Tree Protection News

tpcp

CTHB

Newsletter of the Tree Protection Co-operative Programme and the DST/NRF Centre of Excellence in Tree Health Biotechnology.

VOL: 15 May 2008

MESSAGE FROM THE DIRECTOR

This Saturday morning, as I write this introductory note for the first issue of Tree Protection News, a group of TPCP staff and students are working in the lab extracting nematodes from cultures. Most everyone reading this newsletter will know that the TPCP team has the task of producing sufficient numbers of the parasitic Nematode *Deladenus siricidicola* to inoculate up to 30 000 trees for *Sirex* control this year. While the group has had some years of experience in producing this biological control agent, none of us could possibly have anticipated the enormity of the operation. For those that are interested, more details are provided elsewhere in this document. Suffice to say that this has been a huge effort on the part of many, but also a very successful one. Nematodes have been produced in more than sufficient numbers, despite load shedding and other imponderables. While there have been tensions and some other key research support efforts have needed to be set aside, I think that it is fair to say that there has been a great team spirit. One might even consider this to be an extreme version of a team building exercise, although I was warned this week that I should not take this idea too far!

It is not possible to avoid mention of the nematode rearing and extraction operation up front in this newsletter, because it has penetrated so many aspects of our lives. Yet it is also important to remember that rearing nematodes is only one component of a much larger operation, which includes staff of forestry companies, contractors and others. The nematodes must be inoculated into trees requiring a huge effort in itself. And that step can only begin after trees (all 30 000 of them) have been pre-selected for inoculation. The next step after inoculation (other than hoping for superb results) is to trap emerging *Sirex* wasps later in the year and to evaluate the success of the process. In all,

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Chrizelle Beukes and Francois van der Walt

controlling *Sirex* is a huge priority for a team that goes far beyond the walls of FABI. We approach this with enthusiasm and hope that new nematode strains, additional biological control agents, extended monitoring and refined forestry operations will allow us to reduce the impact of the wasp on our plantations.

In 1997, the idea of moving the TPCP, then in it's eighth year of existence, to the University of Pretoria was raised. This was in effect the start of FABI, now ten years old, and already having a very significant national and international footprint. It is hard to imagine that so much has happened during this decade that has passed incredibly rapidly. This, our tenth anniversary year will be an exciting one for FABI and the TPCP. Conveniently, the FABI tenth Anniversary occurs concurrently with the 100th Anniversary of the University of Pretoria. The upshot is that many of the University celebrations provide an opportunity to celebrate FABI's smaller, yet very significant position in research and education. This year, our TPCP annual meeting will be held alongside a FABI colloquium to celebrate our Anniversary. By definition, this will have a very obvious focus on matters pertaining to tree health. We hope that many TPCP members and friends will attend the event that will be held on he 13th and 14th May.

A little later in the year, July to be exact, South Africa will host the International Congress of Entomology (ICE). This major meeting, which occurs once every five years, will be held at the Durban International Congress Centre and there are already registrations from delegates from 99 (we hope for 100) countries. From a TPCP perspective, this meeting provides a great opportunity to draw together forest entomologists from around the world. In order to capitalise on this opportunity, we are organising an IUFRO Division 7.3 (Forest Insects), symposium and field excursion. The first two days of this event will be held in Pretoria and this will be followed by three days in the field examining and discussing insect pest and disease problems. There has been great enthusiasm for this meeting, which rapidly became fully subscribed by forest entomologists from around the world. It is already clear that this event will add substantial impetus to the TPCP entomology research and collaboration. Just a few weeks ago, the new TPCP insect rearing and quarantine facility was completed on the University of Pretoria experimental farm. Many staff members of TPCP member companies will have seen this facility when visiting the pitch canker screening programme on the farm. The new facility will provide the group with the ability to rear insect biological control agents in a manner that was previously not possible. New genotypes of *Ibalia leucospoides* and other Sirex biological control agents will move through these facilities prior to their release in the field. Further, we expect to be able to bring biological control agents of *Thaumastocoris perigrinus* and *Leptocybe invasa* through this facility and in this way, keep South African Forestry secure from damage due to these and other threatening pests.

If you are not already tired, from reading of the many TPCP activities already operational or planned for this year, I need to share just one more snippet of important news. This is that Dr. Stefan Neser, certainly one of South Africa's most important biological control experts has been appointed by the University of Pretoria. Stefan has a lifetime of experience in working with biological control agents and he has supported the TPCP incredibly strongly during the past year. Just as an example, his advice in building the new insect rearing facility has been invaluable. And he is already providing us with superb support that will lead to our gaining access to important biological control agents for a wide array of pests, not least of which is *Sirex*.

We have an exiting and busy year ahead. In fact a good part is already behind us. I say this thinking that, if one accepts that December is relatively quiet (other than for those rearing nematodes), one third of the year has already past. There is much to do on the research, extension and education fronts. The three main pillars of the TPCP operation. Our success in these efforts relies strongly on superb support and long term friendships with forest managers and field foresters that make the greater TPCP team.

Mike Wingfield

TPCP/CTHB Diagnostic Clinic 2007

This past year has been another record year for the clinic, with more samples being received than ever before.

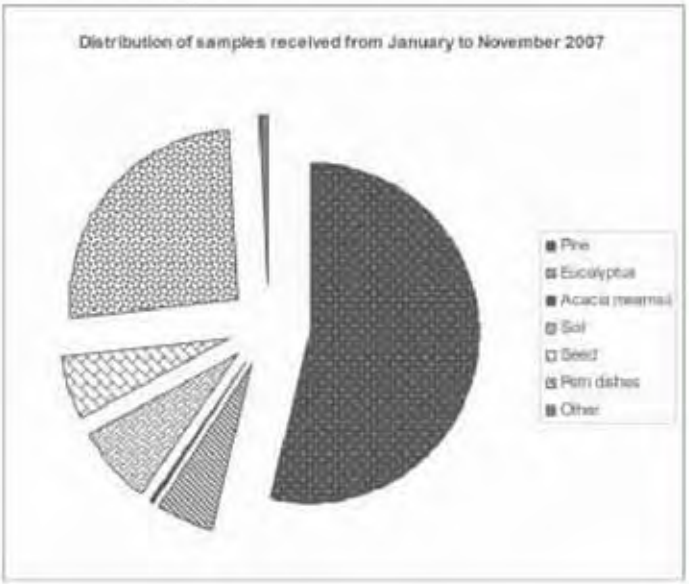
The clinic received a total number of 2913 samples from January until the end of November, with almost 800 of the samples received during October and November alone. Soil, seed and Petri dish samples for *Fusarium* screening comprised most of the 800 samples received. No samples were received in December.

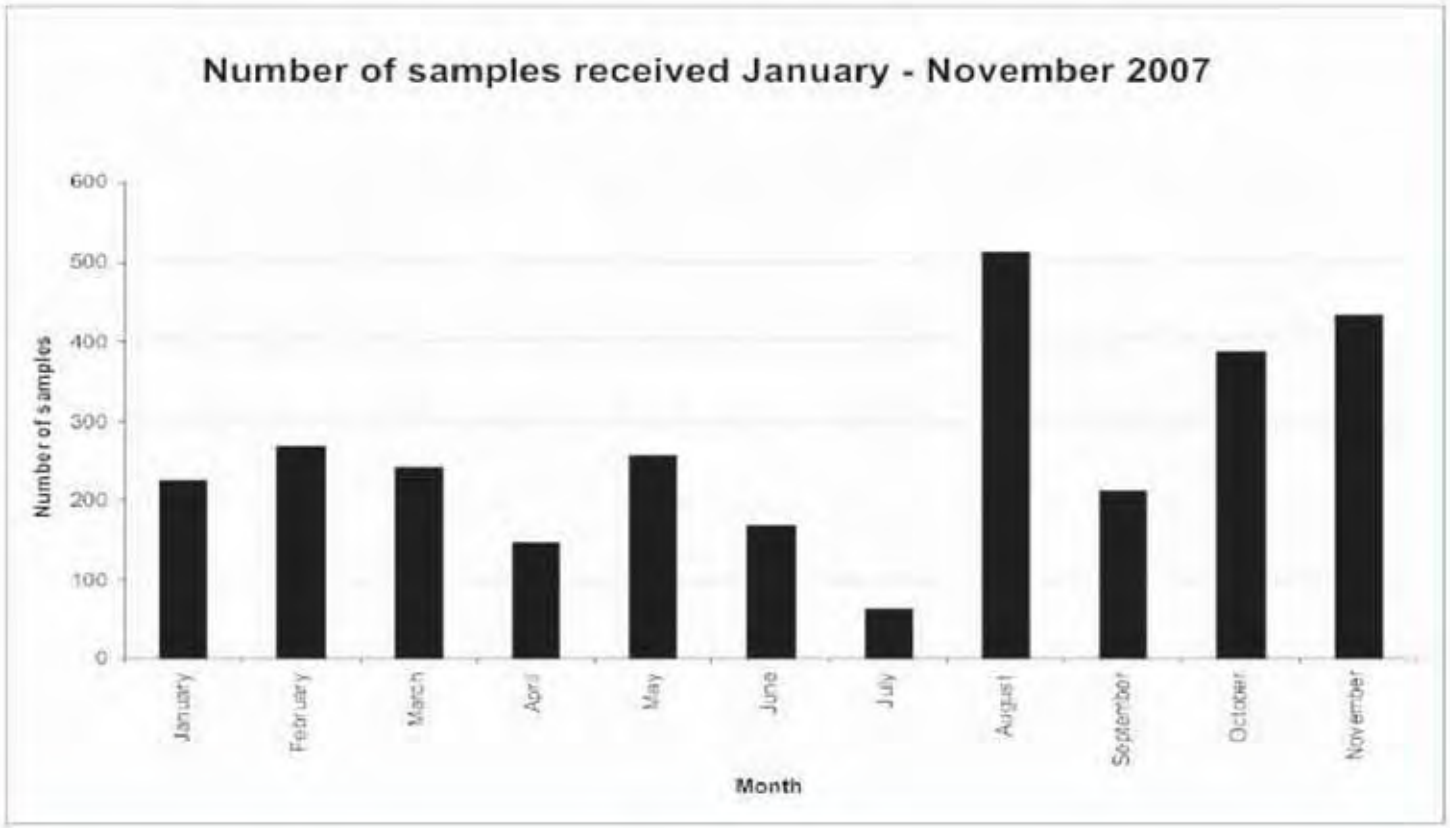
Pine samples comprised 54 % of the total number of samples received, with the majority of these samples received for *Fusarium* screening. *Eucalyptus* samples made up 5 % and *Acacia mearnsii* (Wattle) samples only comprised about 0.5 % of the total amount of samples.

The amount of soil samples received during this period necessitated the move to a new category, with soil samples comprising 7.4 % of all samples received.

Seed samples, received for *Fusarium* screening comprised 6.1 %, while Petri dish samples, for the confirmation of *Fusarium circinatum*, made up 26 % of the total samples.

Samples from non-forestry and indigenous trees as well as water samples are categorized under "other" and these comprised 1 % of received samples.





General clinic news

Each year new student members are drafted into the clinic as part of the training they receive during their respective degrees.

New members for 2008 are f.l.t.r. Donald Chungu, a MSc student from Zambia, Didier Begoude, a PhD student from Cameroon, Guillermo Perez, a PhD student from Uruguay and Shuaifei Chen, a PhD student from China.

In March we went on a fieldtrip as part of disease clinic training, to show members diseases in the field as well as expose them to forestry operations in South Africa. This year's trip was to the Mpumalanga province, specifically the Sabie area. Part of the trip included an indoor information session on how to identify pests and/or disease infestations with York Timbers.

Although many sites were lost to fire we still managed to see the serious damage caused by baboons. *Armillaria* infections as well as various other diseases and pests, including Cossid moth infestations at Lothair.

Contacting the Clinic

If you have any questions regarding possible diseases or would like to send samples to the Clinic, please contact either Izette Greyling (izette.greyling@fabi.up.ac.za) or Jolanda Roux (jolanda.roux@fabi.up.ac.za) for advice on the type and number of samples to send. It also allows us time to prepare properly, especially if a particularly large sample is expected.

Remember to join our online tree health forum, Treehealthnet, for regular updates on pests and diseases, fieldtrip advertisements and other issues related to tree health. More information on pests and diseases as well as sample submission is also available on our website at http://www.fabinet.up.ac.za/tpcp/.

WELCOME to the TPCP and CTHB

We welcome a number of new people to the research teams of the Tree Protection Co-operative Programme (TPCP) and DST/NRF Centre of Excellence in Tree Health Biotechnology (CTHB).

We hope that you will all be very happy in the programmes and enjoy many great interactions with our forestry and conservation colleagues.

PhD candidates

DIVINE SHYNTUM is a Cameroonian who obtained his MSc degree from the USA, where he worked on bacteria. Divine will be doing his PhD project under the supervision of Prof. Teresa Coutinho and Prof. Fanus Venter, working on host specificity factors in the plant pathogen *Pantoea ananatis*. This pathogen is of importance to eucalypt growers in many countries, including South Africa.

PhD candidates

MAYTHASITH KONKARN will be joining the TPCP for three months to work on the identification of the Ophiostomatoid fungi associated with bark beetles from Thailand.

He will be doing this work under the supervision of Prof. Mike Wingfield, Dr. Xhudong Zhou and Kevin Hyde.

MSc. candidate

FRANCOIS BOSHOFF is a new member of the CTHB and will be working on the Diversity of *Bradyrhizobium* species associated with the root nodules of *Melolobium* species.

This work will be done under the supervision of Dr. Emma Steenkamp.

MSc. candidate

LISA-DANELLE DE WET has a BSc honours degree from the University of Pretoria and will be working on the epidemiology of the pine pitch canker fungus, *Fusarium circinatum*, under the supervision of Dr. Emma Steenkamp.

MSc. candidate
LINDA NDOVE joined FABI from Venda and will be looking into the fungi associated with die-back of *Podocarpus* spp. in South Africa, with special reference to the Botryosphaeriaceae. Linda will be working with Dr. Bernard Slippers and Me. Elsie de Meyer.

BSc. Honours candidate
GIDEON GELDENHUYS is a Bioinformatics student working on the root rot pathogen Armillaria sp. and will be doing his honours degree under the supervision of Prof. Brenda Wingfield.

MSc. candidate
SIMON MARTIN completed his ***BSc honours*** in the TPCP and will be continuing with his MSc degree, working towards an understanding of the molecular basis of mating type gene interactions in the pitch canker fungus, *Fusarium circinatum*.

BSc. Honours candidate
LUNGHILE MTHOMBENI
Description of ***Burkholderia*** species nodulating ***Hypocalyptus*** and its relatives.

MSc. candidate
MARC BOUWER Marc is a Chemist by training and joined the TPCP research group for his MSc. under the supervision of Dr. Bernard Slippers and Prof. Mike Wingfield. He will be studying the volatile organic compounds emitted by eucalypt trees in relation to the snout beetle *Gonipterus scutellatus*.

BSc. Honours candidate
GABRIEL DE RIDDER will be using a multigene phylogeny to investigate the existence of cryptic species in the wilt pathogen *Ceratocystis albifundus*. He will be working with Prof. Jolanda Roux to characterize this important wilt pathogen of *Acacia mearnsii* which also occurs on numerous native tree hosts in southern and eastern Africa.

BSc. Honours candidate
JOHAN VAN DER LINDE joined the TPCP group and will be identifying the Botryosphaeriaceae causing wilt and canker of *Acacia mearnsii* in South Africa. He will be working with Prof. Jolanda Roux and will also be looking at possible gene flow between isolates from native *Acacia* spp. and *A. mearnsii* in forestry areas.

BSc. Honours candidate
ERIK BIRKHOLTZ joined the bacterial research group and will be filling in the gaps in the recently determined genome sequence of the eucalypt blight pathogen, *Pantoea ananatis*. He will be working under the supervision of Profs. Fanus Venter and Teresa Coutinho.

BSc. Honours candidate
JAN NAGEL will be working on the Botryosphaeriaceae, characterizing a previously undescribed species in the group.

BSc. Honours candidates
MELANIE VAN DER VAART will be working on a new bacterial disease of eucalypts in South America under the supervision of Profs. Teresa Coutinho and Fanus Venter.

BSc. Honours candidates
DARRYL HERRON will be working with Prof. Emma Steenkamp on the identification of a *Fusarium* species associated with pitch canker like symptoms in Colombia.

BSc. Honours candidates
PRIVYEN PILLAY will be working with Profs. Brenda Wingfield and Mike Wingfield on the characterization of a new species of *Ceratocystis*.

Secrets of cooperation between Trees and Fungi revealed

News from the VIB (the Flanders Institute for Biotechnology) - Gent, Belgium

Trees and fungi have constructed a close relationship with the passing of the ages. Fungi like to grow between the roots of trees and the arrangement is beneficial to both partners. Their delicate balance is now being revealed for the very first time. VIB researchers at Ghent University in collaboration with an international team have succeeded in unraveling the genetic code of the *Laccaria bicolor* fungus. This new information is crucial to our knowledge. It will lead to a better understanding of how fungi help trees to grow and how together they can be indicators of climate change.

Trees and fungi live happily together

Trees are the lungs of the earth. They draw CO2 from the atmosphere and convert it into sugars, which then become a source of energy. In the process they breathe O2 back into the atmosphere. This "green" production of biomass - trees account for 90% of the planet's land-based biomass - is a major influence on the health of our planet.

Trees grow better and faster when certain specialized micro-organisms occur in their root systems. One such organism is the *Laccaria bicolor* fungus. The symbiotic relationship of the fungus and the tree root systems is advantageous to both. The fungus facilitates the uptake of scarce nutrients such as phosphates and nitrogen and protects the roots against parasites in the soil. In return they are able to draw on the sugars in the roots. 85% of all plants and trees are dependent on symbiotic processes of this kind for their growth.

Genetic code of symbiotic fungus yields up first secrets

An international collaborative project was set up to characterize the genome of the soil fungus *Laccaria bicolor*. VIB scientists Pierre Rouzé and Yves Van de Peer, working with France's renowned INRA and JGI of the US, have sequenced the DNA of the fungus. They have been able to identify 20,000 genes in the fungal genome. Their analyses immediately resulted in new knowledge, including the discovery of an arsenal of small proteins known as SSPs (small secreted proteins), which are only made at those places where the fungus and the tree root come into contact. The genome study also revealed that the fungus is unable to break down plant cells but does affect the cell walls of pathogens. This could explain how these fungi protect their symbiotic partners. Additionally the researchers identified genes which play a role in communicating with all the players in the surroundings of the roots of the host tree during growth.

Fungi: barometers of climate change?

A better understanding of the genetic secrets of this fungus does not just hold out the prospect of being able to optimize biomass production; research into the delicate balance between fungus and tree may also yield important information that could be used to monitor climate change. Not only has the genome of the *Laccaria bicolor* been fully sequenced, that of the poplar, one of the trees with which it forms a relationship, is also fully known. This will make it possible to find out exactly how tree and fungus cooperate and react to stress factors such as drought or extreme temperatures resulting from climate change. The hope exists that the assembled information will result in concrete applications in which trees and fungi can be deployed to the benefit of both people and the environment. Yves Van de Peer leads the group of Bioinformatics and Evolutionary Genomics in the VIB Department of Plant Systems Biology, UGent - under direction of Dirk Inzé. (more info: www.vib.be/Research/EN/Research+Departments/Department+of+Plant+Systems+Biology/Yves+Van+de+Peer/)
Contact: Joke Comijn (info@vib.be).

Contacting the TPCP and CTHB

Contact numbers & web address:
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In order for us to coordinate services to you please help us by using the following contact address:
FABI
Lunnon Road, University of Pretoria, Hillcrest, Pretoria, Gauteng

Postal address:
Tree Protection Co-operative Programme (TPCP)
Attention: Professor Mike Wingfield,
FABI
University of Pretoria, Pretoria, 0002

Genome sequence of Pantoea ananatis

Pantoea ananatis is the causal agent of bacterial blight and die-back of young eucalypts in South Africa. This bacterium causes spots on the leaves which often coalesce to form larger lesions. Shoot tip die-back may also occur. The disease is particularly problematic in nurseries and mainly on *E. grandis* x *E. nitens* clones. *P. ananatis* not only attacks this host but has also been found to cause a stalk disease of maize locally and cause a variety of other symptoms on onions, melons, pineapple, rice and sudan grass in other parts of the world. It has also been reported to cause infections in humans. Little is known about the ecology and biology of the diseases caused by *P. ananatis*. In the case of eucalypts, we know it is a common epiphyte on leaves, and that high relative humidity and warm temperatures favour disease development.

Genomics is the study of an organism's entire genome. When a genome is sequenced it provides a wealth of information including all the proteins that the organism can make. This helps in the understanding of the ecology, biology and pathology of a pathogen. Through funding obtained from the University of Pretoria and the NRF, the genome of a virulent strain of *P. ananatis* from eucalypts was sequenced in May 2007. This is the first plant pathogen to be sequenced in Africa. The genome is 4.7 megabases in size and is currently in nine contigs (pieces) which we are currently assembling. The genome has been put through a computer system which annotates (identifies) known genes. The genome has been compared to the sequences of other genomes of bacteria belonging to the same family. A number of pathogenicity factors have been identified and their function is currently being investigated. We have discovered several unique elements in this genome which is going to provide a deeper understanding of how *P. ananatis* functions as both a pathogen of plants and humans. This research is being undertaken in collaboration with the Scottish Crops Research Institute in the UK.

Teresa Coutinho, Peter de Maier, Fanus Verster

Pest Alert

Glycaspis brimblecombei - Red Gum Lerp Psyllid

Glycaspis brimblecombei (Psyllidae, Hemiptera), commonly referred to as the red gum lerp psyllid, is a sap-sucking insect that feeds on eucalypts. Native to Australia, the red gum lerp psyllid has been accidentally introduced into the USA, Mexico, Chile, and Brazil, and it has recently been detected in Uruguay.

Females of the red gum lerp psyllid lay between 45 and 700 eggs. The eggs hatch in 10 to 20 days and the nymphs will pierce the plant tissue with their stylet (mouthparts), feeding on the xylem. As the nymphs feed they secrete honeydew with which they construct a waxy cover (called a lerp) around themselves. This cover is whitish and conical in shape and shelters the insects until the adult stage. In Australia there are two to four generations per year. The red gum lerp psyllid is known as an aggressive insect that spreads rapidly. Symptoms of its feeding include dropping of leaves and drying of leading shoots. Infested leaves are covered with waxy

secretions and honeydew, on which sooty mould grows. Heavy infestations can totally defoliate and kill trees. *Eucalyptus* species differ in their susceptibility to attack by the red gum lerp psyllid, with *E. camaldulensis* and *E. tereticornis* being highly susceptible and *E. grandis* being more tolerant.

Both chemical and biological control measures have been used in an attempt to control infestations of the red gum lerp psyllid. Contact insecticides are known to be ineffective as the nymphs are protected by their covers. Systemic insecticides have been used with some success, but their use is of limited value in plantation forestry due to the high cost. In the USA, natural enemies of the red gum lerp psyllid were imported from Australia. One of these, the parasitic wasp *Psyllaephagus bliteus* (Hymenoptera, Encyrtidae) has become established in the USA as a biological control agent for the red gum lerp psyllid.

The red gum lerp psyllid has not *yet* been detected in South Africa. However, with the increased movement of goods and people between countries across the globe in the last few decades, and the accompanying accidental increase of invasive species – be it weeds, fungal pathogens, insects, or other organisms – we need to anticipate the arrival of new pest species. New pests in South America often appear in South Africa soon afterwards (and vice versa). Good examples are the Sirex woodwasp (*Sirex noctilio*), the eucalypt snout beetle (*Gonipterus scutellatus*) and the bronze bug (*Thaumastocoris peregrinus*). This does not necessarily imply movement from South America to South Africa, but there are patterns relating to time of introduction that are worrying. Thus, the red gum lerp psyllid is a serious *Eucalyptus* pest that could appear in South Africa in the near future. The sooner new pests or pathogens are detected, the sooner control measures can be implemented. If you see these symptoms on *Eucalyptus*, or any unusual symptoms on your trees, please report it to:

Brett Hurley
Phone: 012 420 3938
Cell: 082 909 3211
Email: brett.hurley@fabi.up.ac.za

Figure showing psyllid and white lerp on *Eucalyptus* leaf.

Green Monsters on *Eucalypts*

In April reports were received of "big green worms" (the size of a man's finger!) dropping from the tops of trees and devouring the leaves of eucalypts in the Piet Retief area. These larvae were identified by Brett Hurley from specimens lodged in the Transvaal Museum. He confirmed that the larvae found eating the eucalypts are *Pseudobunaea irius* (Lepidoptera: Saturniidae). The adults are commonly known as the Irian Emperor moth. These larvae have been recorded attacking eucalypts previously, as well as Black Wattle, pines and various indigenous trees. It is known to have two life-cycles per year. The larvae can grow up to 8cm long and are known to be very well camouflaged. This is the stage that can cause severe damage by defoliation.

When fully grown, the larvae drop off the trees and pupate in the soil. Emerging adults have a wingspan of 10-12cm and 'eye-spots' on the hind-wings. Natural enemies of this species are present, but in severe infestations, application of insecticide would be needed to reduce the damage to trees. For the insecticide to have maximum effect, it should be applied when the larvae are still small, i.e. before most of the damage is already done. Of interest, is that one of the cultural control methods suggested for control of Saturniid larvae, in the 70's and before, was to use pigs to eat the pupae which they would detect and dig out the ground. This control proved to be very effective (and of course the pigs get fed, and can make a good pork roast after they have finished their job!!).

Leptocybe is spreading: Please help with monitoring

In the previous issue of TPCP news we reported on the discovery of the Eucalyptus gall wasp, *Leptocybe invasa*, in the Pretoria area. The discovery was made by Dr. Stefan Neser in June 2007 during his regular insect surveys on *Eucalyptus* in the area. At that point, the wasp was only known from a few trees in a limited area. Follow-up surveys later in 2007 by members of the TPCP have found galls on *Eucalyptus* plants a few kilometres away, showing that the introduction must have happened well before June 2007 and was spreading. *Leptocybe* has spread incredibly fast throughout other countries where it was introduced. This seems to indeed also be the case in South Africa. Early in 2008 the first infestations were noticed in the FABI nursery at the University of Pretoria experimental farm, some distance from the initial detection sites. Subsequently, infested plants have also been found in more than one location around Johannesburg. Heavily infested *Eucalyptus* was also recently discovered in the Brits area by members of the Plant Protection Research Institute (PPRI) and even in the Upington area! It is of utmost importance that every forester assist to monitor the spread of the Eucalyptus gall wasp. This is especially important for NURSERY MANAGERS. Sending infested plants to the field is one of the fastest routes to spread this damaging wasp. Please note the photographs with typical galls or swellings on the midrib of the leaves, or young twigs. More images are available on the TPCP website (www.fabinet.up.ac.za/tpcp).

IMPORTANT: If you suspect that you have the Eucalyptus gall wasp, please contact us to organize a visit to the site, or send a picture of the observed symptoms. Do not package and send these galls to us. The wasps are very small and could easily escape from packaged material and spread to new areas.

Sirex! control: Nematodes by the millions to stop

There has been a buzz of activities during the past nine months in labs and incubators of FABI, as the Sirex team has been preparing and producing *Deladenus siricidicola* nematodes – a biological control agent against the Sirex woodwasp. One of the major tasks we took on towards the end of 2007 was to produce two billion of these nematodes for industry inoculations in 2008.

These inoculations form a major part of the industry's strategy to control the Sirex woodwasp.

The nematode production has been an enormous and intense operation involving the whole Sirex research team, as well as other TPCP staff and students. This included five full time technicians, part time assistance of four academic staff members, and ad hoc assistance from a further 5 staff members, 8 undergraduate and more than 20 post graduate students.

The production of the nematodes started in the second half of 2007, coinciding with the dissection of 12000 wasps to assess industry inoculations. This busy start continued throughout December and the first quarter of 2008. Only now is the intensity easing off as we approach the final delivery in middle May. To date more than 17000 flask have been prepared using more than two tons of wheat and rice.

The work continued seven days a week, with flask preparation, inoculation, nematode culture treatments and other work being done during the week, while harvesting was done on Saturdays and Sundays, to be ready for the courier consignments on Monday mornings. The good news is that despite autoclaves that broke, load shedding, tennis elbows and more, we will deliver the two billionth nematode by mid-April. We then aim on delivering a further one billion nematodes based on revised targets to the industry.

This success can only be ascribed to the incredible dedication and hard work of the great Sirex team of the TPCP. Congratulations to all of you!

SAIF Symposium

Pest awareness Symposium

The threat that pests and diseases pose to plantation forestry has, in recent years, become more evident. Serious insect pests like *Sirex* and *Thaumastocoris* are causing widespread devastation, emphasizing the need for cross discipline and cross company efforts to control, not only these pests, but other pests and diseases posing threats to our industry.

For effective measures to be in place to deal with pests and pathogens, knowledge is needed.

To this end the South African Institute of Forestry (SAIF) in conjunction with the Tree Protection Co-operative Programme (TPCP) held a Pest and Disease Awareness Symposium for members of the industry. The workshop was held at the Hilton Hotel in Hilton at the end of January 2008.

Dr. Andrew Morris chaired the workshop and Dr. Bernard Slippers (TPCP) gave the first talk, focusing on pests in plantation forestry. His talk centred around both native (e.g. Cossid moth) and introduced (e.g. *Sirex, Leptocybe*) pests in plantation forestry. He also talked about various methods to prevent future introductions as well as measures used to control pest infestations. Grant Boreham (Sappi) spoke about the Cossid or Goat Moth, *Coryphodema tristis*, giving results obtained as part of a study done on the Cossid moth infestations in the Lothair and Carolina areas.

This native pest has only been reported from *E. nitens* so far, and is a good example of a native pest adapting to attack a commercial plantation species.

Ryan Nadel (TPCP) gave his talk on *Thaumastocoris peregrinus*, introducing members to this little pest currently causing major damage throughout *Eucalyptus* growing regions in South Africa. He also gave an update on the research he has been conducting on *Thaumastocoris* as part of his PhD study, which includes a survey covering most *Eucalyptus* growing regions in South Africa. This survey is done with the cooperation of various members of the forestry industry.

Phillip Croft was next to talk about probably the most important pest currently causing major damage and losses in parts of South Africa – the *Sirex* wood wasp. He talked about the biology of the insect, as well as the effectiveness of the nematodes used as biocontrol agent against *Sirex* as well as other biocontrol agents available. He also gave results on various traps set up by the industry to monitor the spread and occurrence of *Sirex* and confirmed that *Sirex* has moved as far north as the Vryheid area with wasps being found in Kwambonambi and Ngome.

The last two speakers were Mike Kruger (Topcrop nursery) and Izette Greyling (TPCP) with their talks focusing on the pitch canker fungus, *Fusarium circinatum* and the TPCP/CTHB Diagnostic Clinic, respectively.

Mike Kruger focused on *Fusarium circinatum* from a nursery aspect, talking about hygiene methods needed to control this devastating pathogen as well as measure to employ for nursery certification.

Izette Greyling gave a general overview of the diagnostic clinic, what to look for and how to send samples to the clinic for identification.

The workshop concluded with all being in agreement that only through cross discipline, company and country cooperation will we be able to effectively control pest and diseases.

Thaumastocoris peregrius (TPCP) monitoring trial

Provisional results from the *Thaumastocoris peregrius* (TPCP) monitoring trial

In an attempt to accurately determine the population size and dynamics of *Thaumastocoris peregrinus*, a network of eight permanent sampling sites were established across South Africa as discussed in Tree Protection News (November 2007).

The weekly monitoring of these plantations, in collaboration with TPCP members, has significantly improved our understanding of *T. peregrinus*. Certain key environmental variables have now been provisionally identified as impacting the population growth of this pest during the reported time period (February 2007 until December 2007). We thus eagerly await the analysis of several more months of data to establish whether these highlighted variables continue to play an impacting role in the establishment and persistence of this pest, as ascertained in the first six months of this trial.

The trial has been set up in collaboration with several industry members spanning the major *Eucalyptus* growing areas of South Africa. These include:

1. Gauteng; Pretoria
   (Set up 19 February 2007)
2. Limpopo; Tzaneen
   (Set up 29 May 2007)
3. Mpumalanga; Sabie
   (Set up 27 June 2007)
4. Northern KwaZulu-Natal; Piet Retief
   (Set up 16 July 2007)
5. KwaZulu-Natal Zululand;
   Kwambonambi (Set up 17 July 2007)
6. KwaZulu-Natal Midlands:
   Pietermaritzburg
   (Set up 3 September 2007)

Provisional results from this study have indicated that over the studied time period (February 2007 (Pretoria) and since June 2007 (other sites) to December 2007) ambient temperature played an important role in determining the population growth of adult *T. peregrinus* individuals. *Thaumastocoris peregrinus* population growth within the colder regions of the country namely those of Pretoria and Piet Retief areas, were affected by minimum and mean temperatures. In contrast, data obtained from Tzaneen revealed that during the dry, winter period *T. peregrinus* populations were correlated to that of maximum temperatures and not mean or minimum temperatures, possibly because the minimal critical thermal limits of *T. peregrinus* were not reached as might have occurred within the colder Pretoria and Piet Retief sites.

These environmental variables are likely to change considerably within the summer months. It has to therefore be stressed that the information presented here is preliminary and that conclusions about environmental factors affecting the population growth of *T. peregrinus* might be inaccurate at this stage. The data obtained only after a full year of sampling will already give a better indication towards the effects of different environmental factors that affect *T. peregrinus*. These indications can then be verified during a second year of sampling.

Acknowledgements

We would like to thank the following for their assistance with this trial: Botha Maree, Sonia du Buisson, Maurits Perold (Hans Merensky, Northern Timbers), Tony Winter (Mondi), Tammy Swain and Denis Oscroft (ICFR), Glen Mitchell (KLF).

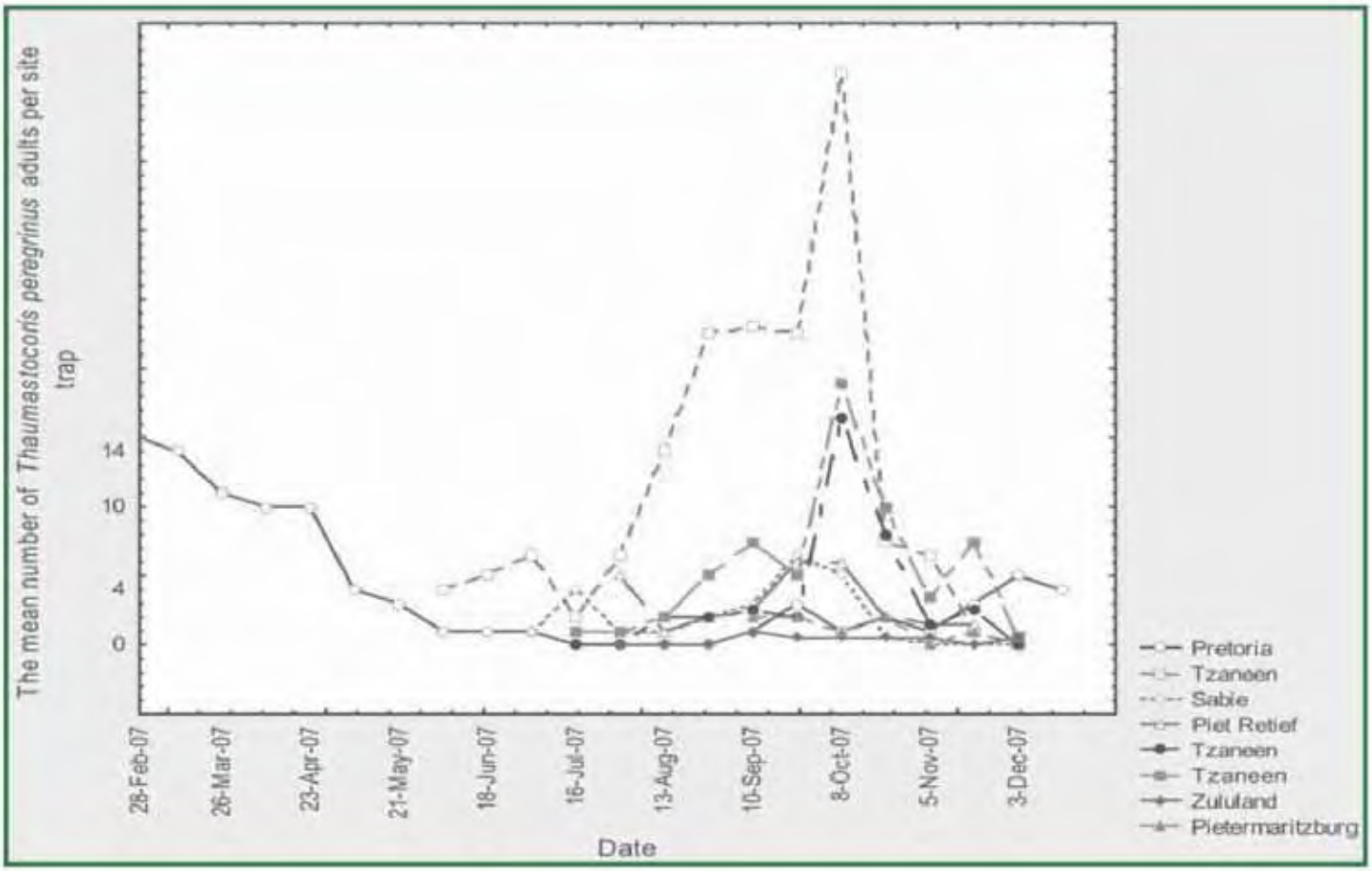

Figure 1: An overview of the mean number of *Thaumastocoris peregrinus* adults caught at each sampling site calculated on a fortnightly basis

Who's Who in the TPCP?

GILBERT KAMGAN NKUEKAM

PhD student

Nationality: Cameroonian

Research / Expertise: My research project focuses on the study of ophiostomatoid fungi, their insect associates and wounds on *Eucalyptus* trees in Australia and South Africa. Ophiostomatoid fungi, which require wounds for infection of trees, are being more commonly reported from *Eucalyptus* trees as important pathogens. These fungi are known to have associations with insects that either make wounds, resulting in fungal infection, or visit fresh wounds and thereby introduce fungi to trees. For my PhD study I will specifically investigate the interaction between wounds, insects and ophiostomatoid fungi on *Eucalyptus* trees. In collaboration with Australian colleagues, I am also investigating ophiostomatoid fungi on *Eucalyptus* in Australia. Although *Eucalyptus* trees are native to Australia, yet very little is known regarding the ophiostomatoid fungi on these trees in their native range.

Hobbies / Interests: I am a big soccer fan. I also play soccer, but due to my studies this does not happen often. Others sports that I enjoy are boxing, athletics and rugby (my interest in rugby has grown considerably since the last World Cup). To keep fit I run and go to the gym.

HARDUS HATTING

Senior Technician

Nationality: South African

Research / Expertise: My work is mainly focused around the biological control of the Sirex wood wasp. I am responsible for the mass production of the parasitic nematode *Deladenus siricidicola*, which is currently used as the primary control agent for Sirex. This includes growing and maintaining healthy cultures of the nematode and the fungus on which the nematodes feed and the long term storage of the nematodes in liquid nitrogen (cryopreservation). The past nine months have been particularly challenging and exciting, as we needed to produce over two billion nematodes! Thanks to an enormous team effort, we achieved this goal, and more. Apart from the nematode production, I am also involved with Sirex and general entomology research and extension work at the TPCP. This includes the preparation and implementation of field trials, collecting samples and presenting at field days.

Hobbies / Interests: Watching rugby, playing golf, having a braai and socializing. I am an outdoors person, and enjoy camping and hiking.

Conferences attended by TPCP/CTHB members

Over the last six months a variety of conferences were attended by members of the TPCP and CTHB. These conferences included the biannual meeting of the South African Society of Microbiology (SASM), the International Union of Forest Research Organisations meeting (IUFRO), the first joint conference of the South African Association of Botanists (SAAB) and South African Society for Systematic Biology (SASSB).

The IUFRO conference entitled "Eucalypts and Diversity: balancing productivity and sustainability" took place from the 22nd to the 26th of October 2007. It was the first time in 15 years that this conference took place in Africa. Delegates included students, researchers, academics and individuals from forest-related fields. Fabians included the following: Derian Echeverri, Ryan Nadel, Brett Hurley, Dr. Bernard Slippers and Proffers Jolanda Roux and Mike Wingfield.

Two other conferences covered a diverse range of topics pertaining to Microbiology, Botany and Systematic Biology and were held in January this year. During 14 to 18 January a joint conference of SAAB and SASSB took place at the Drakensville Resort. Topics included for example Biodiversity and Conservation, Animal and Plant Phylogenetics and Pollination Biology and the last day was set aside for talks specifically covering the Cape Biota. Attendees from FABI included: MSc students (Marija Kvas, Marcele Vermeulen, Chrizelle Beukes), PhD students (Gilbert Kamgan and Albe van der Merwe), Post-doctoral fellow Marieka Gryzenhout and research project leaders Dr. Emma Steenkamp and Prof. Jolanda Roux.

The SASM meeting, 21 – 25 January, formed part of the Bio-08 Conference that also included two other conferences, namely the South African Society for Biochemistry and Molecular Biology and Biotech SA. The combined meeting was held at Rhodes University, Grahamstown. Members from our group which attended this conference included: MSc students (Ariska van der Nest, Amelia Keyser, Aisha Mohammed-Ali, and Francois van der Walt) and project leaders Dr. Gert Marais and Prof. Brenda Wingfield.

IUFRO

Population Diversity and Structure of the *Eucalyptus* snout-beetle, *Gonipterus scutellatus* (Coleoptera, Curculionidae) in South Africa, Spain, Chile and Uruguay

Echeverri D, Slippers B, Hurley BP and Wingfield MJ.

The *Eucalyptus* snout-beetle, *Gonipterus scutellatus*, is native to Australia, but has been introduced into many countries where eucalypts are grown commercially as non-natives. The first record of the pest in South Africa was in 1916 and it has more recently appeared in Uruguay (1943), Spain (1991) and Chile (1998). In these non-native areas, *G. scutellatus* is one of the most important pests attacking *Eucalyptus spp.*, causing defoliation, reduction in growth and sometimes death of the trees. Despite the long history and importance of *G. scutellatus* as a pest of non-native *Eucalyptus* spp. in plantations, little is known regarding the diversity of populations within and between these countries or how this may influence control strategies. In this study, we determined the population diversity and structure of *G. scutellatus* within South Africa, and compared this with representative samples from Spain, Chile and Uruguay. In

South Africa, individuals were collected from all the main areas where *Eucalyptus* species are planted, while individuals from Spain, Chile and Uruguay were collected from individual sites at Pontevedra, Chillan and Paysandú, respectively. A 476 bp DNA fragment of the mitochondrial *cytochrome oxidase I (COI)* gene was sequenced for individuals from the four areas. Analyses of these mitochondrial sequence data showed two distinct clades within the South African *Gonipterus* population, each with very limited diversity and structure. This suggests two independent introductions and subsequent spread throughout the country. One of these South African lineages is also shared by the Chilean, Spanish and Uruguayan populations, reflecting a common origin of the introduction of this lineage of *Gonipterus* into all these regions, or an introduction from the same source population. Finally, a third mitochondrial sequence clade was observed in the Uruguayan population of *Gonipterus,* but not in any of the other countries. The percentage difference between the three mitochondrial *COI* sequence clades was sufficiently high to suggest that the insects represent distinct species. Data from Australia, the country of origin of *G. scutellatus,* is now required to elucidate the uncertainties regarding the taxonomy of *Gonipterus*, and determine its pathways of introduction into other parts of the world.

Distribution and Population Structure of the Bronze Bug, *Thaumastocoris Peregrius* **in South African** *Eucalyptus* **Plantations**

Nadel RL, Slippers B, Scholes MC and Wingfield MJ.

Thaumastocoris peregrinus, also known as the bronze bug, is a recently introduced invertebrate pest on commercially grown *Eucalyptus* trees in South Africa. This sap feeding insect has spread rapidly throughout the country posing a major threat to the local forestry industry by reducing the growth of trees. Intensive, countrywide surveys for *T. peregrinus* were conducted, spanning several *Eucalyptus* species, hybrids and their clones, and various geological and climatic regions.

The main species or hybrids that were found to be attacked by the insect were *E. grandis* x *camaldulensis* and *E. grandis* x *urophylla* clones, although *E. grandis, E. tereticornis* and *E. smithii* were also severely infested. No consistent climatic restrictions on the distribution or population build-up of *T. peregrinus* were observed. Analyses of mitochondrial DNA sequence diversity were used to characterise the population structure and distribution of the pest in South Africa. Results indicate that two haplotypes of *T. peregrinus* are present. One of these is widely distributed in both summer and winter rainfall areas.

The other haplotype is dominant in a small area in the sub-tropical summer rainfall zone. This distribution, together with the known direction of initial spread, suggests that there might have been two introductions of *T. peregrinus* into South Africa.

A Diverse Assemblage of Botryosphaeriaceae **Infect** *Eucalyptus* **in Introduce and Native Environments**

Slippers B, Pavlic D, Maleme H, Wingfield MJ

The Botryosphaeriaceae cause endophytic infections in leaves and bark of various trees, including *Eucalyptus*, and apparently persist for extended periods of time. Under conditions of stress, these fungi cause many different disease symptoms on *Eucalyptus*, of which stem and branch cankers and die-back are the most prominent. Given their cryptic, endophytic nature, the Botryosphaeriaceae are easily overlooked when moving seeds and plants around the world. It is, therefore, not surprising to see a growing number of examples of introductions of Botryosphaeriaceae into new environments. In the past, only two Botryosphaeriaceae were commonly reported from *Eucalyptus*, namely *Botryosphaeria dothidea and Neofusicoccum ribis (reported as B. ribis).* It is now known that these species are generally rare on *Eucalyptus*, and that *N. parvum, N. eucalyptorum, N. eucalypticola, N. australe, N. macroclavatum, N. andinum,*

Dichomera eucalypti, *Pseudofusicoccum stromaticum* and *Lasiodiplodia theobromae* also infect this host. Interestingly, different species dominate on *Eucalyptus* in different regions of the world, irrespective of whether other species occur in that environment or not. As examples, in parts of eastern Australia, *N. eucalyptorum* and *N. eucalypticola* dominate, although *N. australe* is common on *Acacia* spp. in this area, while in western Australia *N. australe* dominates. In South Africa and Chile *N. parvum*, *N. eucalyptorum* and *N. eucalypticola* are common, despite the presence of *N. ribis* and *N. australe* on related hosts such as *Syzygium*. In Venezuela, there are five other species not common on *Eucalyptus* in other areas, but *L. theobromae* dominates. In Colombia, *B. dothidea* and *B. ribis*, and in Uganda and Ethiopia, *L. theobromae* and *N. parvum*, are the most common. These fascinating patterns of distribution are explored, while their pathogenicity and potential influence on *Eucalyptus* plantations and surrounding native plant communities are considered.

SAAB/SASSB

Diversity of *Fusarium* species associated with malformed inflorescences of *Syzygium cordatum*

Kvas M, Steenkamp ET, Wingfield BD, Marasas WFO and Wingfield MJ.

Syzygium cordatum is myrtaceous tree native to Southern Africa. Throughout its natural distribution, the inflorescences of this tree appear to be affected by a malformation disease, typically resulting in abnormally enlarged, excessively branched and sterile inflorescences. On *Mangifera indica*, similar symptoms are associated with at least four *Fusarium* species. The aim of this study was, therefore, to determine the diversity of *Fusarum* species associated with malformed *S. cordatum* inflorescences in South Africa. In order to identify *Fusarium* species recovered from the infected inflorescences, isolates were characterised using DNA sequence data. For this purpose, isolates were grouped using restriction fragment length polymorphism (RFLP) analysis of the amplified ribosomal RNA intergenic spacer (IGS) region. An isolate was then chosen for each unique IGS-RFLP profile and part of the gene encoding translation elongation factor 1 (EF1) was amplified and sequenced. These sequences were then compared with the known Ef1 sequences in the *Fusarium* identification database (http://fusarium.cbio.psu.edu/). Our results showed that malformed *S. cordatum* inflorescences are colonized by highly diverse *Fusarium* species belonging to the sections *Elegans*, *Gibbosum*, *Arthrosporiella* and *Liseola*. Although some of these are well-known saprophytes (e.g. *F. oxysporum*, *F. equiseti* and *F. pallidoroseum*), the majority of the *Fusarium* isolates examined appear to represent novel lineages and/or species of this genus. Hence, previously unexplored ecological niches such as malformed *S. cordatum* inflorescences represent significant sources of novel fungi, not only of *Fusarium*, but most probably also other fungi.

***Cryphonectriaceae* canker pathogens on native and non-native Myrtales in southern Africa**

Vermeulen M, Gryzenhout M, Wingfield MJ and Roux J.

The *Cryphonectriaceae* includes some of the world's most important tree pathogens. Three genera occur in Africa. These include two species of *Chrysoporthe*, *Chr. austroafricana* and *Chr. cubensis*, with *Chr. austroafricana* hypothesized to be native and *Chr. cubensis* introduced. *Chr. austroafricana* infects non-native *Eucalyptus* spp. and *Tibouchina* spp. and native *Syzygium* spp. in southern to central Africa. *Chr. cubensis* infects non-native *Eucalyptus* spp. and *S. aromaticum* in central Africa. *Celoporthe dispersa* is known only from South Africa and has been collected

from *Heteropyxis canescens*, *S. cordatum* and *T. granulosa*. *Holocryphia eucalypti*, known only on *Eucalyptus* spp., is believed to have been introduced into South Africa. Based on the number of new reports, it is clear that the geographical and host distribution of *Cryphonectriaceae* in Africa is incomplete. In this study, Myrtales in Zambia, Swaziland and South Africa were surveyed for the presence of *Cryphonectriaceae*. Isolates were identified based on morphology and DNA sequence comparisons for the partialtubulin gene region and the internal transcribed spacer regions of the ribosomal RNA operon. *Cel. dispersa* was identified for the first time from Zambia on *S. guineense* and *S. cordatum* and on *S. legatti* in South Africa. *H. eucalypti* was found in Swaziland. Results of this study show that *Cel. dispersa* and *H. eucalypti* have a wider geographical and host range then previously thought. Surveys and monitoring of the occurrence of these fungi should continue as they are serious canker pathogens of trees, clearly capable of host jumps between species in the Myrtales.

Polyphasic taxonomy reveals numerous new fungal tree pathogens in the *cryphonectriaceae*

Gryzenhout M and Wingfield MJ

The *Cryphonectriaceae* is a recently established family including well-known canker pathogens of forest trees, some of which have caused devastating diseases. Examples of these pathogens include *Cryphonectria parasitica* that causes chestnut blight in the Northern Hemisphere, and *Chrysoporthe cubensis* that causes cankers on *Eucalyptus* spp. and related trees in the tropics and sub-tropics. Recent DNA and morphology based taxonomic revisions have led to the description of many additional genera and species in this family. Currently the *Cryphonectriaceae* includes ten genera and 22 species, where it prior to these studies, encompassed only two genera and 13 species. Ongoing surveys in several parts of the world and the characterization of existing isolates are constantly revealing new genera and species. For example, a new genus in this group has recently been found to cause a canker disease on *Eucalyptus* clonal hedges in Indonesia. Likewise, another new genus is associated with dying *Eucalyptus* twigs in Colombia. Many of the newly described genera are also monotypic. This clearly indicates that the *Cryphon-ectriaceae* remains a largely under-sampled group. This is also illustrated by the fact that new diseases caused by *Cryphonectriaceae* are continuously found on well-studied hosts such as *Eucalyptus*. These fungi probably occur on the vastly unexplored native vegetation surrounding many of these *Eucalyptus* plantations, and are able to infect trees in commercial *Eucalyptus* plantations.

Bio-08

Botryosphaeriaceae associated with native Acacia spp. with special reference to *Acacia mellifera* **(M. Vahl.) Benth.**

van der Walt FJJ, Marais GJ, Slippers B, Roux J and Wingfield MJ.

Acacia spp. play a significant ecological and economic role in southern Africa. However, there is still a considerable lack of knowledge regarding the diseases associated with these trees. Members of the Botryosphaeriaceae pose considerable risks as pathogens that can jump between native and non-native tree hosts. Because of this threat the identification of, and knowledge of the biology of these pathogens is crucial, especially if they occur on plants that are transported world-wide. Surveys were conducted on *A. mellifera* in the Pretoria area, on *A. mellifera* and other native *Acacia* spp. with similar symptoms in Namibia and in the North Eastern Cape Province.

A large number of Botryosphaeriaceae isolates were obtained. In total, twelve different species were identified based on PCR-RFLP groupings, morphological characterization and sequencing data. Ten new species, including a new genus is described. These included *Dothiorella rosulata* sp. nov., *D. oblonga* sp. nov., *D. capri-amissi* sp. nov., *Neofusicoccum namibiense* sp. nov., *Fusicoccum avasmontanum* sp. nov., *Lasiodiplodia pyriformis* sp. nov., *L. magnistriata* sp. nov., *Diplodia variabilis* sp. nov. and the new genus, *Mucodiplodia* prov gen. with two new species, *M. africana* sp. nov. and *M. papillata* sp. nov. *Dothiorella viticola* and *Botryosphaeria dothidea* which also included a new *Dichomera* synanamorph were also identified. This is thefirst detailed study of Botryo-sphaeriaceae occurring on native *Acacia* spp. in southern Africa and greatly expands the number of species known from the continent.

Chrizelle Beukes, Albe van der Merwe, Marcele Vermeulen, Emma Steenkamp, Jolanda Roux, Marija Kuas, Marieka Gryzehout & Gilbert Kamgan Nkuekam in the Drakensberg.

Ariska Van Der Nest, Francois v. d. Walt & Aisha Mohammed.