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ABSTRACT BOOK

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Plenary speakers

Keynote

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The importance and relevance of ethnobotanical research – science of the future?

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What is the relevance of ethnobotanical research in the 21st century? Are ancient oral-traditional knowledge systems still important in a modern world? To maximize the potential socioeconomic benefits to be derived from indigenous knowledge, high quality primary data, both quantitative and qualitative, are essential. Ethical and legal aspects are equally important. The affordances of ethnobotanical research include the development of new products and services, the empowerment of local communities, the potential role of eco-tourism, and science education. The cultural value of ethnobotany is important in the context of the arts, design, music, cuisine, writing and poetry, and onomastics (the origin of place names). The exceptional botanical and cultural diversity of southern Africa (including the most ancient human cultures on earth) are unique selling points and provide countless opportunities for discovering profound new causal relationships relevant to human welfare, especially in physical and mental health. Furthermore, high quality new digital data will become more and more relevant in a future world of expert systems and automated data mining.

Management of SANParks: The role and development of vegetation classification to make informed decisions (1991-2022).

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Phytosociologists in Europe have been involved in vegetation classification studies following, in particular, the Braun-Blanquet approach since the early 1900's. In South Africa, such studies were undertaken on a limited basis from the early 1970's and have since then steadily increased and improved through modern technology such as computers. The surveying of the enormous diversity of 21 South African National Parks (SANParks) vegetation was one of the author's main assignments when being employed by SANParks in December 1990. The role and development of new methods to do vegetation classification to make informed decisions is still ongoing. The demand for vegetation classification, description and mapping in SANParks has steadily increased over the last 31 years to guide conservation policies, biodiversity studies and ecosystem management. New innovative ideas, which differ from the traditional vegetation classification methods are necessary to meet with these demands. Wildlife is part of the landscape ecosystems and is mostly favoured to be seen by tourists visiting SANParks. However, wildlife management is of the utmost importance to prevent vegetation and soil degradation, with events such as drought and sometimes fire that cannot be predicted. The impact of wildlife on the vegetation and soil (habitat) and its variability over space and time is receiving high priority in national parks. In the past, wildlife management decisions were often based on wildlife numbers based on agricultural carrying capacity and informal field observations alone, which resulted in the need to include other more robust methods to be able to make informed decisions. There are a number of measures of vegetation condition which include both qualitative and quantitative methods and these different methods provide different perspectives at different spatial and temporal scales in SANParks.

Above is a team effort in collaboration with specialist colleagues Dr Sam Ferreira, Dr Nkabeng Mzileni, Dr Charlene Bissett, Lufuno Munyai, Nthabeliseni Munyai, Professor Abel Ramoelo and Professor Leslie Brown.

Being ready when opportunity knocks: From DNA barcoding and honey to the phylogenomics of New Zealand *Leptospermum* and *Kunzea* (Myrtaceae)

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DNA barcoding work undertaken in preparation of myrtle rust arriving in New Zealand, the development of a Mānuka honey standard and a chance event while attending a conference, set the scene for phylogenomic work on *Leptospermum* and its sister genus, *Kunzea*. We provide an overview of the main results with a focus on the phylogenomics of *Leptospermum*. In New Zealand up to six morphologically distinct varieties of this species have been described, although until recently only two were formally recognised. Anchored Hybrid Enrichment was employed to acquire sequence data from orthologous low-copy nuclear loci. Our analyses suggest that a combination of low and incompatible data signal limits resolution of relationships among New Zealand populations of *L. scoparium*. Relationships within New Zealand *L. scoparium* appear to lie at the interface between phylogenetics and population genetics. There was no clear genetic support for previously recognised morphologically defined intra-specific taxa. Our analyses are consistent with genetic diversity being geographically structured, with three groups of *L. scoparium* recovered. In retrospect, the following observations to inform future research practice: 1) Be ready when opportunity knocks. 2) The commercialisation of phylogenomics provides easy access to well-resourced laboratories (but, at a price). 3) The phylogenomic pipeline is complex—a team approach assures quality of outcomes and work. 4) The Nagoya and related regional protocols present a challenge for time-constrained projects.

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Sorghum responses to abiotic stresses: fitting one puzzle piece at a time

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Sorghum is naturally well-adapted to hot and dry environments and has diverse germplasm with an untapped gene pool. It is an ideal model for studying plant responses to heat and drought stresses and identifying potential marker genes for stress adaptation. Our research group and collaborators have been working towards understanding this crop's physiological and molecular changes in response to abiotic stresses using cell suspension cultures and seedlings of various varieties. This talk will present our findings in response to drought, heat or their combination, and exogenous abscisic acid (ABA). While the puzzle is far from complete, some trends in stress responses are emerging – offering clues on how contrasting varieties may perceive and respond to the prevailing water deficits and/or high-temperature stresses. The selective influence of exogenous ABA on sorghum's intra- and extracellular protein expression patterns also enlightens us on the crop's complex subcellular proteomics under stress. However, we acknowledge that the puzzle is far from complete, and more work still needs to be done as we broaden our knowledge in plant stress biology.

Spatially explicit tests of species independence using genome-scale markers

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Wright's (1943, 1946) isolation-by-distance (IBD) theory describes population-level patterns of gene flow where proximal individuals share genes more frequently than remote ones. Under geographically limited dispersal, Wright's theory predicts that genetic differentiation among individuals should vary as a positive linear function of geographic distance. Patterns of IBD can be extended for testing species limits, which Hausdorf and Hennig (2020) recently formalized in their regression-based tests of equality. These tests use population- or individual-based estimates of genetic differentiation, such as those provided by genome-scale molecular markers, to assess evidence for the independence or genetic cohesiveness of two candidate species based on their geographical separation. The regression-based tests, implemented in the R package *prabclus*, represent a compelling, spatially explicit framework for testing species limits in a way logically decoupled from Multi-Species Coalescent (MSC)-based approaches. The basic logic behind Hausdorf & Hennig's tests is explored, and results of the tests presented for evolutionary independent species in the Cape-centred complex *Seriphium plumosum* L. (Asteraceae: tribe Gnaphalieae). Also discussed are biologically realistic ways of quantifying gene flow distances between and within candidate species using least cost paths, and advantages and practical limitations of regression-based tests for discriminating between population structure and more defined genetic discontinuities consistent with species boundaries.

Anatomy

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Nectar spur development in *Nemesia* Vent. (Scrophulariaceae)

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The floral nectar spur is regarded a “key innovation”, causing increased species diversification within clades that contain them. These tubular outgrowths of floral organs generally contain nectar or other rewards for attracting pollinators. Therefore, nectar spurs have ecological and evolutionary importance, as they influence pollinator specificity, pollination efficiency and the reproductive success of the plant. Nectar spurs have evolved independently, through convergent evolution, in several angiosperm families, with substantial developmental differences between spurred taxa. For example, in *Aquilegia* (Ranunculaceae) and *Linaria* (Plantaginaceae), nectar spur development occurs due to cell division, which is followed by cell elongation. However, differences in nectar spur length in *Aquilegia* are largely due to cell elongation, while in *Linaria*, cell number, and therefore, cell division, contribute to spur length variation. Despite the importance of nectar spurs, their development in *Nemesia*, a genus endemic to southern Africa, is still unknown. *Nemesia* consists of approximately 77 species, which show great diversity in floral colour, shape, size and nectar spur morphology. We used a comparative approach to study spur length and width variation among *N. barbata*, *N. macrocarpa* and *N. strumosa*, which have conical, tubular and saccate spurs, respectively. Using scanning electron microscopy of the outer nectar spur epidermal surface, throughout different stages of spur development, the relative roles of cell division and cell expansion (anisotropy) are investigated. Understanding the developmental basis involved in nectar spur morphological variation in *Nemesia*, will inform us of the genetic mechanisms involved in floral trait evolution and diversification in this genus.

Biotechnology

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First report of arbuscular mycorrhizal fungal communities associated with the roots of indigenous legumes of South Africa

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Arbuscular mycorrhizal (AM) fungi are crucial for the survival of plants in harsh, nutrient-limited environments, such as semi-arid ecosystems. However, there is a lack of information on the diversity of AM fungi colonising the roots of indigenous legumes of South Africa. Thus, this study assessed the diversity of AM fungi associated with the roots of eleven indigenous legumes in two South African provinces and investigated the impact of soil abiotic factors on the community composition of AM fungi. Root colonisation by AM fungi was determined by staining and microscopic assessment, while the diversity of AM fungi in roots was examined by Illumina Miseq sequencing of the partial small subunit ribosomal ribonucleic acid (SSU rRNA) gene. The results showed 172 operational taxonomic units (OTUs, at 97% sequence similarity) classified into eight genera; *Acaulospora*, *Ambispora*, *Archaeospora*, *Claroideoglomus*, *Diversispora*, *Glomus*, *Paraglomus*, and *Scutellospora*. The community composition of AM fungi varied among legumes within and between provinces, but the genus *Glomus* predominated the communities. Canonical correspondence analysis indicated that nitrates, pH, manganese, and organic C were the soil properties that significantly influenced the community composition of AM fungi. Overall, findings from this study demonstrated the ecological significance of AM fungi on the indigenous legumes and highlighted that soil properties are important abiotic factors that shape legume-AM fungal symbiosis in semi-arid ecosystems.

An *rbcl* and *trnL* (UAA) reference database for the identification of food items from herbivorous diets based in a South-African savanna

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A successful metabarcoding study is underpinned by a comprehensive and accurate DNA reference database. Since 2019, only ~8% of vascular plants on BOLD Systems originate from Africa, although this continent comprises ~20% of the global landmass and ~15% of the global plant diversity. The compilation of a DNA reference database for herbivore dietary analyses is therefore critical. This will require a list of potential food items and their abundances in the study area, and the selection of suitable barcoding markers that reveal a high level of species discrimination from degraded and fragmented DNA to finally allow for successful taxonomic assignments by their phylogenetic groupings and coverage. *RbcL* and *trnL* are chosen as a two-locus plant identification system for plant DNA from herbivore faeces due to high rates of sequence recoverability from degraded DNA as opposed to the recommended barcode standard for plant identification, which is *rbcl* and *matK*. Species constituting the reference database in this study, which are all representatives of a South African savanna and potential food items of herbivores, represent 99 families, 373 genera and 731 species in the phylum Tracheophyta. Here, we evaluated the efficiency of species discrimination of the barcodes in the compiled reference database based on estimated community phylogenies as well as the prediction of the presence or absence of barcode gaps for each marker, which is suggested to enhance reliable differentiation between species.

Morphological and physiological response of salinity stress on *Glycine max*

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Glycine max (soybean) is a globally important, high protein crop used for human and animal nutrition, soil-nitrogen fixing in organic farming, and raw material in the biofuel production industry. South Africa is a semi-arid country that relies on irrigation, which has 0.1 Million hectares of saline-affected land resultant from irrigation and natural weathering, and, has 9% of the world's saline soils share. Excessive salt accumulation result in a great yield loss to salt-sensitive crops, like cultivated soybean. This study evaluated the physiology and morphology of *Glycine max* (cultivar PAN 1521R) under salinity stress to elucidate model biological processes and demonstrate how salt stress stimulates plant response and adaptation mechanisms. Our results indicate how morphological and physiological traits were affected by salt stress. At large, salt stress-affected morphological traits such as plant height, leaf numbers, shoot and root (length and biomass). On the other hand physiological attributes such as relative water and chlorophyll content, photosynthesis and stomatal conductance were highly affected by salinity. These findings may be used as a guidance on how to conserve crop growth under salinity stress and improve food security in soybean and related crops. In addition assist in model responsive pathways used to engineer or reverse engineer plant cell death.

Ecology

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Effects of *Prosopis velutina* invasion on soil characteristics along the riverine system of the Molopo River in North-West, South Africa

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Many invasive plant species, such as *Prosopis* species, have been introduced around the world and can alter the soil properties of invaded ecosystems. *Prosopis* is one of the most aggressive invasive plant in the North-West province of South Africa, but little information is available about its influence on soil properties. This study was conducted to investigate the effect of *Prosopis velutina* invasion on selected soil properties at five different sites along the riverine system of the Molopo River in North West. At each study site, soil characteristics were measured from soil samples taken under *P. velutina* canopies, between canopies and in the benchmark stands free of *Prosopis*. In all the sites, almost all soils collected from under the canopies had a significantly higher soil exchangeable Ca, K, Mg, and Na, OM, TN, P, EC, and CEC than the other sampling positions, except for the pH which had the high value in inter-canopies. Significantly higher ($p > 0.05$) values of almost all soil properties were found on the densely invaded sites (Tshidilamolomo I and Tshidilamolomo II) compared to lightly invaded sites (Mabule, Black Heat Farm, and Bray). However, it was difficult to generalise as the effects often appear to be site-specific. Soil textural classes ranged between sand, silt, and clay in all study sites with a higher proportion of sand in the benchmark than under-canopies and inter-canopies. The site effects observed in this current study provide evidence that this species may occupy a relatively broad soil niche.

Bush encroachment in the Thornbush Savanna of Central Namibia is not widespread, but habitat-specific

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The thornbush savanna of Central Namibia is dominated by a mixed low tree and shrub layer of various *Acacia* species, mainly *Senegalia mellifera*. Bush encroachment is regarded as wide-spread and a major threat to cattle farming in this landscape. When comparing historical photographs (dating from 1956) from the farm Erichsfelde with the present situation, this general perception on shrub thickening in this landscape seemed confirmed. We however found it difficult to quantify the degree of encroachment from the few photographs, and decided to compare present-day drone imagery with historic aerial photographs (the oldest being from 1968) of three selected sites on different habitats. On shallow soils on calcretes, within the *Boscia foetida*–*Leucosphaera bainesii* association, we found a massive degree of encroachment, transforming a grassland to a closed shrubland. Similarly, within the *Monechma genistifolium*–*Acacia tortilis* association on alluvial soils near an ephemeral river, bush encroachment occurred within an originally open woodland landscape. Here we also found that in an effort to combat bush encroachment, several old trees which already stood in 1968, were felled. The *Aristida congesta*–*Senegalia mellifera* association occurs on sandy soils on the eastern part of the farm and is the dominating vegetation type in the flat landscapes of the southern Thornbush savanna of central Namibia. Here we found that the phanerophytic population was stable, or even slightly declining, over the past 53 years. This casts serious doubt over the popular perception that the Thornbush savanna is widely encroached.

Possible effect of community composition on pollination rewards in three *Nemesia* species from Namaqualand

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With ±77 species, *Nemesia* Vent. is one of the most diverse genera in the Hemimerideae (Schrophulariaceae). However, little is known about the drivers of speciation within the genus. Previous phylogenetic studies have demonstrated diversification in *Nemesia* associated with multiple transitions from the summer to winter rainfall regions of southern Africa. *Nemesia* species also differ in the length and structure of their nectar spurs and ability to produce nectar. Other environmental factors such as edaphic conditions may have equally important effects on diversification of the genus. Starting to address the question of factors driving diversification in *Nemesia*, we conducted a comparative study on the ecology and pollinator rewards of three *Nemesia* species from Namaqualand – *N. anisocarpa*, *N. macroceras* and *N. pulchella*. A phytosociological study was performed using the Braun-Blanquet cover abundance scale during the flowering season. The pollinator rewards offered by the three species was assessed using nectar volume and sugar concentration measurement as well as histology and scanning electron microscopy to compare secretory structures in the nectar spurs of the three species. Our results showed that the three *Nemesia* species are associated with different plant communities. Of the three study species, only *N. macroceras* produced sufficient quantities of nectar to be measured. This nectar secretion was associated with a dense covering of glandular trichomes at the tip of the nectar spur. These results suggest that a combination of environmental factors associated with vegetation communities and pollinators impact the diversification of *Nemesia* in Namaqualand.

Succulent Karoo vegetation change in a new conservation area of Goegap Nature Reserve, South Africa, over eleven years, including a period of severe drought

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In the arid landscapes of the Northern Cape moisture and grazing pressure are important drivers of vegetation community composition and structure. This also applies to the Succulent Karoo vegetation of the western winter rainfall region of the province. Changes in vegetation community variables were investigated over an eleven-year period (2010-2021) on new conservation land adjacent to Goegap Nature Reserve in Namaqualand. The reserve is home to an assemblage of large herbivore wildlife species. The monitoring project was initiated at the end of an above normal rainfall period, whereas the second half of the monitoring period coincided with a severe drought. A gradual influx of large herbivores occurred into the newly included conservation area after fence boundaries were removed. The aim of the monitoring project is to inform management decisions in line with the reserve's conservation objectives. The monitoring program also enables an investigation into vegetation dynamics in this winter-summer rainfall transitional section of the reserve. Annual vegetation monitoring surveys were conducted in spring along fixed line transects using the descending point method. Over the first half of the monitoring period vegetation condition and perennial plant composition remained constant. As the severity of the drought increased a reduction in overall perennial cover abundance was observed, which prompted game reduction interventions. All habitats, dominant plant species and plant growth forms experienced a reduction in cover abundance. Plains habitats were more severely impacted than rocky mountainous habitats. The study shows the importance of long-term monitoring but also uncertainty of drought impacts on vegetation in future.

The reproductive biology of the grassland endemic, *Aloe verecunda*

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Aloe verecunda is a summer-flowering grass aloe, with a disjunct occurrence along the northern rocky ridges of South Africa. The history of habitat loss and fragmentation of *A. verecunda*, along with population size declines, points to the necessity to conserve these small isolated populations. Understanding the aloes' reproductive ecology may be beneficial towards its future conservation. This study aims to determine the level of self-compatibility, primary pollinators, and predators/parasitoids of *A. verecunda*. Breeding system, pollinator exclusion, and camera trap analysis revealed that *Aloe verecunda* is self-incompatible and that pollination is primarily effected by the membrane bee, *Hylaeus heraldicus*. However, nectar volume ($27.5 \pm 1.8 \mu\text{L}$) and concentration ($15.9 \pm 0.4 \text{ \%w/w}$) are indicative of a bird pollination syndrome. Evidence of ineffective pollination by insects is seen in visitors (8.0 ± 1.44 visits/plant/hour) carrying a low pollen load (18 grains/bee). Furthermore, flowers from the open treatment produced lower seed sets of decreased viability and percentage germination compared to the hand-pollinated flowers. Insect emergence observations revealed that 18% of fruit and 30% of seeds contained predators/parasitoids; predominately the wasp, *Eurytoma aloineae*. Results suggest that *Aloe verecunda* is dependent on less effective insect pollinators in the absence of birds. Pollinators are generally less attracted to smaller populations resulting in reduced population fitness, indicating a possible Allee effect. This suggests that these small, sparse populations are vulnerable to long-term decline, and hence suitable monitoring and adaptive management should be explored.

South African health clinic gardens as models for social-ecological systems: plant diversity and potential utilization

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Gardens are essential types of green infrastructure providing multiple ecosystem services (ES) in urban and rural areas. Health clinic gardens in South Africa are gardens on government land attached to health clinics and are intended to provide fresh vegetables and fruits to patients in the community struggling to keep a healthy immune system. Government supported community gardens fulfil residents' strong preferences for vegetables and flowers in many developing countries. However, opportunity still exists to explore the plant distribution, diversity, and utilization in these types of gardens, specifically health clinic gardens, to position them as potential alternatives to other types of gardens. Consequently, this study sought to determine the floristic diversity and utilitarian plant species diversity at health clinic gardens in the Bojanala District Municipality, North-West Province, South Africa. Results showed that health clinic gardens are like home/domestic gardens in terms of dominant families, genera and species representation. They have high numbers of ornamental, medicinal, and food plants which are consistent with privately owned gardens. There was greater diversity in fruit-bearing plant species and most of the medicinal plant species can be used as a tonic. Plant species composition of micro-gardens differed to a great extent where species found in the natural area (*naga*) were mostly native and those in the ornamental micro-garden were exotic. Health clinic gardens have the potential to be alternative sources for various ES goods as they overcome many challenges faced in the establishment and management of other types of gardens. Given the envisaged purpose of health clinic gardens, future research could investigate how traditional knowledge on useful indigenous and naturalized species can be illuminated and restored to position health clinics as spaces that encourage their use, and champion their supply.

Environmental drivers of fine-scale variation in vegetation patterns in a Drakensberg grassland

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Current shifts in climatic conditions are having large ecological and socio-economic impacts, with more pronounced negative effects predicted for the future. Alpine systems, which provide key ecosystem services to large human populations, are forecast to be particularly sensitive to shifts in temperature and precipitation regimes. Several alpine ecosystems globally are co-dominated by grasses, which contribute strongly to several ecosystem services including grazing provisioning. In this study, we apply a correlative approach to fine-scale field-quantified vegetation and abiotic data collected in the Drakensberg, testing if grass assemblage characteristics are related to soil temperature and moisture after accounting for several ecophysiological-relevant soil and topography variables. Fine scale variation in total grass cover was not related to microclimatic variables, but varied with topographical position and rock cover. In contrast, the occurrence of dominant grass species was related to microclimatic, topographic and edaphic variables. Specifically, the occurrence of eight out of ten dominant grass species was correlated with microclimatic conditions, with soil moisture having a negative relationship with the occurrence of five species. Soil temperature was positively related to the probability of occurrence of five species and negatively to the occurrence of another two species. These results suggest that future changes in temperature and rainfall are likely to affect the fine-scale distribution of grass species in this system, but that species responses will also be constrained by non-climatic environmental conditions. Future research in this system will be testing the generality of these findings, specifically examining higher altitude sites.

The use of trapping structures for the housing of mutualistic microbial communities in the “carnivorous” aquatic plant *Utricularia stellaris* (Lentibulariaceae)

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It has been assumed that traps are important to *Utricularia* because of their ability to capture and digest zooplankton prey. However, studies have found that these plants may rely more on the benefits derived from mutualistic microbial communities than zooplankton prey. This study investigates whether the benefits derived from mutualistic microbial trap communities are more important to *Utricularia stellaris* than the benefits derived from capturing of zooplankton prey. Plants were collected from sites in the Eastern Cape, South Africa. The relative biomass allocation and growth patterns were determined. Trap contents and microbial trap communities were assessed. A substantial proportion of total biomass was allocated to traps and plant development levels off by node 12. A substantially greater proportion of traps contain microbial communities than zooplankton prey. The abundance and species richness of trap algae increase with trap age despite a decline in trap functionality. The high biomass investment in traps, ubiquity of trap microbial communities, and the scarcity of zooplankton prey indicate that traps are beneficial to *U. stellaris* due to benefits derived from trap microbial communities and not the benefits derived from the capture and breakdown of zooplankton prey. The stability and reproductive success of trap algal communities indicates a possible mutualism between *U. stellaris* and trap microbial communities. The hypothesis that traps are important to *U. stellaris* due to benefits derived from trap mutualistic microbial communities and not the benefits derived from the capture and breakdown of zooplankton prey is supported by the results of this study.

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Are the fairy circles the tombstones of declining populations of succulent *Euphorbia* species on sandy deposits in the pro-Namib Desert?

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Previous studies reported that the pollen quantity of *Euphorbia* species declined sharply in fossilised dung of herbivorous hyrax (*Procavia capensis*) and dassie rat (*Petromus typicus*) about 1400 years ago in the Kuiseb Valley (Namib Sand Sea area). During this time Namibia experienced periods of extreme temperatures and also reduced precipitation, which might have played a role in the decline in the populations of *Euphorbia* species in the central Namib Desert. The pollen fluctuations in some of the species/families are probably the result of climate change. Many thousands of fairy circles (FCs) and no big succulent *Euphorbia* species are currently found in the sandy areas where the midden samples were collected for the pollen analyses. It is hypothesised that these FCs were caused by populations of big succulent Euphorbias, probably *E. gummifera* and/or *E. damarana*, which died out in sandy areas because of climate change. The nearest current location of *E. damarana* is about 100 km north of the midden collection site. Analyses showed that *E. damarana* and *E. gummifera* are currently dying in alarming numbers in the lower-lying sandy areas of Brandberg and Garub (Namibia), respectively. Thousands of FCs are also found in these sandy areas.

Edaphic endemism and metal accumulation along an eroded ultramafic catena in Sekhukhuneland

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Eroded sites are regarded as degraded ecosystems which require restoration effort. In mining areas, such as the Platinum-belt in Sekhukhuneland, eroded areas are often target sites for tailings dams. However, in this region the eroded areas were formed naturally due to the underlying ultramafic geology. This process is ascribed to the 'serpentine syndrome', i.e. spatially patchy and stunted vegetation associated with soil that is nutrient poor, enriched in heavy metals and magnesium, and with low water-holding capacity. Lack of vegetation cover and a dispersive soil structure explain the prevalent erosion. This raises the question whether the associated vegetation is adapted to this harsh environment. We propose that the presence of any endemic species or metal accumulators would indicate local scale adaptation to this unique ecosystem. We analysed soil and vegetation data of more than 100 sample plots from the Steelpoort River valley. Results confirmed high metal content and natural dispersive properties of the soil, bioaccumulation of metals by plants, presence of 21 Sekhukhuneland endemics of which five were restricted to the naturally eroded areas, and stress-tolerant indigenous species that are suitable for phytoremediation of mine sites and tailings. This is the first study to highlight the need to protect and maintain a naturally degraded savanna ecosystem to ensure the persistence of rare and threatened species of value to green technologies.

Composition changes over the past 20 years in the grass sward of Thornbush Savanna of Central Namibia

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The thornbush savanna of Central Namibia is dominated by a mixed low tree and shrub layer of various *Acacia* species, mainly *Senegalia mellifera*. The grass sward consists of a variety of species, both perennial and annual, with *Stipagrostis uniplumis* and various species of *Eragrostis* dominating. Bush encroachment is regarded as a major form of land degradation in this landscape. Two long-term biodiversity observatories, being Omatako Ranch (game farm) and Erichsfelde (cattle farm), have been established in 2001 in this Thornbush savanna to monitor long-term trends in species composition in relation to land use and global climate change. Species composition has regularly been determined on fixed sample plots within these observatories. There was no evidence of bush thickening ('bush encroachment') over the past 20 years; comparison of aerial imagery from 1968 to present demonstrated a very stable shrub population. Yet the composition of the grass sward gradually changed from dominated by perennial *Stipagrostis uniplumis* to annual *Aristida rhiniochloa*. Broad-leafed palatable perennial grass species like *Schmidtia pappophoroides* and *Brachiaria nigropedata* have become rare, possibly even locally extinct, on these biodiversity observatories. The trend is faster on the game farm compared to the cattle farm. Even drastic destocking during drought years did not help in the regeneration of perennial grass species. The degradation is linked to a prolonged drought period starting in 2013, but in the long term also to increased aridity due to increased temperatures and subsequent higher evapotranspiration.

Composition and status of encroaching woody plant species along riparian zones of the Molopo River, North-West Province, South Africa

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Woody plant encroachment has become increasingly problematic in the arid and semi-arid ecosystems with the potential to pose serious threats to natural vegetation composition and structure. Although several studies have documented the structure of woody plants in a riparian zone, little attention has been given to quantifying woody plant encroachment along the Molopo River. This study was conducted to assess the composition and structural patterns of woody species in the encroached and benchmark sites situated along the Molopo River near the Tshidilamolomo, Makgori and Loporung villages. Three 20 x 20 m plots were evenly laid out along five 200 m transects spaced 30 m from each other to best represent woody vegetation variability at each site. Woody species were recorded and categorized into six height classes and tree densities were calculated. A total of ten woody species were identified, but not all were a manifestation of woody encroachment. However, *Prosopis velutina* and *Senegalia mellifera* with a cover above 40% and densities exceeding 2 000 TE ha⁻¹, were considered as woody encroachment in the study sites. With regard to woody density, there were significant differences between the encroached and the benchmark sites. Woody species near the Makgori and Loporung villages, exhibited an inverted J-shaped distribution. While in an encroached site near the Tshidilamolomo village, plant species dominated by the invasive alien *P. velutina* revealed a bell-shaped pattern. Knowledge on the composition and structure of woody plant species near the three villages proves to be a relevant indicator of the ecological status of riparian ecosystems that could be used to identify high-risk areas, which may provide future opportunities for reducing and managing invasions.

Ethnobotany

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Antibiofilm, antioxidant and anti-inflammatory activity of selected indigenous South African plants used in the treatment of diarrhoea.

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Biofilms are complex structures formed when bacterial colonies group together within an extracellular matrix, providing protection and aiding antimicrobial resistance. Antioxidants protect cells against free radicals. Inflammation is a biological response mechanism to harmful stimuli. Macrophages contribute to the initiation and resolution of inflammation by producing cytokines. Medicinal plants have proven to be a good source of biologically active compounds. This study was designed to determine the aforementioned biological activities of selected *Bauhinia* and *Searsia* species. The crystal violet method was deployed to establish the antibiofilm activity against *Escherichia coli* (ATCC 25922). Antioxidant activity was determined using an *in vitro* assay with 2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) and 2, 2-diphenyl-1-picrylhydrazyl (DPPH). Anti-inflammatory activity was determined via inhibition of nitric oxide (NO) production in lipopolysaccharide (LPS)-activated RAW 264.7 macrophages and 15-lipoxygenase (LOX) inhibitory assays. The percentage of biofilm development inhibition of all the tested plants was above 50%. *Bauhinia bowkeri* had the best antibiofilm activity. However, the plants were not effective against pre-formed biofilms. The antioxidant values ranged from 0.74 to 36.45 µg/ml and 14.313 to 532.86 µg/ml for the ABTS and DPPH assays, respectively. *Searsia batophylla* had the best antioxidant activity and anti-inflammatory activities. All the plant extracts inhibited NO production to varying degrees in a concentration-dependent manner. *Searsia batophylla* had the best NO inhibition of 74.79%, with no adverse effects on cell viability. Therefore, the selected plant extracts have good antioxidant and anti-inflammatory activities and prevent biofilm formation. They were, however, not able to destroy pre-formed biofilms.

Phytochemical, antioxidant and functional group analysis of South African *Evolvulus alsinoides* (L.) L.

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Evolvulus alsinoides (L.) L. is a forb species that is highly valued in traditional medicine for its memory-enhancing effects. The present study was designed to investigate the aqueous and ethanolic extracts of South African *E. alsinoides* using phytochemical and antioxidant assays. The phytochemical contents were determined using spectrophotometric and colorimetric methods while the antioxidant activities were determined using *in vitro* antioxidant (DPPH, ABTS, and FRAP) assays. Additionally, the functional groups present in the extracts were analyzed using FTIR spectroscopy. Results revealed that the ethanolic extracts of *E. alsinoides* gave the highest content of total phenol (17.07 ± 2.28 mg GAE/ g extract), total flavonoid (82.44 ± 0.93 mg QE/g of extract), and proanthocyanidin (32.98 ± 2.10 mg CE/g of extract). The total alkaloid and saponin quantification of the crude extract of *E. alsinoides* gave mean percentage values of $2.6 \pm 0.2\%$ and $33.7 \pm 0.00\%$ respectively. The ethanol extract had the highest DPPH scavenging ability (IC_{50} value: 0.4367mg/mL), ABTS scavenging ability (IC_{50} value: 0.099 mg/mL) and the best ferric reducing antioxidant power (138.16 ± 3.27 mg TE/ g). The total phenolic, flavonoid, and proanthocyanidins contents were highly and positively correlated with DPPH and ABTS activities confirmed by Pearson correlation analysis. Spectral data of FTIR analysis of *E. alsinoides* aqueous and ethanolic extracts revealed the presence of functional groups such as O–H, –OH, C=O, and C≡C. The results of the study justify the usage of *E. alsinoides* in folkloric medicine and can be explored further for pharmaceutical plant-based products.

Molecular identification of medicinally important *Greyia* species and investigation of a white variant's anti-tyrosinase activity

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The taxonomic identification of medicinally important species within the *Greyia* genus is an ongoing debate. Previous studies have found extensive overlap between the morphology and metabolic profiles of *Greyia sutherlandii* and *Greyia radlkoferi*. The ethanolic leaf extract of wild-type red flower plants exhibits anti-tyrosinase activity and is being developed into a cosmeceutical product to treat hyperpigmentation. Therefore, it is essential to accurately identify the species grown in orchards to prevent adulteration. The anti-tyrosinase activity of rare white flower variants were investigated. DNA barcoding targeting the internal transcribed spacer (ITS) region was used for molecular identification, after which a colorimetric enzyme assay was performed to determine the tyrosinase inhibitory activity. DNA barcoding results indicated that selected *Greyia* plants were identified incorrectly using morphological traits. ITS sequences from both *G. radlkoferi* and *G. sutherlandii* were present in some plants. Our working hypothesis is that there was a previous hybridization event, followed by concerted evolution in the ITS region. The anti-tyrosinase activity of cultivated white flower- *G. radlkoferi* plants were found to be higher (IC₅₀=61.96 µg/ml) than that of the tested red flower- *G. radlkoferi* (158.69 µg/ml). White and red flowered trees growing in the same environment did not have significantly different activities (30.94 µg/ml and 32.27 µg/ml, respectively). The study concluded that the anti-tyrosinase activity is dependent on environment and plant age, and not a mutation abolishing the red pigment in leaves (upon senescence) and flowers. Future work will include plastid gene barcoding, and a greater diversity of plants will be screened.

Ethno-medicinal use and antimicrobial activity of potential plant extracts against pathogens causing ear infections

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Medicinal plants are used for the treatment of ear infections in South Africa and across the globe. An ear infection is a common disease caused by bacteria, fungi and viruses in the middle ear. The study aimed to investigate medicinal plants used for the treatment of ear infections in Bushbuckridge Local Municipality, Mpumalanga Province by the local people and traditional health practitioners. In this study, plant materials were extracted with various solvents such as acetone, hexane, methanol, and water. Antifungal activity of eight selected medicinal plants (*Carpobrotus edulis* (L.) L.Bolus, *Cotyledon orbiculata* L., *Sansevieria hyacinthoides* (Linn) Druce and *Carissa edulis*, *Dichrostachys cinerea* (L.) Wight & Arn., *Erythrina lysistemon* Hutch., *Psidium guajava* L., *Ricinus communis* L.) were determined against *Aspergillus fumigatus* and *Candida albicans*. Furthermore, *in vitro* antibacterial activity of the plant extracts was investigated against *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas aeruginosa*. Of eight plant species, acetone extracts *C. edulis* and *S. hyacinthoides*, *D. cinerea* and *S. hyacinthoides* had excellent antifungal activity against *A. fumigatus* and *C. albicans* with MIC values of 0.02 and 0.08 mg/ml. All plant extracts were active against the tested bacteria. Chemical components of different plant extracts were investigated using thin-layer chromatography (TLC). Different coloured bands were observed on TLC chromatograms, indicating the presence of various compounds in the plant extracts. Therefore, the study supports the traditional use of this plant to treat ear infections and its potential use in antimicrobial drug production.

The ethnobotany, taxonomy and anatomy of the genus *Plecostachys* (Asteraceae)

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Plecostachys is a genus of two species distinguished by their tangled growth habit and small, sericeous leaves. The genus is endemic to South Africa and belong to the Asteraceae. Ethnobotanical information is only available for one species (*P. serpyllifolia*). The present study aimed at documenting the taxonomy and ethnobotany of *Plecostachys* and to also explore the anatomy of the genus along with that of the genus *Tenrhynea* (sister taxon) and *Anaxeton* (outgroup). Published literature with related taxonomic and ethnobotanic content of *Plecostachys* were explored and some of the PRE herbarium specimens were studied for interpreting morphological descriptions. The stem, leaf and inflorescence of both the *Plecostachys* and *Tenrhynea* species were anatomically explored to compare some potentially useful anatomical features. Studied anatomical sections revealed potential similarities between *Plecostachys* and *Tenrhynea*. *Plecostachys serpyllifolia* is used to treat various respiratory ailments and is taken as an infusion. Unpublished ethnobotanical data show that *Tenrhynea phyllicifolia* has been used as a tea in the Limpopo Province. Future studies should focus on the anatomy, essential oil chemistry and ethnobotany of the entire group, including *Plecostachys polifolia* and *Tenrhynea phyllicifolia*.

***Capsicum* extracts with the potential to inhibit skin cancer growth and proliferation**

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Melanoma is the 19th most common cancer worldwide, with an estimated 300,000 cases diagnosed globally each year. In South Africa, approximately 11,784 cases were reported between 2005 and 2013. This study evaluated the antiproliferative potential of seventeen *Capsicum* extracts and three capsaicinoids against melanoma (A375). Four plant extracts showed reduced A375 cell viability at 400 µg/mL namely, ethanolic extracts of *Capsicum annuum* 'Serrano' (13.65 ± 3.58%), *C. frutescens* 'Mayhem' (4.09 ± 1.03%), and *Capsicum*. sp (4.16 ± 16.90%) and one sequential aqueous extract of *C. annuum* 'Jalapeno' (21.16 ± 7.30%). No antiproliferative activity was observed against human keratinocytes (HaCat), with fifty percent inhibitory concentrations (IC₅₀) >400 µg/mL. Capsaicin (CAP), dihydrocapsaicin (DHC) and nordihydrocapsaicin (nDHC) showed IC₅₀ values of 97.6 ± 9.80, 78.2 ± 8.01, and 175.2 ± 3.78 µg/mL against A375 cells and 82.9 ± 7.02, 77.4 ± 7.28, and 149.6 ± 4.58 µg/mL against HaCaT cells. Quantitative thin layer chromatography (TLC) confirmed the presence of these capsaicinoids in ten extracts. Capsaicinoids and extracts which reduced A375 cell viability were investigated for inhibitory activity against melanotransferrin (MFI2), a protein which is upregulated in melanoma cells and plays a pivotal role in proliferation and migration. No statistically significant difference in the production of MFI2 was detected in A375 cells treated with capsaicinoids or extracts, compared to the untreated control, however, the capsaicinoids showed an inhibition of MFI2 whereas the extracts showed an increase of MFI2 production. These results correlate with previous literature which reported the inhibitory activity of capsaicinoids against melanoma while *Capsicum* species showed a stimulation of melanoma proliferation. Future considerations should be to evaluate the antiangiogenic and anti-metastatic potential of capsaicinoids as well as different markers of angiogenesis.

Antimycobacterial effect of encapsulated polyherbal poly-(lactide-co-glycolide) nanoparticles' on *Mycobacterium tuberculosis*

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Plants have shown to be great additives to existing treatments as adjuvants that may assist the body in fighting against disease. The current study investigated the evaluation of adjuvants that may show potential for tuberculosis (TB). This study investigated the ethanolic extracts of five indigenous South African plants, *Lippia scaberrima* Sond., *Salvia aurea* L., *Barleria repens* Nees, *Sphedamnocarpus pruriens* (A.Juss) Szyszyl, and *Euclea natalensis* A.DC., alone and in a polyherbal combination, to determine and compare whether the polyherbal formulation may have enhanced activity. Furthermore, poly-(lactide-co-glycolide) (PLGA) nanoparticles were formulated for all plants, alone and in combination and compared to the ethanolic extracts made. The polyherbal formulation as well as the polyherbal's nanoparticle did not exhibit antimycobacterial activity (1000 µg/mL) against *Mycobacterium smegmatis* (*M. smegmatis*) at concentrations ranging from 15.62 to 1000 µg/mL. However, the polyherbal and its respective nanoparticle formulation might still potentially show activity against *Mycobacterium tuberculosis* (*M. tuberculosis*) as previous results indicated that the plants in the polyherbal formulation had significant antimycobacterial activity against *M. tuberculosis* (*Euclea natalensis* and *Lippia scaberrima* both had minimum inhibitory concentration (MIC) values of 125 µg/mL, *Salvia aurea* had a MIC of 31.25 µg/mL, and *S. pruriens* a MIC of 62.5 µg/mL). Initial studies have also shown that two plants, *S. pruriens* and *L. scaberrima*, had no cellular toxicity against hepatocellular carcinoma (HepG2) cells, which further suggests that the polyherbal and its subsequent nanoparticle formulation may have enhanced antimycobacterial activity and less toxicity.

Phytochemical profile of the extracts of *Bidens pilosa* and its correlation with cell proliferation, antioxidant, and larvicidal activity

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Biden pilosa has been traditionally used in foods, medicines and in agriculture as biopesticides, without the report of obvious adverse effects. The aim of this study was to monitor the cytotoxic effects using real-time cell analysis (RTCA), antioxidant and the larvicidal activities of *B. pilosa* (water and 70% ethanol extracts) and to correlate these activities with the phytochemical profiles using a Q-TOF mass spectrometer (Waters Synapt G2). The quantitative estimate of the phenolic compounds in the crude extracts was done with LCMS based methodology using 15 standards and compared with the HPLC measurements. Both extracts exerted cytotoxic activity through dose-dependent impairment of human liver, HepG2, (cancerous) and African monkey kidney, Vero, (non-cancerous) cell lines. However, the ethanol extracts expressed higher cytotoxic effects and hence were considered toxic to the cell lines. The larvicidal activity also followed a similar pattern to the cytotoxicity as the ethanol extracts showed stronger potency. The ethanol extracts expressed more pronounced antioxidant, cytotoxic, and larvicidal activities, probably due to the presence of a higher concentration of phytochemicals as shown in its phytochemical profile in the study. Because *Biden pilosa* is rich in bioactive compounds and minerals, the use of ethanol as an extracting solvent could yield better results than water when the plant is used traditionally or for other purposes.

An Ethnobotanical study of indigenous knowledge and uses of medicinal plants by traditional healers in the rural communities of Nkomazi Local Municipality, Mpumalanga province

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In South Africa, traditional healers are the custodians of invaluable knowledge which is valuable to their indigenous communities. The traditional way of life in villages is rapidly transforming due to economic development, western education, urbanization, and climate change. These factors could lead to the degradation of important indigenous knowledge on the use of medicinal plants. The available research on the traditional use of medicinal plants by the rural communities of Mpumalanga is highly fragmented and under-researched. A detailed account of the medicinal plants and associated indigenous knowledge for the Nkomazi Local municipality is not available. Therefore, the present study aimed to investigate aspects related to the collection, legislation, and utilisation of medicinal plants by traditional healers at Nkomazi Local Municipality, Mpumalanga province. Semi-structured interviews and questionnaires were used to collect ethnobotanical data from 10 traditional healers. The results showed that a total of 140 medicinal species, which belong to 84 families are used by traditional healers to treat various ailments. Roots were the most used parts, accounting for 45% of parts used. The majority of the traditional healers depend on herbalists on street markets for accessing some medicinal plants such as *Vepris reflexa* and *Combretum imberbe*. The data showed that the majority of the traditional healers had limited knowledge of the national environmental legislation. The present study has successfully recorded essential ethnobotanical information on medicinal plants used by traditional healers in Nkomazi Local Municipality in the Mpumalanga province. Therefore, the study could contribute towards the preservation the Indigenous knowledge.

Variation in biological activity of *Ptaeroxylon obliquum* collected in two different geographic regions of South Africa

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Ptaeroxylon obliquum (Thunb.) Radlk. (Rutaceae) is traditionally used to treat microbial and inflammatory diseases in humans and animals. *Ptaeroxylon obliquum* leaves were collected from Nelspruit and Pretoria and extracted using water and organic solvents. Antifungal and antioxidant activity as well as toxicity to cancerous and normal cells was evaluated. Antimicrobial activity was determined against three fungal clinical isolates (*Aspergillus fumigatus*, *Candida albicans* and *Cryptococcus neoformans*). All leaf extracts had MIC values ranging from 500 to 2500 µg/ml with acetone extracts being most active. Solvent-solvent fractionation was used to fractionate acetone extracts of both leaf samples based on polarity. The chloroform fractions (MIC = 310 µg/ml) had the best activity. Radical scavenging activity of the extracts was tested using 2, 2-diphenyl-1-picrylhydrazyl (DPPH) and 2, 2'-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS) assays. Cytotoxicity was tested against Vero cells while anti-cancer activity was tested against human breast cancer (MCF-7), hepatocarcinoma (HepG2) and lung adenocarcinoma (A549) cells using a colorimetric assay. Extracts and fractions scavenged DPPH and ABTS radicals with IC₅₀ values as low as 46.07 and 29.06 µg/ml respectively. Water extracts had the best scavenging activity. The acetone extract and chloroform fraction from trees growing in Pretoria were toxic to Vero cells. The extracts and fractions were not toxic to the cancer cells with the exception of the chloroform fraction from Nelspruit which was toxic to HepG2 cancer cells with LC₅₀ of 13.60 µg/ml. Results demonstrated clear differences in the activity of the leaf extracts based on their geographic origin.

Antiplasmodial properties of extracts from *Pappea capensis* Eckl. & Zeyh. (Sapindaceae)

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Pappea capensis, traditionally used to treat malaria by VhaVenda people of South Africa was subjected to biological screening. Ground twigs were extracted using dichloromethane: 50% methanol (1:1, v/v), separated and dried, yielding dichloromethane (I) and aqueous (II) extracts. Extract I was partitioned using dichloromethane: methanol (1:1, v/v) and dichloromethane (III) and methanol (IV) extracts were recovered. A water-based decoction (V) was also prepared. Extracts II, III, and IV were subjected to silica column chromatography using different solvents (hexane: dichloromethane, dichloromethane: ethyl acetate, ethyl acetate: methanol, and methanol: water) with increasing polarity. *In vitro* antiplasmodial tests using *Plasmodium falciparum* (NF54) and cytotoxicity screening on L-6 rat, skeletal cells were performed on extracts I-V and fractions A-Y. Their respective selectivity indices (SI) were calculated. Of all the assayed extracts, II, III, and IV demonstrated significant activity ($IC_{50} \leq 5 \mu\text{g/ml}$) and high selectivity indices ($SI \geq 10$). From the fractions only D, E and M showed significant antiplasmodial activity less than $1 \mu\text{g/ml}$ and $SI > 10$. Chemical profile analysis using ^1H NMR and GC-MS spectroscopy displayed a prevalence of terpenoids, and tentatively identified lupeol and β -amyryn triterpenoids with can be attributed to the observed activity. NMR-based metabolomics on the fractions successfully predicted antiplasmodial activity by showing a clear clustering pattern among the selective fractions. The study has partly validated the ethnomedicinal use of *P. capensis* for malaria treatment. It also demonstrates the great potential of discovering novel antiplasmodial constituents that could serve as potential drug hits.

Antioxidant and *in vitro* antifungal activities of selected traditional plants used in ethnomedicine

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Indigenous knowledge based on medicinal plants is lacking worldwide due to the ever-increasing dependency on modern western medicine. Twelve selected medicinal plants (*Kleinia longiflora* DC. *Berchemia discolor* (Klotzsch) Hemsl., *Persea americana* Mill., *Sansevieria hyacinthoides* (L.) Druce, *Dichrostachys cinerea* (L.)Wright & Arn, *Withania somnifera* Dunal (Ashgandh), *Momordica balsamina* L., *Lonchocarpus capassa*, *Pappea capensis*, *Rhus lancea* L. fil, *Peltophorum africanum*, *Maytenus heterophylla* (Eckl. & Zeyh.) Robson) were investigated for antifungal activities using the microdilution method against *Candida albicans*, *Cryptococcus neoformans* and *Aspergillus fumigatus*. The qualitative 1,1-diphenyl-2-picrylhydrazyl (DPPH) method was used to determine the antioxidant activities of plant extracts. Plant extracts of *P. americana* and *D. cinerea* had exhibited good activity against *C. albicans* and *A. fumigatus* with MIC values of 0.02 and 0.08 mg/ml. In bioautography assay, active compounds were observed in TLC chromatograms developed Benzene: ethanol: ammonia hydroxide (BEA) R_f values of 0.44 and 0.58 against *C. neoformans*. The acetone and DCM extracts of *S. hyacinthoides* were the most active radical scavenger in the DPPH assay. The results support the use of these plants by traditional health practitioners and the local people for the treatment of various ailments related to fungal infections using water extracts.

Antibiofilm, antioxidant and anti-inflammatory activity of selected ethno-veterinary plants used in the cure of veterinary dermatological infections

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Biofilms are multifaceted structures formed when bacterial colonies group together within an extracellular medium, providing antimicrobial resistance. Antioxidants protect cells against free radicals, which are implicated in various pathological conditions. Inflammation is a biological response mechanism to harmful stimuli. Therapeutic plants have high quality resources of antioxidant, anti-inflammatory and antibiofilm compounds. This investigation aimed to establish antioxidant and anti-inflammatory activities, as well as the ability of six selected plants to inhibit biofilm formation. The plant species, namely Elephantorrhize elephantine, Eucalyptus camaldulensis, Aloe marlothii, Euphorbia tirucali and Musa acuminata are used in ethnoveterinary medicine to treat dermatological conditions. A modified crystal violet method was deployed to establish the antibiofilm activity against *Staphylococcus epidermidis* ATCC 35984, a known biofilm-former. In vitro assay of 2, 2'-azino-bis(3-ethylbenzthiazoline-6-sulfonic acid) (ABTS) and 1,2 -diphenyl-2-picrylhydrazyl (DPPH) were used to determine the antioxidant activity of the plants. Inhibition of nitric oxide (NO) production in lipopolysaccharide (LPS)-activated RAW 264.7 macrophages was used to estimate the anti-inflammatory activity. The percentage biofilm development inhibition of all the tested plant extracts were above 50. Eucalyptus camaldulensis and Musa acuminata had the best anti-inflammatory activity. All the plant extracts inhibited NO production to varying degrees in a concentration-dependent manner. Musa acuminata had the best NO inhibition of 98.29% with no adverse effects on cell viability at a concentration of 100 µg/ml. Therefore, the chosen plant extracts had good antioxidant, anti-inflammatory activities, but few were able to destroy pre-formed biofilms. A traditionally prepared ethnoveterinary remedy for dermatological ailments has better activity than single plant extracts.

Traditional medicine as an alternative source for the treatment of dental diseases in Limpopo Province

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The application of plants to treat dental diseases is being practiced by the local people and traditional health practitioners in Limpopo Province. Indigenous knowledge regarding the specific plants used for the treatment of various diseases and the methods of preparation is passed from generation to generation. An ethnobotanical survey was conducted to identify plant species used for the treatment of oral diseases and related ailments in Mopani, Capricorn and Vhembe districts. The local people and traditional health practitioners were randomly selected via the snowball method. Furthermore, a questionnaire was designed to gather information on the common names, plant parts used, preparation and administration. The study revealed that more than fifty plant species belonging to different plant families were identified to be used by the local people and traditional health practitioners for the treatment of dental diseases in humans. The dominant families were Euphorbiaceae and Rutaceae. The most frequently used plant species were *Solanum elaeagnifolium* Cav., *Carica papaya* L. and *Peltophorum africanum* Sond. Specific parts of the plant used for medicinal purposes vary from species to species and from one traditional health practitioner to another. The mode of administration of medicine was mainly oral. The observed transfer of indigenous knowledge to younger generations was an important practice for preserving useful medicinal plants.

Phenolic characterization of *Protea* pure and hybrid cultivars by liquid chromatography-ion mobility-high resolution mass spectrometry (LC-IM-HR-MS)

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In this study we report the phytochemical composition of *Protea* pure and hybrid cultivars. Aqueous extracts of leaf and bract tissues were analyzed by ultra-high pressure liquid chromatography hyphenated to photodiode array and ion mobility-high resolution mass spectrometric (UHPLC-PDA-IM-HR-MS) detection. Metabolites identified based on their relative reversed phase (RP) retention, UV-Vis spectra, low and high collision energy HR-MS data and collisional cross section (CCS) values included 41 phenolic acid esters and 25 flavonoids, including 5 anthocyanins (delphinidin-*O*-hexoside, cyanidin-*O*-hexoside, petunidin-*O*-hexoside, peonidin-*O*-hexoside, and malvidin-*O*-hexoside). A new hydroxycinnamic acid-polygalatol ester, caffeoyl-*O*-polygalatol (1,5-anhydro-[6-*O*-caffeoyl]-sorbitol(glucitol)), was isolated and characterized by 1D and 2D NMR. This compound and its isomer are shown to be potential chemo-taxonomic markers. Metabolomics tools were used to identify metabolites linked to the post harvesting disorder, leaf blackening, observed in *Protea* species, selections, and cultivars. Metabolites which comprised of benzenetriol- and hydroquinone-glycoside derivatives (such as benzoyl-hexoside of hydroxyquinol, and arbutin (hydroquinone-*O*-hexoside), respectively) were found to be associated with the instigation of the blackening process. On the other hand, species, selections, and cultivars not prone to blackening presented metabolites with known protective properties, such as protocatechuic acid derivatives, vanillic acid-*O*-hexoside, feruloyl-*O*-hexoside, and flavonol derivatives. Lastly, 4 out of the 5 anthocyanins detected (delphinidin-*O*-hexoside, petunidin-*O*-hexoside, peonidin-*O*-hexoside, and malvidin-*O*-hexoside) were distributed in pure species, selections, and cultivars with black-bearded (tufted) flowerheads, suggesting the presence of these anthocyanins in the tufts. The findings from the study, may provide preliminary insights to guide the development of new *Protea* cultivars not susceptible to leaf blackening.

An overview of South African SMMEs contribution to the expanding global cosmeceutical market

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Cosmetics industry generates billions of dollars as new advanced and specialized products enter the market. The global skincare market was valued at US\$ 98.83 billion in 2020 and was projected at US\$ 100.13 billion for 2021. Among the main cosmetic market contributors, Asia-Pacific, Latin America, Middle East, and Africa are considered as the prominent growth markets. In South Africa, the upsurge in the cosmetics market is due to consumer demand for new and innovative products of natural origin. Consumer desire for herbal-based cosmeceuticals has seen a number of South African SMMEs entering the industry. This is a relatively new untapped domain for South African companies to contribute to the economy. Thus, we aim to provide a critical assessment of some of the progress made in South African herbal cosmeceuticals with emphasis on online companies. Local brands (e.g. Portia M, Crede Natural Oils, Africa Organics and Isivuno Naturals) have been well-received by consumers. This is possibly due to several factors such as the use of well-known indigenous African plants (aloe, baobab, marula, rooibos, etc.), formulation of skin and haircare products that cater for the diversity of South Africans, and development of products without the use of parabens and synthetic chemicals. Lastly, the shift to online shopping has made it relatively easy for consumers to access products while allowing SMMEs to promote their products. Based on the increasing number of new products, the cosmetics sector can contribute to the economy, job creation, entrepreneurship skills, socio-economic development and intellectual property generation.

An inventory and analysis of medicinal plants use in the Ixopo area of KwaZulu-Natal, South Africa

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The aim of the study was to record and analyze the patterns of plant use in three villages in Ixopo area under the Ubuhlebezwe Local Municipality, in KwaZulu-Natal, South Africa. Matrix methods was used to guide question and analyze data to understand species popularity index (SPI) and ethnobotanical knowledge index (EKI), further analysis was done using regression analysis and homogeneity index (HI) to understand the relationship between EKI and gender or age, and to understand similarities between the villages. The flip-file of photo plates, re-created from the one used at Amandawe but with a selection of 143 plants, based species popularity index values of 0.87 and above, and this flip-file was used for the interviews in the Ixopo area. Amandawe is the village under Umdoni Municipality in KwaZulu-Natal province of South Africa, and it is the neighboring Municipality to Ixopo area. This talk will be limited to documented medicinal plant use. The total of 200 ethnobotanically relevant species have been recorded for various uses, and 133 species were recorded as medicinal plants. In terms of medicinal plant species popularity, the results show the SPI value of 0.47. The average EKI values of men and women respectively were 0.45 and 0.48 for medicinal plants, and the overall EKI value of 0.47. Age is correlated with the level of knowledge (p-value <0.05) but the low R^2 value of 0.2381 show that the model has a low predictive value and that factors other than age have a strong influence.

Ethnoveterinary study of medicinal plants used for treating and managing cattle diseases on Nkomazi Local Municipality, South Africa

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Farmers frequently use a combination of ethno-veterinary knowledge and contemporary veterinary medicine to cure their cattle's. The sustainability of livestock production in local communities is continuously under the threat due to different types of diseases. The current study documented the medicinal plants used to treat various cattle diseases in Nkomazi local municipality, Mpumalanga province, South Africa. The ethnobotanical data was collected using semi-structured interviews (face to face). The collected data was presented descriptive data and ethnobotanical indices such as frequency of citation and Use Value (UV). In the study area, 23 plant species belonging to various families were currently used for their curative purposes. *Cissus quadrangularis*, *Euphorbia cupularis* and *Acacia mearnsii* were the most cited medicinal plants, respectively. *Acacia mearnsii* (0,08) and *Ceratonia siliqua* had the highest (UV). Maggots (30%) was the most frequently cited cattle disease treated by *Cissus quadrangularis*. The plant parts commonly preferred in the preparation of remedies for various cattle diseases were stem (40%), followed by bark (16%), and flowers (1%) being the least used plant part. Moreover, methods of preparation that were described by the participants were, poultice (62%), Decoction (22%), Maceration (11%) and infusion (5%). Majority of the plants (64%) were administered orally. This study indicates that a wide range of remedies are used by farmers and herder for the treatment of several complications and problems associated with cattle issues. There is a need for scientific evaluations such as antimicrobial activities to prove the empirical use of these plants in the improvement of cattle diseases as well as validating their traditional uses.

***In vitro* antioxidant, anti-inflammatory, and wound healing potential of *Pentanisia prunelloides* and *Dicoma anomala* polyherbal extract**

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Pentanisia prunelloides and *Dicoma anomala* concoction is used in South Africa to treat wounds. However, no scientific evidence exists on the efficacy of this concoction as a wound healing agent. Consequently, the study sought to evaluate the antioxidant, anti-inflammatory and wound healing potential of this concoction as a first step towards its scientific validation as a wound healing agent. Assays 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity, nitric oxide (NO) radical scavenging activity, ferric reducing antioxidant power (FRAP), and β -carotene-linoleic acid model system (CLAMS) estimated the antioxidant activities whilst xanthine oxidase, cyclooxygenase-1 and -2, and 15-Lipoxygenase enzyme assays estimated the anti-inflammatory activities. The Adenosine triphosphate (ATP), lactate dehydrogenase (LDH) and scratch assay were respectively used to measure the effect of this herbal concoction on WS1 human skin fibroblast cells viability, cytotoxicity, and migration *in vitro*. Polyphenols were also quantified. The concoction displayed significant antioxidant, free radical scavenging, and cyclooxygenase activities. Results from the ATP luminescence assay displayed good cellular viability after 24 and 48 h exposure. The extract displayed no signs of toxicity towards the WS1 cells at concentrations 6.25, 25, and 50 $\mu\text{g}/\text{mL}$. Cells exposed to the extract displayed migration rate and percentage wound closure comparable to that of normal wounded control after 48 h, and complete wound closure was observed. The findings from the *in vitro* wound healing study support the folkloric use of this plant combination as a wound healing agent. However, further studies elucidating the extract's molecular mechanism of wound healing are of significance.

The ethnobotany of Zulu ritual plants recorded in the Amandawe area, KwaZulu-Natal

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The ultimate goal of the full ethnobotanical survey conducted in the area of Amandawe in KwaZulu-Natal was to scientifically document and quantify Zulu ethnobotany. Both formal and informal interviews were used with the aid of a flip-file to gather relevant ethnobotanical information. With the aid of the Matrix Method (De Beer and Van Wyk, 2011), data was quantified and communicated in three indices namely Species Popularity Index (SPI), Ethnobotanical Knowledge Index (EKI) as well as Cultural Importance Index (CII). In total, 338 plant species were recorded to have ritual uses. Fabaceae (28 species) and Asteraceae (24) are the two most species rich families recorded. Overall, ritual plants were grouped into psychological (93 species) and spiritual (45). Psychological rituals were further categorised based on uses into protective (145 species), negative (137), positive (85) and others (36). Spiritual uses were categorised into enhance (47), placatory (33), burials (25), dreams (15), warding off (14) and others (37). The most important ritual plant is *Helichrysum odoratissimum* (SPI=1.00), while the most culturally important species is *Cymbopogon excavatus* (CII=11.93). Average ethnobotanical knowledge recorded has an index value of (EKI=0.46). A total of 146 different individual uses were recorded and the most use-diverse species is *Vangueria infausta* (17 uses). Certain plant signatures such as morphology and chemistry are instrumental in facilitating the success of placebo effect in the use of ritual plants. It is highly recommended that similar ethnobotanical studies with emphasis on ritual plants be explored for the improvement of understanding and for preserving cultural heritage.

An inventory and analysis of medicinal plants used by the Bapedi in the Limpopo Province, South Africa

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The Bapedi are a South African ethnic group occupying the Sekhukhune, Capricorn and Waterberg Districts within the Limpopo Province. The Bapedi traditional healthcare system has not yet been fully documented. Dr Sebua Semanya and co-workers have made a substantial contribution to our knowledge on the Bapedi traditional medicine with attention given mainly to Bapedi traditional healers. Mogale et al. (2019) published an ethnobotanical survey conducted in Central Sekhukhuneland, also known as “*the heartland of the Bapedi*”, focusing on the Bapedi local Sekhukhune community. The results indicated a total of 107 new medicinal plant use records. A medicinal plant checklist, based on published records from various references and my own records obtained from ethnobotanical surveys, indicates that there are currently 286 plant species known to have medicinal uses to the Bapedi. The major ailments treated by the Bapedi include respiratory (121), blood related (87) and gastrointestinal ailments (56). Other ailments treated by Bapedi include fertility complications (33) skin infections (17) and “other” complications (69) e.g., fatigue, toothaches, body aches, epilepsy and several others. A comparative analysis showed similarities and differences between traditional healers and community members, including vernacular names, choice of plant species in traditional treatments and methods of administration. The occurrence of new records shows the need for more quantitative ethnobotanical studies in other areas occupied by the Bapedi, not only to better understand the Bapedi traditional healthcare system but also to ensure the preservation of Bapedi traditional medicinal knowledge.

Effects of elevated CO₂ on the phytochemical profile, antioxidant and antimicrobial activities in *Moringa oleifera* Lam. and *Moringa peregrina* Forssk: A comparative study

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Atmospheric CO₂ concentration has been increasing at an alarming rate due to anthropogenic activities. The increase in CO₂ could affect medicinal plants such as *Moringa oleifera* and *Moringa peregrina*. *M. oleifera* and *M. peregrina* have medicinal and nutritional properties, and economic value in South Africa. In the present study *M. oleifera* and *M. peregrina* plants were subjected to short term elevated CO₂ concentration treatments such as 400 ppm, 600 ppm and 800 ppm up to 3 months. The three-antioxidant assay conducted include DPPH, H₂O₂ and Metal chelating. The antimicrobial tests were done on *E. Coli*, *S. aureus* and *S. albulus* using agar well diffusion. The leaves were collected to perform phytochemical screening, antioxidant and antimicrobial assays. The results showed an increase in phytochemical accumulation at 600 ppm in both species, with higher accumulation of phytochemicals detected in *M. peregrina*; and a slight decrease at 800 ppm in both species. The antioxidant activities decreased in both species to an average of 10µg/ml under 600ppm and 800ppm as compared with the control under 400ppm (5µg/ml). The antimicrobial activity improves with increasing CO₂ concentration in both species by an average of 17mm zone of inhibition with *M. peregrina* displaying higher activity (19mm) than in *M. oleifera* under all elevated CO₂ treatments. The study has demonstrated that elevated CO₂ could have potential impact on the phytochemical profiles and biological activities in *M. peregrina* and *M. oleifera*. The responses could have implications on the commercial value and medicinal uses of both species.

An inventory and analysis of medicinal plants of the Free State Province, South Africa

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For many years, extensive research on the ethnobotany of the Basotho people of Lesotho and the Free State of South Africa has been conducted. Further research over the last decade has yielded a comprehensive picture of the most important plant species used for medicine, food, and cultural purposes. This provides an opportunity for a comparative analysis and compilation of the Free State's medicinal plant checklist. The flora of the Free State Province has been extensively studied in recent years, resulting in a comprehensive inventory that includes the regional distribution of all plant species within the province's major vegetation units. Furthermore, the study of medicinal plants in South Africa has received a lot of attention, with exponential increase in the number of publications. However, the extent the medicinal plants of the Free State have been analysed for their main vegetative units and chemical compounds has not been reviewed. The purpose of this research is to examine the regional distribution and create an inventory of all the medicinal plant species found in South Africa's Free State Province. Moreover, the phytochemistry literature of the most important indigenous medicinal plants will be reviewed while identifying and studying their main chemical compounds. A study of the geographical distribution of all known medicinal plants within the province's six major vegetation units, as well as a study to add to the inventory of these species will be conducted. Three priority species, chosen based on the literature, will be studied phytochemically using standard chromatography and organic chemistry methods.

Induced antibiotic resistance and staphyloxanthin as a prospective target for treatment against pathogenic antibiotic resistant staphylococci

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The past century saw the discovery of antibiotics as an effective treatment for bacterial infections. Life-threatening infections that were previously untreatable could now be neutralized by antibiotics with few side effects. However, in recent years this once effective treatment has become less efficient as the bacteria it once treated are becoming increasingly resistant. Antibiotic resistance presents a major concern in human public health not just on a national level, but potentially at a global level. The development of bacterial resistance to antibiotics is a natural response to stress. Plants have always been an important source of medicines and treatments, particularly against bacterial infections. There have also been notable successes in re-establishing antibiotic effectiveness when they are combined with antibacterial plant extracts. The aim of this study was to compare the metabolites of an initially susceptible wild-type (WT) *Staphylococcus xylosus* with its mutated strain which had lost its antibiotic susceptibility. The mutated antibiotic resistant strain was made by exposing the susceptible WT strain to starvation stress. Metabolites from both strains were then extracted in methanol and analysed by ¹H-NMR. Multivariate analyses of the NMR spectra showed significant differences in metabolites between the WT and resistant strains. Staphyloxanthin, a carotenoid that aids in pathogenesis and protects against oxidative stress was partially isolated from the WT strain. This comparative model of a WT and its consequently resistant strain could also be used as a platform to test the effectiveness of different plant treatments against bacterial defence mechanisms like the synthesis of staphyloxanthin.

Inventory of medicinal plants used to treat and manage childhood diseases and wellbeing in North West Province, South Africa

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In South Africa, childhood diseases such as diarrhoea, gastroenteritis and pneumonia remain common and contributes to the increasing high rate of child mortality. Despite the popularity and value of traditional medicine, the systematic documentation of medicinal plants that are prescribed for diseases in children remains poorly explored. This study was aimed to documenting medicinal plants used for the treatment and management of diseases and the wellbeing of children in the North West province, South Africa. An ethnobotanical survey was conducted with 101 participants through semi-structured interviews (face-to-face). Information such as local name of plants, medicinal use, method and part were collected and analyzed. A total of 44 medicinal plants from 25 families were identified. *Aptosinum elongatum* with 0.69 Cultural importance index (CI) and 70.2 Fidelity level (FL) values, *Bulbine frutescens* 0.20 CI and 20.7 FL and *Euphorbia prostrata* 0.316 CI and FL 30.6 were found to be commonly used in the treat and manage of a variety of childhood diseases in the study area. *Aptosinum elongatum* also showed the highest Fidelity Level (FL), as compared to other medicinal plants. Leaves (23%) are mainly used, followed by roots (23%). Decoction and maceration were the main methods of preparation and plant remedies were dominantly administered orally. Skin-related disease, sunken fontanelle, urinary related diseases were the most prevalent childhood diseases treated and managed by the participants. The traditional health practitioners possess rich ethno-pharmacological knowledge and largely depend on naturally growing plants. Evaluation of the biological activities, phytochemical profiles and the safety of the identified popular plants remain an essential aspect for future research.

Two Celastraceae and two alien invasive Crassulaceae plant species with potential against mastitis: antibacterial, anti-inflammatory and antioxidant studies

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Mastitis, an infectious disease that affects cattle and humans with significant health and financial challenges has inflammation as its major feature. Drug resistance has developed in the major causative organism, *Staphylococcus aureus*. The antibiogram profiles of six strains of *Staphylococcus aureus* isolated from clinical cases of mastitis were determined. Using a broth microdilution assay, the antimicrobial activity of ethanol and acetone extracts of *Maurocena frangula*, *Maytenus undata*, *Kalanchoe pinnata* and *Bryophyllum pinnatum* were investigated against the clinical strains and an ATCC strain of *S. aureus*. Cytotoxicity and selectivity index (SI) values were determined. Antioxidant activity was evaluated using radical scavenging 2, 2-diphenyl-1-picrylhydrazyl (DPPH) and electron reducing 2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) assays. Anti-inflammatory activity was determined via inhibition of 15-lipoxygenase (15-LOX) and nitric oxide (NO) production using lipopolysaccharide (LPS)-activated RAW 264.7 macrophages. Antibacterial susceptibility tests showed that each of the clinical isolates were resistant to at least two antibiotics. Minimum inhibitory concentration values of the extracts ranged from 0.02 to 0.63 mg/ml. SI values ranged from 0.04 to 24.58. *Kalanchoe pinnata* extracts had the best antioxidant potency with IC₅₀ values ranging from 0.06 to 0.42 µg/ml. *Kalanchoe pinnata* extracts also had potent 15-LOX inhibitory activity with IC₅₀ values of 1.25 and 2.03 µg/ml for acetone and ethanol extracts respectively. At the highest concentration (100 µg/ml), the acetone extract of *B. pinnatum* had the best NO inhibition of 80.48% and cell viability of 96.75%. Further work is ongoing to investigate the active extracts in therapeutic formulations and to isolate active compounds.

Evaluation of proximate, mineral, amino acid and antinutritional constituents of *Evolvulus alsinoides* (L.) from northern South Africa

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Evolvulus alsinoides (L.) L. is a commonly browsed forb species in South Africa. The present study aimed at determining the proximate, elemental, amino acid, and antinutrient compositions of *E. alsinoides* collected from the Limpopo province of South Africa. The proximate composition was determined using the muffle furnace and micro-Kjeldahl methods, and the elemental composition was determined using atomic absorption spectrophotometry. The amino acid and antinutrient compositions were determined by high-performance liquid chromatography and titration methods, respectively. Proximate analysis revealed the presence of 10.82% protein, 75.34 %, 4.50% moisture, 1.75% crude fat, 29.21% crude fibre, 8.52% ash content, 44.50 % neutral detergent fibre and an energy value of 365.67 Kcal/100g. The plant is rich in C, N, K, Fe and Zn and essential amino acids such as valine, phenylalanine, methionine, isoleucine, leucine, histidine, lysine, tryptophan, and threonine. The antinutrients oxalate (0.00438 ± 0.12 mg/g) and phytate ($1.16 \pm 0.02\%$) were below toxic levels. This study is the first to record antinutritional constituents of the northern South Africa *E. alsinoides*, and it concluded that *E. alsinoides* has high-quality nutrient resources for livestock.

The qualitative phytochemical screening and antioxidant activity of *Carpobrotus edulis* as a response to extreme temperature extreme

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Carpobrotus edulis is an evergreen indigenous medicinal plant that is a facultative halophyte common to the Cape, KwaZulu-Natal, and Gauteng. Its phytochemical constituents contribute to their medicinal properties and are generally used for skin treatment of burns, wounds, and eczema and also used in the management of HIV/AIDS-related infections. In order to investigate the effect of extreme temperatures on *C. edulis*, it is important to understand how various abiotic environmental factors such extreme temperatures affect the composition of secondary metabolites. *Carpobrotus edulis* plants were placed in extreme temperature treatments [45/35°C, 40/30°C, 15/10°C, 7/4°C (day/night)] and leaves collected after (48, 96, 144, and 192 hours). Qualitative phytochemical screening of the methanol extracts was conducted. Antioxidant activity was quantified using the following methods: hydrogen peroxide assay and 2,2-Diphenyl-1-Picrylhydrazyl (DPPH) scavenging assay. In summary, flavonoids and saponins were not observed after 144 at 4/7°C treatment, coumarins after 192 hours (4/7°C). Tannins were not observed in the normal low temperature (15/10°C) after 96 hours, coumarins were not detected at 48 hours in the 40/30°C treatment. At 45/35°C, tannins were not detected only at 48 hours and steroids only disappeared at 192 hours. The lowest IC₅₀ was found in the high temperature extreme as compared to the low temperature extreme. DPPH yielded the best antioxidant activity as compared to the hydrogen peroxide method and this might because these methods could be targeting different reactive oxygen species. These results show strong evidence that extreme temperatures have a significant impact in the production of secondary metabolites.

South African medicinal plants used for the treatment of syphilis in Vhembe District, Limpopo Province

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Medicinal plants are widely used in some countries as a primary source of prevention and control of various diseases in animals and humans. Syphilis is a sexually transmitted disease with a variety of symptoms caused by the *Treponema pallidum* bacterium. The study investigates medicinal plants used for the treatment of syphilis by the local people and traditional health practitioners. An ethnobotanical survey was conducted to document medicinal plants used to treat syphilis in the Vhembe District, Limpopo Province. A semi-structured questionnaire was used to identify medicinal plants used, mode of preparation, dosage and other relevant information. The survey revealed that more than 40 plant species were recorded to combat syphilis. The most highly used plants were *Heteromorpha trifoliata* (H.L. Wendl.) Eckl. & Zeyh., *Anthocleista grandiflora* Gilg., *Capparis tomentosa* Lam., *Aloe marlothii* A. Berger and *Maerua angolensis* DC. Of the forty-six plant species documented, trees were most dominant (64%), followed by shrubs (18%), herbs (12%), creepers, grass and climbers (2%). The most plant parts used were the roots (55%), followed by bark (35%) and leaves (10%). Traditional health practitioners preferred decoction as a mode of preparation. Traditional medicine still plays an important role in the healthcare needs of the rural people in the Makhado Local Municipality.

An overview of the ethnobotany and pharmacology of southern African Lamiaceae.

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The Lamiaceae is a plant family of global importance encompassing ca. 7,800 species in 250 genera. Notable genera include *Lavandula* L., *Mentha* L., *Ocimum* L., *Salvia* L., and *Thymus* L., with numerous species playing important roles in the cosmetic, culinary, nutraceutical and pharmaceutical industries. Members of this family have been used as traditional and folk medicines across the globe, largely due to their biologically active aromatic compounds and other secondary metabolites. The southern African Lamiaceae comprise 297 species (in 42 genera), 94 of which have documented medicinal uses within the region. The top five use-categories reported are: respiratory; ear, nose and throat; topical; digestive and analgesic, with the most cited genera including *Salvia*, *Leonotis* (Pers.) R.Br., *Stachys* L., *Mentha* and *Ocimum*. Traditional uses have prompted further investigations, such as pharmacological studies, in an attempt to understand the mode of action and activity of the species. Our data reveal that there have been 365 *in vitro* and 79 *in vivo* studies conducted with the majority of the studies focusing on antimicrobial-, antioxidant- and anti-inflammatory activity. *Salvia*, *Coleus* Lour., *Leonotis*, *Plectranthus* L'Hér., *Mentha* and *Tetradenia* Benth. are the six most well-studied genera. The link between ethnobotanical uses and pharmacological activities for southern African Lamiaceae are presented.

Ethnobotany and antibacterial activity of medicinal plants used in the Maseru District of Lesotho for the treatment of Tuberculosis and other respiratory tract infections

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Treatment of diseases is a major challenge in Lesotho due to several factors such as limited availability and affordability of western medicine, as well as accessibility to healthcare facilities. Consequently, traditional medicine plays a vital role towards the well-being of a majority of the population in the country. Many studies conducted on medicinal plants in Lesotho have primarily focused on their uses. However, the therapeutic potential and safety of a majority of these plants are still unknown. The aim of the study was therefore to evaluate the antibacterial activity and toxicity of plants used in the Maseru District, Lesotho, to treat Tuberculosis and other respiratory tract infections (RTIs), using the minimum inhibition concentration (MIC) micro-titre plate dilution assay. Only two species (*Gerbera piloselloides* and *Withania somnifera*) displayed moderate activity against *Mycobacteria* strains (both plants displaying MIC value of 0.67 mg/ml against *M. fortuitum*). *Thesium costatum* showed a wide range of antimicrobial activity by inhibiting four of the five RTI test pathogens, even though the highest activity was observed in *W. somnifera* against *Moraxella catarrhalis* (MIC value of 0.33 mg/ml) and *Staphylococcus aureus* (0.21 mg/ml). However, *W. somnifera* was found to be toxic using brine shrimp lethality assay (BSLA). The antibacterial activity of *Ajuga ophrydis*, *Metalasia muricata*, *Thesium angulossum* and *T. costatum* is being reported here for the first time, which could probably mean that their medicinal use is limited to the Basotho people. The antibacterial results partially support the traditional utilization of the plants for the treatment of RTIs.

An evaluation of insecticidal activities and phytochemical properties of selected members of the family Meliaceae used traditionally as insecticides.

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The environmental and health risks associated with the use of synthetic pesticides have increased the demand for botanical insecticides as safer and biodegradable alternatives to control insect pests in agriculture. Hence in this study, nine previously unstudied plants belonging to the family Meliaceae were evaluated for their insecticidal activities against the fall armyworm and the diamondback moth as well as chemical constituents. Repellence, feeding deterrence, and topical application bioassays were employed to test their insecticidal activities. Chemical analysis using various techniques was undertaken to identify the secondary metabolites present in each plant. The repellence bioassay indicated that *M. azedarach* extracts exhibited the highest repellence percentage against the fall armyworm (95%) and the diamondback moth (90%). The feeding deterrence bioassay showed that *C. odorata* and *Turraea dregeana* extracts displayed excellent antifeedant activity against the fall armyworm larvae (deterrent coefficient, 115.85) and diamondback moth larvae (deterrent coefficient, 112.25), respectively. The topical application bioassay demonstrated that *Ekebergia capensis* and *Khaya anthotheca* extracts had the highest larval mortality against the fall armyworm (LD₅₀ 0.14 mg/kg), while *M. azedarach* extracts showed the highest larval mortality against the diamondback moth (LD₅₀ 0.14 mg/kg). Standard phytochemical methods showed that three species (*Cedrela odorata*, *Melia azedarach*, and *Turraea floribunda*) possess six secondary metabolites; alkaloids, coumarins, flavonoids, phenols, saponins, and tannins. GC-MS analysis revealed the presence of two noteworthy compounds: phenols and terpenes, in all the studied plants. Overall, this study lends scientific credence to the folkloric use of Meliaceae species as potential biocontrol agents against insect pests.

Spatial patterns and selection of medicinal plants in South-eastern Africa.

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Cultures in close geographic proximity are said to influence each other when it comes to selecting useful plants. This phenomenon is referred to as Galton's problem or spatial autocorrelation. Thus, the aim was to determine whether spatial autocorrelation is significant when it comes to the selection of medicinal plants in the south and south-eastern parts of Africa (Mozambique, Eswatini, Lesotho, KwaZulu-Natal, and the Vhembe district of the Limpopo Province – the traditional home of the Venda people), or whether medicinal plants are selected based on the available flora. Galton's problem was investigated by following a method similar to that presented in the Nepal study of cultural groups, including distance matrices, Sorensen index, Pearson product-moment coefficient (r), and the corresponding significance values (p). The Mantel test was done using the 'vegan' package in R. Regression analyses were used to identify floristic and cultural preferences in medicinal plant selection. Spatial autocorrelation was not significant for the correlation between geography and the available taxa ($r = -0.681$). There was also no correlation between geography and the medicinal taxa ($r = -0.770$). All regression analyses showed a significant relationship between the available taxa and the medicinal taxa. The selection of medicinal plants in the southern and south-eastern parts of Africa was based on the floristic environment, and thus spatial autocorrelation was not significant in these parts of Africa. Cultural similarities, ancestral linkages, and close geographic proximity were found not to influence the selection of medicinal plants.

Comparative phytochemical analysis, antioxidant and antibacterial activities of the leaves, underground stems and roots of *Bulbine abyssinica*

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Bulbine abyssinica is a succulent medicinal plant indigenous to South Africa. The species has been commonly used traditionally by indigenous people for the treatment of various skin related ailments. To date, limited studies have been conducted on the underground stems and roots, as well as comparative analyses across the different plant parts of the species. Thus the aim of this study was to determine the qualitative and quantitative phytochemical analyses, antioxidant and antibacterial activities of the different plant parts of *B. abyssinica*. Potted plants were maintained in the greenhouse until they fully acclimatised. Methanolic extracts were used to perform phytochemical screening, quantitative phytochemical analyses, three antioxidant assays (DPPH, hydrogen peroxide and metal chelating) and antibacterial tests against *Escherichia coli*, *Staphylococcus aureus* and *Streptomyces albulus* using agar well diffusion. The results showed that the leaves contained most of the phytochemical groups tested, compared to other plant parts. The total phenolic and total flavonoid contents showed a higher quantity in the leaves compared to the other plant parts. The antioxidant activity results showed that the roots exhibited the strongest scavenging power against DPPH, whereas the leaves showed a higher antioxidant power against hydrogen peroxide and metal chelating radicals. All three plant parts showed intermediate zones of inhibition (10-19 mm) against all three tested bacterial strains, with the exception of the stems against *S. albulus*. The current study validates the use of different plant parts of *B. abyssinica* in traditional context, and suggests the plant's potential application in pharmaceutical and cosmetic industries.

Invasive Plant Biology

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Progress made towards eradicating an invasive shrub, *Cistus ladanifer* in Western Cape, South Africa

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Cistus ladanifer is a woody, perennial shrub native to the Mediterranean. Three small naturalising populations (all smaller than 0.1 ha) of *C. ladanifer* were discovered in 2012 in native heathland vegetation (“Fynbos”) of the Western Cape Province, South Africa. *Cistus ladanifer* poses a significant threat to the local endemic vegetation, largely due to its pre-adaptation to similar environmental conditions, and, if left unmanaged, it has the potential to invade further. *Cistus ladanifer* is currently not listed according to South African legislation (i.e. the National Environmental Management: Biodiversity Act 10 of 2004 [NEMBA] Alien and Invasive Species Regulations, 2020) and a detailed species risk analysis is required to inform future legislative and management efforts. We conducted delimitation surveys, monitor the response of *C. ladanifer* to mechanical and chemical control and track spread over a nine year period. A detailed risk analysis was also produced using the Risk Analysis for Alien Taxa framework. Here, we present findings on *C. ladanifer* extent and densities and provide an insight about the feasibility of eradicating the species following nine years of control and monitoring. We also present results for the risk analysis produced for *C. ladanifer*.

The reproductive system of *Campuloclinium macrocephalum* — implications for biocontrol implementation

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Campuloclinium macrocephalum is an alien invasive species currently threatening the grassland biome of South Africa. Without intervention, the species will invade the entire grassland biome and threaten food security. The weed's success might be linked to its reproductive strategies and ability to overwinter via a woody xylopodium. We therefore investigated the modes of reproduction adopted by South African populations of *C. macrocephalum*. This was accomplished by identifying potential pollinators, assessing pollen grain size and viability, determining whether pollen tubes reach the ovules, assessing genetic variation and exploring the link between reproductive strategies and ploidy level. Preliminary data suggest that the species can reproduce via apomixis and/or self-fertilization. The implications of this are that the populations should have low genetic diversity and biocontrol agents should be effective on all individuals within a population. These reproductive modes also have poor evolutionary potential, with low proportions of heterozygotes – a factor which would ensure that the biocontrol agents are also effective against future populations. Conversely, high average pollen viability percentages of 98% and 90% found in four Gauteng populations of *C. macrocephalum* indicate high male fertility which could enhance evolutionary potential by allowing gene flow between populations. Pollen load and pollinator observations suggest that some insects may be effectively transferring pollen between individuals thereby indicating that this species is capable of cross-pollination. These outcomes are relevant for the development of biocontrol management plans. The link between reproductive strategies and consequent genetic variation is a strong predictor of the success of future management plans.

Physiology

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Effects of exogenous application of five antioxidants on vigour, viability, oxidative metabolism and germination enzymes in aged cabbage and lettuce seeds

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Ageing induces modifications capable of compromising seed vigour and viability. Some of these changes can be reversed to an extent by soaking treatments that can restore vigour. This study investigated the effects of exogenous application of five antioxidants: ascorbic acid (AA), gallic acid (GA), reduced glutathione (GSH), trolox and glycerol, on physical and biochemical lesions induced by controlled deterioration (CD) in *Brassica oleracea* (cabbage) and *Lactuca sativa* (lettuce) seeds. The antioxidants were applied at 0.2, 0.4 and 0.6 mM to fresh seeds and seeds subjected to CD: 75% viability (P75), 50% viability (P50) and 25% viability (P25); deionised water (DW) served as control. Controlled deterioration resulted in more abnormal seedlings production in cabbage than in lettuce seeds. Additionally, CD increased electrical conductivity (EC) and protein carbonylation (PC), lowered antioxidant and germination enzymes activities in both species but increased lipid peroxidation in lettuce seeds only. Exogenous antioxidants enhanced seed viability of cabbage at P25 and lettuce at P50 and P25; seedling vigour of fresh, P75 and P25 cabbage; P50 and P25 lettuce seeds. Electrolyte conductivity and lipid peroxidation were reduced by AA, GA, glycerol, GSH and trolox in lettuce seeds, while PC was reduced by GA, glycerol and GSH in both species. Antioxidant and germination enzymes activities were heightened by GA, glycerol, GSH, and trolox in cabbage seeds; AA, GA, glycerol, GSH and trolox in lettuce seeds. The study resolved that CD-induced lesions differ in both species, and the exogenously applied antioxidants are beneficial in quelling injurious spin-offs of oxidative reactions.

Investigating the *in vitro* medicinal compound production and restoring a lost function of *Erythroxylum emarginatum*, an indigenous southern African coca plant species, using genetic complementation and tissue culture feeding approaches

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Erythroxylum emarginatum, *E. pictum* and *E. delagoense* are endemic to southern Africa and belong to the Erythroxylaceae family. These three species are phylogenetically related to the South American *E. coca*, known for its synthesis of the tropane alkaloid cocaine. Interestingly, the southern African species does not produce this psychoactive substance, although the presence of key precursor compounds have been detected. Cocaine synthase (EcCS) functions as the final enzyme in the biochemical pathway, responsible for converting ecgonine methylester (EME) into the plant defence-related secondary metabolite, cocaine. A mutated version of the EcCS gene sequence has been detected in *E. emarginatum*, which could be the cause for the inability to produce cocaine. The aims of this study were to investigate the *in vitro* production of medicinal compounds using tissue culture feeding strategies, and to observe the downstream biochemical effects of re-establishing this lost biosynthetic step by the genetic complementation of the mutant gene in *E. emarginatum* with a functional allele of EcCS from the donor *E. coca*. *E. emarginatum* was chosen as the candidate species because of its biochemical relatedness to the donor species and its ability to produce EME. Tissue culture feeding results highlight the potential of using *E. emarginatum* for the *in vitro* production of medicinal compounds. With successful genetic transformation, we will be gaining insight into the role of lost biochemical functions within a southern African coca species.

Genotype by Environmental Interaction: An experimental strategy for genotype selection for a target environment

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Chickpea (*Cicer arietinum* L.), the third most important legume crop globally after common bean (*Phaseolus vulgaris* L.) and field pea (*Pisum sativum* L.), is rich in nutrients. South Africa has been importing chickpea over the years, worth over US\$ 1.1 million annually, highlighting the need to increase production. An earlier study in Limpopo, South Africa, conducted over two seasons at three sites with known differences in temperature and rainfall, identified genotype and heat tolerance traits from four desi type chickpea genotypes. The study assessed the effects of genotype and environment, and their interaction (GEI) on plant growth and seed yield. The results showed no interaction between genotypes and environment on plant growth at flowering, and seed yield in both seasons after a two-Way ANOVA. However, the environmental effects showed that the cooler site recorded higher grain yield and seed size than the warmer site in one season. The genotypic effects were observed on grain yield with one of the genotypes recording lower yield than the other three that were similar. The lack of significant interaction has implications on the genetic diversity and vulnerability of the genotypes to both abiotic and biotic stresses. A comprehensive assessment of GEI using 12 heat and drought tolerant and susceptible chickpea genotypes, at least six sites in the region, and for two seasons is recommended. Appropriate statistical analyses for GEI studies such as Linear Regression and Additive Main Effects and Multiplication Interaction (AMMI) models are also recommended for measurement of genotypes stability and adaptability.

Review of zooxanthellae and their response to environmental stress

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Global warming has become a serious problem, which recently showed an increased threat to both terrestrial and aquatic ecosystems. In marine ecosystems, sea surface temperatures (SST) have risen approximately 1 - 2°C since pre-industrial times. This has led to unprecedented mass coral bleaching events. Coral bleaching is the process by which these zooxanthellae (algal endosymbionts) are expelled from their respective coral hosts, causing the coral to lose colour and become white. Consensus is that mass coral bleaching is due to the photosynthetic dysfunction in the zooxanthellae. Numerous protective mechanisms are present within both the coral host and its representing zooxanthellae to counteract thermal challenges. The aim of this study was to review the information available on the protective mechanisms present in zooxanthellae cells when they experience environmental stress conditions, such as temperature fluctuations. We only focused on heat shock proteins, chaperone proteins, and the enzymes; super oxide desmutase (SOD), ascorbate peroxidase and catalase (CAT) for their main role in neutralizing produced noxious hydrogen peroxide in the chloroplast of the zooxanthellae. Unfortunately, these mechanisms have proven to be inefficient in recent mass bleaching events around the world which suggests that other mechanisms are required to actively battle coral bleaching. Despite the lower likelihood of bleaching in South African coral reefs, environmental bioengineering, such as the use of the CRISPR- Cas 9 system presents a useful tool to understand and adapt traits within zooxanthellae genotypes to help mitigate coral bleaching in the future.

Utilization of post-harvest chemical treatments for extending shelf-life of selected South African potato cultivars

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The non-renewal of Chlorpropham (CIPC), a common sprout suppressant for potato preservation, came into effect in 2020. This ban on CIPC use, due to toxicity to humans and environmental concerns, necessitated the search for natural anti-sprouting agents. This study aimed to provide a scientific basis for developing postharvest storage protocols for using sprout suppressants to improve the shelf-life of popular South African potato cultivars, i.e., Mondial, Electra, and Panamera, for the export market. The efficacies of selected suppressants in extending shelf-life stored at 24°C were evaluated. The selected sprout suppressants included 1,4-dimethylnaphthalene, SmartBlock, *Aloe vera* gel, S-carvone, and Caraway essential oil. The efficacy of sprout suppressants was monitored by assessing the selected parameters. Also, the effects of different application techniques on the efficacies of the selected suppressants were studied. In both cultivars, Caraway essential oil delayed sprouting by 21 days. Sprouting and storage performance was found to be cultivar-dependent. Anatomical analysis revealed tissue necrosis in the meristematic zones, suggesting suppression through the induction of meristematic tissue necrosis. Regarding the application method, SmartBlock inhibited sprouting for 84 days. The results of this study show that the application method has a significant influence on the efficacy of Caraway essential oil. These results suggest that application techniques are important factors to consider when developing protocols for tuber preservation. The tested molecules were effective at controlling sprouting, but to varying degrees. To meet the dual challenge of keeping stocks sprout-free for prolonged periods, these molecules should be combined with other innovative storage strategies.

The photosynthetic efficiency and carbohydrate responses of edamame cultivars under drought stress

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Edamame (*Glycine max* L. Merrill) is a vegetable-type soybean with high nutritional value. The crop was recently introduced to South Africa. However, there is lack of information on its physiological responses to drought stress conditions. This study was conducted to investigate the photosynthetic efficiency and carbohydrates responses of six edamame cultivars under drought stress. The photosynthetic efficiency parameters such as chlorophyll fluorescence and stomatal conductance were determined using non-invasive methods while the pigments, non-structural and structural carbohydrates were quantified using destructive methods. Drought stress significantly increased the Fv/Fm and Plabs of AGS429 and UVE17 at pod filling stage. Chlorophyll-a was significantly reduced in AGS429 and UVE17. Chlorophyll-b was relatively stable in most cultivars, except UVE17, which showed a significant decline at flowering. AGS429 had increased Chlorophyll-b at pod filling. The carotenoid content was significantly reduced in UVE17 at pod filling. Drought stressed UVE17 further showed a substantial reduction in the stomatal conductance at pod filling. Drought stress significantly increased trehalose, sucrose and starch at pod filling, but decreased glucose content. Chl-a positively correlated with starch. The Fourier-transform infrared and X-ray diffraction results suggest that the cell wall of UVE14 was the most intact during drought stress followed by UVE8 and AGS429. The study shows that the carotenoids, performance index of the photosystems, starch, hemicellulose, and lignin can be used as physiological mechanisms of drought tolerance in edamame, which is essential for screening cultivars for drought tolerance.

Growing swiss chard and lettuce on an organic medium enclosed trough system improves growth, yield, and water-use efficiency

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Lettuce and swiss chard are commonly consumed vegetables in South Africa and their yield is dependent on soil water and nutrient availability. The Organic Medium Enclosed Trough (OMET) system was developed as a non-drainable growing technique which improves crop yield by reducing water and nutrient seepage. The objective of this study was to investigate the impact of the OMET system on growth and yield of common leafy vegetables [swiss chard and lettuce (cv 'Lara' and 'Elisa')]. Four weeks old swiss chard and lettuce seedlings were transplanted on an established OMET and non-OMET system following a complete randomised block design with three replications for a period of 60 and 45 days, respectively. At harvest, yield components including biomass, aerial mass, root mass, dried aerial mass and dried root mass were higher in the OMET treatment than the non-OMET for both swiss chard and lettuce cv 'Lara' and Elisa'. Swiss chard growth attributes including plant height (40 cm), leaf length (25 cm) and root length (15 cm) in OMET treatments were two-fold higher than those found in non-OMET. Leaf length in both lettuce cv 'Lara' and 'Elisa' ranged between 26 and 30 cm either in OMET or non-OMET. Chlorophyll content was higher in non-OMET treatment. The OMET growing technique offers improved yield and crop quality at the disposal of low irrigation water. Therefore, this technique is recommended for small-scale and organic farming.

The impact of dark-adaption duration on the chlorophyll a fluorescence signature of *Amaranthus cruentus* leaves

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Photosynthesis is how plants use sunlight, carbon dioxide and water to make sugars (for enzymes and growth) and oxygen. Biochemically speaking, photosynthesis is a somewhat complicated process within plant cells and requires many enzymes and the transfer of electrons. If any of these photosynthetic systems is disrupted for any reason, the process will shut down and the plant will die. The measurement of Chlorophyll a fluorescence is one of the most widely used methods to probe photosynthesis. Among the existing methods, chlorophyll a fluorescence imaging can probe systematic changes in photosynthetic reactions. Chlorophyll a fluorescence measurements were taken with a Handy PEA chlorophyll fluorimeter. After eight hours after dark adaptation, there was no difference in the F_v/F_m and PI_{total} values. The result show that F_v/F_m increases with time until a certain point where it starts to decrease, meaning that the quantum efficiency of PSII is reducing. Attached leaf recorded the highest F_v/F_m and PI_{total} over detached leaf from 1 hour to 8 hours of dark adaptation. This results shows that attached leaf was performing better in terms of photosynthetic rate as compared to detached leaf.

Development of fungi decontamination protocols for *Trichilia dregeana* Sond. seeds: a representative of recalcitrant seeds

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Recalcitrant seeds survive in storage in the short- to medium-term only under hydrated conditions and at temperatures above zero. A major problem restricting storage lifespan is the presence and proliferation of fungi. The present study was designed to improve the hydrated storage life span of *Trichilia dregeana* seeds by curtailing/eliminating fungal contaminants, and to assess the impact of the treatments on seed viability and subsequent performance of seedlings. Six fungal species prevalent in *T. dregeana* seeds in hydrated storage and in vitro were isolated and identified from axenic cultures and 18S DNA analysis. After in vitro testing of the efficacy of chosen biocontrol agents and chemical compounds for inhibiting the growth of axenic cultures of the test isolates, the following treatments were applied: surface decontamination alone or followed by treatment with a strain of *Trichoderma harzianum*, Eco77®, and Biotaine™ (active ingredient chlorhexidine gluconate) with or without Nipastat® (a mixture of parabens) as a powder applied to seeds, and encapsulation in alginate gel, incorporating Nipastat or not. Under in vitro conditions treatments with Eco77, Biotaine and Nipastat encapsulated in an alginate gel were best for controlling contamination levels and affording higher germination of seeds after 30 days in culture. Six months after removal from storage and planting in the greenhouse trials, significant changes in biomass allocation to leaf area, leaf length, stem biomass and root length were recorded.

Tracking chlorophyll a fluorescence as an indicator of crop growth efficiency in *Amaranthus cruentus*

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The effects of climate change are compelling, and they have led to renewed interest in resilient orphan crops like *Amaranthus cruentus* for alternative food production systems. To enable assessment of plant acclimation to these climate change scenarios, analysis of changes in chlorophyll a fluorescence, arising mainly from PSII, provide valuable information about the photochemical performance of the plant. The accuracy of these measurements depends on the correct selection of physiologically mature stalk leaf at the different plant growth stages. Thus, this research aims on developing a protocol to identify correct leaf for these measurements at different growth stages of *Amaranthus cruentus*. *Amaranthus cruentus* plant was grown under conditions free of biotic and abiotic stress. The plant was dark adapted for 1 hour and a total of six measurements were taken on each stalk leaf using a Handy PEA chlorophyll fluorimeter. The results show that the maximum quantum yield of PSII chemistry, the performances indexes (PI_{total} and DF_{total}) decrease as the plant reaches the booting stage. This observation also coincides with a decline in the reduction of $NADP^+$. Thus, it can be concluded that, as the plant reach booting stage, there is a general shift in the photosynthetic performance, with the stalk leaves formed during vegetative phase becoming stronger sink leaves, while the newly formed stalk leaves (within the top half portion of the stem) becoming the main source leaves. Hence, after booting, chlorophyll a fluorescence measurement can be taken on stalk leaves within the top half of the stalk.

Growing Okra on an organic medium enclosed trough system improves growth, yield and water-use efficiency

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Okra is an indigenous vegetable consumed in Southern Africa for its nutritious and health benefits. Its growth and yield are negatively affected by water and nutrients deficit. Organic Medium Enclosed Trough (OMET) system was developed as a non-drainable growing technique which improves crop's yield by reducing water and nutrients seepage. The objective of this study was to investigate the effects of the OMET system on growth and yield of okra (Clemson spineless) grown under micro-plots. Four weeks old okra seedlings were transplanted on established OMET and non-OMET micro-plots systems using RCBD for a period of 110 days, with three replications. At harvest, yield components including mass of pod yield per plant (44.3 g), number of pods plant (14), fresh pod length (12.8 cm) were two-fold higher in OMET than non-OMET treatment. Okra growth attributes including plant height (152.1 cm) and stem diameter (16.9 mm) were also improved in OMET as compared to non-OMET (134.2 and 7.6 cm). The OMET treatment enhanced physiological maturity, which was 15 days earlier than in non-OMET. The OMET growing techniques offers improved yield and crop's quality at the disposal of lower irrigation water as compare to the non-OMET. Therefore this technique is recommended for small-scale and organic farming hence is it cheap and sustainable.

Improvement of yield and growth in Amaranth species using the organic medium enclosed trough system

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Amaranth species are a highly popular group of vegetables. The organic medium enclosed trough (OMET) system is a non-drainable growing technique. The aim of the study was to investigate the effects of the OMET system on growth and yield Amaranth species: *A. caudatus*, *A. cruentus* and *A. species*. Data for growth attributes are seedling height and stem diameter; whereas for yield attributes are, mass of the aerial parts and root. The amount of water used on OMET and Non-OMET was recorded until termination. T-test at the significance level of 5% was used to analyse data using computer statistical software, Statistix 10.0. The study revealed that the OMET system increased the stem diameter and plant height of *A. caudatus* by 2.8% & 10.5%; *A. cruentus* by 25.2% & 21.6%, while for *A. species*, it increased by 32.4% & 41.2% respectively. The mass of the aerial parts and roots increased significantly for *A. caudatus* by 24.4% & 8% and *A. cruentus* by 12.6% & 50.1% respectively, while for *A. species*, increased by 91.9% & 94.5% respectively on OMET. The study has also shown that the OMET system utilized less water estimated than in non-OMET, making it a brilliant strategy to conserve and utilize water sustainably and effectively.

Vegetative growth response of common consumed vegetables in response to different concentrations of an amino acid based bio stimulant

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Lettuce and swiss chard are commonly consumed leafy vegetables in South Africa. They accumulate non-nutritive nitrates as a result of more than optimum N fertiliser application. However, through improved root surface to enhance water and nutrient uptake mechanism, amino acid- based plant biostimulants could serve as an alternative of N-application to improve vegetative growth of leafy vegetables. The objective of this study was to determine the responses of lettuce (cv 'Lara' and 'Elisa') and Swiss Chard exposed to different concentrations of a Phytostim® biostimulant. Four weeks seedlings were transplanted following a complete randomised block design under greenhouse conditions. Different concentrations (0, 1, 3 and 6%) of a commercially available Phytostim® biostimulant were foliar applied at two weeks interval from planting, three times before experiment termination. Application of 3% biostimulant improved the vegetative growth (fresh biomass, aerial mass, dried aerial mass, plant height, number of leaves) in concomitant to the improved root length, fresh and dried root mass in both lettuce (cv 'Lara' and 'Elisa') and swiss chard vegetables. The improved vegetative growth may be attributed by the amino acid constituents of the biostimulant which act as a signalling molecule to induce phytosynthesis and root development associated with improved water and nutrient uptake. Application of 3% phytostim biostimulant could be used in the organic farming system or as an alternative for N application to improve vegetative growth of leafy vegetables. More research to investigate the effects of the biostimulant on nutrients and secondary metabolites are urgently needed.

Drought and heat stress decreases the photochemical efficiency of *Amaranthus* species

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Climate change progressively results in high temperatures and drought. Drought and heat stress hamper plant growth and productivity of many crop species by limiting photosynthesis and other related metabolic processes. Understanding the photosynthetic response of economically important crops like *Amaranthus* species is therefore of great importance. This study investigated the photochemical response of *Amaranthus caudatus*, *A. hypochondriacus*, *A. cruentus* and *A. spinosus* to combined drought and heat stress and these stresses individually. Four weeks after emergence, the plants were subjected to drought stress (10% field capacity) and different temperature regimes (30, 35 and 40 °C) for two hours. Chlorophyll *a* fluorescence parameters derived from OJIP transient were subsequently analysed. The combined drought and heat stress caused a decline in the PI_{total} (total performance index) and its partial parameters. All four parameters that comprise the PI_{total} index decreased in response to combined drought and heat stress at 40°C. Of the four parameters, the density of the active reaction centers (RC/ABS) and the probability to move an electron beyond Q_A^- ($\psi_{E_0}(1 - \psi_{E_0})$) were the most sensitive. RC/ABS decreased by 70.3%, while $\psi_{E_0}(1 - \psi_{E_0})$ decreased by 69.7% in *A. caudatus* and *A. hypochondriacus*, respectively. Drought, heat stress and combined stress decreased the PI_{total} and its partial parameters, causing damage to the photosynthetic apparatus of these species. This study shows that *A. cruentus* and *A. spinosus* are better adapted to drought and heat stress and a combination of both.

The response of onion (*Allium cepa* L.) to amino-acid based plant biostimulant

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Onion is loved for its richness in flavor and aroma when it is included in stews and salad. Its yield and bulb development are affected by the application of plant hormones. This study was aimed at evaluating the effects of different concentrations of a commercially available Phytostim® biostimulant on onions grown on micro-plot. Data was collected from 90 days old onions crops that was exposed to biostimulant through foliar application at different concentrations (0, 1, 3 and 6%). The onions were grown on a micro-plot following a complete randomized block design with three replications. Vegetative growth (leaf length, biomass, and aerial mass), and roots development (bulb diameter, roots length) were recorded. The results revealed that all these parameters were highly influenced by the application of 1% biostimulant concentration. The application of 1% biostimulant improved leaf number (7) and highest leaf length (61.2 cm), while 6% had the lowest leaf length (35.2 cm). The application of 3% biostimulant enhanced the roots length (43.5 cm), while 6% also had the lowest (30.5 cm). Applying 1% also had the most bulb development with the largest diameter (32.32 mm) and 6% had the lowest (26.9 mm). The application of the biostimulant enhanced the rooting system which improved vegetative and bulb development. However, at high concentration, 6% biostimulant, it acted as a growth inhibitor as all measured parameters were below the control (0%). Onion production with the application of Phytostim® biostimulant improves bulb development and such sustainable strategy can be beneficial to improve yields of organic farmers, small scale and commercial farmers.

Pollination Biology

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Reproductive barriers in bird-pollinated *Erica* species

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The group of Cape *Erica* species is an example of an evolutionary radiation that produced a large number of closely-related species that continue to coexist. In such plant systems, species boundaries can be maintained by pre-pollination and/or post-pollination barriers. Even when post-pollination barriers are in place, however, reproductive interference and pollinator dependence may strengthen selection for pre-pollination barriers. We assessed which reproductive barriers enable coexistence of bird-pollinated *Erica* species in south western part of the Fynbos biome where they are pollinated by the Orange-breasted Sunbird *Anthobaphes violacea*. We also tested for reproductive interference and pollinator dependence. Hand-pollination experiments were conducted in natural populations of *Erica plukenetii*, *E. curviflora* and *E. coccinea*. At least two of the three pre-pollination barriers assessed (distribution ranges, flowering phenology and flower morphology) were weak in each species pair. Seed set from heterospecific pollination (average 8%) was significantly lower than seed set from outcross pollination (average 50%), supporting the hypothesis that species boundaries are maintained through post-pollination barriers. Two of the three species were self-sterile, and therefore likely pollinator dependent. Reproductive interference, assessed in one population by applying outcross pollen three hours after applying heterospecific pollen, significantly reduced seed set compared to outcross pollen alone. This may drive selection for traits that enhance pre-pollination barriers. This study suggests that post-pollination reproductive barriers could facilitate the coexistence of congeneric species, in a recent radiation with weak pre-pollination reproductive barriers.

Seed dispersal by monkey spitting in *Scadoxus* (Amaryllidaceae): fruit selection, dispersal distances, and effects on seed germination

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Fleshy fruits are usually associated with ingestion of seeds but can also serve as a reward to animals that discard seeds without ingesting them. We investigated the seed dispersal systems of two South African *Scadoxus* lilies. Like some other genera in Amaryllidaceae tribe Haemantheae, seeds of *Scadoxus* are non-orthodox, reputedly poisonous, and enclosed within fleshy fruits. The bright red ripe *Scadoxus* fruits attract monkeys which consume the fleshy fruit and spit out the seeds. De-pulping increases the rate of seed germination. Monkeys spit some seeds out in the immediate vicinity of the plant and carry others further away in their cheek pouches (84% of *S. multiflorus* subsp *katherinae* seeds and 78% of *S. puniceus* seeds were dispersed further than one meter away from the parent plant). Both species have very specific spatially-restricted habitats; *S. multiflorus* subspecies *katherinae* is confined to patches of swamp within forests while *S. puniceus* is confined to small bush clumps in a grassland mosaic. Monkey-mediated seed dispersal may be advantageous for these two *Scadoxus* species as it ensures that some seeds are spat out in the spatially restricted habitats of the parent plants, while others are carried through cheek-pouching to more distant habitat patches favoured by monkeys.

To bee or not to bee – floral rewards and pollination efficiency in South African Macadamia.

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Too Bee or Not Too Bee – unlike Shakespeare, the Macadamia pollination industry cannot afford for this question to go unanswered for much longer. Much controversy exists between scientists, beekeepers and farmers alike as to whether having costly bee hives placed in orchards during the Macadamia flower season, is justified by a notable increase in yield. The possibility of bees preferring certain cultivars over others for various reasons could also assist in better future orchard plans. No clear-cut answers have yet been established, but we hope to provide more clarity to these problems. In this project, 15 different cultivars were compared in terms total pollen production, tepal surfaces, nectar sugar concentration and nectar volume. These factors were assessed in order to determine if certain cultivars might be preferred by bees as a source of food and nectar whereby these cultivars end up receiving more visits from bees and are consequently better pollinated. Pollen Transfer Efficiency experiments were also conducted to determine if pollination is more efficient on farms that have kept hives vs. farms that do not bring hives into their orchards during the flowering season. Even the smallest improvement in understanding either of these problems would lead to large financial gain for producers.

Contrasting patterns in flowering phenology between species adapted for pollination by generalist compared to specialist flower-visiting birds

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Two separate bird-pollination syndromes have been proposed based on different floral traits and nectar properties. Specialist bird-pollinated plants are adapted to the specialised morphology and foraging behaviour of specialist birds such as sunbirds and hummingbirds, while generalist bird-pollinated plants are adapted to a wider range of opportunistic bird visitors. Flowering phenology is an important but often overlooked floral trait that may represent an adaptation to the biology of pollinators and is hence crucial for understanding their coevolution. Based on ecological theory of facilitation, competition and resource partitioning in pollination systems, flowering phenology patterns are expected to differ between specialist and generalist bird-pollinated species. Flowering records of bird-pollinated species occurring in the south-eastern summer rainfall region of southern Africa were used to investigate patterns in flowering phenology using circular statistics. The mean flowering times of specialist bird-pollinated plant species were distributed across all seasons. This staggered flowering between species may reduce competition for pollinators and could be essential for sustaining specialist pollinators which depend on nectar year-round. In contrast, the flowering of most generalist bird-pollinated species peaked in late winter to early spring. This allows these plant species to make the most efficient use of their opportunistic bird pollinators by providing nectar as a food source when other typical food sources are scarce. These findings provide further support for generalist bird-pollination and specialist bird-pollination being recognised as two distinct classes of ornithophily and suggests that flowering phenology may be an important trait to consider in pollination syndromes.

Sending private messages: Floral ultraviolet signals are associated with pollination syndromes in *Erica*

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The occurrence and function of ultraviolet (UV, wavelengths between 300-400 nm) reflectance in insect-pollinated flowers is only recently emerging, but even less is known about its prevalence in other pollination systems and African species. We chose the genus *Erica* for studying the prevalence of UV because of its extreme radiation (c. 680 species) in the Cape, with a diversity of pollination syndromes. This study quantified the prevalence and intensity of UV reflectance for different *Erica* pollination syndromes and tested pollinator preferences for UV reflectance in the two groups with the highest prevalence: sunbirds and long-proboscid flies. Our results show that UV colouration is absent or rare in *Erica* species pollinated by unclassified insects, rodents, or wind. About 17 % of bird-pollinated species reflected UV but choice experiments revealed that free-ranging sunbirds showed no preference for UV colouration. All sampled long-proboscid fly-pollinated species reflected UV and its experimental removal decreased seed set drastically. This suggests that long-proboscid flies in the Cape have a strong preference for UV and thereby contributed to selecting for the evolution of this signal.

**Birds or bees for fruits and seeds: key pollinators of the summer flowering
Aloe reitzii var. *reitzii***

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Previous research showed that both birds and bees are frequent visitors to *Aloe reitzii* var. *reitzii*, a summer-flowering South African grassland endemic, but their role as pollinators was not established. The pollinator effectiveness of birds and bees at *Aloe reitzii* var. *reitzii* flowers and the relative importance of specialist and generalist nectarivores in its pollination were therefore investigated. Pollen loads between birds and bees did not differ significantly, indicating that both birds and bees play a role in the pollination of *Aloe reitzii* var. *reitzii*. Bird-excluded treatments produced significantly lower seed set compared to treatments that included all visitors, showing *Aloe reitzii* var. *reitzii* is predominantly bird-pollinated, while bees appear to play a minor role in its pollination. High pollen loads present on Cape Weavers (*Ploceus capensis*) and Malachite Sunbirds (*Nectarinia famosa*) indicate both are effective pollinators of *Aloe reitzii* var. *reitzii*. The long, tubular flowers produce copious dilute nectar; this combination of floral traits of *Aloe reitzii* var. *reitzii* is intermediate between a generalist and specialist bird pollination syndrome, enabling effective pollination by generalist and specialist nectarivores. Effective cross-pollen transfer by both bird species is facilitated by the aloe's strongly exerted style and filaments, coupled with each species' feeding behaviour. The consistent abundance and frequent visitation of Cape Weavers to *Aloe reitzii* var. *reitzii* flowers in multiple years, indicate they are its primary pollinators.

Does an active plant-focused restoration approach restore pollinator visitation to flowering plants?

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Pollinator visitation can ensure the long-term success of vegetation restoration, as it will prevent the eventual decline of introduced plant species by ensuring their reproduction and genetic variability. Over a period of two years, we have tested how successful pollination is restored by an active plant-focused restoration approach, in Cape Fynbos in South Africa. By comparing a recently restored and fragmented site with two reference sites (fragmented and intact) we aimed to determine whether pollination was restored by comparing the visitation frequency and species richness of pollinators as well as the seed set of generalist and specialist plant species. Pollinator visitation on generalist plant species was the highest at the fragmented reference site and restoration site at 78% and 33% higher than the intact reference site. Pollinator species richness was 33-53% higher at the intact reference site. Natural seed set was 19 – 33% higher at the intact reference site. Specialist plants that were incapable of autonomy were absent from fragmented study sites and even when introduced at these sites, pollination was still sub-optimal. Our results show that generalist plant species have a greater potential to facilitate ecosystem recovery in terms of pollination. However, providing sustained nectar sources and stepping-stones for specialist pollinators in Cape Fynbos, is important for ecosystem function, especially in small, urban nature reserves.

Floral morphology, visitors and scent chemistry of *Agave sisalana* in Nkomazi local Municipality, Mpumalanga, South Africa.

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Agave sisalana is a plant of economic importance grown mostly for fibre production. However, it is an introduced plant classified as a category 2 invasive in South Africa and requires a permit to cultivate. There is dearth in knowledge on reproduction of *A. sisalana* making it difficult to control the plant from spreading. Understanding the floral morphology, traits and visitors can aid in understanding the reproduction biology of this plant. We investigated the floral morphology and traits that play a role in determining plant pollination systems. We further investigated the presence of floral rewards (nectar and scent), and observed the floral visitors at different times of the day. *A. sisalana* has a large paniculate inflorescence with small tubular flowers that are greenish-yellow in colour. The anthers were dehiscent before stigma receptivity. Nectar production occurs at night and it was present until morning. Gas chromatography-mass spectrometry analysis revealed that floral scent profile of *A. sisalana* consist mainly of terpenoids, aliphatics and benzenoids. Sulphur containing compounds were few with dimethyl disulphide and S-Methyl 2-propenethioate only occurring at night. The floral visitors were abundant in the morning and the visitation declined in the afternoon and evening. Birds were the most abundant visitors followed by bees and houseflies. Behavioural observation suggests that birds could be the possible pollinators and bees and houseflies possible pollen and nectar robbers. We suggest that cross pollination is possible in *A. sisalana* and birds are the probable pollinators.

Quantifying pollen transfer between cultivated and wild *Cyclopia* species, South Africa

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Honeybush, *Cyclopia* Vent., is an endemic genus in the fynbos biome with commercial value in the tea industry of South Africa. With the shift in supply from wild to cultivated biomass, it is important to consider how best to develop biodiversity-friendly farming practises. The potential for gene flow between cultivated populations and their wild relatives through cross-pollination needs to be considered. My objectives are to determine the primary pollinators of commercially important *Cyclopia* species, and establish whether these pollinators have the potential to cross-pollinate and initiate the flow of genetic material between cultivated and wild populations. Preliminary results indicate there are five species of carpenter bees (of the genus *Xylocopa*, Latreille) responsible for the pollination of *Cyclopia* in the Overberg, Western Cape and the Langkloof, Eastern Cape. Pollinators were observed moving between cultivated and wild patches of *Cyclopia* during foraging bouts, indicating their potential to transfer pollen. Between species crosses produced seeds (*C. subternata* x *C. genistoides*), illustrating the ability of hybrids to form as a result of geneflow. Such data are important in determining safe planting guidelines for the honeybush tea industry.

Systematics

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Systematics of southern Africa species of *Rhynchosia* (Cajaninae, Phaseoleae, Fabaceae)

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Rhynchosia is the largest genus in the subtribe Cajaninae with ca. 79 species in southern Africa. Due to a lack of a recent taxonomic revision in type section *Rhynchosia*, the species are poorly known and as a result are difficult to identify, while their distribution ranges and the correct typification have not been properly established. A systematic study was carried out to provide a comprehensive taxonomic revision of the type section *Rhynchosia* in southern Africa using vegetative and reproductive morphology. Phylogenetic analyses were also carried out to assess the phylogenetic relationships within the genus and the subtribe Cajaninae, as well as to determine whether the current sectional classification is supported, using a combination of morphological and molecular data from DNA sequences of nuclear (ITS) and plastid (*rpl32-trnL*) regions. A comprehensive taxonomic revision resulted in the recognition of 47 species, of which two are newly described and several others synonymised. The phylogenetic study revealed that the genus *Rhynchosia* is paraphyletic based on molecular data sets from the two DNA markers, the genera *Eriosema* and *Bolusafra* are embedded within the genus *Rhynchosia*. Furthermore, the phylogenetic tree obtained did not support Baker's sectional classification as none of the sections was recovered as monophyletic in the phylogeny. Ancestral node reconstruction of selected morphological characters revealed significant overlaps between the genera *Bolusafra*, *Eriosema*, and *Rhynchosia*. Hence, there are no useful diagnostic apomorphies to separate the genera, and as well reinforce the need for a new infrageneric and intergeneric classification.

Phylogeny of the southern African marigolds: the daisy tribe Calenduleae

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The African marigolds (tribe Calenduleae) are a small tribe of ~ 130 species in the daisy family Asteraceae (Compositae) that are widely used in horticulture and as pharmaceuticals. The taxonomy of the southern African members has been neglected since the last comprehensive revision in 1945, leaving much of the variation undescribed. Until recently, the tribe comprised 12 genera; this was reduced to just three based on a phylogenetic hypothesis that included only 24 terminals (20 % of species). As currently defined, *Garuleum* (8 spp.) occurs mainly in the summer-rainfall zones of southern Africa, while *Osteospermum* (~ 80 spp.) and *Dimorphotheca* (~ 25 spp.) are centred in the winter-rainfall Greater Cape Floristic Region, with a few species reaching other temperate zones, including a small radiation in the Northern Hemisphere (the genus *Calendula*). Here, we present a molecular phylogeny based on both nuclear and plastid gene sequences and incorporating the largest sample of Calenduleae species to date (>50% of species). Our phylogeny confirms the non-monophyly of many genera in the pre-2012 taxonomy. The current generic taxonomy is supported with the exception of one species of *Dimorphotheca*, which is placed within *Osteospermum*. Field and molecular studies continue to discover undescribed variation in the tribe.

In search of the true identity of *Nemesia ligulata*

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Before any proper conservation of a specific taxon can be addressed, the foundational knowledge of the circumscription needs to be well understood. To this end a taxonomic study of the genus *Nemesia* is needed. The genus consists of ±77 species of annuals or perennial herbs to sub-shrubs, endemic to Africa, distributed in southern tropical Africa including Zimbabwe, Mozambique and southern Africa. The centre of highest diversification is in the Western Cape Province, South Africa. *Nemesia* are attractive annual plants with variously coloured flowers, and are therefore ideal for cultivation and hybridization, which has led to numerous hybrids being produced. Plants are popular with gardeners as bedding plants, and used in baskets, window boxes and containers. *Nemesia* has been identified in the *Strategy for taxonomic research in South Africa 2015–2020* as one of the genera in need of revision. As this study has just commenced, in this presentation the true identity of *Nemesia ligulata* will be discussed as an example illustrating the confusion that results from a blurred circumscription of a taxon.

The taxonomy and systematics of sections *Compressogemmatae* and *Sericanthae* in the genus *Millettia* (Millettieae: Fabaceae) in Africa and Madagascar

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The genus *Millettia* belongs to the tribe Millettieae in the legume family Fabaceae. As traditionally circumscribed, the tribe comprises over 50 genera and nearly 903 species and is predominantly pantropical in distribution with a few species extending to subtropical and one extending to temperate areas. *Millettia* was erected by Wight and Arnott to accommodate two species, *Millettia rubiginosa* and *M. splendens*. Since then many species have been added to the genus whereby there are now ca 150 species in tropical and subtropical Africa, Asia, and Australia. In his revision of the genus in 1912, Dunn placed these species in 15 sections, namely; *Afroscandentes* (8 spp.), *Albiflorae* (4 spp.), *Bracteatae* (1 sp.), *Compressogemmatae* (6 spp.), *Efulgentes* (25 spp.), *Eurybotryae* (*Austro-millettia* [3 spp.]) (12 spp.), *Fragiliflorae* (19 spp.), *Macrospermae* (4 spp.), *Otosema* (10 spp.), *Podocarpae* (5 spp.), *Polyphyllae* (3 spp.), *Robustiflorae* (1 spp.), *Sericanthae* (23 spp.), *Truncatocalyces* (5 spp.), and *Typicae* (9 spp.); however, some species from Madagascar have no sectional placement. These sections were mainly distinguishable based on the presence or absence of the stipels and indumentum on the petals, base of the standard petal (auriculate or exauriculate), and other floral characters such as the pedicels, bracts, petal colour, and pods. Recent molecular evidence however has shown *Millettia* to be polyphyletic with an estimated number of species between 90-200. The aim of this presentation is to present preliminary results on the taxonomic revision of sect. *Compressogemmatae* and *Sericanthae*, in Africa and Madagascar.

Systematics of *Cotula*: Reflections and challenges of working on a cosmopolitan genus

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Phylogenetic analyses of nuclear (nrITS) and plastid DNA (*psbA-trnH* and *trnC-petN*) of *Cotula* have shown that *Cotula* is paraphyletic, comprising four main clades, with *Leptinella* and *Soliva* embedded within. Based on these phylogenetic and morphological data *Cotula* is re-circumscribed to form one large genus, i.e. *Cotula sensu lato*, comprising almost 100 species with a cosmopolitan distribution. An infrageneric classification of this large and diverse genus is proposed with *Cotula sensu lato* divided into two subgenera: subgenus *Soliva* and subgenus *Cotula*. Subgenus *Soliva* includes all the South American species previously circumscribed in *Soliva* as well as the closely allied South America *Cotula* species, *C. mexicana*. This subgenus is supported by the presence of only male disc florets and hairy receptacles. Subgenus *Cotula* includes all the Australian and New Zealand species previously circumscribed in *Leptinella* as well as the remaining species of *Cotula sensu stricto*, distributed mostly in southern Africa but with some species extending to or endemic to tropical Africa, Eurasia and Australia. This subgenus is supported by the absence of spiny appendages on the disciform florets. A sectional classification is also proposed. In this presentation, the challenges, and main findings of working within such a diverse cosmopolitan genus will be discussed.

A taxonomic revision of the *Thesium scirpioides* species complex (Subgenus *Frisea*, Santalaceae) near endemic to South Africa

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The genus *Thesium* L. is in need of revision and has been identified by the South African National Biodiversity Institute as a high priority for taxonomic research in South Africa. As part of an ongoing effort to revise this large genus (ca. 350 species worldwide and ca. 170 species in South Africa), a group of nine morphologically similar taxa from Subgenus *Frisea* was studied. The aims were to 1) reassess and confirm taxon concepts within the group (referred to as the *T. scirpioides* complex), 2) provide a full taxonomic treatment for each taxon, 3) provide an up-to-date identification key for the group, and 4) reassess the conservation status of each of the taxa. Species of the *T. scirpioides* complex share the following characters: (1) inconspicuous unarmed herbs or suffrutices, (2) scale-like leaves, (3) indeterminate spike-like inflorescences, (4) post-staminal hairs attaching the anthers to the perianth and (5) twisted placental columns. Based on morphological discontinuities and geographical distributions, seven species are recognised, including one species new to science, *T. atratum* N.Lombard & M.M.le Roux. Both the taxonomic status and conservation status of three taxa hitherto classified as Data Deficient are now resolved. A status of Endangered is suggested for *T. lisae-mariae* Stauffer, while *T. junceum* Bernh. var. *plantagineum* A.W.Hill and *T. junceum* var. *mammosum* A.W.Hill are synonymised under *T. junceum* var. *junceum* (classified as Least Concern). An identification key, updated nomenclature and typifications, descriptions, diagnostic characters, distribution maps and conservation notes are also presented.

Taxonomy of *Epichroxantha*, a recent segregate of the genus *Gnidia* (Thymelaeaceae)

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Phylogenetic studies have shown that *Gnidia* L. is polyphyletic and that there is a need to refine the circumscription of the genus and recognise several additional segregate genera. One such segregate includes species previously accommodated in the genus *Epichroxantha* Eckl. & Zeyh. ex Tiegh. The species in this genus are recovered in a clade sister to the Australian genus *Pimelea* and can be distinguished by the presence of four membranous petaloid scales, 8 stamens, and the few-flowered heads with yellow hypanthia. Although several species were listed under *Epichroxantha* by Ecklon and Zeyher, no formal name combinations within *Epichroxantha* currently exist for any of the species. We here clarify the generic circumscription of *Epichroxantha*, formally transfer the species with new combinations and present a taxonomic revision of this South African endemic genus. Eleven species are here recognised including a newly described species. These species occur in the Western Cape in the Cape fynbos biome and in the Eastern Cape in the Albany thicket and grassland biomes and they are distinguished by growth form, flower morphology and ecology. Diagnostic characters, ecology and distribution of the species are also discussed.

Taxonomic revision and molecular phylogenetic analysis of *Nemesia* species in the Free State

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Nemesia Vent. is one of sixty-two genera from the Scrophulariaceae family. The genus is part of the Hemimerideae tribe, which is the earliest branching phylogenetic lineage of Scrophulariaceae. *Nemesia* comprises seventy-seven species of annual and perennial herbs, and sub-shrubs which are all native to southern and tropical Africa. The genus was listed as one of many priority South African genera that are in need of taxonomic revision by SANBI in 2015, on a list compiled based on time since last revision, level of endemism, collecting effort, proportion of taxa included in revisions, and specimen identification confidence. The only published molecular phylogenetic study of *Nemesia* was conducted by Datson, et al. in 2008 using DNA nucleotide sequences of the nuclear internal transcribed spacer (ITS), external transcribed spacer (ETS) and the chloroplast *trnL*-spacer regions. The study included twenty-three *Nemesia* species, and only five of them were species occurring in the Free State. In this study a systematic analysis of *Nemesia* species occurring in the Free State was conducted, with the molecular studies done using the same gene regions as Datson et al in 2008. Descriptions and typification of these species were corrected, and as many Free State *Nemesia* species as possible were added to the *Nemesia* phylogenetic tree.

Speed talks

Impact of flow conditions on the dynamics of phytoplankton groups in the Sabie River, South Africa.

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Climate change is one of the emerging (since 2006) factors that impact on and threaten freshwater biodiversity and aquatic ecosystem health. The effect of climate is more difficult to quantify since precipitation and temperature affect the flow of water, whilst nutrient loading is known to alter phytoplankton assemblage, diversity, and biomass. In this study, we have considered the impact of different flow conditions, on the presence of phytoplankton groups in the Sabie River. Although the chlorophyll-a concentrations were higher during low flow conditions, a slightly lower number of genera were observed for the class Chlorophyceae. During higher flow conditions the number of genera of the class Cyanophyceae decreased together with chlorophyll-a and total cell concentrations. Low flow conditions favoured filamentous genera that are common to mesotrophic conditions while higher flow conditions favoured single-celled small genera more common to oligotrophic waters with higher disturbance. This study showed that flow has the potential to impact on phytoplankton community dynamics in a river system and thereby may have further impacts on the health and activities of surrounding communities.

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Flowering Plant Families of southern Africa

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The publication of 'Guide to Plant Families of southern Africa' (2013) led to an expansion of the project to cover all the flowering plant families of the region. The expanded publication will be available soon. Each family is introduced by images of the key characteristics of the family, a distribution density map, and general information about the size of the family, growth forms, habitats and significance of members of the family. Larger families are further illustrated by representative images of species. For the families with more than 500 species the second level classification is discussed and separately illustrated. The forthcoming publication covers 240 plant families and includes introductory information about classifications, statistics and identification of plant families.

Effect of geology on the tree species composition of the olive forest in the Vredefort Dome

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The Vredefort Dome is the largest and oldest impact crater worldwide. The Dome contains complex geological stratigraphic layering, each with its own plant communities comprising of distinct species assemblages. Although wild olive forests are common on southern aspects of the Vredefort Dome, limited geobotanical studies have been conducted to determine whether geology and associated mineral composition influences their plant diversity. Therefore, the aim of this study was to investigate the relationship among geology, soil mineralogy and species composition. This will provide useful information in soil-plant associations regarding soil and geology in the wild olive forest. The belt transect method was used for counting all woody species on each geology and to sample rock and soil. PERMANOVA results of floristic data indicated that wild olive forests had distinct species assemblages which were linked to the underlying geology. Soil variables influenced the indicator species for each geological substrate, such as *Grewia occidentalis* and *Olea europaea* subsp. *africana* for epidioritic and granitic soil with high calcium content. *Grewia flava* assembled on soil high in phosphorus and potassium, while *Celtis africana* was associated with soil high in chromium and manganese, and *Combretum molle* assembled on soil high in barium and strontium. Thus, a clear relationship was observed between the soil and the associated species composition of the different geological units in the wild olive forest. This study provides the first assessment of soil-plant associations in this economically important wild olive forest and practices provide valuable baseline information for future monitoring and restoration.

Taxonomic studies of the genus *Dalbergia* (Fabaceae) in southern Africa.

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Dalbergia is a genus of trees that belongs to the Fabaceae family. The genus consists of 250 species and occurs in the tropical and subtropical regions of the world. *Dalbergia* belongs to subfamily Papilinoideae within Fabaceae. Fabaceae is a large and economically important family of flowering plants. *Dalbergia* includes several species of high economic and ecological value. These species can be amongst the most threatened due to illegal logging and the timber trade. To help with protecting and ensuring effective conservation of *Dalbergia* species, the identity of species needs to be well known. The aim of this study was to revise and update the nomenclature, taxonomy, and geographical distribution of *Dalbergia* species (indigenous and alien) occurring in southern Africa. In southern Africa, seven species are recognized of which six species are indigenous and one non-indigenous species naturalized in South Africa.

The effect of geology on the prominence of *Olea europaea* subsp. *africana* in the Vredefort Dome

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The Vredefort Dome is the oldest and largest meteor impact site on Earth and comprises unique geological structures that host an edaphically determined indigenous flora. A prominent species in the dome is *Olea europaea* subsp. *africana* (wild olive), an evergreen tree which is widespread across South Africa. Along the core-collar boundary of the dome it forms dense forest clusters on south-facing slopes. Large individuals of this species are frequently harvested and the wood exported, as it is economically valuable. Previous studies of the wild olive forests indicated a facultative association with epidiorites. Our study tested this assumption by comparing the performance of wild olive trees on adjacent geological strata (i.e. epidiorite, granite and quartzite). Transects were laid out according to geological units, wild olive trees were counted and their stem circumference measured. Rock and soil samples were also assessed, for chemical characterization. Comparisons were made across strata with Kruskal-Wallis tests ($\alpha = 5\%$), which indicated that both abundance and stem circumference on epidiorite differed significantly from those found on quartzite, but not with granite. Our study therefore confirms that epidiorite is not obligate for wild olives, as they also occur readily on granite. Exploratory analyses with PERMANOVA suggested that epidioritic and granitic soils were more favorable as both abundance and stem circumference correlated with higher calcium, pH and CEC, but that the observed stunted growth on granite could possibly be ascribed to higher-than-normal strontium levels. This aspect requires further investigation.

Two unlisted suspicious alien plants in KwaZulu-Natal to look out for

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The Directorate on Biodiversity Evidence (DBE) in SANBI (funded by the Department of Forestry, Fisheries and the Environment) assesses alien taxa starting to show invasive potential. For the purposes of this talk, two such species, referred to as “emerging” invasive alien plants (IAPs) known to occur in KwaZulu-Natal (KZN), will be discussed. *Alstroemeria psittacina* (Alstroemeriaceae), commonly called “Princess lily”, is a multi-stemmed plant (<1m) native to South America. It has underground tubers that can survive winter, and spoon-shaped leaves arranged in a rosette. Plants are crowned with attractive funnel-shaped flowers in different shades of red, orange and purple, that can be flecked, striped, or with brownish spots. Erect capsules split open to release tiny seeds. It can also reproduce vegetatively from cuttings and rhizomes, forming dense groundcover posing a threat to forests and forest margins. One population is known from Westville. The second species, *Brasiliopuntia brasiliensis*, also native to South America is one of the tallest Cactaceae members. Growing up to nine meters, it can resemble a tree with an erect trunk. Armed with reddish-brown spines, this species produces large, bright yellow flowers, and red bell-pepper shaped fruit when mature. Populations are known from northern KZN and Limpopo. Commonly called “Brazilian Prickly Pear,” this cactus can reproduce via seed and vegetatively, posing a threat to native forest species by reducing habitat availability and accessibility. Sighting reports are encouraged (invasivespecies@sanbi.org.za) to improve species assessments, which will result in more accurate risk analyses, improved decision-making, and subsequently, effective management and even eradication.

Non-marine algae and cyanobacteria of South Africa, Kingdoms of Eswatini and Lesotho: taxonomical checklist preparation launched

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Main priorities in algal systematics research are to develop a checklist of all known species in South Africa, database associated information, make this information accessible online, and to stimulate research projects addressing taxonomic problems (Bolton 2018). As an initial step in realizing of this aim we started preparation of taxonomic checklist of algae and cyanobacteria recorded in freshwater, terrestrial and aerophytic ecosystems of South Africa, Kingdoms of Eswatini and Lesotho. This checklist will include all taxonomical groups known for this area from all known scientific literature since Gottlob Ludwig Rabenhorst published his work on South African algae in 1855. As an example we can show preliminary information about diversity of desmids (Charophyta, Zygnematophyceae, Desmidiiales) in freshwaters and soils of studied area. In total, according to the published literature data, 563 species (948 intraspecific taxa including type taxa) were recorded in Southern Africa. Three hundred and seven of these taxa were described as new for science i.e. 110 new species, 1 new subspecies, 165 new varieties, 29 new forms, 1 new name, 1 new combination. All documented with detail geographical and ecological information of sampling sites. Preliminary number of algae and cyanobacteria in terrestrial and aerophytic ecosystems is 265 species (including cyanobacteria – 80 species, Chlorophyta - 45, Charophyta – 59, Xanthophyta – 2, Eustigmatophyta - 2, Bacillariophyta – 74, Euglenophyta – 2, Dinophyta - 1). All documented with detail geographical and ecological information of sampling sites.

First record of the green algae *Hormotilopsis gelatinosa* Trainor et Bold in Africa and taxonomical status of its name

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In serpentine soils collected near Kaapschehoop (Mpumalanga Province, South Africa) we found an interesting green alga which was identified as *Hormotilopsis gelatinosa* Trainor et Bold 1953 (Chlorophyta, Chaetopeltidales). Intensive literature searches showed us what this is the first record of *H. gelatinosa* in Africa. Previously it was found in USA, Ukraine, Russian Federation, Spain, UK, Bulgaria, Iran, and Brazil. However, there are some dispute about the name, since Aleksandr Korschikov described *Gloeophyllum fimbriatum* in 1953. These two algae have similar morphology and life cycles, and we propose that the name *Gloeophyllum fimbriatum* must be used and *H. gelatinosa* must be regarded as a synonym. Differences in ecology and general appearance of colonies are considered insignificant and if we assume that these taxa are one species, it is unclear why the name "*Hormotilopsis gelatinosa*" is the preferred name, since both descriptions date back to 1953. An article of Francis Trainor and Harold Bold was published in December 1953, whereas Korschikov's book was signed for printing on 15 December 1952 and printed at the beginning of the next year! In addition, it is known that this book was written before the WW2, but published later, after the author's death (he died in Nazi concentration camp Dora Mittelbau in 1945). Later *G. fimbriatum* underwent another nomenclature change (*Phyllogloea fimbriata* (Korschikov) by R. Silva in 1959) but this generic name already was used for a basidiomycete fungus. In our opinion the name *Gloeophyllum fimbriatum* Korschikov 1953 is much more legitimate and without a doubt *H. gelatinosa* Trainor et Bold 1953 and *Ph. fimbriata* (Korschikov) Silva 1959 must be reduced to synonyms.

Vitality evaluation of urban oaks (*Quercus robur* L.) using physiological parameters

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Trees in an urban setting, such as the exotic Oak (*Quercus robur* L.) trees in Potchefstroom (North West province, South Africa), provide several ecosystem services on tree, street and city level. Since urban trees are very favorable to urban life, it is pivotal to study their health and conserve these trees as long as possible. *Quercus robur* trees in the historic Oak Avenue in this city have been showing a decrease in vitality since 1980, for which the cause is still largely unknown. This study aimed to determine the vitality of a section of the trees in the avenue with different types of land cover by estimating visual tree conditions and measuring leaf water potential, nutrient analysis and chlorophyll *a* fluorescence. Data obtained indicated that the parameters and indicators used to determine tree vitality were beneficial in monitoring plant health; however, this data alone does not account for the health decline of the trees. It was determined that trees with the dominant land cover of compacted bare soil pose a greater risk to urban residents in terms of structural failure than trees with natural / vegetative cover. The fluorescence parameters PI_{TOTAL} and F_V/F_M proved little support in identifying trees with health issues or declining vitality. Oak trees in the avenue in Potchefstroom should be re-evaluated and monitored carefully in the future to establish trends in changes to vitality and ensure successful conservation by scientists, city residents and municipal authorities.

Metals in soil and plants of South Africa: a not-so-complicated relationship

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South Africa has a myriad of geologies and mineral compositions. Not surprisingly, its indigenous flora is also represented by great diversity and species-specific features. One of these is the hyperaccumulation – the ability to uptake metals in concentrations usually harmful to most of the species. In the last two years, we have evaluated the relation between the concentrations of metals in different soil types and the presence (i.e. abundance) of indigenous, exotics and endemic plants in specific areas of South Africa. Our findings revealed that metal concentration in plant leaves can be generally determined by the concentrations in the atmosphere and the soil, especially in those enriched by anthropogenic land use (e.g. mining activities). Moreover, many of the investigated plants are metallophytes – species capable of thriving on soils with high concentration of PTM's (potentially toxic metals), including through bioaccumulation. Besides the health risks related to the consumption of useful plants – medicinally and as food, it was also possible to identify potential phytoextractor species, which could be employed for phytoremediation and phytostabilization of contaminated soils. As perspectives, we intend to dive deeper in the relationship metals-soil-plants, adding primary and secondary consumers (i.e. herbivore and their predators) of the food chain, especially in areas with naturally metal-enriched soils (e.g. serpentines), to understand the mechanisms (i.e., ecological, physiological, behavioral, etc.) behind the survival of the individuals and the maintenance of the ecological processes in such challenging environments.

Distribution and spread of *Ulmus parvifolia* (Chinese elm) in South Africa

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Ulmus parvifolia known as Chinese elm, in the family Ulmaceae, is native to Asia. It is a deciduous tree and is planted as an ornamental tree in many countries. Its distinctive features are dark green leaves, multicolored exfoliating bark, and samara seeds. It can withstand harsh environments and requires a lot of water supply due to its root-system that can extend in great distances from the main trunk. In South Africa, *U. parvifolia* has been recorded naturalized in several habitats including riparian areas and is showing signs of being invasive in some areas. Riparian areas extend from the river margins to the limits of the area between the aquatic system of the river and terrestrial ecosystem. The aim of the study is to investigate the distribution and spread of *U. parvifolia* in South Africa and to quantify the impacts in urban riparian habitats. We investigated the distribution in South Africa, the reproductive strategy, and the impacts on plant communities. We studied distribution and spread from collection records obtained from PRE, NH, NBG, Southern African Plant Invaders Atlas, supplemented by field observations made between 2012 and 2021 by field biologists and the authors. Preliminary results indicate that *U. parviflora* has spread across six provinces of South Africa and the first record of a naturalized population dates back to 1979 from Potchefstroom Municipal area along railway reserves. It spreads mainly by seeds (winged and wind-dispersed), self-propagation and may resprout from root suckers.

Current status of the invasive shrub *Berberis julianae* Schneid. (Berberidaceae) in Golden Gate Highlands National Park (Free State Province, South Africa).

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Berberis julianae, commonly known as Wintergreen barberry, is a shrub species native to China which has been introduced into South Africa and is found growing along the riverbanks of a few highveld locations. Keet *et al.* (2016) identified a need to put control measures in place to manage an increasing population in Golden Gate Highlands National Park (GGHNP) in the Free State Province. After assessing the plant demographics within the park, cut stump and seedling removal control measures were implemented. The authors recommended that follow-up research should be done to check if their control measures were a success or not. This study aimed to map and compare the current distribution of *B. julianae* in GGHNP and to investigate the change in *B. julianae* population demographics to that recorded by Keet *et al.* (2016). The study showed that the number of seedlings had multiplied in number from 6 seedlings in 2014 to 588 in total in 2020, while the number of mature shrubs had decreased. In conclusion the control measures that were put in place by Keet *et al.* (2016) were successful in reducing the adult population however follow-up control of seedlings is still required.

***Desmodium* Desv. (Leguminosae, Papilionoideae) species naturalised in southern Africa and key to distinguish between native and alien species.**

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The genus *Desmodium* is widely distributed globally, comprised of three sections. Although the genus and its impacts are well understood in other parts of the world, the distribution and invasive impacts of some species in southern Africa region is not well studied. This might be due to the difficulty in distinguishing the species in the genus. Identification keys and risk analysis are necessary for the early detection and rapid response to manage alien invasive species. There is a need for an updated identification key to identify *Desmodium* species. Furthermore, the need to assess the invasion status of the naturalised species. Herbarium specimens and literature sources were studied to develop an identification key of southern African *Desmodium* species. Risk analysis for three naturalised species of *Desmodium* was conducted using the Risk Analysis for Alien Taxa framework developed for South Africa to assess and analyse their invasion risk. Combining vegetative and reproductive characters resulted in the most complete dichotomous key available for *Desmodium* species in the region. Preliminary results for the risk analyses classified all three naturalised species, *D. tortuosum*, *D. uncinatum* and *D. incanum*, as category 1b under NEMBA. This work makes a first contribution in understanding indigenous and alien *Desmodium* species as well as understanding the invasion risks of naturalised taxa in South Africa. Since the last revision of the genus *Desmodium*, the number of recognised species in the FSA region has increased from ten to 15, of which 12 are indigenous and three naturalised.

Effects of different scarification treatments on seeds germination and seedlings growth of *Prosopis velutina* and *Vachellia erioloba*

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In South Africa, the alien invasive *Prosopis velutina* and the protected *Vachellia erioloba* are two arid woody species belonging to the family Fabaceae. They produce large amounts of seeds which are known to have hard seed coat resulting in poor germination. This study was undertaken with the aim to evaluate seed germination and seedling growth of these two species to different scarification treatments. All the experiments were conducted at North-West University. Seeds were subjected to a total of 5 treatments such as the immersion in sulfuric acid (98%) for 30 and 60 minutes respectively, the slight and intensive abrading with sandpaper and control. Each seed was sown in a perforated black plastic bag containing sandy soil and watering was done every three days. A completely randomized design with 4 replicates was used. Four variables were measured as follows: germination days, germination percentage, number of leaves and seedlings height. All data were analyzed using ANOVA to identify significant difference among treatments at $p < 0.05$. The results showed that treating both species with H₂SO₄ for 30 minutes resulted in faster germination (2 to 3 days). *Prosopis velutina* was the first that germinated in 2 days and *Vachellia erioloba* germinated on the third day than the control (36 days). Slight Sandpaper Scarification (SSPS) treatment had achieved the highest germination percentage with *P. velutina* (16.7%) and *V. erioloba* (27.8%), whereas the lowest was recorded in untreated seeds with *P. velutina* (2.8%) and *V. erioloba* (5.5%). Seed germination of the two species was significantly ($p < 0.05$) affected by different treatment methods but with an overall low germination percentage. Furthermore, no treatment performed significant differences in the number of leaves and seedlings height.

Seed biology and dispersal of *Cotoneaster pannosus* in grasslands of eastern Free State.

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South African grassland vegetation is threatened by various invasive woody plants of which *Eucalyptus* and Pine species have dominated and have therefore demanded most of the attention regarding research and management. However, members of the Rosaceae family are becoming problematic and should be considered. Due to the increased spread and abundance, *Cotoneaster* species are posing a major risk to ecosystem services and biodiversity in KwaZulu-Natal, Eastern Cape, and eastern parts of the Free State. Despite this, there has been very little research conducted that focuses on the drivers of *Cotoneaster* invasions in South Africa and their impacts. Here we explored the seed biology of *Cotoneaster pannosus* Franch., native to China, that has established in montane grasslands of the eastern Free State, by investigating fruit availability, seed production and seed dispersal. Results show that *C. pannosus* has a lengthy fruiting duration that persists through winter into spring. Furthermore, *C. pannosus* has a large fruit crop size with an average of 4,000,000 seeds produced per m³ with a significant positive relationship between shrub volume and seed production. Fruits are consumed by frugivores including Cape white-eyes, red-eyed bulbuls, mice, and livestock which suggests that long distance seed dispersal is facilitated by indigenous birds and mammals. High seed abundance, extended fruit availability and efficient avian dispersal are associated with weediness which adds to the invasive potential of *C. pannosus*. It is hoped that these findings will be useful in addressing infestations, especially if populations are small and detected at an early stage of invasion.

Indigenous livestock husbandry and ethnoveterinary practices under low input farming systems of the Eastern Cape Province.

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The use of medicinal plants is an alternative option for livestock owners who cannot afford to use allopathic drugs for animal health problems. Eastern Cape is characterised by a wide range of climatic and ecological conditions thereby possessing enormous diversity of fauna and flora. However, available evidence on indigenous ethnoveterinary plants is highly fragmented. Very few studies conducted on medicinal plants used for the treatment of livestock diseases in the province. The aim of the study was to conduct the overview of the medicinal plants used for the treatment of livestock diseases in the Eastern Cape Province. Data for this review were generated from published peer reviewed journals, conference proceedings and theses. There were no restriction on the period of publications and no discriminations were made in relation to methodologies applied when collecting data. Focus was only on flora found in the Eastern Cape Province. Data were analysed using descriptive statistics and to calculate frequencies, percentages and generate graphs Microsoft Excel (2016) was used. Publications were searched using the following scholarly search engines; Pub Med, Google Scholar, Research Gate, Open Access Journals. This search produced 19 articles. Most studies were done within the radius of 200 km around Fort Hare University. The results showed a total of 56 plant species belonging to 33 families. Helminths was the most prevalent disease. The most commonly used plant families were Asphodelaceae 10.7%, Fabaceae 8.9%, Lamiaceae 8.9%, Rubiaceae 5.4%, Rutaceae 5.4%, Asteraceae 3.6%, Apiaceae 3.6%, Hypoxidaceae 3.6% and Asparagaceae 3.6%. The most widely used plant parts for the preparations of remedy were leaves (38.7%), followed by roots (24.2%) and bark (19.4%) and used in solution form by using water as a most cited solvent. From this review, it can be safely concluding that, medicinal plants play a major role in healthcare of livestock in rural communities. Therefore, there is an urgent need to conduct further research to document, screen and perform clinical trials on medicinal plants.

Assessing the likelihood of major plant species distribution under climate change scenarios along an aridity gradient in Namibia

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Climate change is expected to have major impacts in altering the distribution of plant species worldwide. Global rainfall and temperature averages have changed over the past years; in southern Africa an increase in temperature and reduced precipitation has been recorded. Plant species are affected by these changes in one of three ways: the timing of seasonal activities (phenology), physiology under increased stress conditions and distribution space. Namibia, which is the driest country in southern Africa, is projected to have an increase in temperature of 2°C by the end of the 21st century. This will exacerbate the aridity conditions in the country. Namibia's vegetation is characterized by open woodland in the north-eastern parts of the country (Kavango) to low open shrubs with scattered trees in the southern part of the country (Karas). These differences are a result of increasing aridity from north to south. North-eastern Namibia receives an average rainfall of 600 – 800 mm per annum, whilst southern regions receive as little as 100 – 160 mm rainfall on average per year. The climatic envelope of present-day vegetation has been established, from here this study aims to predict the effect of shifting climatic conditions on the major vegetation units and their dominant species along this aridity gradient. Environmental variables from two general circulation models – the IPSL-CM5A-LR developed by Institut Pierre Simon Laplace and HADGEM2-ES developed by the UK Hadley Centre are to be used in this modeling approach.

Effects of legume forb encroachment on an abandoned ploughed grassland: legacy of ploughing, or risk to productivity?

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Grasslands in South Africa support extensive agriculture, from crop production to valuable forage provision for livestock and game. The intactness of natural grasslands is, however, affected by direct and indirect effects of agricultural practices, particularly ploughing. Ploughing is considered an exogenous disturbance that permanently alters soil properties, which may have various effects on plant community composition, diversity and productivity. The link between historical ploughing and encroachment by indigenous, competitive legume forb species in grasslands is not well understood. Here, we assessed the potential negative effects of high densities of a native herbaceous legume, *Indigofera cryptantha*, on the productivity and diversity of a previously ploughed Dry Highveld Grassland ecosystem in the North-West Province. Comparisons to an adjacent natural grassland site confirmed *I. cryptantha* densification in abandoned ploughed fields (APF), where floristic diversity, veld condition and biomass production were significantly lower. Soil chemical analyses revealed a nutrient spike in the APF, specifically P, even after >10 years since fertilized/cultivated. Legumes perform particularly well under locally enhanced soil P conditions, especially in P-limited systems, explaining the expansion of this N-fixing, palatable forb into APF. Our results could not support obvious negative effects imposed by higher densities of *I. cryptantha*, since they seem to facilitate the growth of high-quality forage grasses, such as *Digitaria eriantha* through localized N-enrichment in the degraded APF. More studies on long-term effects of ploughing on soil properties, floristic diversity, productivity and ecological function in grassland ecosystems are needed to improve management recommendations to perceived encroachment risks.

Saving *Marasmodes undulata*: Reintroduction of a Critically Endangered species

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Marasmodes is a small genus in the Asteraceae family and is considered to be one of the most threatened genera in South Africa. *Marasmodes undulata* is listed as Critically Endangered and is only known to occur on one 6ha conservation area in Paarl, Western Cape. Historical records suggest that the population has declined steadily from over 500 plants in the early 90's to 3 plants in 2018. A project was initiated to collect seeds and cuttings to propagate the species and reintroduce the species. Seeds were sourced from the Millennium Seedbank at KEW gardens in the UK and propagated at the Kirstenbosch and Stellenbosch Botanical Gardens. In addition to the reintroduction of this species, other associated plants were grown to facilitate the improvement of the natural habitat. We will share the results of our propagation trials and highlight the challenges of reintroducing species with limited source material and knowledge of successful propagation methods. We will also make recommendations for threatened species ex-situ conservation based on lessons learnt from this project.

***In vitro* propagation of *Lippia javanica* (Burm.f.) Spreng: an important medicinal plant**

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Lippia javanica is an important medicinal plant of economic value used as herbal tea to treat ailments such as in colds, cough, fever, malaria, wounds, diarrhoea, chest pains, bronchitis, and asthma. Due to its popularity in the medicinal plant markets, unsustainable overharvesting in the wild has been noted. Therefore, an alternative supply of medicinal plant material is required. In the present study, *in vitro* plant propagation of *L. javanica* was conducted as an alternative propagation method in order to meet the medicinal demands of this species. For direct organogenesis, nodal explants were cultured on MS medium (control-no PGR), whereas for indirect organogenesis, leaf explants were cultured on MS medium containing cytokinins (BAP&KN: 2 mg/l, 4 mg/l, 6 mg/l) alone or in combination with auxins (Picloram and NAA: 1 mg/l, 2,4-D: 0.5 mg/l, 1 mg/l). The results showed that 80% of the nodal explants produced axillary shoots within two weeks of culture, with an average of 3.7 shoots, 7.1 nodes per explant after four weeks, and 70% of the shoots produced adventitious roots (3.1) and were successfully hardened. All leaf explants produced callus after four weeks of culture in all the tested PGR combinations. The study indicates that both direct and indirect organogenesis of *L. javanica* can serve as an alternative supply of plant material, as well as the production of secondary metabolites for commercial purposes.

The influence of serpentine soils on the early development of *Berkheya radula*, a non-serpentine species.

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Serpentine soils are rich in heavy metals, which are potentially toxic to plants. Although serpentine-adapted species have been studied in detail to determine their ability to tolerate these harsh conditions, there is a lack of information on non-serpentine congeners' ability to survive on serpentine soil. As the relationship between phylogeny and metal hyperaccumulation is well documented, we test whether serpentine tolerance is also inherited. *Berkheya radula*, a non-serpentine congener, was grown in a pot experiment to compare early developmental features (i.e., germination rates, leaf count/length, biomass) and photosynthetic parameters in serpentine versus non-serpentine soils than that of the control. The heavy metal content was found to be significantly higher in the serpentine treatment. Over 90 days, no differences were observed for germination rates leaf counts, nor did lengths among treatments. However, seedlings of serpentine exceed the control in terms of the photochemical efficiency. This early success of the seedlings of a non-serpentine *Berkheya* species suggested that (1) the germinated plants may inadvertently uptake metals as they scavenge for the nutrients in the nutrients-poor serpentine soil, with consequences to be manifested later in the development; and/or (2) that the genus possibly harbours an inherent ability to tolerate soil rich in heavy metals, which might be a facultative ability of species not found in serpentine soils. The next step is to analyze leaf elemental levels at maturity to test for hyperaccumulation, a well-known trait of the genus.

***Prosopis*: an ethnobotanical relevant plant or an environmental problem? An assessment of indigenous knowledge**

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Prosopis is an alien woody legume plant species that occurs as shrubs and mature trees. It is the second largest invasive plant in South Africa after Australian *Acacia*. Although these alien species are invasive and have negative impacts on natural ecosystems, indigenous communities still derived basic benefits from some such *Prosopis* spp. The study was aimed on determining whether *Prosopis* is a possible solution for grazing or an environmental problem in the community, through assessing the indigenous knowledge of the species. An ethnobotanical survey was conducted using a semi-structured questionnaire accompanied by photographs of *Prosopis* among residents of Mahikeng Local Municipality in 7 selected villages. 37 participants from all the villages form part in the survey. 94.6% of the participants identify at least one of the *Prosopis* spp. with related ethnobotanical uses. The main uses of the species are among the community members are cultural, medicinal, forage for cattle, source of energy, fencing and nutrients. Majority of the participants (81%) view it as a solution within communities while only about 16% of the participants know *Prosopis* species as an alien species. There is a need for more data to be generated through future studies in order to establish if it is a solution or a problem and to educate the community members of the plants which are actually alien species through community engagement forums.

A morphological revision of six *Adenogramma* species (Molluginaceae) in South Africa

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Adenogramma Rchb. is a genus within the Molluginaceae. It is distributed mainly on the coastal plains and lower mountain slopes in the Western and Eastern Cape and some parts of the Northern Cape of South Africa. It comprises ±10 species of annual or perennial herbs. The genus was first described by Reichenbach (1828). Between 1828-1957, ten species were described, followed by a description of one new species in 2011. The genus has, therefore, received little attention resulting in outdated species' description and distributions, lack of identification keys, and few specimens in herbaria. The red list status of three species (*A. congesta* Adamson, *A. physocalyx* Fenzl and *A. teretifolia* (Thunb) Adamson) can also not be assessed due to a lack of taxonomic data and *A. rigida* (Bart.) Sond. is listed as critically rare as it was last seen in 1970. This genus is, therefore in dire need of a taxonomic revision to update the species' circumscriptions, revise the distribution, assess the red list statuses of the species and provide a key for identification. This project aims to update and revise the description of six *Adenogramma* species, namely *Adenogramma capillaris* (Eckl. & Zeyh.) Druce, *Adenogramma glomerata* (L.f.) Druce, *Adenogramma mollugo* Rchb., *Adenogramma sylvatica* (Eckl. & Zeyh.) Fenzl, *Adenogramma lichtensteinana* (Schult.) Druce and *Adenogramma diffusa* Fenzl.