

# TREE PROTECTION NEWS

Newsletter of the Tree Protection  
Co-operative Programme -  
University of Pretoria

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## MESSAGE FROM THE DIRECTOR

An ex student of FABI and member of the TPCP team recently visited us and then went home to write an e-mail to express his amazement at the “hive of activity” that he had witnessed. Certainly the Tree Protection Co-operative Programme has flourished in recent years. This is largely, perhaps exclusively, due to a highly motivated and passionate team of researchers and support staff. I remain convinced that enthusiasm and passion is the key to accomplishments of the TPCP. If I were to begin to list the awards and special accomplishments of TPCP team members, I would certainly fall into the trap of omitting some, and there would be little space for any other news in this editorial.

As I write this piece, the final day of February has just past. Just two months and yet so much has already happened this year. There have been field trips just about every week, various members of the team have attended the annual meetings of the Southern African Society for Plant Pathology and the programme to deploy the biological control nematode for *Sirex* wood wasp control is moving into top gear. Meetings have been held with many of the TPCP members to discuss research plans for the coming year and a very extensive pitch canker screening activity has been undertaken. The TPCP team spent nearly five hundred person days in the plantations last year and this seems to be the norm for the group. I certainly expect little different this year and with the *Sirex* biological control programme running at “full throttle” the TPCP flag will certainly be flying high in plantations.

The annual research meeting of the TPCP is always one of the premier events of the year. This year the meeting will be somewhat different but very special. We have planned the meeting to be held on Wednesday 26<sup>th</sup> of May and thus following a day-long series of lectures and other events to mark the inauguration of the new FABI facilities. In effect, the TPCP meeting might be seen as a two-day event, the first of which will include a great deal of material of importance to forestry, but it will not be exclusively focused on this industry. Various forest pathologists and forest entomologists will be attending and presenting lectures during these two days. They include Bill Dyck from New Zealand who conducted a review of Forestry Research in South Africa about ten years ago, and who now acts as a

technology vendor to the New Zealand Forestry Industry.

Prof. Tim Paine, well known forest entomologist with a strong interest in Eucalyptus pests, from Riverside California will be this years Hans Merensky Fellow and will be attending the event. We will also be hosts to Dr Thomas Kirisits, specialist on bark beetle/fungal pathogen interactions, Dr Tom Gordon, Pitch Canker authority from Davis, California and Dr. Angus Carnegie forest protection specialist and Sirex control expert from State Forests, NSW, Australia. All in all a fantastic line-up of speakers to join our team in presenting some of the newest ideas in the field of forest protection. We certainly look forward to sharing this special event with many staff working for TPCP member companies.

The TPCP team and other FABIANS are working furiously to complete outstanding elements of the new FABI facilities. The new three storey building that is physically attached to the initial single storey building is known as FABI SQUARE BIOINFORMATICS. Two floors of the building essentially extend the facilities previously available to the team. They include world class plant quarantine greenhouses, facilities for research on genetically modified plants and tissue culture, and totally new DNA sequencing facility, additional laboratories, offices, lecture rooms and more. Most of the third floor of the building houses the new Bioinformatics facilities that include amazing Computer Hardware and which will be most important to the TPCP in coming years. Our group took occupation of the new facilities towards the end of last year and we are now dealing with the customary "teething problems". As anyone who has built a house will know, such problems are the norm, but the complexity of some of the facilities have clearly added a special dimension to completing all elements of the project. We anticipate that by the time of the inauguration, these will not require further discussion.

This year, FABI including the TPCP will host two important workshops. These will be the International Fusarium Workshop that is traditionally held at Kansas State University and a special workshop on fungal pathogen

population genetics. The Fusarium workshop is especially pertinent given the importance of the pitch canker pathogen in South Africa. This workshop will be presented by at least four overseas guests as well as by a team of South African specialists. The planned population genetics workshop will be more finely focused on the needs of researchers in the TPCP and will probably not be open to others, due to a need for hands-on activities. This will be presented by Prof. John Leslie of Kansas State University and Dr. Andre Drenth of the Queensland (Australia) Department of Primary Industries.

I am sure that some readers of Tree Protection News have already noticed the new and impressive FABI Web Site with the greatly improved TPCP site to go with it. Much effort has gone into making the site interesting and useful to TPCP members. Please do visit the site when you have the opportunity to do so. We would also welcome feedback and suggestions for improving the site further. To access the site use the following URL: <http://www.fabinet.up.ac.za>

I am sincerely hoping to see many of you at this years annual research meeting and FABI Square inauguration. If not at this event, we will surely meet in the plantations. Please do not hesitate to contact us if we can help with disease or insect pest problems. The line of communication via Dr. Jolanda Roux and Prof. Teresa Coutinho has worked well this last year. For enquiries relating to field problems please contact Jolanda at 0829093202, [jolanda.roux@fabi.up.ac.za](mailto:jolanda.roux@fabi.up.ac.za) and for diagnoses of disease and pest problems Teresa at 012 4203934/8, [teresa.coutinho@fabi.up.ac.za](mailto:teresa.coutinho@fabi.up.ac.za). Although we use these two portals of access to the Programme to ensure effective response, you are of course free to contact any other team members. Specifically for Sirex questions, it is best to contact Brett Hurley 0829093211, [brett.hurley@fabi.up.ac.za](mailto:brett.hurley@fabi.up.ac.za) directly. I am also always happy to hear from TPCP colleagues and can be contacted at Pretoria 012 420 3938, [mike.wingfield@fabi.up.ac.za](mailto:mike.wingfield@fabi.up.ac.za).

**Mike Wingfield, Director of TPCP, FABI**

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## **Identification and development of microarray markers linked to *Cryphonectria cubensis* tolerance in *Eucalyptus grandis***

*Cryphonectria* canker caused by *Cryphonectria cubensis* is one of the more serious diseases affecting *Eucalyptus* plantations in South Africa. This disease has severely limited the development of plantations of susceptible *Eucalyptus*

spp. in areas where climatic conditions favour infection. Deployment of *Cryphonectria* canker resistant *Eucalyptus grandis* planting stock is considered the only feasible means of controlling the disease. The South African forestry industry, therefore, requires rapid, yet reliable screening methods that reflect disease susceptibility and tolerance. One approach could be through the application of PCR-based genotyping techniques.

PCR-based methods for trait diagnosis have become available during the past 15 years. Among these, RAPD, PCR-RFLP, SCAR and SSRs are increasingly being used for fingerprinting and gene-tagging in breeding populations. All of these methods are technically accessible and quickly provide polymorphic markers. However, due to their dependence on gel electrophoresis, the number of fragments that can be analyzed limits throughput. This limitation can

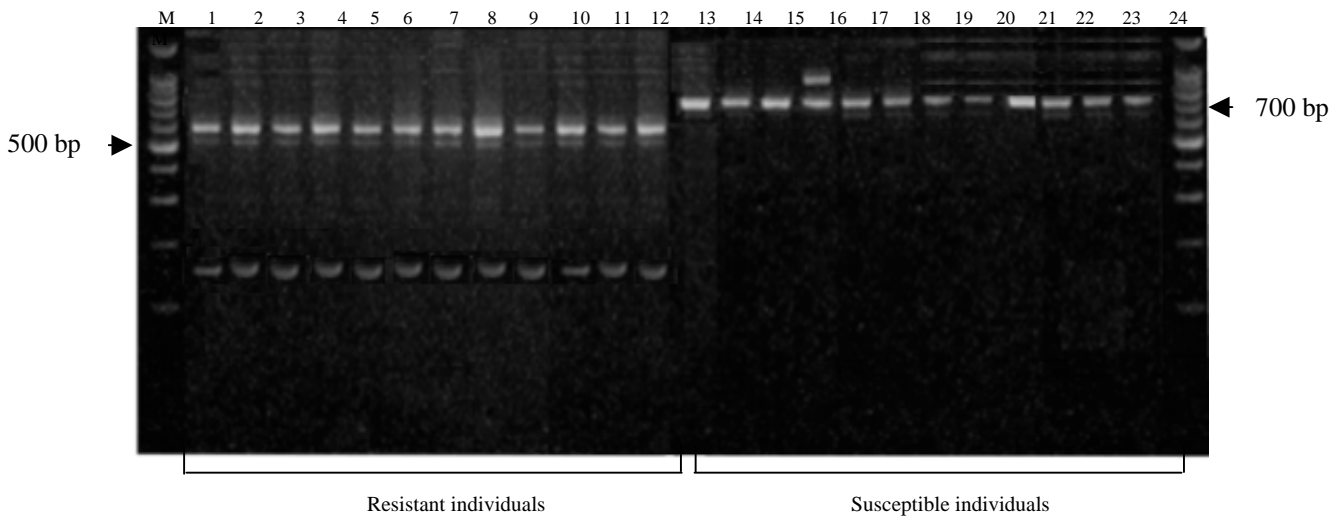
be overcome by using microarrays. The successful application of microarray technology to identify and develop markers associated with *Cryphonectria* canker will form a basis to reduce the impact of the disease.

As part of the Forest Molecular Biology Co-operative (FMBC) which has now come to an end, a DNA microarray chip has been developed for the identification of genomic DNA markers associated with *Cryphonectria* resistance in *Eucalyptus grandis* following a bulked segregant analysis (BSA) approach. Putative microarray markers were identified by comparing tolerant and susceptible bulks in 2-dye experiments on a 384-probe array. BSA revealed a total of 109 scorable polymorphic loci, of which 11 polymorphic features (2.9%) were associated with *Cryphonectria* canker tolerance.

Microarray analysis is still very expensive and not suitable for routine marker-based tagging of individual genes in large

tree populations. To avoid these limitations and to obtain single markers that could be useful in tolerance screening programmes, we converted two of the putative array markers associated with tolerance into PCR-based markers. The two PCR-based markers may provide valuable tools for the accurate and early assessment of tolerance in *E. grandis* trees. However, before these two markers can be used in routine breeding applications, the linkage of the markers to *Cryphonectria* canker tolerance will have to be confirmed in a larger set of phenotyped individuals of the same population, and the population-wide linkage disequilibrium confirmed in other families of *E. grandis*.

This research forms the focus of the PhD project of Ms. Sabine Lezar and was supervised primarily by Prof. B.D. Wingfield, and co-supervised by Dr. A.A. Myburg, Dr. D.K. Berger and Prof. M.J. Wingfield.



Agarose gel showing the PCR marker phenotypes of the *Eucalyptus grandis* individuals screened for *Cryphonectria* canker tolerance and used to perform bulk segregant analysis.

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**THE RESEARCH TEAM OF THE TREE PROTECTION CO-OPERATIVE PROGRAMME**

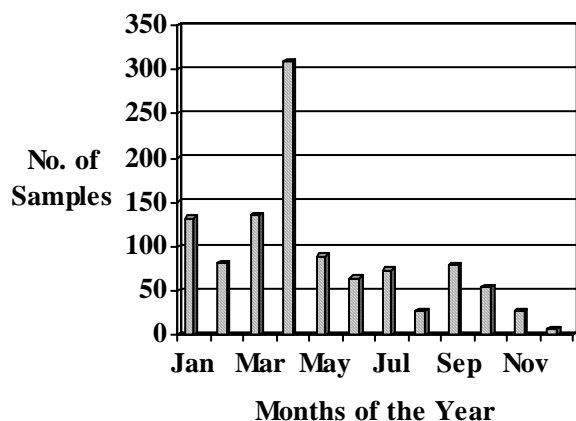
The research team of the Tree Protection Co-operative Programme is varied. It includes full time staff of the University of Pretoria (Prof MJ Wingfield, Director and Mondi Professor, Prof B Wingfield, Prof TA Coutinho, Dr J Roux , Dr P Govender, Brett Hurley, Rosemarie Visser, Sonja de Beer, Eva Muller, Hardus Hatting, Helen Doman, Valentine Nkosi, Martie van Zyl, Pritty Khumalo, Lydia Twalo and Martha Mahlangu). Colleagues and students attached to other organizations such as the ICFR, technical assistants funded by the University or through membership fees and post graduate students who are mainly funded by the NRF. Staff from various Departments in the University provides advice and support where this is required.

**Contributions by:**  
**Teresa Coutinho, Jolanda Roux, Brett Hurley, Irene Barnes, Sabine Lezar**

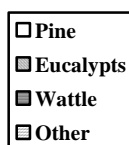
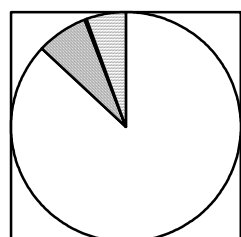
# DIAGNOSTIC CLINIC 2003

The Diagnostic clinic received a total of 1077 samples during 2003. Most samples were received in the first half of the year [Fig. 1]. Of the samples sent, the majority were from pine (87%) [Fig. 2] and more than 50% were research samples. The research samples involved determining the reason why seedlings/cuttings were dying in specific trials which, for example, included testing fungicides and

biological control agents for efficiency against *Fusarium circinatum*. The number of pine samples received in 2003, were almost double those received in 2002. Only 7% of the samples received were from eucalypts and 5% were classified as other. The category "other" included insect, seed, growth media and water samples. Less than 1% of the samples received were from black wattle.



**Fig. 1**  
Number of diagnostic and research samples received (from Jan to Dec 2003)



**Fig. 2**  
Number of samples received from different hosts (from Jan to Dec 2003)

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## TPCP EXPLORES EAST AFRICA A RISK ASSESSMENT

**Species of exotic *Eucalyptus*, *Acacia* and *Pinus* have been grown in Africa for more than 100 years.**

Relatively little information is, however, available regarding the pests and diseases affecting these trees in countries other than South Africa. Most reports of tree diseases in Africa are from the 1960's and 70's. As political stability has returned to countries in East Africa, there has been a

resurgence of interest in commercial plantation forestry in these countries in the last 10 years especially.

Pathogens and pests are often moved across borders with plant and other infected material. In Africa this would be especially easy as people commonly move across borders to find work and visit relatives. It is thus likely that any pest or pathogen that is introduced into one country, will readily make its way to other countries in Africa. One need only to look at the spread of the cypress aphid a few years ago. This devastating insect pest of cypress trees made its way from Europe and North Africa, all the way across the African continent to finally reach South Africa. The spread of pests and pathogens across Africa provides an especially serious threat to countries that rely on large-scale commercial plantation forestry. Knowledge of pests and diseases in

neighbouring countries is important as it provides prior warning of impending threats.

In 2003, the TPCP, under the leadership of Dr. Jolanda Roux, initiated surveys of plantation forestry species in countries north of South Africa. The aims of these surveys were to:

- Establish collaboration with other research institutes, forestry companies and universities Learn more about forestry in other African countries
- Survey *Eucalyptus*, *Pinus* and *Acacia* plantations for diseases
- Survey native Myrtales for *Cryphonectria* spp.
- Investigate fungal infection after bark collection

Two survey trips were undertaken in 2003 using a Landrover Defender partly sponsored by the Department of Trade and Industry (THRIP). The first survey was a two-week trip to Southern Mozambique in August. For the surveys, the



*Ronald Heath (PhD student) and Jolanda Roux searching for Ceratocystis albofundus in Kenya.*

TPCP collaborated with the Mozambique Forestry Research Centre in Maputo. The survey started at the KosiBay border post in the South of the country and surveyed *Eucalyptus* plantations and *Syzygium* trees as far North as Chimoio and Beira. The days consisted of travelling and surveying, while the evenings and late afternoons consisted of the processing of samples and setting up of camp. In order to allow us to complete our work successfully two microscopes and isolation media were included in our supply package. This first, short trip served to prepare the group for their longer trip later in the year.

The second survey trip required five-weeks and extended all the way up to Kenya. Countries visited on this trip included Malawi, Tanzania, Kenya and Zambia. This trip included two stops to work on a British funded project to investigate fungi that infect bark wounds made from harvesting for medicinal purposes. This project is done in collaboration



*TPCP team members with researchers from the Mozambique Forestry Research*

with Dr. Coert Geldenhuis, a forest scientist very well known to foresters in South Africa.

The group visited the Forestry Research Institutes of Malawi, Kenya and Zambia, as well as two private forestry companies in Tanzania, namely the Tanganyika Wattle Company and the Kilombero Valley Teak Company where surveys of *Acacia mearnsii*, *Eucalyptus* and teak trees were conducted. As was true in the case of the Mozambique trip, the group spent days either driving, or surveying plantations and native forests for pathogens. Evenings were spent putting up tents and preparing working areas necessary for processing samples collected during the day. Because of the length of this trip some time was included for relaxation and to do some sight seeing over weekends.



*Camping in the backyard of a hotel in Chimoio, Mozambique.*

The survey trips described here have been highly informative, even though the data collected are of a rather preliminary nature. Yet in only seven weeks (more than 17000km travelled in Mozambique and East Africa), we were able to gain a clear view of forestry in our

neighbouring countries, to meet with foresters and research personnel and to gain knowledge of the most important disease and pest problems present in these countries. From these surveys, and work done by TPCP students from Uganda and Ethiopia for example, it seems that there are no serious pathogens currently threatening South African forestry.



There is, however, a serious insect pest of *Eucalyptus* spp. in Uganda and Kenya, which has moved down from Northern Africa, that needs to be monitored carefully.

It is our hope that we will be able to strengthen ties built with colleagues in other African countries during these surveys. This will allow us to establish an active protection network across Africa and ensure healthy plantations for all forestry organisations and groups on the continent. We would thus welcome suggestions and particularly names of foresters from other African countries. Feed back or comments from South Africans who have worked in other African countries would also be very welcome and will help us greatly in keeping track of forest health problems on the African continent.

*Hard at work, isolating fungi in our field lab*

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# RESTORING THE BALANCE

## Biological Control of Sirex and other Insect Pests in Forestry

**The South African forestry industry is dependant on exotic tree species as a source of fibre and solid wood products.** The most commonly planted trees are species of *Pinus*, *Eucalyptus* and *Acacia*. One of the benefits of growing exotics is the absence of the vast array of herbivores that feed on these trees in their native countries. Some indigenous South African insects have adapted to feed on our exotic forestry species, for example the pine emperor moth and the brown lappet moth, which have adapted to feed on pines. However, our exotic forestry species still have fewer insects feeding on them compared to their native countries and although some of our indigenous insects do become pests on the exotic trees, natural enemies normally limit their density.

A severe pest problem usually arises when an insect that feeds on pines, eucalypts or wattle in the tree's native range, is accidentally introduced into the country. Such accidental introductions are not uncommon and with the increase in the

movement of people and goods between countries, these introductions are likely to increase. Many of the insects accidentally introduced into South Africa are not able to establish and thus are never detected. In contrast, those that do establish in the new environment have the potential to become serious pests as there would not be natural enemies to limit their population increase.

Often, the best way to control exotic pests is by means of biological control. Biological control entails the rearing and release of biological control agents that will maintain the population of the pest at low densities. Various organisms are used as biocontrol agents, including fungi, insects, nematodes, viruses, mammals, etc. Biocontrol agents are generally predators or parasites of their hosts. Biological control aims to restore balance by introducing or enhancing these natural enemies. Of course, not all attempted biological control programmes succeed. The initial phase of a biological control programme involves a considerable investment in finance, effort and time, and then it is still not guaranteed to succeed. However, those biological programmes that do succeed offer a far better alternative to the continued expense and environmental hazard of chemical control

A number of biocontrol agents have been introduced into South African forestry to control exotic pests. These include: the parasitic wasp *Dendrosoter caenopachoides* introduced to control the European bark beetle, *Orthotomicus erosus*; the parasitic wasp *Pauesia* sp. to control the black pine aphid, *Cinara cronartii*; the parasitic wasp *Anaphes nitens* to control the Eucalyptus snout beetle (ESB), *Gonipterus scutellatus*; and the parasitic wasps, *Megarhyssa nortoni* and *Ibalia leucospoides*, and nematode *Beddingia siricidicola*, used to control the sirex woodwasp, *Sirex noctilio*. Prior to

the release of *Pauesia* sp., the black pine aphid was considered the most serious pest on pine in South Africa, but due to the efficacy of this control agent, the black pine aphid is no longer a serious pest. ESB had also become a serious pest throughout South African eucalypt plantations, but due to the introduction of *Anaphes nitens*, ESB is not generally considered a serious pest besides at high altitude sites where the parasite has decreased efficacy.

It is evident that biological control of insect pests in South African forestry has been a crucial component of maintaining tree health and thus a viable forestry industry.

Much research is required to initiate biological control programmes and thus the full support of the forestry industry is needed. Such industry support has been clearly demonstrated in the biological control programme against the siren wood wasp – an exotic insect pest currently posing a serious threat to the pine industry. The introduction of more insect pests into South Africa in the future, some of more concern than siren, is very likely. Only the continued support of and investment into tree protection research will ensure that these introductions do not devastate South Africa's forestry industry.



*The Eucalyptus snout beetle, Gonipterus scutellatus: showing the adult and larval stage and the damage it causes to eucalypts*

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# Snow, wasps and fungi in Sweden

**The TPCP team has made significant contributions to the understanding of the *Sirex notilio* – *Amylostereum areolatum* pest complex in the past.** During the past year, the research and control effort has been substantially increased following the wasps' spread to the Pine plantations of the Eastern Cape and KwaZulu-Natal. As part of this program we are collaborating with the Forest Mycology, Pathology and Entomology departments of the Swedish University of Agricultural Research (SLU). One of the main aims is to characterize this pest complex and its parasites in their native range and compare these data to introduced populations.

This should reveal patterns of introduction, establishment and spread of *Sirex* around the world, which will help to

guard against future introductions, or help with control of introductions that could not be stopped. For example, the specific native origin of the introduced populations of *Sirex*, its symbiont and pests in South Africa (and other Southern Hemisphere countries) is not known. This information is important to select pests that are best adapted to attack the populations of the wasp and fungus present in South Africa and elsewhere.

In order to understand the geographic distribution and spread of *Sirex*, I (Bernard Slippers) am attempting to unravel molecular phylogeographic patterns of the wasp and fungus. This simply means characterization of phylogenetic relationships amongst populations of the pest complex in different locations, including endemic and exotic populations. For this purpose, we are collecting wasps and fungi from different parts of Europe (native), while collaborators are collecting wasps on our behalf in South

America and Australia (both introduced). We also now have access to prior collected fungal collections from Europe and America to compare with collections from South Africa. Currently, however, the focus is on lab work and characterizing already collected populations. New collections in Europe will only happen once the snow melts and it is possible to work outside.

Speaking of this frozen world: My wife and daughter, Jana and Yvonne, and I arrived in Sweden at the beginning of

September 2003. The days were long and the day temperature a wonderful 20 °C or thereabouts. Soon, however, things changed dramatically. Snow started to fall in October and now often covers the ground to 50 cm or more for weeks; days became progressively shorter until Christmas when they were about 6 hours long; and the temperature dropped below 0 °C and hardly ever rose above that level until March, often staying at -15 °C for days. Having said this, everything in Sweden is extremely well geared for the cold and the snow, so life goes on almost as usual. The frozen landscape is also very beautiful, especially when the sun comes out, and so we have really enjoyed this wonderful opportunity. I work mainly with Prof. Jan Stenlid and Dr. Rimvis Vasiliauskas, who have excellent experience, collections and connections regarding *Sirex* work. There have also been ample opportunities to interact with other Forest Pathologists and Entomologist, do relevant courses and to learn new techniques and gain new ideas. So although the landscape is currently frozen, the research is anything but chilly.

[Compiled by Bernard Slippers]



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## ***CONGRATULATIONS***

The TPCP once again had a very successful 6 months since the previous newsletter. We wish to congratulate the following students on obtaining their degrees:

### **PhD:**

**Martin Coetzee:** Molecular phylogenetic studies on *Armillaria* with special reference to southern hemisphere species.

**Eduard Venter:** The molecular characterisation of interaction between *Fusarium circinatum* and *Pinus patula*.

**Schalk van Heerden:** Studies on *Cryphonectria cubensis* in South Africa with special reference to mycovirus infection.

### **MSc:**

**Ronald Heath (Cum Laude):** Studies to consider the possible origin of three canker pathogens of *Eucalyptus* in South Africa.



# SIREX BIOCONTROL PROGRAMME

The sirex woodwasp, *Sirex noctilio*, is an exotic insect pest first detected in South Africa at Tokai plantation, Cape Town in 1994. Since its accidental introduction into South Africa, sirex has spread rapidly and is now well within the pine growing areas of KwaZulu-Natal and moving towards Mpumalanga and onwards. Pine mortality resulting from sirex is increasing and this pest currently poses a serious threat to the pine industry.

A biological control programme has been established to combat the threat of sirex. The biological control agent being used is a parasitic nematode, *Beddingia siricidicola*. This nematode together with two parasitic wasp species were introduced as biocontrol agents previously in South Africa soon after sirex was first detected, but failed to spread with the wasp into the Eastern Cape and KZN. We are now in the process of re-introducing the nematode, the primary biocontrol agent of sirex, on a much larger scale and more continual basis than previous to ensure that it becomes established throughout South Africa, Swaziland and Lesotho. The nematodes are being mass-reared in the millions in the facilities of the Forestry and Agricultural Biotechnology Institute (FABI) and the first releases into the plantations are due this April. These releases will continue on a yearly basis in sirex-infested areas. When established, the nematode can achieve over 90% parasitism rates, thus maintaining sirex density below economically important levels.



Countries in which *Sirex noctilio* occurs as pest of *Pinus* spp.



Top: Hardus Hatting and Pritty Khumalo working on the Sirex Biocontrol Programme

# A SABBATICAL IN BELGIUM FOR OUR CLINIC MANAGER

By Prof. Teresa Coutinho

I was given the opportunity last year to spend three months in the Laboratorium voor Microbiologie, University of Gent, Belgium. My first impression of the Laboratorium was of vibrancy. The atmosphere was very similar to FABI and I felt at home. Academics, students and technicians in the Laboratorium conduct focused research in the broad field of bacteriology. They also have a commercial bacterial culture collection and a diagnostic service available. The topics they cover in the field of bacteriology are broad – from probiotics to identifying lactic acid bacteria found in cheese. The group consists of 60 people, a third are academics and researchers, a third students and a third are technicians linked to the BCCM/LMG collection. I shared a laboratory with a visiting Serbian professor working on lactic acid bacteria and Dr. Marc Vancanneyt, a researcher and my “hands on” collaborator on this project.

The objective of my research was two-fold. Firstly, I attempted to identify and characterise *Pantoea* spp. infecting eucalypts in South Africa and countries where symptoms

I thoroughly enjoyed doing hands-on research again. Having no responsibilities other than getting results was pleasurable. Gaining some experience in molecular techniques has boosted my confidence. I have now learnt how to do techniques I had previously only had an understanding of: REP-PCR and DNA:DNA hybridisations. I was also given opportunities to see other bacterial diagnostic techniques performed. These included FAMES, SDS-PAGE and DGGE.

While at the University of Gent, I attended the PhD defences of two candidates from the Laboratorium voor Microbiologie. I was fortunate to meet with the gurus of bacterial taxonomy who were the external examiners of the two candidates. A “Feest” was held for Professor Monik Gillis to celebrate her retirement from the Laboratorium. It



*Top: Centre of Gent*



*Left: Faculty of Biological Science Building. The laboratory for Microbiology is on the 4<sup>th</sup> floor*

similar to those observed in South Africa have been reported. Preliminary results suggested that more than one member of the Enterobacteriaceae are involved in this disease. Secondly, I attempted to identify and characterise two species of *Pantoea* associated with Coniothyrium canker of eucalypts in South Africa. These two bacterial species are believed to have a synergistic interaction with the fungal pathogen, *Coniothyrium zuluense*.

was in the form of a mini-symposium with invited speakers. I was invited to present two seminars; one entitled: “Coniothyrium canker of eucalypts: a phytopathological enigma” at the Centraal Bureau voor Schimmelcultures (CBS) in Utrecht, the Netherlands on the 5<sup>th</sup> of November 2003 and the other entitled: “South Africa, Forestry and Pathogens” in the Laboratorium on the 26 November 2003.

Belgium is an incredible country to visit. It has two provinces – the Flemish and Wollemia provinces. In Flanders they speak Dutch and its various dialects and in Wollemia, French. The Belgium chocolate was wonderful as were the small percentage I got to taste of the 350 beers produced there.

# Deodar weevil - *Pissodes nemorensis*

The search for bio-control agents of *Pissodes nemorensis* yields some promising results.

Since it was first detected in South Africa in 1942, the deodar weevil, *Pissodes nemorensis*, accidentally introduced from North America, has become widely distributed in all pine growing regions. In recent years, damage by the deodar weevil has led to considerable concern. This is because there has been an increase in attacks by the insect and in some cases these infestations are causing tree death.

Generally *P. nemorensis* is considered to be a secondary pest, having close association with stress factors such as marginal sites, fire and drought. However, the insect has also been observed in many instances attacking vigorously growing young pines in South Africa. In some cases, attack by the weevil following hail damage of young pines has resulted in a need to clear fell entire compartments due to trees failing to recover from the combined effects of hail damage and *P. nemorensis* infestation. In other instances, attraction of the weevil to stressed trees on margins of compartments, mainly due to fire damage during weed control or slash burning, has resulted in healthy trees being attacked. Hence *P. nemorensis* has risen in priority to become an important component of pest protection efforts in South African forests.

Research on *P. nemorensis* started in FABI when Solomon Gebeyehu joined the TPCP team in January 2002 as a Postdoctoral fellow of the University of Pretoria. One of the

focus areas of Solomon's research has been to investigate possible control options to manage *P. nemorensis* in the context of South African forestry practices. This work has included a search for possible bio-control agents. Careful searches and observation, combined with some luck, has resulted in the discovery of insect parasites feeding on *P. nemorensis* in the field. One of these insects is a small wasp belonging to the order Hymenoptera, family Pteromalidae and genus *Pycnetron*. The second insect is a small fly belonging to the order Diptera, family Stratiomyidae and genus *Gobertina*. Neither of these parasites has been reported having associations with *P. nemorensis* in South Africa, and they are not known to occur in its native range. This means that these parasites are most likely indigenous to South Africa, originally feeding on some other insects, but having adapted to feeding on an exotic pest species. Although such a phenomenon is not very common in the insect world, it is certainly a desirable scenario from pest management point of view, especially in checking exotic pest species from reaching outbreak proportions. Perhaps these parasites may be the reason why *P. nemorensis* has not reached outbreak levels in South Africa, despite the fact that it was introduced more than 60 years ago.

Now that potential bio-control agents have been discovered in the field, the next step is to continue research into their efficiency of parasitism and ways of mass-rearing them in the laboratory on artificial media. Solomon has continued his research in FABI to this end, and the future seems to hold some promise for effective biological control of *P. nemorensis* in South African pines plantations.



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## young tpcp researcher shines

Dr. XuDong Zhou, a post-doctoral fellow at FABI, who successfully completed his PhD within the TPCP programme has recently successfully secured a "ChunHui Project" from the Ministry of Education in China. The award is made to excellent overseas Chinese scholars, visiting or working full time or part time in China. Dr. Zhou was also invited to an interview at the University of Tibet in October 2003, and has been

appointed as associate professor and assistant rector (vice rector level) at the university for the period 1 January 2004 to 1 January 2005. Currently, Dr. Zhou also serves as the president of ACSSEA (Association of Chinese Scholars and Students in South Africa), which includes more than 3000 Chinese scholars and students in South Africa.

# WELCOME

The following people have recently joined the TPCP. We wish you a happy and prosperous association with the TPCP.

**Gilbert Kamgan Nkuekam:** Gilbert is originally from Cameroon where he grew up and obtained his BSc degree. He then came to Pretoria where he obtained his honours degree with the Department of Microbiology and Plant Pathology. Gilbert has joined the TPCP for his MSc and will be working on fungi associated with wounds on forestry and other tree species in Africa.

**James Mehl:** James is currently busy with his BSc (Hons) in Plant Pathology after completing his BSc in Microbiology at the University of Pretoria. His honours project is to

determine the sensitivity of a molecular diagnostic method for identifying seedlings infected with *Fusarium circinatum*.

**Joha Grobbelaar:** Joha completed her BSc honours in the department of Genetics at Pretoria University and joined the TPCP for her MSc degree in 2004. For her honours project she looked at mutations in microsatellites in the wilt pathogen *Ceratocystis fimbriata*. For her MSc she will be developing microsatellites for another Ophiostomatoid fungus.

**Magriet van der Nest:** Magriet has re-joined the TPCP for her PhD after completing her MSc in Industrial Biotechnology at the University of Newcastle upon-Tyne in 2003. For her PhD, Magriet will be studying the interaction between pines and *Fusarium circinatum*.

**Dina Gomez:** Dina comes from Colombia and has joined the TPCP for her MSc. She will be working on Ophiostomatoid fungi from various hosts, worldwide.

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## ANNUAL CONGRESS OF THE SOUTHERN AFRICAN SOCIETY FOR PLANT PATHOLOGY

Annually, members of the Southern African Society for Plant Pathology (SASPP) meet in order to exchange scientific knowledge, share ideas and form collaborations. This year, the society met for the 42<sup>nd</sup> time in January at Cathedral Peak Hotel in the picturesque Drakensberg mountains.

A wide range of topics were covered. In total, there were 34 oral papers presented and 28 posters that were on display in the disciplines of disease control strategies, disease surveys in South Africa, disease resistance and breeding, cytology, histology and mycotoxins, pathogen identification and characterisation, disease detection and losses, genetics and research opportunities. The TPCP group from FABI contributed significantly to the congress presenting 14 papers and 3 posters, mostly in the area of pathogen identification and characterisation. All the presentations were well received. The conference also presented us with a great opportunity to see what is happening in the broader context of plant pathology in South Africa and to learn more about the different research programmes in the country. There was also a short lecture on the do's and don't of poster making. This was beneficial to a number of members as this

is one of the more difficult mediums of scientific communication, but one that is used quite extensively at international meetings.

Once again, FABI's participation in the meeting was very successful with Lerato Matsaunyane (MSc student of David Berger) winning the best paper presentation for her talk entitled "Characterisation of apple polygalacturonase-inhibiting protein 2. At the gala dinner, Marieka Gryzenhout and Irene Barnes received the 'Young Plant Pathologist



*Irene Barnes, Prof. Z.A. Pretorius and Marieka Greyzenhout at the gala dinner, after receiving their awards.*

Award' and Irene Barnes was also awarded the student travel grant donated by CS Africa.

On the social side, as is tradition, the Mildenhall stakes were fiercely contended in the male, female and over 50ties categories. The art of balancing a beer on ones head while trying to do a backward summersault presented itself to be a particularly difficult scientific technique that has only been mastered by a rare few. Readers of Tree Protection News that have attended the annual meetings of the TPCP and have witnessed this exercise will understand this well. At the SASPP meeting, we

had the Guru himself, Professor J.P. Mildenhall demonstrating the technique of suppleness and control (not to be forgotten is the fact that he is over sixty years old!). While the ladies event was again a landslide victory for Jolanda Roux who won with ease and agility, it proved to be a bit more challenging for the males. A bit of morning horse riding and evening walking in the beautiful settings of the mountains rounded off a very successful and enjoyable conference.



Top: Maria-Noel Cortinas (left, standing) and Jolanda Roux (right, sitting) competing in the womens event.  
Left: Prof. John Mildenhall demonstrating the "Mildenhall Stakes" technique,

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## ABSTRACTS FROM RECENT SASPP CONGRESS

### **DNA based phylogeny of *Dothistroma septospora* reveals a species partition separating North American isolates.**

*Dothistroma septospora* is a needle pathogen of many pine species and causes serious defoliation of trees in many parts of the world. The pathogen is thought to have originated from the subtropical, high-altitude cloud forests of Central America, but now has a worldwide distribution. It is particularly well known in countries of the southern hemisphere that plant *Pinus radiata* and where severe damage due to needle blight has occurred. Three different varieties of this pathogen have been described based on differences in the average length of the mitospores. There is, however,

debate as to whether spore size represents an adequate characteristic to distinguish between forms of *D. septospora*. The aim of this investigation was to consider phylogenetic relationships between *D. septospora* isolates from different countries and to determine whether the separation of this fungus into different varieties deserves support. Isolates from 11 different countries representing five continents were chosen for this study. Three DNA regions including the ITS,  $\beta$ - tubulin gene and elongation factor 1-alpha gene were sequenced and analysed. In general, the isolates from different countries had very similar sequences for all the DNA regions studied and no obvious groups based on origin were observed. However, isolates from the U.S.A., were distinctly separated in their own clade. This partition separating the North American isolates from all

others could indicate the presences of a discrete species. Results further show that there is no phylogenetic support for the separation of *D. septospora* into three different varieties.

### **Genetic comparison of *Cryphonectria cubensis* isolates from native and exotic hosts in South Africa**

*Cryphonectria cubensis* is one of the most important canker pathogens of *Eucalyptus* trees in tropical and sub-tropical regions. Recent studies have shown that *C. cubensis* in South Africa is distinct from the fungus of the same name, occurring elsewhere. This and the recent discovery of *C. cubensis* on native *Syzygium* spp. has led to suggestions that the South African form of *C. cubensis* is native to this country. The purpose of this study was to compare the genetic structure of South African isolates of *C. cubensis* from native *Syzygium* spp. (62 isolates) to those from exotic *Eucalyptus* (34) and *Tibouchina* (37) spp., using vegetative compatibility tests and polymorphic DNA (SSR) markers. The *Tibouchina* population had the highest gene diversity (H) (0.57) followed by the *Eucalyptus* population (0.43) and the *Syzygium* population (0.17). The populations from the two exotic hosts also produced the highest genotypic diversity (33% and 45% respectively) compared to that of the *Syzygium* population (5%). Low levels of gene flow were observed between the *Syzygium/Tibouchina* and the *Eucalyptus/Tibouchina* populations, compared to the levels of geneflow present between the *Syzygium/Eucalyptus* populations. This corresponded with the mode of reproduction with geneflow only occurring between the two sexually reproducing *Eucalyptus* and *Syzygium* populations. There were more unique genotypes amongst isolates from *Eucalyptus* and *Tibouchina*, than from the native *Syzygium* spp. Results indicated that the two exotic hosts have been colonised more recently than the native host, and possibly by isolates originating from the native *Syzygium* spp. The high

level of gene diversity also suggests that the fungus is native to this country, in contrast to previous suggestions that it was introduced from Asia.

### **Diseases of plantation forestry species in Southern and Eastern Africa**

Plantations of exotic tree species such as *Eucalyptus*, *Pinus* and *Acacia* spp. have been established throughout Southern and East Africa. These trees provide valuable alternative sources of timber for construction, furniture and fuel wood, thus reducing the negative impact on rapidly dwindling native tree species. Other than in South Africa, the last comprehensive disease surveys on these tree species were conducted ~ 30 years ago. In order to gain a perspective of the current disease situation on these trees, surveys were undertaken in Mozambique, Malawi, Tanzania, Kenya, Ethiopia and Zambia. Diseases were identified based on symptoms, morphology of pathogens collected, studies in pure culture and using comparisons of DNA sequence data. The most commonly observed disease of *Eucalyptus* spp. was *Botryosphaeria* canker, caused by a number of *Botryosphaeria* spp. Other diseases included *Cryphonectria* canker, *Coniothyrium* canker, *Armillaria* root rot and *Mycosphaerella* leaf blotch disease. On *Pinus* spp., *Diplodia* canker and die-back, *Armillaria* root rot and *Dothistroma* needle blight were identified, while on *A. meurnsii* the most commonly encountered disease was canker and wilt caused by *Ceratocystis albofundus*. A number of these diseases represent new reports from the countries in which they were collected. The fact that they have not previously been recognised in these environments has important implications for quarantine and should now provide a basis to exclude them from countries that are currently free of them. Government quarantine organizations, forestry companies and researchers in these African countries are encouraged to develop strong collaborative alliances to ensure the future health and sustainability of plantation forestry in this region.

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## **IMPORTANT : PLEASE READ THIS**

In order for us to co-ordinate our services to you please help us by using the following contact address:

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