PLANT PROTECTION NEWS

AGRICULTURAL RESEARCH COUNCIL - PLANT PROTECTION RESEARCH

Summer 2021

No 118

In this issue

NEW: YourSoil™ – Complete soil health package created for clientele

Adult tephritid gall-fly.

Mass-rearing facility open day

General enquiries

Plant Protection Research,
Private Bag X134, Queenswood,
0121 South Africa

e-mail: infoppri@arc.agric.za • web-site: http://www.arc.agric.za

YourSoil™ – Complete soil health package using biotic and abiotc indicators

Soil health is the capacity of soil to function as a vital living system, within ecosystem and land-use boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality, and promote plant and animal health (Doran & Zeiss, 2000). Soil plays an important role in agriculture, and is the foundation of successful crop production. It is therefore important to determine the health status of a soil to ensure healthy crop production. Healthy soil means healthy crops. If a producer can improve the health status of the soil, input costs will be reduced and the sustainability of the soil increased.

There are biotic (living) and abiotic (non-living) means of determining soil health. Soil organisms play an important role in the biotic systems of soil, and they are frequently used as biological indicators of soil health for various reasons. Abiotic characteristics of the soil, including soil structure and its physical and chemical status, are directly linked to the biotic aspect of soil, as many abiotic characteristics influence the presence and abundance of soil organisms. YourSoil™ is a new package service offered by ARC- Plant Health and Protection (PHP) and ARC-Natural Resources and Engineering (NRE) that analyses biotic and abiotic components in soil.

Biotic: the first set of tests is based on the presence of biological indicator organisms, which include a number of widely accepted tests used in determining soil health. Biological indicator organisms play an important role in nutrient cycling and mineralization in soil ecosystems. They also form part of the soil food web as primary and secondary consumers of organic matter and other organisms. Soil organisms such as fungi, bacteria and nematodes react rapidly to environmental change and are therefore well suited as biological indicators of soil health. Because soil is such a complex system, the use of soil organisms as biological indicators is a unique and effective tool used to determine the biotic aspects of the health status of the soil. Clients are free to choose one or more of the following tests offered as part of the YourSoil™ package:

- **Nematodes:** Plant-feeding nematodes are particularly important in agriculture due to the damage they cause to crops. However, the largest percentage of nematodes found in the soil are beneficial. These nematodes play an important role in various soil systems and are involved in different levels of the soil food web, as a food source for other organisms, and as predators of various organisms. Nematodes are often used as biological indicators of soil health because they occur in large numbers and respond rapidly to any changes in the soil ecosystem. A faunal analysis of nematodes forms part of the YourSoil™ package and includes the identification of plant-feeding nematode species, and free-living nematode genera. In this test, the number and type of nematodes are determined and the soil is classified into one of four specific groups, based on different indices. Each group has specific characteristics that indicate the health status of the soil.

- **Rhizobacteria**: These bacteria, also known as the phyto-friendly soil microbes, occur in the rhizosphere (area around the root system) of plants and therefore represent the part of the soil with the highest microbial activity. Rhizobacteria have a high root colonization ability and facilitate vegetation through various modes of action, including solubilization of phosphorus, production of phyto-hormones, chelation of iron (Fe) and its availability to plants, as well as the production of metabolites such as ACC deaminase activity that alleviates various abiotic stresses in plants by regulating the level of ethylene. Rhizobacteria also play an important role in improving the soil ecosystem functioning and services, and the biogeochemical cycling of soil nutrients. Common Rhizobacteria include species belonging to the genera *Pseudomonas, Bacillus, Burkholderia, Azospirillum, Rhizobia* and *Azotobacter*. The presence of these microorganisms in large quantities in a given soil is a good microbial indication of soil health. The YourSoil™ package includes the following tests of Rhizobacteria analysis: microbial profiles in the soil using metagenomics, identification of important bacterial indicators of soil health, community level physiological profiling (CLPP) using the Biolog, which measures the utilization of 31 carbon sources to compile a profile, and microbial enzyme activities.

- **Soil borne plant pathogens:** Root diseases are without a doubt one of the most important yield-limiting factors in crop production. These diseases are often caused by a complex of pathogens that seriously harm the health of the root, as diseased roots cannot absorb water and nutrients optimally. Although root health is an important component of soil health and is essential to optimize crop yields, farmers are often unaware of the root status of their crops. The reason for this is that many root diseases do not cause visual symptoms on the soil. The YourSoil™ package includes a root-health assessment test, done under greenhouse conditions. The test is done by planting annual crops in the client's soil to detect any pathogens; as a control the pathogens are eliminated from the same soil, planted to the annual crop and the severity of root rot is rated.

- **Fungi:** Fungi are important within soils, facilitating various processes which contribute to the status of soil health. Most fungi support plant growth; fungal hyphae physically bind soil particles together, creating stable aggregates that help increase water infiltration and soil water-holding capacity. Various fungal species are pathogens of other soil organisms that ensure balance in the soil ecosystem. Fungi are also decomposers of organic matter and play a major role in the circulation of nutrients in the soil. The following tests are included in the YourSoil™ package: Morphological identification of fungal species; measurement of disease load in a sample while providing species specificity for the possible pathogen; molecular identification and phylogenetic analyses of fungal species.

Abiotic: the second set of tests (abiotic) is based on the physical and chemical condition of the soil. Here the focus is on soil as a natural resource: its formation, classification and mapping, in addition to physical, chemical, biological and fertility properties, which are linked to the sustainable use and management of soils. The following tests are available to clients:

- Sample preparation: drying and milling (10 g - 250 g); washing, drying and milling; milling only; digestion (open tube); microwave digestion;
- Farmer Package: top or middle soil analyses: P (Bray No 1 or 2); K, Ca, Mg, Na (Ammonium Acetate); pH (water); Exchangeable acidity; Resistance; Texture (3 Fractions);
- Determination on sample as received: ashing and moisture content;
- Determinations of Dry-Oxidation: total nitrogen and total nitrogen and carbon
- Determinations on digested samples of various element: Ca, Mg, P, K, Na, Fe, Mn, Zn, Cu, Al, B; ICP-OES trace elements, (e.g. As, Pb, Cd, Ni, V, Co, Cr, Mo); Semi-Quantitative scan (ICP-MS) (Li, Be, Ti ,V, Cr, Mn, Co, Ni, Cu, Zn, As, Se, Rb, Sr, Mo, Cd, Sn, Sb, Te, Cs, Ba, La, W, Pt, Hg, Tl, Pb, Bi, U)
- Determinations of nitrate, nitrite, chloride, fluoride, sulphate; pH and electrical conductivity (EC) on extracted samples.

The YourSoil™ package also includes an option to receive recommendations from a specialist. These recommendations may include selection of follow-up crops in a rotation cycle to minimize soil borne plant diseases; integrated management strategies for soil borne plant diseases; effects of conservation agricultural practices on soil borne plant diseases; effects of nursery practices on soil borne plant diseases; and recommendations for fertilizers on most vegetable, grain crops and fruit crops. A combination of these tests enables the client to identify aspects in their soil samples that require attention and thus promote crop production. Not only are the pests and pathogens identified during these routine tests, but also the beneficial organisms, such as free-living nematodes, fungi and bacteria in the soil that promote plant growth. This type of analysis, targeting a combination of organisms, provides the client with a holistic assessment of soil health status.

Contact: Dr Chantelle Girgan at YourSoil@arc.agric.za or call 012 808 8266.

Polymorphomyia basilica Snow (Diptera: Tephritidae), a new biological control agent against *Chromolaena odorata* R.M.King & H.Rob. (Asteraceae) in South Africa

The invasive alien shrub *Chromolaena odorata* R.M.King & H.Rob. (Asteraceae), known locally as triffid weed, has been brought under a reasonable level of biological control in some parts of South Africa. However, in many parts of the country, infestation levels are still unacceptably high. For this reason, a stem-galling tephritid fly, *Polymorphomyia basilica* Snow, was imported into quarantine at the Agricultural Research Council-Plant Health and Protection (ARC-PHP), Cedara, from Jamaica in 2012. Jamaica was targeted because the 'biotype' of chromolaena invading southern Africa originated from this region, and therefore no mismatches between the host-plant and its potential biocontrol agent were expected. A colony of the gall-fly was successfully established in quarantine, and host-specificity trials were undertaken to determine whether it would be safe to release in South Africa. Studies of the gall-fly were also undertaken to elucidate its life history. Much of this work has been described in a previous edition of *Plant Protection News* (Dube 2017).

Ms Thandeka Mahlobo, MSc student (UKZN) collects data from her trial, investigating the impact of the gall-fly on chromolaena

These host-range trials indicated that the gall-fly has a very limited host range in the laboratory (published as Dube *et al*. 2020). Adult progeny were obtained from a few species of closely related weeds of South American origin, but this was of no concern due to their invasive alien status. However, some development of the gall-fly was also recorded on two indigenous species of Asteraceae, which was of concern. *Stomatanthes africanus* is a low-growing forb in high-altitude grasslands in Mpumalanga, and is very closely related to chromolaena. *Felicia amelloides* is a small shrub, but less closely related. Both these species appeared to be inferior hosts for development of the gall-fly (smaller and fewer galls, smaller adult progeny with a short lifespan); consequently, a release application was submitted to the Department of Agriculture, Land Reform and Rural Development (DALRRD) in September 2018. This was sent to three external reviewers (local and international) for comment. While two were happy with the work conducted, the third reviewer felt that further work was merited in order to demonstrate more conclusively that the gall-fly would only form a population on chromolaena.

Stomatanthes africanus growing in situ in grassland near the town of Sabie, Mpumalanga

Further host-range trials were therefore conducted. To better simulate more 'realistic' field conditions, we undertook trials in a large 'walk-in' cage, 4m long, 2m wide and 2m high, into which several plant species were placed.

Mature gall from which adult gall-fly has emerged

Young gall – note initial helical mining by larva

Gall induced by *Polymorphomyia basilica* on pompom weed (*Campuloclinium macrocephalum*) in the laboratory

These included chromolaena, as well as most of the species on which galling had been recorded in previous trials. 'Continuation' trials were undertaken thereafter, to determine the fitness of the F1 generation. No galling occurred on *F. amelloides*, but galls were recorded again on *S. africanus*. However, once again, galls were fewer and smaller than those on chromolaena, and the small adult progeny emerging from galls on *S. africanus* had a very short lifespan and could not establish an F2 generation. For insects, small adults typically have lower fecundity and produce fewer offspring. A 'risk analysis' exercise was undertaken, in which the suitability of each test plant species was calculated relative to chromolaena, using survival to adulthood, gall size, and development rate as parameters. While the South American weeds *Ageratum conyzoides* and *Campuloclinium macrocephalum* (pompom weed) were 66% and 32% as suitable as chromolaena respectively, *S. africanus* was only 4% as suitable. Furthermore, we argued that the habitat in which *S. africanus* grows (much cooler than that for chromolaena, and with frequent winter fires that destroy the above-ground parts of the plant), would mean that *P. basilica* would not be able to sustain a population on the forb.

A revised release application was submitted in March 2021 and DALRRD issued a release permit for *P. basilica* in August. We are currently increasing the size of the culture to a level where we can undertake initial releases, which we plan to do at a few carefully selected sites along the coast of KwaZulu-Natal province. The initial aim is to establish a permanent population of the gall-fly on chromolaena in South Africa, which may take several years. Thereafter we plan to monitor its population increase and its spread from the initial release sites. Given that the gall-fly is diurnally active, and is thus less likely to be affected by the low nighttime temperatures thought to have inhibited the establishment of two species of moths on chromolaena; the longevity of the adults; the short lifecycle; and large number of eggs laid (Dube *et al*. 2020), chances are good that *P. basilica* will establish.

Although it is difficult to predict how effective the gall-fly will be at reducing growth and reproductive rates of chromolaena, and ultimately the population of the weed, another species of stem-galling tephritid fly [*Cecidochares connexa* (Macquart)] has been very effective at controlling the biotype of chromolaena invading other parts of the world. Gall formers are often effective biocontrol agents because their galls act as 'nutrient sinks', reducing the amount of nutrients available to the plant.

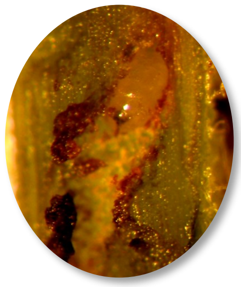
As a preliminary step in determining the efficacy of *P. basilica*, an MSc student, Ms Thandeka Mahlobo, jointly supervised by Dr Caswell Munyai (University of KwaZulu-Natal) and ourselves, and funded by the Centre for Invasion Biology (C·I·B), undertook laboratory-based impact trials. Potted chromolaena plants were exposed to the gall-fly for several months, at two densities (50% and 100% of shoot tips galled, with a control of 0%). Plant height, leaf biomass and flower production were all reduced by galling, although branching increased. For some parameters, both 50% and 100% galling were effective, while for others, only the higher level produced results. Ultimately, effectiveness will depend on factors such as the population level achieved by *P. basilica* under field conditions, and when in the season their numbers are high. The gall-fly may also be more effective in stunting small plants (seedlings) than the larger shrubs, and it remains to be seen whether it prefers shaded or sunny habitats.

References

Dube, N. (2017) Gall forming tephritid fly from the Caribbean possess interesting biological features and seems host specific to triffid. *Plant Protection News* 109: 9-10.

Dube, N., Zachariades, C., Uyi, O., Munyai, T.C. (2020) Life history traits and host suitability of a gall-forming fly, *Polymorphomyia basilica* (Diptera: Tephritidae), for the biological control of *Chromolaena odorata* (Asteraceae) in South Africa. *Arthropod-Plant Interactions* 14: 237–250.

Contact: Dr Costas Zachariades at zachariadesc@arc.agric.za and Dr Nontembeko Dube at duben@ufs.ac.za



Young larva inside a gall

Biocontrol mass-rearing facility open day at PHP Roodeplaat

Mass-rearing facility open day

The Agricultural Research Council-Plant Health and Protection (ARC-PHP) hosted the opening of the Weed Biocontrol Mass-rearing Facility, Roodeplaat West campus, on 14 May 2021. The purpose of the event was to officially open the mass-rearing facility and to provide recognition to the Department of Forestry, Fisheries and the Environment: Natural Resources Management Programmes (DFFE: NRMP) who have generously supported the weeds biocontrol projects at PHP for many years and also provided the funding for the renovation of the mass-rearing facility. The new facility comprises six large greenhouse tunnels that have been refurbished with new polycarbonate roofing, wet-wall padding, fan cooling systems and extensive irrigation infrastructure. The event also provided an opportunity for the Insect Ecology Division to highlight their biological control projects and for PHP to feature the invaluable contribution made to the science of biological control by the National Collections housed at the Biosystematics Division. The event enabled the Weeds Division to showcase their biological control projects on a range of weed species, including *Tithonia, Tecoma, Lantana, Campuloclinium, Anredera, Dolichandra, Prosopis*, Australian Acacias and Hakea species. Staff at the Insect Ecology Division demonstrated their projects on the biological control of diamondback moth, fall armyworm and tomato leaf miner. The beekeeping development unit and the stored-grain pests unit also hosted excellent exhibits of their work. The marvellous exhibition presented by the Biosystematics Division displayed examples of how the collections and capacity within the various units support and develop the science of biological control. The Division's displays highlighted the importance of the National Collections, which represent an archive of the rich biodiversity in South Africa, that biocontrol endeavours rely on for IPM and invasive weed control.

Approximately 50 guests attended the event, from various government departments, local universities, commodity organisations, agro-chemical and biotech companies, as well as an excellent complement of journalists from the agricultural media sector. Our Senior Manager, Dr Ansa van Vuuren, opened and hosted the event, while the official opening of the facilities was undertaken by Mr Nceba Ngcobo, Director Operations: National Programmes at DFFE: NRMP. The Open Day was a great success and visiting delegates expressed their appreciation for such interesting displays and discussions on the potential of biological control. The many months of hard work in the planning and organising of the event certainly paid off. We now hope to put the mass-rearing facility to good use and build up our biocontrol agent cultures for release in the field later in the spring.

Contact: Dr Roger Price at pricer@arc.agric.za

The big-headed Mexican sunflower Tithonia tubaeformis (Asteraceae) is rapidly expanding its distribution in South Africa

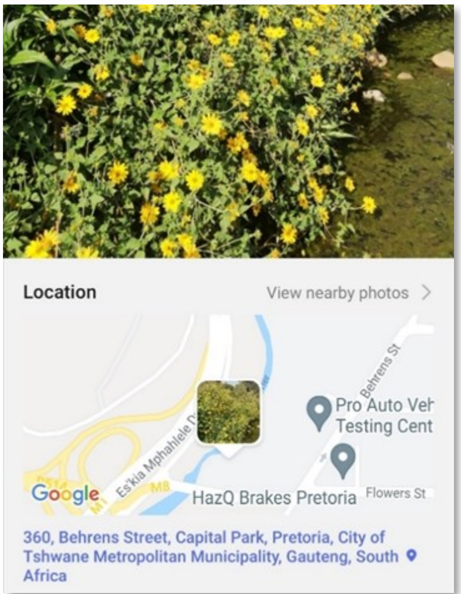
The big-headed Mexican sunflower *Tithonia tubaeformis,* is one of the three *Tithonia* species that are invasive in South Africa. It is distinguished from its relatives by having simple leaves and bright yellow flowers (Fig. 1). *Tithonia tubaeformis* was initially reported invading arable land and road sides at Lomahasha in the North of Eswatini in 2011, but was later discovered at Mbuzini village (Mpumalanga province) along the border of South Africa with Eswatini and Mozambique during 2015/2016. The weed's spread has been monitored annually in the country since 2017, in preparation for an eventual biological control programme against it. The big-headed Mexican sunflower was previously assumed to be restricted to the villages in and around Nkomazi municipality (in Mpumalanga), until new populations of the invader were recently discovered along the Apies River, Pretoria, in Gauteng province in May 2021 (Fig. 2).

Tithonia tubaeformis has the propensity to produce large amounts of seed, easily spread by birds, floods and wind over long distances, making it extremely difficult to eradicate. This weed can grow to over four meters tall creating dense monoculture stands that can easily displace crops and native plant species. In its native Mexico, *T. tubaeformis* is the most widely distributed of the three *Tithonia* species, displaying invasive tendencies in arable land (particularly maize fields), road sides and, urban and rural open spaces. In its early stages of invasion, *T. tubaeformis* poses a great risk, with serious consequences for agroecosystems and biodiversity not only to South Africa but also to other sub-Saharan African countries such as Eswatini, Mozambique, Zimbabwe and Zambia. The big-headed Mexican sunflower is also known to hybridize with its closest relative, the orange-red sunflower *T. rotundifolia*, that is already invasive in South Africa. The resultant hybrid may possibly display a much wider invasion distribution and also be less susceptible to some natural enemies of both *T. tubaeformis* and *T. rotundifolia*, thus creating a conundrum that could be difficult to resolve through biological control. The fact that *T. rotundifolia* and *T. tubaeformis* co-occur in Mpumalanga and Gauteng provinces, presents a greater risk of hybridization of these two plant species. Since *T. tubaeformis* frequently invades maize fields in South Africa and Eswatini, it poses a direct threat to food security in the region.

Although *T. tubaeformis* is currently not regulated in South Africa, its risk assessment is underway. To curb its invasiveness and negative impact on biodiversity, the Agricultural Research Council – Plant Health and Protection (ARC-PHP) has initiated a biological control programme with funding from the Department of Forestry, Fisheries and Environment of South Africa. Should you come across the big-headed Mexican sunflower, please take a picture and send it with geographic locality information to any of the three researchers below.

Contact: Khethani Mawela at Mawelak@arc.agric.za, Dr David Simelane at Simelaned@arc.agric.za and Dr Naweji Katembo at Katembon@arc.agric.za

Top & bottom: Big-headed Mexican sunflower, *Tithonia tubaeformis* (Asteraceae)



Big-headed Mexican sunflower infestation along the Apies River, Pretoria in Gauteng province.

Nematodes associated with crop seeds

Before harvest germination of groundnut seeds due to *Ditylenchus africanus* infestation. (*Photo: Dr S Steenkamp (ARC-GC)*).

Diverse groups of plant-parasitic nematodes are associated with seed, including seed tissue and other propagating plant material. Many plant -parasitic nematodes also survive in, and disseminate through edible roots, corms, rhizospheres, tubers and seedpods. Some nematodes exist in a desiccated state in true seeds and can survive for long periods, in or on infected seed. Examples include species of the genera *Anguina, Aphelenchoides, Robustodorus* and *Ditylenchus*. The infected seed and other propagating material act as a source of inoculum for disease development that may cause both qualitative and quantitative losses to a wide range of agricultural crops. A number of nematodes have been reported from cereal crops seeds, including the seed gall nematode (*Anguina agrostis*). Annual ryegrass toxicity (ARGT) linked to *A. agrostis* is an ongoing concern for livestock farmers, and importers and exporters of hay. The seed gall nematode, *Anguina tritici*, has been reported from oat, rye and wheat, where the seed gall nematode, *Subanguina wevelli*, has been reported from weeping love grass, while the rice leaf nematode, *Aphelenchoides besseyi* and the rice stem nematode *Ditylenchus angustus*, are known from rice.

The seeds of leguminous plants are also susceptible to a number of nematodes such as the stem and bulb nematode *Ditylenchus dipsaci* (*s.s.*) on clover, faba beans, pea and Lucerne; a distinctive symptom of infested Lucerne is white flagging of the leaves. Clover seeds can also be infected with the "giant race" of the stem and bulb nematode, *Ditylenchus gigas*. Due to groundnut pods and seeds initially forming below ground, they are susceptible to a number of nematodes including the peanut pod nematode, *Ditylenchus africanus*; the potato rot nematode *Ditylenchus destructor*; the testa nematode *Robustodorus africanus* (=*Aphelenchoides arachidis*); a stunt nematode *Tylenchorhynchus brevilineatus*; root-knot nematodes, *Meloidogyne* ssp., and lesion nematodes, *Pratylenchus* spp. The stem and bulb nematode, *Ditylenchus dipsaci* (*s.s.*), can infect beetroot, leeks, onion and shallot seeds and the potato rot nematode, *Ditylenchus destructor*, is known to infect carrot seeds. Seed potatoes can be infected by the potato rot nematode, root-knot nematodes and lesion nematodes. *Anguina tritici, Ditylenchus. gigas* and *D. angustus* have not been reported from South Africa.

Nematodes survive within these hosts between growing seasons, which facilitates the introduction of nematodes to new areas, potentially causing cross-border transmission through local markets and international trade. The presence of these pathogens in a sample may lead to phytosanitary regulations and restrictions, and the rejection of shipments.

Staff at the Nematology Unit at Biosystematics, ARC-Plant Health and Protection have the capability to provide identifications of all plant-parasitic, and various other groups of nematodes. Internationally approved techniques, such as the modified Baermann tray and sieving-centrifugation-flotation techniques are used to extract nematodes from seed and other plant propagation material. The aeration method, which prevents plant material from rotting and subsequent increase in bacterial and fungal feeding nematodes, is also used to extract economically important root-knot and cyst nematodes from hard rhizomes, corms and taproots.

The turnaround time for sample analyses depends on the number of submitted samples, and the number of tests required. An analysis takes approximately 10 working days, but the client will be informed should more time be required. The laboratory at the Nematology Unit is registered with the Department of Agriculture, Land Reform and Rural Development as a plant-testing laboratory, with registration number: 72030003.

Contact: Dr Mariette Marais at MaraisM@arc.agric.za or Dr Antoinette Swart at SwartA@arc.agric.za

Post-fire seedling emergence and survival of Acacia longifolia and A. pycnantha under long-term biological control in South Africa

Acacia longifolia (Andrews) Willdenow and *A. pycnantha* (Bentham) are listed under category 1B of the Alien and Invasive Species Regulations of the National Environmental Management: Biodiversity Act, 2014 (NEMBA). These species are therefore recognised by law as invasive, requiring compulsory control, as part of an invasive species management programme. They must be removed and destroyed and are deemed to have such a high invasive potential that infestations qualify to be placed under a government-sponsored invasive species management programme. In addition, no permits may be issued for their propagation.

These Australian acacias have been subject to various management strategies for decades, but biological control has emerged as a long-term and the most sustainable management option. The biocontrol programme against these alien invaders, which is based on the use of insect agents that reduce their reproductive capacity, was instituted during the 1980s, and the agents have since established across the geographic range of the infestations. The biocontrol agents utilized for this purpose consist of univoltine and parthenogenetic bud-galling pteromalid wasps, namely *Trichilogaster acaciaelongifoliae* (Froggatt) and *T. signiventris* (Girault), which were released on *A. longifolia* and *A. pycnantha* respectively. Two seed-feeding weevils, *Melanterius ventralis* (Lea) and *M. maculatus* (Lea), were also subsequently released on *A. longifolia* and *A. pycnantha* respectively, to supplement the two pioneering bud-galling wasps.

As part of a long-term post-release evaluation study on the efficacy of biological control, the post-fire seedling recruitment and survival dynamics of these invader species were investigated. These alien acacia species have invaded and established in fire-prone fynbos ecosystems and therefore have traits to cope with recurrent fires such as fire-stimulated recruitment. However, the fire events, which are natural ecosystem processes, have become increasingly frequent, because of human activity. As a general survival 'strategy', in addition to being prolific invaders, these acacia species produce and accumulate large quantities of seed in the soil, which can remain dormant and viable for many years. The bulk of these seeds accumulate in the top 10 cm of soil. Fire is therefore an important environmental factor that influences the success of these acacia species. The heat shock produced by wildfires in these Mediterranean fire-prone ecosystems, such as the fynbos, is an important external stimulus that breaks seed dormancy and triggers germination.

The occurrence of these fires has presented an opportunity to test the hypothesis that in the presence of long-term biocontrol, at least two fire cycles are necessary before the alien plant population starts to decline. Our results showed that this null hypothesis is true. It was observed that the long-term presence of biocontrol prevented parent alien trees from producing seed that would otherwise accumulate unhindered in the soil seed bank. There is a significant reduction in seed production, as evidenced by the post-biocontrol seed rain compared with pre-biocontrol records, especially for *A. longifolia* (7 646 seeds/m2 pre-biocontrol vs. 235 seeds/m2. post-biocontrol). In the case of *A. pycnantha*, historical records are lacking, but current seed rain averages 1 655 seeds/m2 (inter-site and season range 31 – 8 161 seeds/m2). Observations showed that the bulk of the seeds that escaped biocontrol were deposited on the soil surface or at a shallow depth beneath, and were burnt by the wild fires. This significantly depleted the number of seeds available for recruitment in both species, as a direct consequence of fire-induced seed mortality and subsequent germination of the few surviving seeds. This showed the importance of fire as a potentially useful tool for management or eradication of these woody alien species, where biocontrol is present over a long period.

Over the past eight years since 2014, a series of veld fires (both natural and human-induced) have swept through some of the study sites, affecting both Acacia weed species in the Western Cape (*A. longifolia* sites at Banhoek and A. *pycnantha* sites at Lievland and Houwhoek). At each site, seedlings emerging in 1 m^2 quadrants, 5 m apart along a 50 m transect were tagged and counted monthly. Data on the extent of the soil seed banks before and after fire events (Table 1), rate of biomass accumulation until reproductive maturity, as well as how quickly biocontrol is restored post fire events, were collected. The impact of fires on the extent of soil seed bank showed that there was a significant number of seeds lost to fire-induced mortality, with the remaining surviving seeds germinating subsequent to these fires. At Banhoek, all seedlings of *A. longifolia* in the riparian habitat that were originally tagged in each of all 10 quadrants along the transect, were reduced to zero after 15 months following germination.

The quadrants have since become overgrown with grass species and ferns, and the acacia seedlings that emerged could not compete with these recolonising species. To date, no new seedlings have emerged in those permanent quadrants, which have been continually monitored since the fire in 2016. In the mountain habitat, seedling survival was approximately 35% by the time the plants had reached reproductive maturity. The mortality in both scenarios is attributable to excessive heat due to drought over the years since the fire, in addition to fierce competition from other secondary weeds that emerged soon after the fire. The seedling survival and recruitment scenario remained unchanged in the Banhoek riparian habitat, following the good rains after the drought of 2018, where seedling count remained at zero with no new seedlings recruited. However, on the mountain site, seedling mortality slowed down, with surviving plants having attained reproductive maturity within two years from germination. Within the first year of development of seedlings to saplings, biocontrol was already evident by the presence of vegetative and reproductive galls, in the first year of flowering.

Tagging of Acacia pycnantha seedlings in 1m2 quadrants along a permanent transect at Kortes Hoven (Houwhoek), Western Cape province.

In the case of *A. pycnantha* at Lieveland, the seedling survival was approximately 50% by time of reproductive maturity. Despite a high number of seedlings germinating after the fire, drought, overcrowding and competitive stress thinned out of the infestation; 30% of the trees bore seed for the first time in the 2018/2019 season, indicating a minimum time to reproductive maturity of three years after seed germination in September 2015. This increased to 100% in the subsequent seasons. As early as the first year after germination, vegetative galls were present on the host infestations, while the appearance of reproductive galls coincided with the same year that the species attained reproductive maturity.

Tagged seedlings for monthly monitoring of plant mortality, growth and presence of biocontrol on the post fire infestation.

Table 1 gives a summary of pre- and post-fire seedbank densities for both *A. longifolia* and *A. pycnantha* thickets subjected to wild fires. Overall, the findings from this study show us that while fire plays an important role in the success of these fire-adapted invasive species, it can also play a pivotal role in their management. From a management perspective, the reduction in reproductive capacity due to the long-term presence of biocontrol implies that fewer viable seeds are being added to the seed bank. Intense fires that sweep through these thickets destroy the parent plants as well as a significant number of seeds stored on the soil surface or in the seed bank. This implies that the seedlings or saplings that emerge from the few seeds that germinate can easily be managed by manual clearing or herbicidal control, before they attain reproductive maturity. After a series of subsequent fires and judicious removal of newly recruited plants, coupled with the aggressive recolonization by other species, there is a good likelihood that infestations will eventually be eradicated from these previously invaded habitats, paving the way for restoration of these ecosystems.

Collection of soil cores for seed bank density analysis along permanent 50m transects, following fire on *Acacia longifolia* on a mountain habitat at Banhoek – Stellenbosch, Western Cape Province.

Table. 1. Soil seed bank estimates (average seeds/m2 ± S.E) for A. longifolia and A. pycnantha infested sites burnt in the Western and Eastern Cape provinces of South Africa

Habitat	Western Cape				Eastern Cape	
	Banhoek (*A. longifolia*)		Houwhoek (*A. pycnantha*)		Grahamstown (*A. longifolia*)	
	Before Fire	After Fire	Before fire	After Fire	Before fire	After Fire
Riparian	530,2 ± 116,05	178,54 ± 76,04	-	-	908,92 ± 238,9	18,94 ± 13,03
Mount	25,23 ± 15,01	6,31 ± 6,3	-	-	239,85 ± 59,49	31,56 ± 15,94
Flat/ gentle slope	-	-	3913,39 ± 510,8	145,17 ± 45,11	-	-

Contact: Dr Pride Mudavanhu at MudavanhuP@arc.agric.za

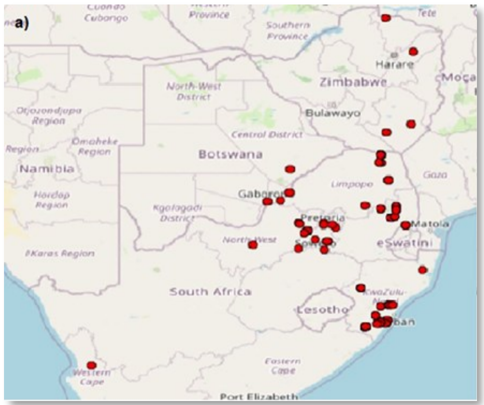
FAW strains investigated

Recent molecular studies investigating phylogenetic relatedness of specimens of FAW (*Spodoptera frugiperda* (JE Smith), (Lepidoptera, Noctuidae) from West, Central and South Africa, indicate that there are two different strains of FAW present on the continent. Although these studies provide insight into the genetic diversity within the species, these results were based on a very small sample size. Consequently, more data are needed to confirm the presence of the two strains in southern Africa and the genetic composition of the current prevailing population.

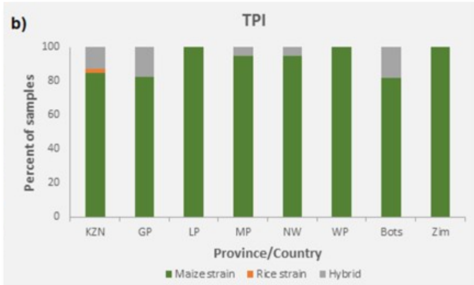
In this research and development project for the Department of Land Reform and Rural Development (DALRRD), to study the genetic composition of FAW in South Africa, 172 COI and 177 TPI marker sequences were produced from FAW larvae and moths from KwaZulu-Natal, Mpumalanga, Gauteng, Limpopo, North West, and Western Cape provinces, as well as Zimbabwe and Botswana.

Phylogenetic analyses of the TPI marker revealed that the maize strain was found in 158 of these samples, 18 samples were hybrids and only one sample contained the rice strain. The study revealed that the rice strain is not as prevalent in South Africa as previously thought. The maize and rice strains are known to have specific hosts. Further work on the biology of hybrid strains, including host range is required.

Contact: Dr Tendekai Mahlanza at MahlanzaT@arc.agric.za



Overview of FAW collection localities



Percentage off rice, maize and hybrid strain presence in FAW samples detected in each province/country, using the TPI gene.

Seed treatment to control soilborne diseases in field crops

Proper establishment of seedlings is essential in improving yield and is an important component of sustainable production. Seed treatment is one of the most effective ways of significantly improving the establishment of seedlings. Many of the pathogens affecting seedlings of field crops have a broad host range; these pathogens cannot be controlled with crop rotation. Since crop rotation is such an important part of conservation agriculture, crops such as maize, sunflower, soybean and dry bean that are susceptible to some of the same pathogens, are often rotated in the same field. In order to protect seedlings against these pathogens with a broad host range, effective seed treatment should be included in an integrated management strategy where it can play a significant role against soilborne diseases. The Soilborne Plant Diseases Unit conducted numerous seed treatment evaluations against the most important soilborne pathogens responsible for poor establishment of field crops. These glasshouse studies demonstrated the ability of seed treatments, which included a cocktail of active ingredients, to effectively target pathogens that are part of the complex of soilborne pathogens associated with seedlings of these crops, and significantly improve plant survival, growth and reduce root rot severity.

Seed treatment evaluation against soilborne diseases

Contact: Dr Sandra Lamprecht at LamprectS@arc.agric.za

ARC-PHP shares research findings on Fall armyworm, [Spodoptera frugiperda (JE Smith), (Lepidoptera, Noctuidae) with other countries in the SADC region

Staff at the Agricultural Research Council-Plant Health and Protection (ARC-PHP) recently presented their work on Fall armyworm (FAW) to various stakeholders and researchers at an international conference hosted by the Food and Agricultural Organization of the United Nations (FAO). Entitled, 'Developing smallholder-oriented IPM strategies for Fall armyworm [Spodoptera frugiperda (JE Smith)]', the conference was hosted online on 24-26 August 2021 and attended by researchers and agricultural extension officials from countries impacted by FAW from around the world.

Funding received from the Department of Agriculture, Land Reform and Rural Development (DALRRD) and the FAO has enabled the ARC-PHP to undertake various studies on aspects of the ecology of FAW, and the presentation of training courses on the identification and management of FAW for smallholder farmers through a farmer field school. Research findings were presented using PowerPoint and poster presentations.

The following presentations where shared with conference delegates:

- Van der Walt, E. Lundall-Magnuson, E., Price, R., Van Niekerk, P. Sgudhla, E. & Mooka, J. Optimal pesticide application efficiency training for FAW control and decreased human and environmental contamination.
- Lyle, R, Van Niekerk, P, Swart, M, Mooka, J & Sgudhla, E. Indigenous knowledge systems – The application of ash to manage FAW on maize. (Poster)
- Lyle, R. & Lundall-Magnuson, E. FAO FAW Farmer Field School: A South African perspective.
- Mailula, T. Overwintering sites of fall armyworm in South Africa.
- Lundall- Magnuson, E. & Price, R. FAW causes food insecurity in smallholder communities in Limpopo, South Africa.
- Lundall- Magnuson, E., Khorommbi, G., Mathibe, S. & Price, R. FAW eats more than its share. (Poster)

Contact: Robin Lyle at LyleR@arc.agric.za

The role of certain mortality factors in the underperformance of Aristaea thalassias (Meyrick) as a biological control agent of Australian myrtle Leptospermum laevigatum F. Muel.

South African ecosystems are prone to alien plant invasions, threatening natural resources and biodiversity, human health and the economy. Of all the provinces, the Western Cape is considered the most severely affected, with several invasive species occurring in the species-rich Fynbos biome. One of the most problematic threats to coastal dunes is *Leptospermum laevigatum*, commonly known as Australian myrtle. *Leptospermum laevigatum* has become invasive in South Africa since its introduction in the 1800s, competing with the native fynbos across the coastal regions of the Western Cape province and into the Eastern Cape. The species is also often considered a secondary invader in sites where another invasive tree, *Acacia saligna*, is removed from the landscape. Due to its aggressive spread, it has become of significant concern and is considered a highly invasive species, necessitating a concerted management plan in South Africa. Australian myrtle is particularly difficult to control, and various methods have been employed with limited degrees of success. For the most

Aristaea thalassias adult on a *Leptospermum laevigatum* leaf. (*Photo: Mr Tony Gordon).*

Ms Thembelihle Mlokoti (*Photo: Dr Candice Lyons)*

The pre-pupal life stage of *A. thalassias* (*Photo: Dr Candice Lyons*)

The mining (larval) stage of *Aristaea thalassias* (*Photo: Dr Candice Lyons*)

The pre-pupal stage of *A. thalassias* damage to *L. laevigatum* leaves. (*Photo: Dr Candice Lyons*)

part, manual clearing and chemical control are considered too expensive, requiring several follow up treatments and are thus, largely unsustainable. As a result, biological control remains the primary goal for providing a long-term, reliable and affordable method to supress the growth of *L. laevigatum*.

Biological control of *L. laevigatum*, using the gracillariid moth *Aristaea thalassias* has been implemented since the 1980s with limited success. In order to understand the underperformance of this leaf-mining moth, imported from Australia and released in 1986, the biology of the immature stages, larval mortality due to parasitoids, predation and overcrowding, and seasonal influences on development and survival, were investigated. Findings showed that *A. thalassias* is always present and has the potential to complete several overlapping generations throughout the year. The overall mortality of larvae, prepupae and pupae measured over a 20-month period indicated seasonal variability, with highest mortality in autumn and winter. Mortality attributed to parasitism (maximum 25%), predation and overcrowding (maximum 15%) was low and thus unlikely to account for the low efficacy of the agent. Despite this, mortality through other unknown factors was at times found to be high, especially for first instar larvae in mined leaves (maximum 58% in one year). The results also showed that the peak oviposition period was not aligned with the peak in production of new leaves, suggesting a possible phenological mismatch between *L. laevigatum* and its biological control agent *A. thalassias*, which could curb population expansion. Whilst mortality resulting from unknown factors (i.e., not investigated in the scope of this study) might be significantly impacting the performance of *A. thalassias*, the specific factors investigated during this study could not explain why the moth is not more prolific.

Determining why any particular biological control agent is ineffective is often not a straightforward process because it may be the result of a complex of interacting factors, which are not easily discernible. The findings of this study show that this is probably true for *A. thalassias* in South Africa and much more work is needed to resolve the reasons for its underperformance as a biological control agent.

Contact: Thembelihle Mlokoti at MlokotiT@arc.agric.za

Continuation of the standards of generally recognised accounting practice (GRAP) activities in the National Collection of Fungi

Type of fungal collections

The National Collection of Fungi (NCF) consists of two collections: the living fungal culture collection (Acronym = PPRI) and the dry specimen collection (Acronym = PREM). The PPRI collection, which currently comprises 29 000 strains, was established in 1981 by Dr. Cecile Roux. The PREM collection was established by Pole Evans in 1905 and currently comprises of 62 000 specimens. The fungal specimens represented in the collections were isolated from plants, insects, soil, environmental samples and other sources South Africa and neighbouring countries.

The collections mandate

The mandate of the National Collection of Fungi (NCF) is to preserve, maintain, and document South African and African fungal diversity. Fungal cultures preserved in the NCF are used in fungal systematics, biodiversity research, quarantine and pathogen diagnostics. Several methods of preservation are used in the collections to safeguard these cultures. PREM specimens are mounted and packed in envelopes and boxes whereas the PPRI cultures are preserved as agar slants covered with paraffin oil, agar punches in sterile water, freeze-dried material and mycelium suspensions stored at ultra-low temperatures.

GRAP implementation in South Africa and activities in the NCF

The Department of Arts and Culture implemented an accounting practice for South Africa's heritage assets known as GRAP 103 and for its biological assets, known as GRAP 27. The practice became effective in 2014/15. In September 2019, the Agricultural Research council (ARC) adopted these GRAP activities, which requires its different institutes to adhere to these requirements. The NCF team started implementing GRAP activities towards the end of September 2019, which is an ongoing process. In April 2021, the NCF team was joined by four colleagues from other units within ARC-PHP, namely Mr Piet van Niekerk, Ms Magda Swart, Mr Joel Mooka and Mr Ezekiel Sgundla, who have been assisting with GRAP activities and other duties in the Mycology unit.

GRAP activities

PPRI-GRAP 27 activities include: verification of available cultures in bottles and cryo-vials, labelling of bottles, cabinets, shelves and steel racks, re-packing cultures into the cabinets, shelves and freezers, and capturing of culture data on the PPRI database. Since April 2021, about 25 865 freeze-dried bottles, 10 125 water and oil bottles and 800 ultra-low freezing tubes have been verified and repacked into the collection. In addition, 1 310 strains have been freeze-dried, 1620 strains preserved in water and oil and 400 strains preserved in glycerol suspensions and all data were updated in the PPRI database.

The GRAP 103 activities included spot-checks to confirm the actual location of 200 specimens within the fungorium cabinets, verification of database entries with accession registers for 20,000 entries and electronic recording of all previously hand-recorded material of 1500 specimens in the van der Byl and Rump collection. Two hundred newly generated labels were placed with material. More than 18,000 specimen fields in the PREM database we updated with cabinet localities. Adding and correcting specimen authorities for 1000 entries.

PPRI preserved cultures and PREM specimens in the NCF

GRAP activities are time consuming, but are beneficial to the functioning of the unit. These activities allow re-packing of cultures and specimens in a consistent manner, which facilitates access to material, indicates items that are no longer available and encourages us to prioritize maintenance activities and ensure effective planning for the future. All these activities assist in the determination of knowledge gaps in taxonomic groups, habitats or host plants, and potentially highlighting research priorities in South Africa.

Contact: Grace Kwinda at KwindaG@arc.agric.za

The establishment of two weed biocontrol agents

Some of the biocontrol agents released against alien invasive weeds in South Africa have shown excellent performance against target weeds in the laboratory. However, once released into the field, they either fail to establish, or do so in very low numbers and do not achieve the desired impact. Despite years of quarantine research and eventual official approval for release of a biocontrol agent, the 'proof of the pudding' is the agent's successful establishment in the field and subsequent ability to thrive under South African conditions.

Field establishment is the most crucial step and we are therefore very pleased to announce the successful establishment of the introduced tortoise beetle, *Physonota maculiventris* Boheman (Chrysomelidae) as a biocontrol agent of invasive Mexican sunflower species (*Tithonia* spp.) at certain sites. High populations of the beetle have been recorded in KwaZulu-Natal, Mpumalanga and Limpopo provinces, following monitoring during winter and into the spring of 2021.

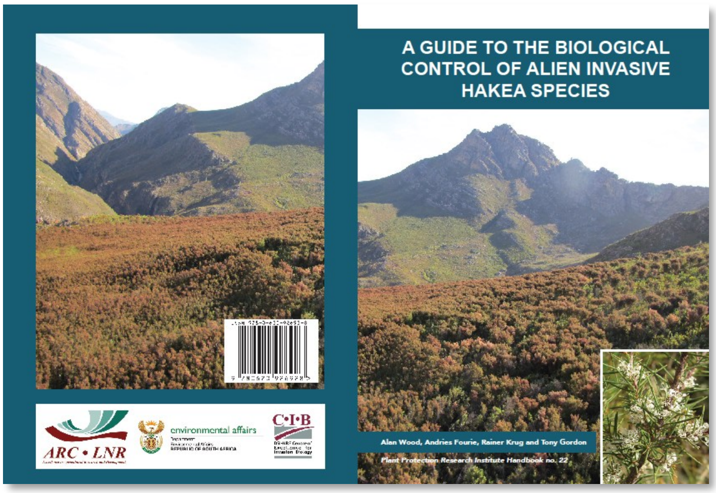
In addition, monitoring of the stem-boring cerambycid beetle, *Aphanasium australe* (Boisduval) against invasive *Hakea* spp. in the Western Cape shows that the beetle is now well established and thriving on trees inoculated with eggs from adults imported from Australia, and released in 2019. The beetle is also being recovered from different sites, which indicates that it is starting to disperse naturally on its own.

Contact: Dr Roger Price at PriceR@arc.agric.za

New book on control of alien invasive hakea

Authored by Alan Wood, Andries Fourie, Rainer Krug and Tony Gordon, this guide has been produced as part of the Plant Protection Research Institute Handbook series. The manual collates and summarises all options for the biocontrol of invasive hakea that have been developed over the past 30 years and provides practical guidelines on management strategies for landowners, conservationists and environmental officers.

Contact: Dr Alan Wood at WoodA@arc.agric.za



Erratum

Swart, A.2021. A quarantine nematode found on "String of pearls". *Plant Protection News* 117: 11-12.

On page 12: The caption of the image should read as follows: Scanning electronic microscope image of *Aphelenchoides ritzemabosi*.

SCIENTIFIC PUBLICATIONS

Bopape, F.L., Gwata, E.T., **Hassen, A.I.** & Zhou, M.M. 2021. Symbiotic efficiency of pigeonpea (Cajanus cajan (L.) Huth) with different sources of nitrogen. Plant *Genetic Resources: Characterization and Utilization* 19(4), 312-315. doi:10.1017/S1479262121000356

Crous, P. W. and Lombard, L. and Sandoval-Denis, M. and Seifert, K. A. and Schroers, H.-J., Chaverri, P., Gene, J., Guarro, J., Hirooka, Y., Bensch, K., Kema, G. H. J., **Lamprecht, S. C**., Cai, L., Rossman, A. Y., Stadler, M., Summerbell, R. C., Taylor, J. W., Ploch, S., Visagie, C. M., Yilmaz, N., Frisvad, J. C., Abdel-Azeem, A. M., Abdollahzadeh, J. and Abdolrasouli, A., Akulov, A., Alberts, J. F., Araujo, J. P. M., Ariyawansa, H. A., Bakhshi, M., Bendiksby, M., Ben Hadj Amor, A., Bezerra, J. D. P., Boekhout, T., Camara, M. P. S., Carbia, M., Cardinali, G., Castaneda-Ruiz, R. F., Celis, A., Chaturvedi, V.,Collemare, J., Croll, D., Damm, U., Decock, C. A., de Vries, R. P., Ezekiel, C. N., Fan, X. L., Fernandez, N. B., Gaya, E., Gonzalez, C. D., Gramaje, D.,Groenewald, J. Z., Grube, M., Guevara-Suarez, M., Gupta, V.K., Guarnaccia, V., Haddaji, A., Hagen, F., Haelewaters, D., Hansen, K., Hashimoto, A., Hernandez-Restrepo, M., Houbraken, J., Hubka, V., Hyde, K. D., Iturriaga, T., Jeewon, R., Johnston, P. R., Jurjevic, Z., Karalti, I., Korsten, L., Kuramae, E. E., Kusan, I., Labuda, R., Lawrence, D. P., Lee, H.B., Lechat, C., Li, H. Y., Litovka, Y. A., Maharachchikumbura, S. S. N., Marin-Felix, Y., Matio Kemkuignou, B., Matocec, N., McTaggart, A. R., Mlcoch, P., Mugnai, L., Nakashima, C., Nilsson, R. H., Noumeur, S. R., Pavlov, I. N., Peralta, M. P., Phillips, A. J. L., Pitt, J. I., Polizzi, G., Quaedvlieg, W., Rajeshkumar, K. C., Restrepo, S., Rhaiem, A., Robert, J., Robert, V., Rodrigues, A. M., Salgado-Salazar, C., Samson, R. A., Santos, A. C. S., Shivas, R. G., Souza-Motta, C. M., Sun, G. Y., Swart, W. J., Szoke, S., Tan, Y. P., Taylor, J. E., Taylor, P. W. J., Tiago, P. V., Vaczy, K. Z., van de Wiele, N., van der Merwe, N. A., Verkley, G. J. M., Vieira, W. A. S., Vizzini, A., Weir, B. S., Wijayawardene, N. N., Xia, J. W., Yanez-Morales, M. J., Yurkov, A., Zamora, J. C., Zare, R., Zhang, C. L., Thines, M. 2021. Fusarium: more than a node or a foot-shaped basal cell. Studies *in Mycology*, 98:100116. pp. 1-184. ISSN 0166-0616

D'Alessandro. P., Iannella, M., **Grobbelaar. E.** & Biondi, M. 2021. Taxonomic revision of the *Calotheca parvula* species group from southern Africa, with descriptions of three new species (Coleoptera, Chrysomelidae). *African Invertebrates* 62(1)315-337. https://doi.org/10.3897/AfrInvertebr.62.62426

Gatabazi, A.; Vorster, B.J.; Mvondo-She, M.A.; Mangwende, E.; Mangani, R.; **Hassen, A.I.** 2021. Efficacy of Peat and Liquid Inoculant Formulations of *Bradyrhizobium japonicum* Strain WB74 on Growth, Yield and Nitrogen Concentration of Soybean (*Glycine max* L.). *Nitrogen* 2, 332-346. https://doi.org/10.3390/nitrogen2030023

Hassen, A.I., Khambani, L.S., Swanevelder, Z.H., Mtsweni, N.P., Bopape, F.L., **van Vuuren, A.,** van **der Linde, E.J.** and Morey, L. 2021. Elucidating key plant growth–promoting (PGPR) traits in *Burkholderia* sp. Nafp2/4-1b (= SARCC-3049) using gnotobiotic assays and whole-genome-sequence analysis. *Letters in Applied Microbiology* 73(5), pp.658-671.https://doi:10.1111/lam.13556

Javal, M., Terblanche, J.S., Conlong, D.E., Delahaye, N., **Grobbelaar, E.**, Benoit, L., Lopez-Vaamonde, C. & Haran, J.M. 2021. DNA barcoding for bio-surveillance of emerging pests and species identification in Afrotropical Prioninae (Coleoptera, Cerambycidae). *Biodiversity Data Journal* 9. https://doi.org/10.3897/BDJ.9.e64499

N'Guessan, K.K., Mamadou, D., Koffi, E.K. & **Lyle, R.** 2021. State of Knowledge on Spiders (Arachnids, Araneae) in Côte d'Ivoire (West Africa). *IAR Journal of Agriculture Research and Life Sciences* 2(2): 9-16.

Oldroyd, B.P., Yagound, B., **Allsopp, M.H.**, Holmes, M.J., Buchmann, G., Zayed, A. and Beekman, M. 2021. Adaptive, caste-specific changes to recombination rates in a thelytokous honeybee population. *Proceedings of the Royal Society B*, 288(1952), p.20210729. https://doi.org/10.1098/rspb.2021.0729

Shubane, A.R., Swart, A., Knoetze, R., Tiedt, L.R. and Fourie, H. 2021. Characterization of Heterodera carotae from the Tarlton area, South Africa, and determination of its life cycle under field conditions: a baseline study. *Journal of Plant Diseases and Protection,* pp.1-19.https://doi.org/10.1007/s41348-021-00503-5

Wood, A.R. & Breeyen, A.D. 2021. Incidence of gummosis disease in silky hakea under natural conditions in South Africa.*South African Journal of Plant and Soil* 38(2), pp.126-133. DOI:10.1080/02571862.2021.1879286