (N. B. Ward. & Harv.) Reber., with emphasis on cardiac glycosides

S. Emamzadeh-Yazdi^a, A. Hussein^b, G. Prinsloo^c and Q. Kritzinger^a (M03)

^a Department of Plant Science, University of Pretoria, Pretoria, South Africa 0002

^b Department of Chemistry of Medicinal Plants. National Research Centre, Dokki, Cairo, Egypt

^c Department of Agriculture and Animal Health, University of South Africa (UNISA), Florida Campus, Private bag x 6, Florida, 1710

The discovery of active compounds in medicinal plants plays a strategic role in the phytochemical investigation of crude plant extracts. According to literature, plants containing cardiac glycosides (CG) show potential for the discovery of more effective new drugs in the treatment of viral infections. The aim of this study is to investigate the antiviral, antibacterial and cytotoxic activities of South African plants containing CG. Six plants were selected for the study of which none of them exhibited anti-herpes simplex virus activity, but only general toxicity. Chromatographic methods were used to identify the compounds and determine activity of the extracts on two bacteria, namely *Escherichia coli* (gram negative) and *Enterococcus faecalis* (gram positive). The results showed promising activity of *Strophanthus speciosus* fruit extract against gram positive bacteria. Isolation and purification of the ethanol extract of fresh fruit of *S. speciosus* was carried out by column chromatography and crystallization. Three compounds including a cardiac glycoside were isolated from *S. speciosus*. Structural determination of the pure compounds was based on physico-chemical properties and various spectral data analysis such as NMR was done in order to elucidate the structures of the compounds. The names of the compounds are yet to be confirmed.

Optimizing germination of *Coleonema album* seeds

O.O. Fajinmi, M.G. Kulkarni, J.F. Finnie and J. van Staden (B07)

Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal Pietermaritzburg, Private Bag X01, Scottsville 3209, South Africa

Coleonema album is a decorative shrub with dainty, small white flowers and a sweet, honey smell. It's an ideal garden plant and an interesting specimen for floral arrangements. Apart from its horticultural value, a wide range of pharmacological activities such as antithrombotic, antiplatelet, vasolidatory, antibacterial, antifungal, antimycobacterial, antioxidant, antimicrobial, phytoalexin, and analgesic activities are attributed to *C. album*. Aromatherapy properties of *C. album* includes: antiseptic, deodorant, insecticide, sedative, restorative, diuretic and febrifuge activities. *Coleonema album* is gaining popularity in South African herbal medicine as a tincture, known as Immunat, is now marketed commercially. This has led to an increase in demand for *C. album* in the traditional medicinal markets. This will cause a threat to the wild population as a result of its intensive collection. Increasing demand of a medicinal plant such as *C. album* can only be met by developing effective propagation protocols. Unfortunately, there is very limited information available on the propagation requirements of this highly used medicinal plant. This necessitates research into the seed germination requirements and general physiological growth requirement *in vitro* and *ex situ* as a crucial step to ensure its survival. The influence of light, temperature,

stratification (cold and warm), smoke-water, KAR (Karrakinolide 1), plant growth regulators (kinetin, GA₃, KNO₃, IAA, NAA and IBA) on seed germination of *C. album* was investigated in this study. Seeds incubated under cold temperatures of 10°C and 15°C showed favourable germination. No germination was recorded at high temperatures (20, 25, 30 and 35°C) even after six weeks, unless they were transferred to cold conditions. Seeds exposed to continuous dark conditions showed equal responses as seeds exposed to alternating light conditions. Seeds with and without endosperm readily imbibed water which rules out physical dormancy and indicates physiological dormancy. Cold temperature conditions play a significant role in germination of *C. album* seeds.

The application of salicylic acid to induce tolerance against *Chrysosporthe austroafricana* in *Eucalyptus grandis* clone ZG14

L. Ferreira, S. Naidoo and A.A. Myburg (B08)

Department of Genetics, University of Pretoria, Forestry and Agricultural Biotechnology Institute (FABI), Pretoria, South Africa 0002

Eucalyptus is a globally important hardwood and fibre crop that provides high quality raw material for industry. One of the most important pathogens of this perennial is *Chrysosporthe austroafricana*, a fungus that is indigenous to South Africa and causes stem canker. This plant-pathogen interaction is used to investigate the activation of defense signalling pathways and the systemic induction of defense mechanisms in *Eucalyptus*. Salicylic acid regulates one such defense signalling pathway resulting in broad spectrum long-lasting resistance throughout the entire plant (systemic acquired resistance) that can be detected through the enhanced expression of particular marker genes such as pathogenesis-related protein 2 (*EgPR2*). The aim of this study is to determine whether the exogenous application of SA will lower the susceptibility of *E. grandis* ZG14 clones to *C. austroafricana* infection. The experimental procedure involved the treatment of the susceptible ZG14 clones with SA 24 hours before inoculations. As controls, ZG14 clones treated with buffer and tolerant TAG5 clones were included in the experiment. In addition to measuring the lesion lengths of the plants, stem tissue surrounding the inoculation sites were harvested at three different time points. Following RNA isolation and purification from the harvested samples, gene expression profiling was conducted using reverse transcriptase quantitative PCR (RT-qPCR) to determine the quantitative expression of *EgPR2*. It is expected that a decrease in lesion size and accumulation of *EgPR2* transcripts, indicative of the SA pathway and SAR, will be observed in the treated ZG14 clones before and after infection with the pathogen. This study will indicate whether the SA signalling pathway is important for defense against *Chrysosporthe austroafricana*. It will also qualify the use of SA to improve the tolerance of *Eucalyptus* against this fungal pathogen.

The identification of desiccation-responsive heat-stable proteins from the fronds of the desiccation tolerant form of the resurrection fern *Mohria caffrorum*

A.T. Gill, J.M. Farrant and M.S. Rafudeen (B09)

Department of Molecular and Cell Biology, University of Cape Town, Private Bag X3, Rondebosch, 7701, South Africa.

Mohria caffrorum (L.) Desv has the unusual characteristic in that it has a seasonal desiccation tolerance (DT), being tolerant in the dry and desiccation-sensitive (DS) in the rainy season. DT fronds acquire protective mechanisms during drying that are similar to those reported for resurrection angiosperms; however, there is limited information on mechanisms of vegetative desiccation tolerance in ferns. Preliminary one dimensional sodium dodecyl sulphate - polyacrylamide gel electrophoresis (1-D SDS PAGE) showed that the DT form produced several new heat-stable proteins, which were not present in the DS form upon dehydration. The current study was undertaken to further characterise the heat-stable proteome of this species by use of 2-DGE in order to identify proteins that might play a protective role against dehydration. Three proteins were detected only in the dehydrated samples and these proteins were referred to as 'de novo' proteins. All three of these protein spots were identified by LC-MS/MS with high significance to known proteins. Two of the protein spots that were detected as 'de novo' were identified by LC-MS/MS as the chloroplast enzyme RuBisCo (ribulose-1,5-bisphosphate carboxylase/oxygenase) and the third 'de novo' protein was identified as an Oxidoreductase. Additionally, five protein spots that were up-regulated (paired t-test, $P < 0.05$) during drying were identified by LC-MS/MS. Only two of the five proteins were matched to known proteins. The two up-regulated proteins were identified as the antioxidant enzyme, superoxide dismutase (SOD) and the molecular chaperone, heat shock protein 70 (HSP70). The functional physiological and biochemical roles of these proteins as protectants in desiccation tolerance are being investigated.

Identification of transposable elements in the pitch canker fungus, *Fusarium circinatum*

J. Grové^a, M.P.A.Coetzee^b and E. T. Steenkamp^a (D04)

Departments of Microbiology and Plant Pathology^a and Genetics^b, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, South Africa

Fusarium circinatum is an economically important ascomycete that causes pitch canker disease of pine trees. Transposable elements (TEs) have been identified in some *Fusarium* species where they may comprise a significant portion of the genomic content. The aim of this study was to identify TEs in *F. circinatum* using the available genome sequence and to evaluate their potential contribution to inter- and intra-specific diversity in *Fusarium*. The *de novo* repeat-finding program, RepeatScout, was used together with BLAST to identify a set of 93 repetitive sequences in *F. circinatum*. Of these, eight were selected for further analyses as they displayed similarity to known TEs or the characteristic structural features associated with these elements. Six of the repetitive sequences represent retrotransposons and two non-autonomous DNA transposons. By making use of similarity searches with known proteins encoded by TEs, a further 58 contig regions with enzyme homology were identified in the *F. circinatum* genome, although only two harboured open reading frames that were not

interrupted by nonsense and frame-shift substitutions. Overall it thus seems that the putative TEs in *F. circinatum* are fragmented and non-functional, which suggests that these elements are in the degradation phase of their lifecycle within the genome. Additionally, at least one of the eight putative TEs in *F. circinatum* appeared to be present in high copy numbers in the closely related *F. verticillioides* and *F. fujikuroi*, but not in the more distantly related members of this genus. PCR-based assays indicated that one of the putative DNA transposons varied considerably in distribution and density among different isolates of each of *F. circinatum* and *F. verticillioides*. These data thus suggests that TEs represent a significant source of intra- and well as inter-specific variability and holds potential for use in epidemiological studies of *F. circinatum* and its close relatives.

Invasive *Crotalaria agatiflora* Schweinf.–ecology and distribution in South Africa

T.P. Jaca (E05)

Early Detection and Rapid Response programme, South African National Biodiversity Institute, Private bag X101, Pretoria 0001, South Africa

Crotalaria agatiflora (canary bird bush) is an evergreen shrub or small tree up to 1-10 m tall; mostly glabrous, with young shoots softly hairy. The leaves are grayish-green, 3-foliolate; leaflets on a short stalk (petiole), petiole mostly longer than leaflets, glabrous to densely hairy. Flowers are lemon-yellow or greenish-yellow, with a projecting greenish or purple beak in many-flowered racemes to 400 mm long. Canary bird bush Native to Tropical East Africa and North-East Africa (Tanzania and Kenya) and was probably introduced in South Africa as an ornamental. The earliest record of this species in South Africa is of a specimen in the Pretoria National Herbarium collected in Johannesburg area at the Railway Horticulture Garden in 1921 and the earliest record of its establishment in the wild is from Rustenburg to Brits. The species is a proposed category 1a in NEMBA regulation. Although no studies have focused on its effects on natural ecosystems canary bird bush may affect the ecology of invaded areas in several ways, for example the enhancement of nitrogen levels in the soil. It threatens watercourses in grassland and savanna biomes; potentially invasive on forest margins and also occupying cleared grassy areas. The distribution of canary bird bush in South Africa was mapped using records from South African Plant Invaders Atlas database, herbarium records and field records. Results indicated that canary bird bush distribution is mostly along road sides and the species has spread into natural areas for example, Colbyn wetland, Wonderboom Nature reserve and Faerie Glen Nature Reserve.

A guide to plant families of southern Africa

M. Koekemoer^a, H.M. Steyn^a and S.P. Bester^{a,b} (D07)

^a*National Herbarium, South African National Biodiversity Institute, Private Bag X101, Pretoria 0001*

^b*School of Environmental Sciences and Development, North-West University, Private Bag X6001, Potchefstroom 2520*

Knowledge of plant families allows one to place unknown plants into broad categories. Once the familial identity is established one can focus on the smaller divisions of genera and eventually species. Southern Africa has a total of 225 plant families. A project to illustrate the diagnostic characteristics of the 51 largest families is currently underway—these families represent ±90% of the total flowering plants for this area. The publication will assist scholars, students, amateurs and professionals to identify these plant families. The aim is to educate the public on the beauty and diversity of our fascinating flora and to expand the base of knowledge of the plant families. If a person can attach a name to a plant, it immediately has more meaning and scientific value, fostering appreciation. It is then easier to communicate about the plant and to find more related information on it. The information presented is uncomplicated and designed for users at different levels of expertise. The focus will be on having more images than text. This product will make the complex process of classification and identification more ‘friendly’ and understandable to scientists and non-scientists alike.

Comparison between grey leaf spot lesion area and *Cercospora zeina* DNA content within maize leaves

J.N. Korsman^a, T.G. Schmidt^a, B. Meisel^b, F.J. Kloppers^c, B.G. Crampton^a and D.K. Berger^a (B10)

^a*Department of Plant Science, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa 0002*

^b*MONSANTO, Petit, South Africa.*

^c*PANNAR SEED (Pty) Ltd, Greytown, South Africa.*

Grey Leaf Spot (GLS) is a fungal leaf disease of maize (*Zea mays*) that can cause severe yield loss. It is caused by two fungal species, *Cercospora zeina* and *Cercospora zeae-maydis*, *C. zeina* being responsible for GLS in Africa. The disease is characterised by elongated tan to grey lesions spreading within the bounds of the parallel leaf veins. We hypothesised that lesion area on a leaf corresponds with the fungal biomass within the leaf. In order to test this, we used an established *in planta* qPCR assay to quantify the amount of *C. zeina* DNA in infected resistant and susceptible leaf tissue. These results were compared with percentage lesion area of the same infected leaves. In this regard, leaves from nine biological replicates of two maize lines differing in susceptibility to GLS were photographed and the percentage lesion area was calculated. The amount of *Cercospora* DNA detected by the qPCR assay correlated well with calculated lesion area (Pearson correlation coefficient = 0.8). The amount of fungal DNA within the leaves of the susceptible and resistant lines were significantly different (t-test, $p < 0.001$). This indicates that the biomass of *C. zeina* corresponds to lesion area in both susceptible and resistant lines.

Diversity array analysis of *Methyltransferase* (MET1) mutants of tomato

B. Kuriakose^a, E. Viljoen^a, A. Killian^b, F. Carriero^c, S. Minoia^d, A. Bendahmane^d and D.K. Berger^a (B11)

^aDepartment of Plant Science, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa.

^bDiversity Arrays Ltd, Yarralumla, Canberra, ACT, Australia.

^cMetapontum Agrobios, SS Jonica, Metaponto (MT), Italy

^dUnité de Recherche en Génomique Végétale, UMR INRA-CNRS, Evry Cedex, France

DNA Methylation plays important roles in growth and development of organisms. In plants they are thought to influence many processes ranging from development to fruit ripening. Methylation of genes and promoter sequences are often related to the silencing of gene expression. DNA methyltransferases are enzymes catalysing the addition of methyl groups during DNA replication and their subsequent maintenance. There are different classes of methyltransferases and their mutations affect normal plant growth and function. MET1 is a maintenance methyltransferase enzyme in plants. A population of 7 TILLING mutants of the Met1 gene in *Solanum lycopersicum* cv. M82 were analysed in the present study. Using bioinformatics tools the effect of each point mutation on protein function were predicted. DNA from the different mutant lines was extracted and Diversity Array analysis was carried out on the mutants to understand the global reduction in methylation due to the effect of mutations. Reduction in methylation of genomic DNA was observed in mutant lines when compared to wild type M82. Specific mutations were further validated using PCR of specific clones selected from a diversity array experiment.

Be warned! Invasive succulent, *Furcraea foetida* (Mauritius hemp) on the loose!

R. Lalla (E06)

Early Detection and Rapid Response programme (EDRR)*, South African National Biodiversity Institute (SANBI)

Furcraea foetida (= *F. gigantea*) is a succulent, with sword-shaped, light green leaves arranged in a basal rosette. Belonging to the Agavaceae family, this species was widely cultivated for fibre in the past. However, in more recent years *F. foetida* has been labelled as a “garden escapee” in some countries, as its invasive potential is being recognized. *F. foetida* thrives in cliff-like habitats, and is tolerant of very shallow soil. This species reproduces prolifically, with approximately 1250 bulbils (vegetative reproductive structures) produced per flowering stalk. Carpets of offspring which result from the mass production of bulbils, transforms into impenetrable thickets over time. *F. foetida* poses a threat to indigenous vegetation, and its invasive potential must be investigated. In 2009, *F. foetida* was proposed to the EDRR programme as an EMERGING Invasive Alien Plant (IAP) in KZN, and at the time, not much was known about the distribution of this species in the country. Efforts of the EDRR programme have led to a significant increase in the number of known *F. foetida* localities in KZN, and in other provinces in South Africa. This poster highlights the biology and ecology of *F. foetida*, identifying features of the species, and current known South

African localities, as well as challenges that have been experienced in dealing with this species.

*The EDRR programme is funded by Working for Water

Single nucleotide polymorphism (SNP) diversity in xylem and leaf expressed cellulose synthase genes of *Eucalyptus grandis*

B. Lombard^a, C. Hefer^{a,b}, A. van der Merwe^a, E. Mizrachi^a and A. A. Myburg^a (B12)

^a*Department of Genetics, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa, 0002*

^b*Bioinformatics and Computational Biology Unit, Department of Biochemistry, University of Pretoria, South Africa, 0002*

Eucalyptus grandis is a commercially important tree species with excellent wood properties for pulp and paper production. Recent availability of the *Eucalyptus grandis* genome sequence provides possibilities for tree improvement by means of marker-assisted breeding. Genetic variant discovery is important for identifying links between genotypic and phenotypic variation in tree populations. This study focussed on single nucleotide polymorphism (SNP) diversity in xylem and leaf expressed cellulose synthase (*CesA*) genes of *Eucalyptus grandis*. Illumina mRNA-seq reads from immature xylem and young leaf tissues were generated from 30 *E. grandis* individuals representing 28 families in a breeding population. The RNA sequences were aligned to the *E. grandis* reference genome using TopHat software and visualized using SAMtools. Data for eleven characterized *Eucalyptus CesA* genes was extracted and analysed to determine the distribution of SNPs in primary and secondary cell wall related *CesA* genes. Coverage up to 4160X as well as minor allele frequency (MAF) up to 0.29 were detected for the *CesA* genes. SNPs were only called with a minimum SNP quality of 60, minimum coverage of 8X at the SNP position and minor allele coverage of 4X. Synonymous or non-synonymous amino acid changes associated with coding SNPs were recorded. An interesting finding is that it appears that there is far greater diversity of alleles expressed in primary cell wall related genes than secondary cell wall related genes of the same thirty individuals. It was also found that SNP density in primary and secondary cell wall related *CesA* genes ranged from 0.6 to 13.3 and 1.2 to 10.3 SNPs per 100 bp. Methods from this study will be applied to the rest of the transcriptome sequence to describe genetic diversity in protein coding genes of this breeding population, as well as for the development of SNP markers for future molecular breeding efforts.

South Africa's succulents under threat

L.E. Makwavela and J.E. Victor (E07)

National Herbarium, South African National Biodiversity Institute, Private Bag X101, Pretoria, 0001 South Africa.

The Red List of South Africa is a useful tool for assessing conservation priorities, and furthermore, provides insight into areas where information is lacking and future research thrusts are needed. Succulents are prone to threats from overharvesting, and also have an unusually high number of species classified as Data Deficient due to the lack of information. Our analyses show that in South Africa, the Asphodelaceae are the most threatened of the succulent plant families and are therefore most in need of conservation attention.

Mesembryanthemaceae have the highest proportion of (and highest number of) Data Deficient species and are therefore most in need of future research efforts. It is concluded that succulents are particularly vulnerable to threats because of their relatively slow growth compared with other herbaceous plants, coupled with their desirability in the horticultural industry.

Variations in the epidermal structure of *Acacia tortilis* Hayne. grown in different climatic conditions

P.S. Mashile, R.B. Bhat and M.P. Tshisikhawe (D09)

Department of Botany, University of Venda, Thohoyandou 0950

Leaf epidermal structure of *Acacia tortilis* from nine different sites was investigated. The leaves of *A. tortilis* were found to be hypostomatic. The epidermal cell showed a polygonal structure with anticlinal walls either straight or curved. Trichomes were non-glandular, unicellular and conical in shape. Paracytic, anisocytic and stoma with a single subsidiary cell were the stomatal types observed. The frequency of epidermal cells was higher as compared to that of guard cells. The stomatal index and frequency of stomata and epidermal cells were also compared and recorded. Variations in the epidermal structure of leaves and morphological structure of the plant from different habitats were also recorded. Further anatomical investigations are in progress.

Ecological, morphological and ethnobotanical assessment of *Acacia karroo* and *Acacia tortilis* at Nylsvley Nature Reserve, Limpopo Province South Africa

P. Masiagwala^a, R.B. Bhat^a and M.H. Ligavha-Mbelengwa^a (E10)

^aDepartment of Botany, University of Venda, Thohoyandou 0950

The genus *Acacia* is the most significant species of the family Fabaceae. This family was first described by Linnaeus in 1773. *Acacia karroo* also known as the sweet thorn, is a species of *Acacia*, native to southern Africa from southern Angola east to Mozambique, and south to South Africa. This species is widespread in clayey water-logged areas of the Nylsvley Nature Reserve. It is a pioneer species and generally invades disturbed habitats. It grows in a range of soils from sandy to heavy clayey ones. In Nylsvley Nature Reserve it occurs in a few numbers in sandy soils and is common in fertile clayey waterlogged areas. *Acacia karroo* is often associated with symbiotic *Rhizobium* sp. that fixes nitrogen. It is therefore important in the nutrient dynamics of the nature reserve. This plant is of economic importance as fodder for the animals and it has a number of medicinal and other commercial values. *Acacia tortilis* is drought resistant, can tolerate strong salinity and seasonal waterlogging and generally forms open forests in pure stands or mixed stands in these dry lands. It is also adapted to an environment with little and unreliable rainfall, and under these constraints it is as well adapted to anthropogenic pressures as it is the sole source of animal fodder, firewood and shelter for nomads. Combined pressures and their variation are reflected in its growth forms. The aim of the project was to investigate the ecological, morphological and ethnobotanical assessment of *Acacia karroo* and *Acacia tortilis* at Nysvley Nature Reserve, Limpopo province. Data were collected from different habitats of the Nysvley

Nature Reserve such as water logged, sandy and loamy soil areas. Canopy diameter was measured by using the measuring tape, tree height was taken using the height rod, and the trunk diameter by using diameter tape. Morphological variations have been observed within species growing under different habitats. Further ecological and ethnobotanical investigations are in progress.

Medicinal plants of the Blouberg Mountain, Limpopo Province, South Africa

K.M. Mathibela, B.A.Egan, H.J. Du Plessis and M.J. Potgieter (M04)

University of Limpopo Department of Biodiversity, Private Bag x1106, Sovenga, 0727

Blouberg Mountain, Limpopo, is a centre of plant endemism upon which a large rural community depends for numerous natural resources. The mountain itself enjoys no formal protection (Blouberg and Maleboch Reserves protect only a small area on the northern slopes) and few studies have examined the extent to which it provides such services. This study investigates the medicinal plant species used by traditional healers on the mountain in terms of species collected, manner of harvesting and where they are collected. Traditional healers from villages surrounding the mountain were interviewed during questionnaire surveys and participant observation walks. Data was gathered on general and personal information, species collected, collection methods, patient consultation, legislation and conservation. Healers were also interviewed in their consultation rooms. Plant specimens were collected and identified and the conservation status of the species investigated. This was compared to the healers' perceptions of the rarity of the plants. Forty one species of plants were collected from the mountain and 37 species were noted from consulting rooms. None of the healers interviewed had any knowledge of the most important plant conservation legislation in the province, or of the existence of a Red Data list for plants. Most interviewees (71 %) however, felt positive about the proposal of harvesting restrictions. The medicinal plants *Warburgia salutaris*, *Hypoxis hemerocallidea* and *Boophane disticha* are mentioned in the Red Data List (2011) as endangered and declining but are not perceived as rare or declining by the healers. Conversely, a number of least concern plants are seen as rare by the healers. Conservation recommendations were developed.

The use of GIS in documenting medicinal plants of Blouberg mountain, Limpopo Province, South Africa (M05)

K.M. Mathibela, B.A. Egan, H.J. du Plessis and M.J. Potgieter

University of Limpopo Department of Biodiversity, Private Bag x1106, Sovenga, 0727.

A Geographic Information System (GIS) is a tool that combines statistics with geographic locations to create informative maps. The use of GIS technology to document the indigenous knowledge of an area has become more prevalent, however, the analysis of medicinal plant collection pathways (tracks) using this method is ground breaking. This study examined the utility of a readily available and easily implemented application of Geographic Information Systems for describing and analyzing the collection tracks of traditional healers with respect to where the most important collecting sites are located. For the first time, healers' pathways was digitized, geo-referenced and linked to other information on the

medicinal plants around Blouberg Mountain, making this information easier to analyse and access. Sixteen of the most used collection tracks were investigated and co-ordinates taken for the beginning of each track, plant collection per track, change of a track and ending of each track. Forty one medicinal plant species were documented from the 16 tracks surveyed. The collection tracks were mapped and overlaid with topographic and demographic information as well as vegetation types of the Blouberg Mountain. Areas of high use or areas impacting on sensitive vegetation types are clearly evident from the map and can be targeted for immediate remedial action. This information, particularly that regarding species of medicinal plants utilised and most impacted areas of the mountain, will be invaluable to Limpopo conservation officials who are in the process of implementing a conservation plan for the province.

Biodiversity of *Ceratocystis* species infecting wounds on elephant-damaged trees in the Kruger National Park

M. Mbenoun^a, M.J. Wingfield^a, B.A.D. Begoude^b, B.D. Wingfield^a and J. Roux^a (D08)

^a*DST/NRF Centre of Excellence in Tree Health Biotechnology (CTHB), Department of Microbiology and Plant Pathology, Forestry and Agricultural Biotechnology Institute (FABI), Private Bag X20, University of Pretoria, Hatfield, 0028, South Africa*

^b*Institute of Agricultural Research for Development (IRAD), Nkolbisson, B. P. 2067, Yaounde, Cameroon*

It is now widely accepted that fungi represent a major component of the diversity of life on Earth. Their critical role in creating and maintaining biodiversity and in the functioning of terrestrial ecosystems is also well established. Yet, our knowledge of fungal biodiversity remains very limited, the more so when considering the community of micro-fungi in natural ecosystems in Africa. The genus *Ceratocystis* includes important pathogens of mostly woody plants. These micro-fungi exist in symbiotic associations with insects, relying on morphological and semiochemical adaptations to facilitate these interactions. *Ceratocystis* spp. have also received increasing attention globally because of the emergence of serious diseases that they cause in commercial forestry plantations and native forests. Studies in South Africa have suggested the existence of a substantial undiscovered biodiversity of *Ceratocystis* species in natural ecosystems in the country. In this study we investigated the occurrence and diversity of *Ceratocystis* species on elephant-induced tree wounds on trees in the Kruger National Park. Surveys conducted in the park between 2009 and 2010 resulted in the collection of 298 *Ceratocystis* isolates, obtained from 25 tree species in 10 plant families. Isolates were characterized using DNA multigene phylogenies and diversity indices and estimators were calculated for the entire park as well as for various ecozones within the park. An unexpectedly high diversity of *Ceratocystis* was discovered, both at genetic and species levels. We recovered 9 species in 3 *Ceratocystis* lineages and at least 50 distinct haplotypes. Four of the species represent previously unknown taxa. Although there were some differences in species abundance and occurrence between the various ecozones, similarity indices did not show significant departure from complete evenness. This is an indication that factors other than the host species composition determine the distribution and prevalence of these fungi in natural ecosystems.

Grafting method to improve vine production of Orange Fleshed Sweet Potato cultivar 'Resisto' under water stressed conditions

H.C. Mihiretu^a, E. Du Toit^a, J.M. Steyn^a and S.M. Laurie^b (E08)

^a *Department of Plant Production and Soil Science, University of Pretoria, Pretoria, South Africa 0002*

^b *ARC-Roodeplaat, Vegetable and Ornamental Plant Institute, Private Bag X293, Pretoria, 0001, South Africa*

Orange fleshed sweet potato (*Ipomoea batatas* L.) (OFSP) is a rich and cheap source of pro-vitamin A β -carotene. Vitamin A deficiency, which leads to night blindness and infant mortality, can be combated by dietary intervention using OFSP. Shortage of planting material caused by a prolonged dry season is one of the main constraints of OFSP production in Africa. Therefore, this research was conducted to assess the vine production potential of drought sensitive OFSP cv. 'Resisto' (R) grafted onto drought tolerant creamy fleshed sweet potato cvs. 'Tanzania' (T) and 'Zapalo' (Z) under different water stress conditions. Histological analysis conducted to determine the graft compatibility of scion (R) and root stock (T and Z) revealed differentiation of a new vascular cambium on the 10th day after grafting (DAG). By the 23rd DAG the presence of newly restored vascular tissue was observed. A two factor field experiment was conducted to compare three planting materials ('Resisto cutting' (R), 'Resisto x Tanzania' (RT) and 'Resisto x Zapalo' (RZ)) at three deficit irrigation levels (30%, 60% and 100% of measured deficit refilled). RT gave significantly ($\alpha < 5\%$) higher above-ground biomass yield (14.1tha⁻¹) at 30% deficit irrigation. Similarly, RT at 30% deficit irrigation showed significantly ($\alpha < 5\%$) higher water use efficiency (WUE) for above-ground biomass (179.1kgha⁻¹mm⁻¹) and cutting number (7091cuttings ha⁻¹mm⁻¹). Application of drought sensitivity and tolerance indices on vine yield showed that among the three planting materials used, RT was the most productive planting material at 30% deficit irrigation level. Generally, grafting improved vine productivity of OFSP cultivars under low water conditions. This study introduced the use of grafting techniques as a novel method to maintain OFSP vine over prolonged drought conditions.

Comparative anatomy of the galls and equivalent healthy plant organs in *Terminalia sericea* (Combretaceae)

T.E Mphephu and L.I. Ramovha (D10)

Department of Botany, University of Venda, private bag x5050 Thohoyandou, South Africa 0950

Various types of galls and the equivalent healthy plant organs of *Terminalia sericea* were collected from different areas of Limpopo. Leaf, young stem, round axial and long axial galls were mostly recorded in seedlings and young trees whereas flower and seed galls were prominent on adults trees. Collected samples were fixed in 75% FAA. Sections were prepared by hand and studied under light microscope. Digital images were taken by MR5 axio vision microscope for analysis. The main objective of the study was to establish the cause of the galls and resultant changes induced by the gall-forming agent to the associated plant tissues. Larvae of various wasps and moth species were found in long axial galls. Flower and seed galls were formed by fungi. Unidentified insects larvae were associated with Young stem, leaf and round axial galls. Comparative anatomical examination of the galls and healthy

organs showed the proliferation in the former of new parenchyma tissue around vascular bundles and the pith. Galls on stems are characterized by deformed and poorly developed secondary xylem with increased medullary rays. The results indicate that galls develop in response to the formation of new, mainly parenchymatous tissue resulting in local swelling of the particular plant organ. Gall-induced parenchymatous tissues are the main food source for the insect larvae inhabiting the galls.

A survey of invasive alien plants and their current distribution at Wonderboom Nature Reserve

B.T. Mthembu and T. P. Jaca (E09)

Early Detection and Rapid Response programme, South African National Biodiversity Institute, Private bag X101, Pretoria 0001, South Africa

Alien plant invasion in South Africa poses a major threat to the biodiversity and functioning of ecosystems and the services they provide. Nature reserves in the world are threatened by invasive alien plants (IAP's) and South Africa is not an exception. The invasion of nature reserves by alien invasive plants results in ecological succession, where the new alien invaders establish themselves and outcompetes the native species. The most cost effective way to manage IAP's is to prevent invasions before they even occur, or controlling them at the early stages of invasion. An inventory of IAP and their distribution inside the reserve is significant in understanding their control methods and threat to biodiversity. The aim of this study was to identify and determine the distribution of IAP in the Wonderboom nature reserve. This information will be used by the reserve managers to develop management plans for targeted species in the reserve. Stratified random sampling methods were used to determine the extent of IAP's distribution inside the reserve. This study present preliminary findings of an initial survey of IAP's inside Wonderboom Nature Reserve. A total of 17 IAP's were identified, and their distribution was mapped using geographic information system.

An inventory of indigenous medicinal plants used in the treatment of sexually transmitted infections in Vhembe district municipality, Limpopo province, South Africa

T.M. Mulaudzi and N.A. Masevhe (M06)

Department of Botany, School of Mathematics and Natural Sciences, University of Venda, Thohoyandou 0950, South Africa

Ethnobotanical survey was conducted to document medicinal plant species used in the treatment of sexually transmitted infections in Vhembe district municipality, Limpopo province, South Africa. Twenty one traditional healers were interviewed using structured questionnaires. Topics discussed during the interviews were symptoms of the disease, local plant names, plant parts used, mode of preparation, route of administration, dosage etc. A total of 45 plant species belonging to 21 different families were reported as being used for the treatment of sexually transmitted infections such as syphilis, gonorrhoea and herpes. The most common used families were the Myrtaceae and Apocynaceae (9,4%) and the rest contributed 2,3% only. The most frequently used plant parts were roots (68%), stem bark

(17%) and leaves (15%). Preferred methods of preparing the remedy were infusion and decoction, and about 91% of preparations were administered orally in variable doses until the patient was cured. Forty five medicinal plant species recorded in this study were also used by traditional healers to treat other ailments and this may validate their efficacy. Further studies are ongoing in order to establish their antimicrobial activity and to isolate the active chemical components.

***Hypericum pseudohenryi* – an invasive alien plant in the KZN Drakensberg and KZN Midlands**

I. Nänni (E11)

Early Detection and Rapid Response to emerging invasive alien plants programme (EDRR) – funded by Working for Water, South African National Biodiversity Institute

An alien species of *Hypericum* (Clusiaceae), recently identified as *H. pseudohenryi*, is invading stream beds and undisturbed grassland habitats in the KZN Drakensberg and KZN Midlands. Preliminary investigations and risk assessment suggest that this species poses a threat to indigenous biodiversity by out-competing indigenous species, hybridising with local *Hypericum* species and affecting stream flow dynamics in the Drakensberg catchment areas. An initial survey of the KZN Mistbelt revealed that naturalised populations of *H. pseudohenryi* are more extensive and widespread than originally thought. These concerns justify the decision to fund clearing efforts in KwaZulu-Natal while collecting information on reproductive strategy and response to herbicides.

Surveillance of potential emerging wetland alien invaders

M.M. Nxumalo^a, H.J.N. Sithole^a and E. van Wyk^b (E12)

^aEarly Detection and Rapid Response to emerging invasive alien plants programme (EDRR – funded by Working for Water) South African National Biodiversity Institute, P.O Box 52099, Berea Road, 4007, Durban, South Africa

^bPrivate Bag x7 Newlands 7735, Capetown

Hydromorphic and aquatic invasive alien plants pose a great threat to South Africa's wetlands. They form dense stands blocking the movement of water and aquatic fauna causes sedimentation and outcompete indigenous plants. Their ability to reproduce in several ways: vegetatively, sexually, through tubers and turions gives them a competitive advantage over indigenous plants. Tubers and turions have an ability to survive adverse conditions for long period. Six potential wetland invaders are currently investigated by the Early Detection and Rapid Response (EDRR) Programme. Four of them are listed as category 1a on the proposed NEMBA regulation. *Hydrilla verticillata* is a completely submerged aquatic plant, *Hydrocleys nymphoides* floats in water, *Iris pseudacorus*, and *Lythrium salicaria* are rooted in water but emergent above the surface. The remaining two species are newly detected invaders and do not yet have a legal status as invaders. *Sagittaria platyphylla* has two forms: emergent and submerged, *Spartina alterniflora* have an ability to hybridize with indigenous grasses. The varied growth forms and behaviour of these plants makes it difficult to generalize about their management. The poster highlights the outcomes of the work in progress such as mapping, assessment of ecological impacts and current levels of infestation.

Reports of new sightings will add value to the work done. Hence the purpose of the poster is to advise the public about possible dangers, and to call for more sightings of these species.

Indigenous medicinal plants used by traditional healers in the stimulation of appetite of HIV/Aids related patients in Vhembe District Municipality, Limpopo Province, South Africa

K.R. Phalandwa and M.P. Tshisikhawe (M07)

Department of Botany, University of Venda, Thohoyandou 0950.

The study investigated indigenous medicinal plants used by traditional healers in the stimulation of appetite in HIV/AIDS related patients in Vhembe District Municipality. In this study Knowledge of traditional healers was used in order to collect data through structured interviews. Questionnaire collected data on causes, symptoms and treatment of lack of appetite. The preparation techniques and dosages of medicinal plants were also recorded. Thirty-five plant species were recorded to be used in the stimulation of appetite. The plants were identified and information about their other medicinal uses were also investigated. The large amount of information on medicinal plants used in treatment of lack of appetite suggests the importance of traditional knowledge in appetite stimulants amongst.

Characterization of wetland systems and –zones based on vegetation composition on the Maputaland Coastal Plain

L. Pretorius^a, L. Brown^a, G. Bredekamp^b and T. Mostert^c (E13)

^a*Applied Behavioural Ecology and Ecosystem Research Unit, UNISA, Florida*

^b*Department of Plant Science, University of Pretoria, Pretoria*

^c*Department Botany, University of Zululand, KwaDlangezwa*

The Maputaland Coastal Plain (MCP), located in north-eastern KwaZulu-Natal, is dominated by cover sands and is therefore regarded a sandy coastal aquifer. According to the Department of Water Affairs' wetland delineation manual delineating wetlands on these aquifers is complex due to problematic wetland soils. It is believed that an understanding of the vegetation composition of wetlands on the MCP can aid wetland delineators. The aim of this study was to classify and describe the vegetation of the different zones of the various wetland systems to determine if differences in vegetation could assist in delineation and understanding of these complex ecosystems. Thirteen wetlands that can be grouped into four wetland systems located between Tembe Elephant Park in the west and Kosi-Bay in the east were surveyed. In each wetland the permanently wet, the seasonally wet, and the terrestrial zones were sampled. A total of 73 relevés of 4 m² size were placed using the modified Braun-Blanquet cover abundance scale. The relevés were captured into Turboveg for Windows 1.97 and imported into JUICE 7.0. A modified TWINSpan was performed to determine a first approximation of the vegetation. A phytosociological table was compiled. Synoptic tables and dendrograms were used to aid in the identification of vegetation clusters. Preliminary results identify 11 vegetation communities which are strongly related to the wetland types as defined by the hydrological regime, geohydromorphological setting, and substrate type. Variation of the vegetation composition within the specific wetlands gives a strong indication of zoning, which is related to the wetness gradient within each system. These results

contribute to the understanding of the relationship between vegetation and environmental drivers within wetland systems, and can be used to aid wetland delineation on the MCP.

Biological activity of methyl ursolate from *Funtumia africana* (Apocynaceae) leaf extracts

T.E. Ramadwa^a, E.E. Elgorashi^b and J.N. Eloff^a (M08)

^a*Phytomedicine Programme, Department of Paraclinical Sciences, Faculty of Veterinary Sciences, University of Pretoria, Private Bag X04, Onderstepoort 0110, South Africa*

^b*Currently ARC-Onderstepoort Veterinary Institute, Private Bag X05, Onderstepoort 0110, South Africa*

Funtumia africana is traditionally used across Africa to treat diverse ailments including infectious diseases, burns and others. The aim of the study was to evaluate the antibacterial, antifungal, and anti-inflammatory activities of the acetone extract of *F. africana* leaves, together with its fractions (Hexane, chloroform, 30% in methanol, butanol, and water) and the isolated bioactive compound. The antimicrobial activity was evaluated using serial microdilution assay and bioautography. The anti-inflammatory activity was determined using both cyclooxygenase (COX)-1 and cyclooxygenase (COX)-2 enzymes. The four nosocomial bacterial pathogens tested were *Escherichia coli*, *Enterococcus faecalis*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*. The fungal test organisms used were *Aspergillus fumigatus*, *Cryptococcus neoformans*, *Candida albicans*, *Fusarium oxysporum*, *Penicillium janthinellum* and *Rhizoctonia solani*. The crude extract of *F. africana* had minimal inhibitory concentration (MIC) as low as 0.08 mg/ml against both bacterial and fungal test organisms. On bioautography, *F. africana* crude extract had consistent antibacterial activity against all the bacteria with clear zones of inhibition on TLC bioautograms. The chloroform fraction had an MIC as low as 20 µg/ml against *P. aeruginosa*. The crude extract together with hexane and chloroform fractions had moderate activity against both COX-1 and COX-2. The chloroform fraction was more active than the crude extract ($59.7 \pm 1.4\%$) with an inhibition of $68.2 \pm 6.6\%$ against COX-1. One antimicrobial compound was isolated from chloroform fraction using column chromatography and chemically characterized as methyl ursolate using NMR and Mass Spectroscopy. MU had an MIC as low as 63 µg/ml against *F. oxysporum*. Although MU was less active against all the bacteria with MIC values >250 µg/ml, the MIC value of chloroform fraction (20 µg/ml) was more than hundred times lower than the MIC of MU. This provides strong evidence of synergistic activities and may be a model to investigate how synergism contributes to antimicrobial activities.

An inventory of indigenous medicinal plants used in the treatment of thrush and related infections in Vhembe district municipality, Limpopo province. South Africa

S. Ramulifho and N. A. Masevhe (M09)

^aDepartment of Botany, School of Mathematics and Natural Sciences, University of Venda, Thohoyandou 0950, South Africa

Ethnobotanical survey was conducted to document indigenous medicinal plants used in the treatment of thrush in Vhembe district municipality, Limpopo province, South Africa. Twenty one traditional healers were interviewed using structured questionnaires. Some of the topics discussed with the traditional healers included symptoms of the disease, local plant names, plant parts used, mode of preparation, route of administration and dosage. A total of 55 plant species belonging to 23 different families were recorded. The most common used families were Celestraceae and Rutaceae (11,4 %) and the rest of the families contributed (2,1 %) each. Plant parts frequently used were roots (62 %), then followed by stem bark (27%), leaves (9 %) and seeds (2 %) . The most common preparations were infusion and decoction. Oral application had the highest mode of administration (83.7%) while the combination of both oral and external applications was (12.5%). A total of 55 plant species used to treat thrush by traditional healers support the importance which the medicinal plants have in the primary health care of the rural people. These plant species were also found to be used in other parts of the world for treating a variety of diseases and further investigation is going on to determine their antimicrobial activity.

SNP-based genetic linkage maps of cassava (*Manihot esculenta*)

S.M. Reynolds^a, M.M. van Dyk^a, M. Luo^b, E. Okogbenin^c, L.A. Becerra Lopez-Lavalle^d, M. Ferguson^e, A.A. Myburg^a and P. Rabinowicz^f (B13)

^aDepartment of Genetics - Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, 0002, South Africa

^bUniversity of California, Davis CA, USA, ^cNational Root Crops Research Institute, Nigeria,

^dInternational Center for Tropical Agriculture (CIAT), Columbia, ^eInternational Institute for Tropical Agriculture (IITA), Nairobi, Kenya ,

^fInstitute for Genome Sciences, University of Maryland School of Medicine Baltimore MD, USA.

Cassava is a highly heterozygous staple crop grown in the tropical regions of Africa, Asia and Latin America. To date, six genetic linkage maps have been published for cassava, using mainly AFLP, RFLP and SSR markers. However, to our knowledge no genetic maps incorporating SNP markers have been reported. Our aim was to construct genetic linkage maps for a cassava mapping pedigree using SNP markers developed from expressed gene sequences and BAC-end sequences. Three custom designed GoldenGate Genotyping (Illumina) kits were used to genotype the CTS-1A drought tolerance mapping population (235 progeny) resulting from a cross between MCOL1734 (female, drought tolerant) and VEN77 (male, drought susceptible). This population was grown in a CIAT field in Columbia as part of a collaborative project funded by the Generation Challenge Program (GCP). Both parents and 226 progeny were successfully genotyped with 718 SNP markers, of which 556 (68%) were polymorphic in the parents. GenTrain scores ranged from 0.39-0.97, with an

average of 0.82. JoinMap (Kyazma) was used to construct linkage maps consisting of 430 SNP markers distributed in 19 linkage groups (LOD 4.0), each one containing between 3 and 63 markers and spanning 2.9 to 162.7 cM (Kosambi) in length. The total map length was 1219 cM with an average marker interval of 2.94 cM. The coverage and resolution of these maps will be increased by the addition of 124 SSR markers. In addition, using the same SNP markers, genome-wide SNP diversity patterns will be assessed using 217 cassava germplasm accessions from Africa and Latin America. These maps will also be useful for future QTL studies of drought tolerance, a trait that is expected to segregate in this population.

Expression analysis of two genes during three different wheat-*Puccinia* interactions using qPCR

J.J. Scholtz and B. Visser (B14)

Dept. Plant Sciences, UFS, PO Box 339, Bloemfontein, 9300

Two unidentified genes that could play a role during the defence response of wheat (*Triticum aestivum*) following infection by *Puccinia triticina* were recently cloned. Real-time PCR (qPCR) was used to evaluate the expression levels of both genes during infection of wheat by *P. triticina*, *P. graminis* f.sp. *tritici* and *P. striiformis* respectively. Normalisation of qPCR data requires stable reference genes under all environmental and physiological circumstances. Since limited information on reference genes during wheat-rust interactions was available, the stability of six candidate reference genes during the three respective interactions was determined. The geometric means of multiple reference genes most suitable for normalization for each interaction were identified using the geNorm Plus algorithm. For *P. triticina* infected wheat, the *18s rRNA* and *RLI* genes showed the best stability and for *P. striiformis* infected wheat, *CDC* and *RLI* were the most stable combination. *P. graminis* f.sp. *tritici* infected wheat required three reference genes, namely *CDC*, *18S rRNA* and *GAPDH*. Using the appropriate reference genes, the differential expression of the cloned genes was confirmed.

***Catharanthus roseus*: Extraordinary Bapedi medicinal herb for gonorrhoea**

S.S. Semanya^a, L.J.C. Erasmus^b and M.J. Potgieter^a (M10)

^a*Departments of Biodiversity and* ^b*Physiology and Environmental Health, School of Molecular and Life Sciences, University of Limpopo, Private Bag X1106, Sovenga 0727, South Africa.*

Catharanthus roseus is a widespread exotic perennial herb, with numerous variations in flower colour. It is world renowned for its contribution to the development of specific anti-cancer drugs, as well as its hypoglycaemic activity. Existing South African findings indicated poor anti-STI activity from leaf extracts. However, Bapedi traditional healers exclusively utilize root extracts to treat gonorrhoea. Support for this use is found in the vernacular name for *C. roseus*; lepolomo le pinki la *drop*, which alludes to the presence of an abnormal urethral discharge in the symptomatic presentation of gonorrhoea. Current uses and research, focuses predominantly on aerial parts of this species. The unique Bapedi custom to

exclusively use roots warrants further investigation to validate its efficacy in the treatment of gonorrhoea.

Expression and characterisation of *EgrSND2*, a *Eucalyptus grandis* transcription factor potentially involved in secondary cell wall regulation

P. Singh^{a,b}, S.G. Hussey^{a,b}, C. Maritz-Olivier^a, E. Mizrachi^{a,b} and A.A. Myburg^{a,b} (B15)
^aDepartment of Genetics, ^bForestry and Agricultural Biotechnology Institute (FABI),
University of Pretoria, Pretoria, 0002, South Africa

The transcriptional regulation of secondary cell wall (SCW) biosynthesis has been studied extensively in *Arabidopsis thaliana*, revealing a hierarchy of predominantly NAC and MYB transcription factors that regulate the formation of cellulose, hemi-cellulose and lignin, the three main SCW biopolymers. *AtMYB103*, *AtSND2* and *AtSND3* transcription factors have been found to regulate SCW-associated cellulose synthase (*CesA*) genes in *A. thaliana*. Cellulose fibres from *Eucalyptus grandis* wood are one of the most commonly used short fibre sources for pulpwood in the paper industry. Our understanding of the regulation of cellulose biosynthesis is vital to the pulping industry and will have great economic impact. Currently, the transcriptional network regulating SCW biosynthesis in *E. grandis* is poorly understood. We aimed to identify and clone an *E. grandis* ortholog of *AtSND2*, heterologously express the protein and evaluate its promoter binding specificity. The *EgrSND2* coding sequence was cloned into the pET160 vector and transformed into the *Escherichia coli* BL21Star strain, which was used to optimise expression using temperature, time and inducer concentration parameters. We obtained exclusively insoluble protein when expression was induced at temperatures of 25°C, 30°C and 37°C and IPTG concentrations of 0.1 mM, 0.5mM and 1 mM over 24 hours. Hence, we adopted a protein unfolding/refolding approach by purifying inclusion bodies and denaturing and refolding the heterologous protein. Soluble protein and candidate *E. grandis* gene promoter fragments will be analysed by electrophoretic mobility shift assays to identify direct targets of *EgrSND2*.

A Study of a Population of a *Thamnochortus* species at the Sundays River, Eastern Cape

S. Situngu and R.A. Lubke (D11)

Department of Botany, Rhodes University, Grahamstown, 6140

A population of a *Thamnochortus* species has been discovered in the Sundays River mouth region. There were many species planted in the area by the Department of Forestry to stabilise the mobile dune field but *Thamnochortus* species were never used in the stabilisation programme. The population of plants seemed to be indigenous to the area and is confined to the limestone cliffs along the old and present river valley away from the stabilisation site. *Thamnochortus* species of the Western Cape growing on alkali sands and limestone were studied and compared with the population at the Sundays River region. A multivariate analysis was performed on both quantitative and qualitative characters to test whether the Sundays River species was different from the Western Cape species. The Sundays River *Thamnochortus* species showed affinity with *T. erectus*, *T. muirii* and *T. spicigerus* in some characters and affinity with *T. insignis* on other characters. The Sundays River species

formed a single group in the cluster analysis and in the PCA analysis suggesting that species is different from the other *Thamnochortus* species. Thus the Sundays River species appears to be morphologically different from the Western Cape species and should perhaps be considered a new species. However, more studies on genetic variation between these species are required to substantiate these findings.

Analysis of genetic variation in *Moringa oleifera* (lam.) using RAPDs and SSRs

R. Smit, E.S. du Toit and B.J. Vorster (B16)

Department of Plant Production and Soil Science, University of Pretoria, Pretoria South Africa, 0002

Moringa oleifera is potentially an economically important tree species. It has gained interest globally for its multipurpose uses, in particular as a source of nutrition and oil as well as various medicinal properties. Moringa is native to India, Malaysia and the Middle East, but have been introduced to many countries throughout Africa ranging from Niger to South Africa. There is however limited knowledge regarding the genetic variation of both native and introduced populations of Moringa, although phenotypic observations suggests the presence of significant genetic diversity. In this study we aim to determine the level of genetic variation found between different populations of Moringa from locations including India, South Africa, Malawi and Hawaii. Molecular marker such as Random Amplified Polymorphic DNA (RAPD) and Simple Sequence Repeats (SSR), will be used to analyse the genetic diversity based on their success in other tropical tree population studies. Their low capability for SSR conservation, make them ideal for the application at the intra-specific level. Various RAPD primers and 20 SSR primer pairs will be used to generate amplification profiles that can be used in a diversity analysis. Early screening has identified markers that show significant genetic diversity amongst the population. Furthermore the study will try to identify markers related to quantitative traits such as seed oil content and yield that could potentially be useful in future selection and breeding programs aimed at tree improvement.

How generalized is the pollination system of *Hypoestes aristata*?

C-J. Thorne, C. Gornall and C.I. Peter (E03)

Department of Botany, Rhodes University, Grahamstown, 6140

Generalized pollination systems are underrepresented in the literature. Observations have shown numerous insects and birds visiting *Hypoestes aristata* (Vahl) Sol. Ex Roem. & Schult var. *aristata*. We therefore set out to test hypothesis that this species is an extreme generalist by 1) recording the suite of insects and birds visiting the flowers 2) determining pollen loads of the most frequently collected insect visitors and 3) the visitation rates of different pollinators. We recorded a total of 45 species of insects visiting the flowers. These represented 19 families and 4 orders. In addition, four species of sunbirds have been observed visiting the flowers opportunistically. The orders Diptera and Hymenoptera were shown to have the greatest pollen loads, in particular species from the families Acroceridae, Nemestrinidae and Apidae. Although late in the season honeybees were by far the most abundant visitors and showed the highest visitation rate. In conclusion, a wide variety of

pollinators visit *Hypoestes aristata* and contribute to this generalized pollination system but late in the season honeybees predominate.

The treatment of persistent headache in HIV/aids related patients by the traditional healers of Vhembe district municipality, Limpopo province (RSA)

T. Tshilande and M.P. Tshisikhawe (M11)

Department of Botany, University of Venda, Thohoyandou 0950

The recent study was about indigenous medicinal plants used by traditional healers in the treatment of persistent headache in HIV/AIDS related patient in Vhembe District Municipality, Limpopo Province, South Africa. In this study knowledge of traditional healers was used in order to collect data, and data was collected through the use of interview, and question such as the cause, symptoms and treatment of persistent headache were asked, and also the preparation and dosages were asked through interviews. In this study, traditional healers reported that forty-five plant species were used in the treatment of persistent headache and of those plants; thirty-one were identified given their family, scientific name and given information about their other medicinal uses. It was concluded that indeed knowledge of traditional healers is of importance in the treatment of persistent headache as well as for the patient who are infected by HIV/AIDS.

Landscape functionality and plant diversity in urban and rural grassland fragments in the Tlokwe Municipal area

L. van der Walt, S.S. Cilliers, K. Kellner and M.J. du Toit (E14)

School of Environmental Sciences and Development, North-West University, Potchefstroom, South Africa

Grassland habitats around the world have been identified as a biodiversity conservation priority. South Africa's grassland biome, covering 16.5% of the country's surface, is one of biologically richest in the country. Unfortunately nearly 30% of it has been transformed by activities such as urbanisation, cultivation, mining, and forestry. The remaining untransformed grassland areas are fragmented to a critical extent, such as the Rand Highveld grassland vegetation type. A mere 1% of this endangered vegetation type is currently being conserved. The fragmentation, transformation and exotic species invasion of landscapes worldwide are threatening biodiversity and altering ecological processes, spatial organization of physical landscape attributes, as well as species distribution and composition. Fragmentation of the Rand Highveld Grassland may also influence the functionality of such landscapes. To understand how a landscape functions, and why a landscape may be deemed functional or healthy, knowledge about the processes taking place in landscapes (to enable it to function effectively as a biogeochemical or biophysical system) must be obtained. This can be done using the Landscape Function Analysis (LFA) method. Patches play a vital role in landscapes by capturing and utilising natural resources such as water, nutrients, and soil sediments and are thus indicators of the extent to which landscapes are functional or dysfunctional. Generally habitat remnants in urban areas are considered to be extensively disturbed and no longer functional or species rich, leading to a perception that these

fragments need not be as actively conserved as fragments in rural or more “natural” landscapes. In this proposed study we aim to quantify an urban-rural gradient, and describe differences and similarities between urban and rural grassland fragments in terms of species composition, species richness, functional diversity and landscape functionality, in the Tlokwe Municipal area, situated in the Rand Highveld Grassland vegetation type. This comparison will allow for recommendations to be made in terms of conservation priorities of grassland remnants.

The effect of four medicinal plants on oral *Candida albicans* isolates

C. van Wyk^a, F.S. Botha^a, V. Steenkamp^b and J.N. Eloff^a (M12)

^a*Phytomedicine Programme, Department of Paraclinical Sciences, Faculty of Veterinary Sciences, University of Pretoria*

^b*Department of Pharmacology, School of Medicine, Faculty of Health Sciences, University of Pretoria*

Crude acetone, water and hexane extracts of four plant species, used as traditional medicine to treat fungal related diseases or shown to contain antifungal activity, were evaluated so as to identify new sources of chemical entities which can be used in the treatment of fungal infections. Antifungal activity against *Candida albicans* standard strain (ATCC 10231) and six clinical isolates was determined using the serial microplate dilution method. Antioxidant activity was determined using the TEAC and DPPH assays and cytotoxicity against mouse fibroblast cells. MIC's below <1 mg/ml were observed for the acetone and water extracts of *Sclerocarya birrea* (stem-bark), as well as the acetone extracts of *Harpephyllum caffrum* (leaves), *Ptaeroxylon obliquum* (leaves) and *Rapanea melanophloeos* (leaves). Bioautography confirmed the presence of compounds with antifungal activity in *S. birrea* and *P. obliquum*. *S. birrea* contained the highest antioxidant activity with the lowest cytotoxicity. These results implicate that the extracts contain compounds with therapeutic potential against oral *C. albicans* isolates.

Author Index

Abdillahi, HS	95	Beukes, CW	129
Adamu, M	33	Bezeng, BS	40
Adewumi, RO	89	Bhat, RB	38, 73, 144
Adewusi, EA	33	Biye, EH	130
Ahmed, AS	82, 99	Boatwright, JS	41, 48, 72
Alberts, WG	108	Bond, W	29
Alexandersson, E	124	Bond, W.J	76, 106
Alimi, AA	89	Bossinger, G	47
Al-Zaidi, AM	34	Botha, FS	42, 157
Amoo, SO	35, 127	Botha, J	130
Anthonissen, R	86	Bredenkamp, G	150
Anyasi, RO	35	Bredenkamp, J	42
Archer, RH	36	Brown, L	150
Archibald, S	106, 121	Brown, LR	136
Aremu, AO	35, 36, 127	Burger, JT	104
Arena, G	37	Burrows, JE	58
Aroke, A	76	Byrne, M	79
Atagana, HI	35	Bytebier, B	62, 75
Awosika, OS	53	Campbell, EE	113
Ayeni, OB	38	Carriero, F	142
Bagla, V	42	Castelyn, HD	43
Bairu, MW	36, 127	Castillo, RR	131, 132
Balkwill, K	130	Chakauya, E	78
Baloyi, O	38	Chen, S	43, 64
Barker, NP	108	Chikwamba, R	78
Baskaran, P	128	Chinsamy, M	44
Bayley, A	115	Choi, YH	31
Becerra Lopez-Lavalle, LA	152	Cilliers, SS	134, 156
Becker, CH	128	Cingo, P	44
Beck-Pay, SL	39	Coetsee, C	128
Begoude, BAD	146	Coetzee, MPA	45, 84, 139
Bello, AO	39	Coetzee, Z	124
Bellstedt, DU	109	Coetzer, N	132
Bendahmane, A	142	Cordier, W	46
Berger, DK	42, 56, 83, 132, 141, 142	Coutinho, TA	59, 105
Berrington, D	40	Cowling, RM	29, 113, 128
Bessong, PO	100	Cozien, RJ	46
Bester, MJ	102	Crampton, BG	133, 141
Bester, SP	141	Creux, N	130
		Creux, NM	47
		Cromarty, AD	46

Cromarty, D	102	Fitza, K	53
Cron, G	79	Folorunso, AE	53
Cron, GV	44, 130, 133	Fourie, H	55
Cushman, KC	115	Fourie, L	54
Daniels, RJ	48	Foxcroft, LC	117
Danson, JW	80	Freeborough, MJ	104
Daru, BH	48	From, MM	55
Davies, TJ	123	Gaertner, M	55, 57
Davoren, E	134	Gazendam, I	56
de Canha, M	135	Geerts, S	57, 81
De Castro, MH	47	Gere, J	57
de Rapper, S	49	Gill, AT	139
Deloire, A	124	Gillson, L	93
Difford, M	113	Gornall, C	155
Dockrall, SJ	69	Goulet, M-C	120
dos Santos, A	135	Grace, OM	67
Du Plessis, HJ	77, 145	Grové, J	139
Du Plooy, CP	94	Gruèzo, WSM	58, 131, 132
Du Preez, PJ	136	Gryzenhout, M	59
Du Toit, E	147	Gulumian, M	46
du Toit, ES	155	Haddad, WA	58
du Toit, MJ	156	Hakizimana, JD	59
Duffy, KJ	50	Hamer, M	41
Duodu, KG	102	Hamilton, C	91
Dutta, SR	50	Harder, LD	62
Ebrahim, I	111	Hefer, C	143
Egan, BA	77, 145	Hefer, CA	114, 135
Elgorashi, EE	151	Hendriks, CBS	60
Eloff, JN	33, 42, 51, 74, 76, 80, 82, 90, 99, 151, 157	Herman, PPJ	60
Emamzadeh- Yazdi, S	137	Herselman, L	119
Engelbrecht, FA	51	Heyman, HM	61
Engle, NE	115	Heystek, A	61
Erasmus, BFN	106	Hills, PN	70
Erasmus, LJC	78, 100, 153	Hirschberg, J	30
Fajinmi, OO	137	Hobbhahn, N	62
Farrant, JM	139	Hoffman, MT	76, 93, 95
Ferguson, M	152	Human, H	92
Ferreira, L	138	Hussein, A	101, 105, 137
Figlan, S	52	Hussey, S	130
Finnie, JF	36, 44, 78, 85, 89, 137	Hussey, SG	154
		Ivey, P	41, 121
		Jaca, TP	63, 140, 148
		Jacob, RM	80
		Jacobs, SM	66

Jacobson, D	124	Le Roux, JJ	57, 109, 117
Janse van Rensburg, R	64	le Roux, MM	69
Jimu, L	64	Lebouteiller, B	70
Johnson, SD	46, 62, 65, 67, 76, 106, 111	Lerch-Henning, S	71
Jones, N	114	Ligavha-Mbelengwa, MH	144
Joubert, F	115	Little, DP	97
Kabera, G	78	Lombard, B	143
Kalwij, JM	65	Lötter, MC	54
Kamatou, GPP	49, 66	Lubke, RA	154
Kambaj, OK	66	Luo, M	152
Kanzler, A	114	Magee, A	92
Kellner, K	156	Magee, AR	71, 72
Kemp, G	64	Magwede, K	73
Kenyon, C	61	Maharaj, R	78
Khan, J	34	Maharaj, V	61
Kiepiel, I	67	Mahlanza, T	73
Killian, A	142	Maize eQTL project consortium	74
Kim, HK	31	Makhafola, TJ	74, 80
King, CW	61	Makome, L	80
Klopper, RR	67	Makunga, NP	52, 124
Kloppers, FJ	141	Makwarela, LE	143
Koekemoer, M	141	Mankga, LT	75
Koen, K	39	Manning, JC	48
Köhler, A	68	Mantlana, KB	66
Korsman, JN	141	Manyama, P	121
Kossmann, J	79	Marais, C	29
Kossmann, JM	52	Maree, JE	118
Kritzinger, Q	137	Maritz-Olivier, C	47, 130, 154
Kritzinger-Klopper, S	57	Martos, F	75
Kulkarni, MG	115, 137	Masevhe, NA	76, 148, 152
Kullan, ARK	114	Mashile, PS	144
Kunert, KJ	116	Masiagwala, P	144
Kuriakose, B	142	Masubelele, ML	76
Kuzmina, M	123	Mathibela, KM	77, 145
Lall, N	40, 91, 101, 103, 135	Maurin, O	40, 41, 48, 57, 75, 123
Lalla, R	142	Mavundza, EJ	78
Landry, M	55	Mawela, KG	78
Lashbrooke, J	124	Mayekiso, KTV	80
Lashbrooke, JG	69	Mayonde, SG	79
Laurie, SM	147	Mbenoun, M	146

McBride, J	104	Naidoo, S	42, 53, 83, 88, 138
McGaw, LJ	74, 76, 80, 90, 99	Naidoo, V	33, 90
McGeoch, M	57, 93	Nakhooda, M	84
Meisel, B	141	Nänni, I	121, 149
Meyer, JJM	60, 61, 105	Ncube, B	85
Mhora, TT	80	Ndhlala, AR	86
Michaud, D	120	Nefefe, T	100
Midgley, J	112	Nemakanga, R	86
Midgley, JJ	111	Netshivhulana, TA	87
Mihiretu, HC	147	Newman, D	114
Mills, A	29	Nicolas, AN	71
Mills, AJ	113	Nicolson, SW	68, 71, 87
Minnaar, A	102	Nottebrock, H	55
Minoia, S	142	Nxumalo, MM	149
Mizrachi, E	115, 130, 135, 143, 154	Nzama, SN	88
Mncwangi, NP	118	O'Connor, T	95
Moodley, D	81	Oates, CN	88
Mostert, T	150	Odiwe, AE	89
Motaung, SCKM	95	Oelofse, D	56
Moteetee, AN	75	Ogunsanwo, O	89
Motlounge, RF	82	Okem, A	89
Moyo, M	127	Okogbenin, E	152
Mphephu, TE	147	Oladipo, OT	39
Mthembu, BT	148	Olaokun, OO	90
Mthombeni, L	129	Oosthuizen, CB	91
Mtunzi, FA	82	Opoku, AR	104
Mucina, L	72	Osborne, CP	96
Mulaudzi, T	100	Oyedeji, AO	104
Mulaudzi, TM	148	Pailer, T	75
Muleya, E	82	Parker-Allie, F	91
Muofhe, C	57	Pauw, A	61
Murray, S	104	Payn, K	53
Murray, SL	133	Peter, CI	48, 155
Muthelo, VG	45	Phalandwa, KR	150
Mwenje, E	64	Phephu, N	92
Myburg, AA	47, 53, 83, 88, 107, 115, 130, 132, 135, 138, 143, 152, 154	Pierce, S	29
Mycock, D	84	Pietersen, G	119
Naidoo, K	84	Pinard, D	130
Naidoo, R	83	Pirk, CWW	68, 92
		Plunkett, GM	71
		Pool, JR	93

Potgieter, MJ	77, 100, 145, 153	Sharma, R	101
Powell, M	93	Shelembe, JS	102
Powell, RF	93	Siebert, SJ	134
Pretorius, JC	64	Sigwela, A	29
Pretorius, L	150	Singh, M	104
Pretorius, ZA	43, 120	Singh, P	154
Prinsloo, G	94, 137	Singh, S	103
Privett, SDJ	55	Sipamla, AM	82
Puttick, J	95	Sithole, HJN	88, 103, 149
Pyšek, P	30	Situngu, S	154
Rabinowicz, P	152	Six, DL	111
Rafudeen, MS	139	Slippers, B	84, 88
Ramadwa, TE	151	Smit, R	155
Ramovha, LI	147	Smith, GF	67
Ramulifho, S	152	Snyman, SJ	73
Ranik, M	47	Solofoharivelo, MC	104
Razwinani, M	95	Soundy, P	108
Rebelo, AG	81	Soyingbe, OS	104
Reynolds, SM	152	Spokevicius, A	47
Richardson, DM	55, 57, 81, 109, 117	Stafford, GI	86
Ripley, BS	93, 96	Stanton, SL	105
Robertson, MP	65, 82	Steenhuisen, S-L.	106
Roets, F	43	Steenkamp, E	84
Rouget, M	54, 58, 82	Steenkamp, ET	53, 119, 129, 139
Rousseau, P	97	Steenkamp, V	33, 46, 157
Roux, J	43, 64, 98, 111, 146	Stephan, D	104
Ruiters, AK	98	Stevens, N	106
Rutherford, RS	73, 80	Steyn, HM	141
Saheed, SA	38, 39	Steyn, JM	147
Sakong, BM	99	Strauss, R	96
Samie, A	100	Sutherland, R	107
Sandasi, M	66, 119	Swanepoel, KM	108
Sasa, SR	86	Sweby, DL	80
Savolainen, V	123	Symes, C	37
Schafer, H	123	Szabo LJ	119
Schmidt, TG	141	Taylor, CL	108
Scholtz, JJ	153	Thompson, GD	109
Seipethlo, L	136	Thorne, C-J.	155
Semenya, SS	77, 100, 153	Tilney, PM	71, 98, 109
Sethusa, MT	41	Treurnicht, M	110
Shaik, S	101	Tschaplinski, TJ	114
		Tshikalange, TE	95

Tshilande, T	156	Victor, JE	143
Tshisikhawe, MP	38, 73, 87, 144, 150, 156	Viljoen, A	107
Turner, R	112	Viljoen, AM	49, 66, 118, 119
Turner, RC	111	Viljoen, E	142
Tuskan, GA	114	Viljoen, R	119
Udom, OO	80	Visser, B	43, 64, 119, 153
van de Peer, Y	115	Vivier, M	124
van den Berg, N	59, 107	Vivier, MA	69
van der Bank, M	40, 41, 48, 57, 75, 97, 123	Vorster, BJ	116, 120, 155
van der Linde, JA	111	Vorster, P	97
van der Merwe, A	143	Watt, MP	73, 84
van der Merwe, CF	60, 105, 109	Whitecross, M	121
van der Niet, T	112	Wilson, JR	57, 82, 109, 117, 121
van der Vyver, M	29	Wingfield, BD	45, 84, 146
van der Vyver, ML	113	Wingfield, MJ	43, 45, 64, 84, 98, 111, 146
van der Walt, A	104	Winter, PJD	122
van der Walt, L	156	Witkowski, ETF	37, 121
van Dyk, MM	114, 152	Würger, G	51
van Jaarsveld, I	115	Yessoufou, K	40, 123
van Rensburg, BJ	64	Young, P	124
van Rooy, J	92	Young, PR	69
van Staden, J	35, 36, 44, 78, 86, 89, 95, 115, 127, 128, 137	Yuliana, N	31
van Vuuren, SF	49, 66, 98, 124	Zonyane, S	124
van Wyk AE	58, 67, 92, 109		
van Wyk, B-E	69, 71, 98, , 116		
van Wyk, C	42, 157		
van Wyk, E	149		
van Wyk, SG	116		
van Zyl, E	129		
van Zyl, RL	66		
Vardien, W	117		
Venter, HM	63		
Venter, SN	106, 129		
Vermaak, I	118		
Verpoorte, R	31		
Verschaeve, L	86		