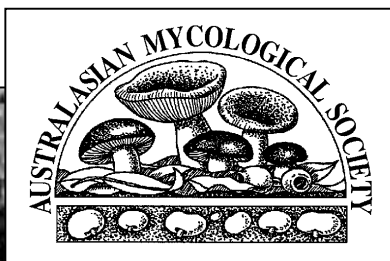


**SCIENTIFIC MEETING OF THE AUSTRALASIAN  
MYCOLOGICAL SOCIETY  
BRISBANE 21-23 APRIL 2014**

**PROGRAM AND ABSTRACT BOOK**



## Scientific Meeting of the Australasian Mycological Society 21-23<sup>rd</sup> April 2014

Welcome to Brisbane and the **2014 Australasian Mycological Society Scientific Meeting**. We are meeting back-to-back with the Australian Fungimap Society conference in order to encourage significant cross pollination (or should that be spore dispersal!) between the two groups resulting in stronger societal linkages. The AMS scientific program includes two outstanding plenary speakers in Tania Sorrell, Director of the Sydney Institute of Emerging Infectious Diseases and Biosecurity and Roy Halling from the New York Botanical Garden. The program covers a wide variety of mycological themes, from infectious fungal diseases, molecular mycology and plant pathology to fungal ecology, systematics and applied mycology. We hope you find these sessions interesting, beneficial and enjoyable. We also hope you take the opportunity to reacquaint with colleagues and form new research linkages. We invite you to conference drinks from 5pm in the Ecosciences Precinct courtyard on Tuesday 22<sup>nd</sup> April and the conference dinner at the iconic “Jetty Restaurant” overlooking the Brisbane River and CBD on the evening of Wednesday 23<sup>rd</sup> April. We would particularly like to thank the co-operation of Ecosciences Precinct for providing an exceptional venue for this meeting.

**Diana Leemon**  
**John Dearnaley**  
**Sandra Abell-Davis**  
**Nigel Fechner**  
**James Fraser**  
**Peter McGee**  
**Richie Robinson**

*AMS 2014 conference committee*



Cover design by Peter McGee (negative image of fungal mycelia growing into leaf litter)

## Monday 21<sup>st</sup> April

1200-2000 **Field trip to Anstead Bushland Reserve**

Meet at 1200 at Diana Plaza foyer, Woolloongabba

## Tuesday 22<sup>nd</sup> April

0800 **Registration desk open** Outside Room GA604 Ecosciences Precinct

0900 **Welcome and Introduction:** President Diana Leemon Room GA604

0910 **Invited Plenary Talk I** – Professor Tania Sorrell, Director of the Sydney Institute of Emerging Infectious Diseases and Biosecurity. *Cryptococcus neoformans* – a designer fungus with serious intent

1000 **Morning Tea**

### 1030 **Symposium I: Infectious Fungal Diseases** Room GA604

**Chair: James Fraser**

**Presentation Title**

Monica Slavin	25 min	New approaches to guiding antifungal therapy
Leona Campbell	25 min	<i>Cryptococcus</i> strains with different pathogenic profiles secrete diverse protein cohorts
Ana Traven	25 min	<i>Candida albicans</i> and the innate immune response: understanding how <i>Candida</i> causes macrophage cell death
Asa Perez-Bercoff	25 min	Genome sequencing, assembly and annotation of <i>Scedosporium aurantiacum</i>

1210 **Lunch** (60 min)

1310 Symposium II: Molecular Mycology Room GA604

**Chair: Ana Traven**

**Presentation Title**

Julie Djordjevic	25 min	Contribution of a novel branch of phospholipase C1-mediated signalling, via inositol polyphosphate kinases, to fungal pathogenesis
Kylie Boyce	25 min	Tyrosine catabolism and pyomelanin production in the human pathogen <i>Penicillium marneffe</i>
Mark Wilkins	25 min	Building the protein methylation network of <i>Saccharomyces cerevisiae</i>
Anthony Borneman	25 min	Characterisation of intra-specific genomic diversity in industrial yeasts by whole-genome sequencing

1450 **Afternoon Tea/ Posters** (30 min)

1520 Symposium III: Plant Pathogenesis Room GA604

**Chair: Liz Aitken**

**Presentation Title**

Don Gardiner	25 min	How does <i>Fusarium</i> deal with the chemical defences of wheat
Liz Dann	25 min	Making the most of fungal pathogenesis
Sue Thompson	25 min	When dead is alive - a complex of <i>Diaporthe</i> species identified from broad-acre crop and weed residues
Celeste Linde	25 min	Effect of a weedy host on pathogen evolution

1700 **Conference Drinks** – *Ecosciences Precinct courtyard*

**Wednesday 23<sup>rd</sup> April**

0800 **Registration desk open** Outside Room GA604 Ecosciences Precinct

0855 **Welcome and Housekeeping:** President Diana Leemon Room GA604

**0900 Symposium IV: Proffered papers** Room GA604

**Chair: Sandra Abell-Davis/Richie Robinson**

**Presentation Title**

Susan Nuske	20 min	Ecosystem services of mycophagous mammals
Tom May	20 min	Australian fungi have wide distributions that are explained by climate
Celeste Linde	20 min	Extreme mycorrhizal specificity in <i>Chiloglottis</i> , <i>Drakaea</i> and <i>Paracaleana</i> orchids
Greg Bonito	20min	The root myco-biome: disentangling the influence of host and soil on fungal community structure
Katharina Schwabenbauer	20 min	<i>Scedosporium aurantiacum</i> – an emerging fungal pathogen with extensive genetic diversity

1040 **Morning tea** (30 min)

**1110 Symposium V: Systematics** Room GA604

**Chair: Nigel Fechner**

**Presentation Title**

Morwenna Boddington	25 min	New Russulaceae species in SE Qld
Rachel Mapperson	25 min	Molecular taxonomy of Australian endophytic Pezizales
Jeff Powell	25 min	Towards a trait-based ecology of fungi
Laszlo Irinyi	25 min	Quality controlled ITS database for human and animal pathogenic fungi

**1250 Lunch** (60 min)

**1350 Symposium VI: Applied Mycology Room GA604**

**Chair: Diana Leemon**

**Presentation Title**

Lisa Guilino	25 min	Gut inhabiting anaerobic fungi
Lesley Francis	25 min	Resistance of naturally-durable timbers to fungal decay
Vic Galea	25 min	Using fungi as bioherbicides for woody weeds
Richie Robinson	25 min	Comparing molecular and morphological species concepts for <i>Cortinarius</i> from long-term monitoring in Western Australia

1530 **Afternoon Tea/Posters (30 min)**

**1600 Symposium VII: Fungi & Restoration Room GA604**

**Chair: Peter McGee**

**Presentation Title**

Jess Mowle	20 min	Improving the recovery outcomes of the critically endangered Wollemi pine: is success determined by soil microbes?
Cathal Danes	20 min	Arbuscular mycorrhizal fungi actuate the aggregation of mine spoil
Ash Martin	20 min	Working with mycorrhizal fungi in the field
Tendo Mukasa Mugerwa	20 min	Some melanised root-associated fungi may increase organic carbon in soil
Kate Newman	20 min	Can inoculation with fungi improve the rehabilitation of a mine site?

1745 **Invited Plenary Talk II** – Dr Roy Halling, New York Botanical Garden  
Location, Location, Location: Input from Boletography

1835 **Closing Address and Prizes**

1930 **Conference dinner at Jetty Restaurant, South Bank**

### ***Cryptococcus neoformans* – a designer fungus with serious intent**

Sorrell TC

*Marie Bashir Institute for Infectious Diseases and Biosecurity, University of Sydney*

**Abstract.** Cryptococcosis is primarily a disease of the lung and central nervous system and is the commonest cause of fungal meningitis, which is fatal if untreated. Pathogenic species in the *Cryptococcus* complex, *C. neoformans* and *C. gattii*, have distinctive ecological niches, biochemical pathways, host preferences and clinical manifestations, despite the fact that they produce the same major virulence factors. These virulence determinants include the polysaccharide capsule, melanin, ability to grow at physiological temperature and the enzymes, phospholipase B and urease. Understanding the pathogenesis of lung infection and especially of central nervous system infection, and the respective roles of the pathogen and the host response in disease pathogenesis, is critical to developing new therapeutic strategies and/or preventing severe sequelae. Gene deletion and reconstitution experiments have shown that both phospholipase B and urease are essential for *C. neoformans* to cross the blood brain barrier and cause neurological disease, but it is still debated as to whether transmigration occurs as free cryptococci or within phagocytes via a Trojan horse mechanism in initial infection. There is direct evidence for the former, but only indirect evidence for the latter. Furthermore there is emerging evidence that *C. neoformans* and *C. gattii* may differ in their predilection to cause neurological disease and that within the species *C. gattii*, this may be genotype-dependent and influenced by host factors. This presentation will place new evidence regarding disease pathogenesis in the context of the concept of the fungal virulence composite, the contribution of genomics and of the host response.

## Plenary talk II

### Location, Location, Location: Input from Boletography

Halling R

*Institute of Systematic Botany, New York Botanical Gardens, USA: rhalling@nybg.org*

**Abstract.** Distribution patterns of macrofungi have been postulated by several different methods. Up until the mid to late 20<sup>th</sup> century, if a fungus had been observed in one or more sites, it was thought to truly occur in those places. Of course, macro- and then, micromorphological congruence were the traditional observational tools for lending support to those hypotheses. After all, airborne spores were produced and dispersed around the globe in wind currents. Hypothetically then, everything could be everywhere. Bringing living fungi to the laboratory and manipulating them on various growth media spawned investigations in sexual compatibility. The confrontation of viable, haploid single spore isolates in all possible combinations became a technique to assess whether or not a flourishing dikaryon could be produced. By extension then, if this “mating” worked, the two (or more isolates) were sexually compatible and belonged to the same species regardless of origin. The saprobic lifestyles of many fungi were quite amenable to such manipulation. Not so with obligate mycorrhizal fungi such as bolete mushrooms. If it looked the same, it was the same despite population location and geographical isolation. With the refinement of techniques to isolate and compare DNA sequences, new analyses were employed to infer degrees of similarity and difference. Such tools have helped clarify taxonomic relationships and global distribution of boletes.



## Symposium I: Infectious Fungal Diseases

**Chair: James Fraser**

### **New approaches to guiding antifungal therapy**

Slavin MA

Peter MacCallum Cancer Centre and Victorian Infectious Disease Service, Royal Melbourne Hospital, at the Peter Doherty Institute for Infection and Immunity, Parkville, Victoria, AU.

**Abstract.** Invasive fungal infections remain a leading cause of mortality in patients with haematological malignancies. Risk groups for fungal infection are expanding with the introduction of new therapies. Traditional approaches to reducing the incidence and mortality of these infections such as antifungal prophylaxis and empiric antifungal therapy have had limited success in reducing mortality. Most antifungal usage in hospitals is empiric and is associated with cost and toxicity. Recently non-culture tests, such as *Aspergillus* galactomannan and PCR, have been evaluated for surveillance and directing early treatment of fungal infection in both serum and bronchoalveolar lavage fluid. However, local epidemiology and prevalence of fungal infection, impact of construction and building works, and the availability of diagnostic testing need to be considered in a decision as to whether prophylaxis, screening or a diagnostic driven approach is justified. New imaging techniques such as PET scan are also useful. Personalised approaches to quantifying risk for infection are under investigation including genetic testing, immunological profiling and evaluation of the microbiome.

**Keywords.** antifungals, diagnostics, fungal infection.

## ***Cryptococcus* strains with different pathogenic profiles secrete diverse protein cohorts**

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**Abstract.** Closely related strains of *Cryptococcus gattii* and *C. neoformans* exhibit different degrees of pathogenesis in the mammalian host. As the fungal secretome is likely to be important in modulating host-pathogen interactions, the secretomes were determined for a hypovirulent and hypervirulent strain of *C. gattii* and a virulent strain of *C. neoformans* when grown under *in vivo* mimicking conditions. The three strains secreted significantly different protein cohorts. In total 67 proteins were identified with different numbers of proteins secreted by each strain. Only one protein was secreted by all strains, a putative glycosyl hydrolase. The secretomes of the virulent strains were limited and included uncharacterized proteins containing catalytic regions involved in carbohydrate degradation. The hypovirulent strain secreted a more diverse set of proteins including many canonical cytosolic proteins. Some enzymes secreted by the hypovirulent strain were immunogenic, eg. enolase, a known fungal allergen that binds IgE. These findings indicate that virulence and the secretome are linked in *Cryptococcus*. The limited protein cohorts secreted by the virulent strains suggests they may better avoid detection once within the mammalian host lung. The allergenic proteins secreted by the hypovirulent strain suggest it triggers a more effective immune response, leading to clearance of the pathogen.

**Keywords.** *Cryptococcus*, secretome, virulence.

# ***Candida albicans* and the innate immune response: understanding how *Candida* causes macrophage cell death**

Uwamahoro N<sup>1</sup>, Verma-Gaur J<sup>1</sup>, Shen H-H<sup>2</sup>, Qu Y<sup>1,2</sup>, Lewis R<sup>3</sup>, Vince JE<sup>3</sup>, Naderer T<sup>1</sup> &  
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**Abstract.** Phagocytosis by macrophages is an important component of the innate immune response to *Candida albicans*. *Candida* can counteract by causing macrophage cell death and escaping. It is known that the morphological switch of *C. albicans* from yeast to hyphae is important for macrophage killing, but the mechanistic reasons for this remained elusive. The current model is that the highly polarized growth of hyphae causes mechanical destruction of macrophages. We have devised a new time-lapse microscopy assay to monitor the interaction of *C. albicans* with macrophages, and used it to show that *Candida* kills macrophages in two temporally and mechanistically distinct phases. Early post phagocytosis, *C. albicans* triggers pyroptosis, a proinflammatory macrophage programmed cell death. Hyphal morphogenesis is important for triggering pyroptosis. In the absence of the pyroptotic caspases 1 and 11, macrophage killing by *C. albicans* is significantly impaired, despite the formation of hyphal filaments, showing that mechanical destruction of macrophages by hyphae is not the sole mechanism of macrophage killing. We identified a *C. albicans* mutant that formed hyphae of wild type morphology, but was impaired in causing early macrophage death. The mutant hyphal filaments showed a breakdown of surface architecture as assayed by atomic force microscopy, and lower levels of exposed 1,3  $\beta$ -glucan on the hyphal cell wall. These results implicate proper hyphal cell wall organization in triggering early macrophage death. The second phase of macrophage killing starts at around 8 hours post phagocytosis under our experimental conditions. This phase requires robust hyphal filament formation, but not the pyroptotic caspases, showing it is mechanistically distinct from pyroptosis. We propose that the model for how *C. albicans* causes macrophage cell death should be revised to include a new key function of hyphal structures: hyphae are necessary for triggering pyroptotic macrophage cell death. A key question now is what the consequence of pyroptosis is in the context of disease. Pyroptosis might serve to augment the inflammatory response to *C. albicans* infection, but the pathogen might hijack pyroptosis to escape and thus evade immune destruction.

# Genome sequencing, assembly and annotation of *Scedosporium aurantiacum*

Pérez-Bercoff A<sup>1</sup>, Ramsperger M<sup>2</sup>, Huttley GA<sup>1</sup> & Meyer W<sup>2</sup>

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**Abstract.** *Scedosporium aurantiacum* is an emerging pathogen of humans and animals. It is commonly present in urban environments. Animal studies, using the *Galleria mellonella* model, have revealed virulence differences among strains. To establish a molecular basis for future studies to understand the origin of those differences, we have sequenced and assembled for the first time the genome of *S. aurantiacum* strain WM 09.24 using different genome assemblers. To project the genome annotation and develop gene models we performed whole-genome alignment with the next closest species for which an annotated genome was available, *Trichoderma virens*. However, this species is still so distant that only approximately 30 % of the two genomes could be aligned through whole-genome alignment. Therefore, neither direct-gene projection nor BLAST searches could be applied in order to annotate the *S. aurantiacum* genome. To overcome this problem we performed RNA-sequencing of strain WM 09.24 under different growth conditions (to maximise the number of transcribed genes), and used the JAMg pipeline to annotate the genome. The annotated genome of strain WM 09.24 was then used to annotate three additional *S. aurantiacum* strains with varying virulence to enable a whole-genome comparison to identify genes or genetic signatures that are associated with virulence.

**Keywords.** *Scedosporium aurantiacum*, genome assembly and genome annotation.

**Chair: Ana Traven**

**Contribution of a novel branch of phospholipase C1-mediated signalling, via inositol polyphosphate kinases, to fungal pathogenesis**

Djordjevic JT<sup>1</sup>, Lev S<sup>1</sup>, Li C<sup>1</sup>, Desmarini D, Chayakulkeeree M<sup>2</sup>, Traven A<sup>3</sup>, Sorrell TC<sup>1,4</sup> & Saiardi A<sup>5</sup>

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**Abstract.** We established that fungal phospholipase C1 (Plc1) is crucial for cellular homeostasis and virulence of *Cryptococcus neoformans* (*Cn*) and now investigate the mechanism of *Cn*Plc1-mediated signalling, and whether signalling components downstream of *Cn*Plc1, contribute to the *Cn* virulence composite. We established that recombinant *Cn*Plc1 is similar to the mammalian PLC $\delta$  isoform in that it hydrolyses phosphatidylinositol 4,5-bisphosphate (PIP<sub>2</sub>) to produce inositol trisphosphate (IP<sub>3</sub>) in a radiometric enzyme assay. However, rather than activate calcineurin as in mammalian cells, *Cn*Plc1-derived IP<sub>3</sub> was phosphorylated by an inositol polyphosphate kinase (IPK) called Arg1/Arg82. By comparing the inositol polyphosphate (IP) profiles of the *Cn*IPK deletion mutants,  $\Delta$ *arg1* and  $\Delta$ *kcs1*, using HPLC, we demonstrated that Arg1 (the major IP<sub>3</sub> kinase in *Cn*) and Kcs1, another IPK predicted to function downstream of Arg1 as an IP<sub>6</sub> kinase, convert IP<sub>3</sub> to (IP)<sub>4-5</sub>, and IP<sub>6</sub> to the inositol pyrophosphates (PP-IP)<sub>7-8</sub>, respectively. Loss of Arg1 and Kcs1 function in  $\Delta$ *arg1* and  $\Delta$ *kcs1*, respectively, had a pleiotropic effect on the virulence composite, with virulence attenuation in  $\Delta$ *arg1* being more severe than in  $\Delta$ *kcs1*.  $\Delta$ *arg1* and  $\Delta$ *kcs1* were avirulent and hypovirulent, respectively, in a mouse inhalation model.  $\Delta$ *arg1* lung infection was cleared by 30 days post-infection, and while  $\Delta$ *kcs1* lung infection persisted for up to 50 days post-infection, no dissemination to the brain was observed.  $\Delta$ *kcs1* hypovirulence correlated with reduced uptake of  $\Delta$ *kcs1* by THP-1 and primary monocytes, and reduced monocyte activation, compared to WT. Our findings show that (1) a key biochemical function of *Cn*Plc1 is provision of IP<sub>3</sub> for synthesis of complex IPs, (2) *Cn*IPK enzymes functioning downstream of *Cn*Plc1 are essential for *Cn* virulence and (3) *Cn*IPKs, particularly Arg1, are candidate antifungal drug targets. Phenotypic comparison of the remaining IPK mutants in the *Cn*Plc1-IPK pathway is currently being undertaken, and will enable determination of the relative contribution of IPs and PP-IPs to the *Cn* virulence composite.

**Keywords.** *Cryptococcus neoformans*, pathogenesis, signalling.

# Tyrosine catabolism and pyomelanin production in the human pathogen *Penicillium marneffe*

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**Abstract.** For pathogens to successfully infect a host, two important events must be achieved; the pathogen must evade the host's defence systems and it must be able to utilise the available nutrient sources within the host in order to grow. These factors are significant hurdles for most pathogens, but especially so for intracellular pathogens which have to tolerate the cytotoxic machinery of innate immune cells and scavenge nutrients from this relatively nutrient poor environment. In a screen aimed at identifying pathogenicity genes in the dimorphic fungus *Penicillium marneffe*, a number of metabolic genes were found which are specifically expressed in the pathogenic yeast cells and not in the saprophytic hyphal cells. One of these genes, designated *hpdA*, encodes 4-hydroxyphenylpyruvate dioxygenase (4HPPD) which catalyses the conversion of 4-hydroxyphenylpyruvate to 2,5-dihydroxyphenylacetate (homogentisate), a step in the tyrosine catabolic pathway. Tyrosine is catabolized via a conserved pathway to provide the fungus with both nitrogen and carbon. In addition, the oxidation and polymerization of a tyrosine metabolic intermediate, homogentisate, can generate the brown pigment pyomelanin which can protect against oxidative stress. This study describes the deletion of tyrosine catabolism genes and the characterization of their role in growth, pyomelanin production and pathogenicity in *P. marneffe*.

**Keywords.** melanin, pathogenicity, tyrosine catabolism.

# Building the protein methylation network of *Saccharomyces cerevisiae*

Wilkins MR

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**Abstract.** Methylation of proteins occurs predominantly on arginine and lysine residues in the eukaryotic cell. Until recently, its predominance was unknown and its role obscure. This presentation outlines our efforts to construct the first ‘protein methylation network’ for a eukaryotic cell and presents evidence that arginine methylation can modulate protein-protein interactions in this network. We analysed the yeast methylproteome to identify methylated proteins and precise modification sites. Immonium ion-based scanning and targeted data acquisition - electron transfer dissociation LC-MS/MS was used, as were yeast proteome arrays (containing 4,400 chips spotted on to microscope slides). This showed that protein methylation is widespread in the eukaryotic cell. To build the intracellular methylation network, all known and putative methyltransferases in yeast were knocked out and the methylproteome re-analysed to determine which enzyme was responsible for which methylation event. This led to the discovery of a two new lysine methyltransferases, one which we have named Efm2. Enzyme-substrate links were further investigated by the analysis of recombinant substrate proteins methylated by recombinant enzymes, by *in vivo* methylation assays and/or the incubation of proteome arrays with recombinant enzymes. Validated enzyme-substrate links were integrated with the yeast protein-protein interaction network to generate the first ‘methylproteome network’. Interestingly, this suggested that many protein-protein interactions could be controlled by protein methylation. To test this, we constructed a new ‘conditional’ two-hybrid (C2H) system. Interactions of proteins were tested in the presence of a methyltransferase or in the presence of the same enzyme with active site knocked out. Of all the protein-protein pairs involving arginine methylated proteins, half of those tested to date have shown dramatic increases in interaction in association with methylation. The functional implications of this will be discussed.

# Characterisation of intra-specific genomic diversity in industrial yeasts by whole-genome sequencing

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**Abstract.** Industrial yeasts such as those of the genus *Saccharomyces* that are involved in winemaking, brewing and pharmaceutical industries, represent a diverse collection of species and strains that have been selected for their ability to perform specific biochemical transformations despite exposure to osmotic, nutrient and ethanol stress. However, in addition to these beneficial yeast species, there are common industrial spoilage yeasts, such as the *Dekkera bruxellensis*, that also thrive under the stressful conditions of an industrial fermentation while producing metabolites with detrimental effects on the industrial fermentation process. In many cases, the phenotype of industrial microbes is highly variable across strains of the same species, with individual strains displaying a broad range of desirable, and undesirable characteristics. By understanding the genetic basis of these phenotypic differences, it will be possible to maximise the desirable characteristics within a strain while minimizing potential undesirable characters. We have therefore applied next-generation genome sequencing and comparative genomics to catalogue the variation present across strains of the yeasts *S. cerevisiae* and *D. bruxellensis*. In each case, genomic data have uncovered a significant pool of genetic diversity within each species. Individual strains were shown to contain large amounts of nucleotide variation (SNPs), while also displaying significant differences in gene content due to the presence of large deletions and strain-specific insertions of novel genes. This genomic data will be combined with transcriptomic, proteomic and metabolomic information in order to associate phenotypic diversity with specific genomic variation and to allow for predictions to be made regarding how the introduction of genomic variation will impact upon specific industrial traits in these and other important industrial species.



## Symposium III: Plant pathogenesis

Chair: Liz Aitken

### How does *Fusarium* deal with the chemical defences of wheat?

Gardiner DM<sup>1</sup>, Kazan K<sup>1</sup>, Kettle AJ<sup>1,2</sup>, Batley J<sup>2</sup>, Stephens AE<sup>1</sup>, Munn AL<sup>3</sup> & Manners JM<sup>4</sup>

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<sup>4</sup>CSIRO Plant Industry, Black Mountain, Canberra, ACT, AU

**Abstract.** Plant-pathogen interactions are highly dynamic processes that involve complex defence and anti-defence strategies employed by both partners. Host plants deploy a battery of inducible defences to protect themselves from pathogen attack while pathogens have evolved to overcome these defences to successfully colonize their hosts. We are interested in diseases of wheat and barley caused by *Fusarium* species such as *F. graminearum* and *F. pseudograminearum*. Through genomic analyses of these pathogens we have recently discovered that *Fusarium* pathogens have evolved mechanisms, including via horizontal gene transfer, to deal with small molecule compounds that are likely part of the hosts' chemical weaponry. Inactivation of these pathogen genes often results in reduced virulence towards wheat and/or barley. Specific examples will be discussed, including a *Fusarium* gene cluster that detoxifies a major class of phytoalexins found in wheat, rye and maize and also an ABC transporter that exports an unknown plant defence compound. These findings strongly suggest a great deal is yet to be learned about both fungal virulence and the chemical defences of our major crops and pathogenomics offers invaluable tools for understanding the mechanisms involved plant-microbe interactions.

**Keywords.** phytoalexin, detoxification, crown rot, head blight.

## Making the most of plant pathogenesis

Dann EK

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**Abstract.** Plant pathogenesis may be defined as “the sequence or processes in disease development from the time of infection to the final reaction in the host”. Plants have developed an arsenal of constitutive and inducible defences deployed during pathogenesis to protect them against pathogens. These responses, which include multi-gene activation, biochemical and structural defences, can also be activated by non-pathogenic chemical, physical or biological agents, in a phenomenon known as systemic acquired resistance (SAR). SAR is being utilised in several commercial crop protection programmes to reduce losses caused by diseases. Research by our group over the last several years has demonstrated 1) activation of pathogenesis-related (PR) proteins associated with the resistance shown by pea to *Ascochyta* blight caused by *Mycosphaerella pinodes*, 2) the involvement of PR proteins and cellular fortifications in the response of cotton roots to inoculation with the *Fusarium* wilt pathogen *Fusarium oxysporum* f. sp. *vasinfectum*, and 3) a role for alk(en)ylresorcinol compounds in the defence of mango to anthracnose disease caused by the *Colletotrichum gloeosporioides* complex. While the above examples concern responses conferring resistance to fungal pathogens, SAR is broad spectrum, and protection against bacterial, viral and nematode diseases can also be achieved.

**Keywords.** disease resistance, pathogenesis, integrated disease management.

## When dead is alive - a complex of *Diaporthe* species identified from broad-acre crop and weed residues

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**Abstract.** Herbicides have been favoured for weed control in broad-acre zero and minimum tillage systems for more than twenty years with soil moisture retention, decreased compaction and cost advantages the main benefits. Low tillage cropping systems which rely on herbicides also retain crop and weed stubble on the surface enabling colonisation of this stubble by many fungal species. As well as emerging herbicide resistant weeds, of concern is the increasing incidence of stubble borne fungal pathogens which survive in the unincorporated plant residues. *Diaporthe* (syn. *Phomopsis*) spp. are well known saprobes, endophytes and pathogens responsible for damaging stem cankers on crops such as sunflower, soybeans, mungbeans and lupins. In an ongoing study, more than twenty previously undescribed *Diaporthe* species with a range of virulences have been indentified from crop and weed residues. Twelve new species have recently been described. These findings are highly significant, as it is evident that a number of these species have numerous crop and weed hosts. Additionally, as well as crop stubble, weed residues play a pivotal role in assisting the survival of many pathogenic *Diaporthe* species and although weeds may be controlled by herbicides, the remaining residues can be alive with multiple pathogenic species.

**Keywords.** *Diaporthe*, fungal, herbicides, pathogenic, *Phomopsis*, stubble, weeds.

## Effect of a weedy host on pathogen evolution

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**Abstract.** Scald caused by *Rhynchosporium commune* is commonly found on barley as well as barley grass (*Hordeum leporinum*), a common weed in Australia. Wild hosts of diseases could play a significant role in the epidemiology and evolution of diseases. Barley grass is genetically more diverse than cultivated barley and likely harbours more resistance genes than barley. This heterogeneity could select for a pathogen population that is also genetically diverse with virulences that could render newly introduced resistance genes in barley ineffective. To investigate the effect of barley grass on the evolution of scald, pathogenicity of scald isolates from barley and barley grass was assessed on both hosts. No significant difference in leaf area affected on barley was observed for isolates from barley and barley grass, thus isolates from barley grass are equally likely to affect barley than barley grass. In contrast, isolates from barley resulted in a significantly smaller percentage of leaf area infected on barley grass lines. This suggests that scald populations from barley rarely infects barley grass, however scald populations from barley grass have a high potential for gene flow to barley populations. Barley grass therefore successfully acts as an ancillary host to scald harbouring highly virulent scald populations.

**Keywords.** pathogen evolution, weedy hosts, genetic diversity.

## Symposium IV: Proffered papers

**Chairs: Sandra Abell-Davis & Richie Robinson**

### **Ecosystem services of mycophagous mammals**

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**Abstract.** Ectomycorrhizal fungi are vital components of many Australian ecosystems as they form mutualistic associations with many native plants and provide food for many mammals. As key dispersers for hypogeous ectomycorrhizae, mammals are hypothesised to be important for maintaining ectomycorrhizal diversity and plant-fungi relationships. Extensive loss of Australian mammal abundance and diversity could have consequences for ecosystem functioning through potential major reductions in dispersal of these vital fungi. However, little is known of the direct consequences of mammal dispersal on the fungi-plant relationship, even within intact ectomycorrhizal forests. This presentation will outline the preliminary results of a review that tests this hypothesis by quantifying the extent of mycophagy by Australian mammals and comparing this to ectomycorrhizal communities known from herbaria records. At least 48 native mammals have been recorded to have fungal spores within their scats. Some mammals are more important dispersers than others; bettongs and potoroos (family Potoroidae) specialise in eating fungi and *Potorous tridactylus* consumes over 100 fungal species. Of Queensland sequestrate genera 51% were found within scats of mammals; however an additional 7 genera were recorded in mammal scats but not yet as sporocarps. These results illustrate the significance of mammals for maintaining diverse ectomycorrhizal communities.

**Keywords.** mycophagy, hypogeous, dispersal.

# **Australian macrofungi have wide distributions that are explained by climate**

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**Abstract.** Recent advances in bioinformatics have aggregated an unprecedented amount of publicly available point distribution data. Through the Atlas of Living Australia, we selected observation and specimen data, predominantly from Fungimap and Australia's Virtual Herbarium, for 200 species of macrofungi (total >60,000 records). Mapping showed that species mostly had broad distributions within Australia, but rarely occupied the whole continent. The main patterns were (1) southern, including south-west Western Australia and Tasmania, and often extending in south-eastern Australia to the Macleay-McPherson overlap (MMO a narrow zone in the vicinity of the New South Wales / Queensland border, well-established for other biota), (2) semi-arid, occupying the centre of the continent and (3) northern, across the top of Australia, but also with (4) some species occurring along the entire length of the east coast, across the MMO. We applied environmental niche modelling (ENM, with MAXENT) using climate variables such as mean annual rainfall and temperature, and the seasonality of each. Taking into account under-sampled areas, ENM was very successful in modelling observed distributions. We conclude that the wide distribution of many macrofungi is a result of dispersal out to the limits of climate tolerances, subject to available habitat.

**Keywords.** biogeography, environmental niche modelling, mycogeographic provinces.

# Extreme mycorrhizal specificity in *Chiloglottis*, *Drakaea* and *Paracaleana* orchids

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**Abstract.** Delimitation of species of mycorrhizal fungi is critical to understanding the ecological and evolutionary consequences of mycorrhizal specialisation. Studies of mycorrhizal specificity in orchids primarily rely on quantifying genetic divergence among clades of ITS sequences. Here, we undertake the first rigorous test of mycorrhizal specificity in orchids using eight sequence loci to delimit species. We focused on representatives of the Australian genera *Chiloglottis* (9 species), *Drakaea* (7 species), *Paracaleana* (5 species) and *Arthrochilus* (1 species). Four species of *Tulasnella* fungi were identified, one used by *Drakaea* and *Paracaleana*, one used by *Chiloglottis* and at least two used by *Arthrochilus oreophilus*. The difference between the fungi from *Chiloglottis* and *Drakaea* was supported by the absence of germination in a cross-specificity trial. Sequence divergence among the *Chiloglottis* and *Drakaea+Paracaleana* associated lineages is 18% across the eight loci, with evidence for phylogeographic clustering within mycorrhizal species. Complete sharing of a mycorrhizal fungal species within orchid genera demonstrates that the mycorrhizal partners have not contributed to patterns of speciation and biogeography within these genera. The phylogenetic results from eight loci are strongly congruent with those from using ITS alone, supporting the case for the utility and reliability of ITS in fungal species delimitation.

**Keywords.** *Tulasnella*, specificity, ITS.

# The root myco-biome: disentangling the influence of host and soil on fungal community structure

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**Abstract.** Microbial communities in plant roots provide critical links between above and belowground processes in terrestrial ecosystems. Variation in root communities has been attributed to plant host effects and microbial host preferences, and factors pertaining to soil conditions, microbial biogeography and the presence of viable microbial propagules. To address hypotheses regarding the influence of plant host species, plant host genotype and soil origin on root fungal communities, a trap-plant bioassay experiment was carried out. In this experiment triplicate host plants belonging to *Populus trichocarpa*, *Populus deltoides*, *Pinus taeda* and *Pinus ponderosa* were grown in soils originating from 4 common garden experimental plots in western North America (CA, OR, WA, BC) and 1 site in eastern North America (NC). Genotype effects were assessed by including eight genotypes of *P. trichocarpa* exhibiting a range of phenotypes. Negative control plants were grown in sterile sand. Root fungal community profiles from these 190 plants were assessed through multiplex amplicon sequencing of both ITS and 28S rDNA regions with MiSeq resulting in >4.5 million reads representing approximately 700 OTUs and an average of 1815 quality reads per sample. ITS and 28S rDNA datasets were largely congruent. Soil origin was the strongest determinant of fungal community.

**Keywords.** fungal communities, ITS barcoding, MiSeq amplicon sequencing, rhizosphere fungi.



# ***Scedosporium aurantiacum* - an emerging fungal pathogen with extensive genetic diversity**

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**Abstract.** *Scedosporium spp.* are amongst the most important fungal infections for patients with cystic fibrosis (CF). Invasive *Scedosporium* infections are also problematic after near-drowning accidents and in transplant patients. *Scedosporium aurantiacum* is an emerging human pathogen causing a wide range of invasive infections. To identify possible environmental sources of infections and to determine the spread of this emerging pathogen a better understanding of its epidemiology is required. A total of 188 clinical, environmental and veterinary *S. aurantiacum* isolates from Australia, Europe, Asia and the America were characterised by multilocus sequence typing (MLST). The scheme is based on the combined sequence analysis of six genes: actin (*ACT*), elongation factor-1 $\alpha$  (*EF1 $\alpha$* ), calmodulin (*CAL*), RNA polymerase subunit II (*RPB2*), manganese superoxide dismutase (*SOD2*), and  $\beta$ -tubulin (*TUB*) at: [mlst.mycologylab.org](http://mlst.mycologylab.org). Among this geographically diverse collection 10-25 alleles per genetic locus and 166 unique sequence types were identified. Phylogenetic analysis revealed separate clustering of the Australian and European samples. The high diversity among the Australian strains suggests that *S. aurantiacum* may have originated within Australia and was subsequently dispersed to other regions. A fact revealed by the close phylogenetic relationships between some of the Australian sequence types and those found in other parts of the world.

**Keywords.** *Scedosporium aurantiacum*, MLST, emerging pathogen.

Chair: Nigel Fechner

**New Russulaceae species in South-east Queensland**

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**Abstract.** *Russula* species are ectomycorrhizal basidiomycetes common throughout Australian woodlands. Molecular and morphological examination of *Russula* specimens from various sites in South-east Queensland has uncovered a number of phylogenetically diverse and potentially undescribed specimens. At this stage, most of these remain as individual specimens. However, several specimens have been grouped into project species. In this presentation, two potentially new species of *Russula* will be described. Both species are widespread on red to black clay soils in the Toowoomba region and appear to be phylogenetically related to each other. Project species 1 has a white stipe which developed a pink flush after collection and red to pink centrally depressed cap with white gills. The spores of this species were subglobose, white and ornamented with amyloid warts connected in short chains. The second species, project species 3, is macroscopically similar, differing with its larger size, purple-pink cap colour, and rougher cap texture. Its spores are globose to subglobose, white and ornamented with small amyloid warts connected in short chains. Continued molecular and morphological studies of the *Russula* species of South-east Queensland may uncover more novel Australian species.

**Keywords.** *Russula*, phylogenetics, taxonomy

## **Molecular taxonomy of Australian endophytic Pezizales**

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**Abstract.** The order Pezizales contains a wide variety of striking macro ascomycetes not commonly observed in Australia. Known as cup fungi and false morels due to the prominent, often brightly coloured apothecia that are produced, most samples may be observed on the ground or on decaying leaf and bark material. Recently, 33 likely novel isolates belonging to Sarcosomataceae and Sarcoscyphaceae were found occurring endophytically in leaves collected from a semi evergreen vine thicket in the Brigalow Belt in South-East Queensland. An endophytic life mode is considered rare for these families and so this observation raises interesting ecological questions. The 33 isolates were analysed to determine their phylogenetic relationship with other known Sarcosomataceae and Sarcoscyphaceae and found to belong to three potentially new genera. This observation represents an increase in the observance of Sarcosomataceae in Australia and also a possible range expansion for one genus within the Sarcoscyphaceae.

**Keywords.** Sarcosomataceae, Sarcoscyphaceae, endophytes.

## Towards a trait-based ecology of fungi

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**Abstract.** The development and uptake of next generation sequencing has greatly increased mycologists' abilities to characterise fungal diversity but, by itself, has done little to increase our general understanding of the ecological drivers of this diversity and its consequences. In November 2013 we hosted a workshop including experts in fungal ecology and evolutionary biology from around the world. The workshop participants concluded that a trait-based approach, coupled with next generation sequencing, is necessary to address these questions. Traits are physical (morphological, anatomical) and chemical (physiological, nutritional) characteristics of species, many of which have functional consequences for species survival and growth and influence many biotic interactions. Shifting trait distributions along environmental gradients reflect changes in the relative abundance of species expressing these traits. Constraints on trait distributions within communities can be a characteristic of environmental filtering during community assembly (environmental conditions select for traits that allow species to persist). Trait distributions may also become more dispersed, reflecting assembly processes that limit similarity among co-occurring species and generate functional diversity within communities. Each of these processes has had consequences for the maintenance of ecosystem services in studies of plant communities, but attempts to understand these consequences in fungal communities are in their infancy. The workshop resulted in a conceptual model that incorporates traits into our current understanding of the ecological interactions between fungi and the environment and their consequences for ecosystem functioning in natural and managed ecosystems. By engaging the broader mycological research community, we hope to advance these ideas and identify traits that may have use across broad disciplines, while also agreeing on approaches for collecting trait data, which will lead to a more general framework for developing a trait-based ecology of fungi.

**Keywords.** trait-based approaches, trait databases.

## Quality controlled ITS database for human and animal pathogenic fungi

Irinyi L, Khan MA, Pan G, King W, Meyer W and the global medical mycology ISHAM barcoding working group

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**Abstract.** Mycoses have significantly increased worldwide prompting a need for a rapid and accurate identification of the causative agents. Conventional identification methods are time-consuming, laborious and require specially trained personal. Sequencing based, DNA barcoding is a cost-efficient method to simplify and speed up the identification of organisms to the species level from all kingdoms including fungi without being an expert. However, DNA barcoding is challenged by the lack of quality controlled reference databases and low numbers of reliable ITS sequences (official barcode for fungi) of pathogenic fungi in public databases. As a result of an international consortium of medical mycology reference laboratories, a quality controlled ITS database as part of the ISHAM working group “Barcoding of medical fungi” was established. This new online database currently contains 2700 ITS sequences representing 416 species ([www.mycologylab.org](http://www.mycologylab.org)). Overall intraspecies variation was less than 1.5%, with most of the phylogenetic taxa presenting a barcoding gap. However, for a number of species/species complexes intra- and interspecies genetic distances are overlapping, revealing major limitations of the ITS region for a universal fungal barcode. To overcome these limitations new genetic loci identified from whole genome data are currently under investigation to either be used as secondary or alternative fungal barcode.

**Keywords.** quality controlled ITS database, medical fungi, barcode.

**Chair: Diana Leemon**

**Gut inhabiting anaerobic fungi**

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**Abstract.** Zoospores of anaerobic fungi were first discovered in the rumen in 1910, albeit being incorrectly identified as flagellated protozoa. It wasn't until 1974 that Orpin discovered zoospores isolated from the rumen, were actually expelled from a sporangium, giving rise to the idea that it was in fact an anaerobic fungus. To date, all anaerobic fungi fall under the phylum Neocallimastigomycota, and the order Neocallimastigales, with six genera being identified and 24 species. Since their discovery, Neocallimastigomycota have been detected in ruminant (sheep, cattle) and non-ruminant, herbivorous mammals and reptiles. The role of anaerobic fungi in the gastro-intestinal tract is primarily fibre colonisation and digestion. Due to the fibrolytic/cellulolytic nature of anaerobic fungi, and an increasing interest in sustainable biofuel production, there has been a recent resurgence of interest in anaerobic fungi and their fibre-degrading enzymes. The current taxonomic status of anaerobic fungi will be reviewed and updated, with particular reference to anaerobic fungi of the gut. Results from a new Queensland study, examining gut samples obtained from the rumen of cattle and the forestomach of kangaroos and using a high through-put sequencing approach will also be described, with particular emphasis on the identification of anaerobic fungal populations and fungal-associated fibrolytic enzymes.

**Keywords.** anaerobic fungi, enzymes, high-throughput sequencing.

## Resistance of naturally-durable timbers to fungal decay

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**Abstract.** Saprophytic fungi that decay wood on the forest floor play a vital role in carbon recycling. In the built environment, wood decay is undesirable, causing potentially dangerous failures of structural timbers. Decay fungi produce extracellular lignocellulolytic enzymes and low molecular weight compounds that decompose wood cell walls resulting in three general decay patterns: brown, white and soft rot. Brown rots and white rots are commonly caused by basidiomycetes (e.g. *Gloeophyllum abietinum*, *Trametes versicolor*). Soft rot is often caused by ascomycetes (e.g. *Phialophora richardsiae*) in timber used in ground contact and subject to high moisture and reduced oxygen availability. Timber from sustainably-managed natural forests and plantations has impressive life-cycle environmental credentials. Research is underway at DAFF Forestry Science to measure and understand the causes of natural decay resistance to facilitate timber quality management and tree breeding. Natural durability is attributed to chemical ‘extractives’ such as polyphenols that actively protect timber through toxic effects on decay fungi. In addition to toxic extractives, we have found that some compounds in naturally durable native timbers may provide passive decay resistance, such as waxes that prevent the accumulation of free water, which is necessary for decay.

**Keywords.** wood decay, natural durability, decay fungi.

# Using fungi as bioherbicides for woody Weeds

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**Abstract.** Among the Weeds of National Significance (WoNS), two in particular, parkinsonia (*Parkinsonia aculeata*) and prickly acacia (*Acacia nilotica*) are considered to be among the most important threats to the rangelands and wild lands of northern Australia affecting approximately 3 and 6 million ha respectively. Collectively, these weeds are responsible for significant losses to productivity by hindering access for cattle and creation of thickets and woodlands which shade out pasture and invade riparian zones. Each of these woody weeds is affected by a range of endophytic fungi, which under conditions of stress, can lead to the development of dieback disease. A method for field scale inoculation of these weed species has been developed with the aim of initiating dieback events. The method involves the use of capsules containing inocula which are inserted into, and sealed within the weed tree stem to induce infection. This method is currently undergoing field trials with the objective of producing a commercially viable product.

**Keywords.** woody weeds, bioherbicide, inoculation.



# Comparing morphological and molecular species concepts for *Cortinarius* from long-term monitoring in Western Australia

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**Abstract.** Following long-term macrofungal surveys in permanent plots in jarrah (*Eucalyptus marginata*) forest in Western Australia, 178 voucher collections of *Cortinarius* were made and were clustered in 118 morphospecies. Many morphospecies could be identified in the field, but some represented species complexes that were difficult to separate on morphological characters alone. The analysis of the ITS rDNA region of the 178 voucher collections using a 98% similarity threshold produced 94 molecular species among which 57 were represented by a single collection. The molecular and morphological species concepts were congruent for only eight morphospecies. Ten morphospecies fell together in one clade but were mixed with one or more other morphospecies and 14 occurred in two or three branches in the tree. When morphospecies were split, the segregate molecular species were sometimes closely related but could also be phylogenetically highly divergent. In particular, the morphological group identified as *Cortinarius sublargus* included three phylogenetically unrelated collections. Furthermore, some collections that fell within individual molecular species of the *Cortinarius sublargus* complex had observable morphological differences and therefore were assigned to different morphospecies. Our analysis, however, confirms the high level of diversity in the genus; indicates that further sampling is required to fully inventory the genus; allows calibration of morphological characters to recognise molecular species; highlights groups with morphologically cryptic species; and provides a databank for future molecular identification. When collecting ecological data based on morphospecies, it is better practice to split putative but complex species in order to not underestimate the species diversity — this and other outcomes associated with macrofungal surveys will be discussed.

**Keywords.** *Cortinarius*, species delimitation, cryptic species, biodiversity monitoring

**Chair: Peter McGee**

**Improving the recovery outcomes of the critically endangered Wollemi pine: is success determined by soil microbes?**

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**Abstract.** Wollemi pine (*Wollemia nobilis*: WP W. Jones, K. Hill & J. Allen) is a monotypic species, of which fewer than 100 trees are known in the wild. The Wollemi pine Recovery Team has proposed translocation as a conservation strategy to establish 'back-ups' to the wild population; however, knowledge regarding the environmental/ biotic requirements of individuals planted in new environments is limited. One of the most important limitations to the introduction or reintroduction of tree species is the presence of suitable microbial partners. Plants in novel environments will encounter fewer co-evolved mutualists. WP grows on shallow soils of poor nutrient status and high acidity and is likely to be highly dependent on mycorrhizal fungi, which have been observed associated with the roots of WP, and bacteria that contribute to nutrient cycling. We found that microbial communities associated with soils under WP in the wild differed from those under neighbouring species. We also found that WP seedlings were slightly larger at 5 months when interacting with their own microbes than with microbes associated with these neighbours, suggesting that generalist pathogens and/or a lack of host-specific beneficial associations may be an important factor limiting WP recruitment.

**Keywords.** Wollemi pine, translocation, microbial community analysis.

# **Arbuscular mycorrhizal fungi actuate aggregation of mine spoil**

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**Abstract.** Arbuscular mycorrhizal (AM) fungi play a key role in plant growth and development. Less well known is their role in soil functions. Extensive hyphal exploration of soil leads to the possibility that AM fungi play an important role in formation of top soil, especially soil structure. Aggregate and pore development underpin the structure, water characteristics, carbon storage and root penetration of the soil profile. Mine spoil lacks structure, organic matter and a diverse microbial community. The importance of the inclusion of AM fungi for the development of soil structure was examined in two experiments. Compost, plants and AM fungi were essential for the formation of topsoil in spoil from Mt Own (Hunter Valley). The contribution of plants was superficial, and relied on the presence of compost and AM fungi to establish resilient structure within the soil. Subsequently the contribution of each AM fungus was examined. Each fungus had a different impact on various components of soil structure. Overall, no single AM fungus yielded as good an outcome as all eight fungi together. The data demonstrate the importance of including many AM fungi, as well as diverse plant communities and compost, for the formation of structured topsoil from mine spoil.

## **Working with mycorrhizal fungi in the field**

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**Abstract.** Mycorrhizal fungi are an important indicator of soil ecosystem health in natural environments, and their abundance and diversity can be used to gauge ecosystem health in restored environments, agriculture and forestry systems. Working with mycorrhizal fungi in restoration projects and field experiments can present some specific challenges unless these are understood and planned for beforehand. One such challenge is the use of inoculum, which can sometimes be of poor quality and contain unsuitable species, and must be applied correctly to achieve success. There are a number of soil characteristics and management practices that must be avoided or minimised to allow mycorrhizal fungi to establish and proliferate successfully. Measurements of mycorrhizal abundance and diversity are another challenge. Mycorrhizal quantification is available from commercial soil microbiology laboratories, but it is important to follow sampling methods specific to mycorrhizal fungi to obtain representative and accurate results. Measuring mycorrhizal fungi in your own laboratory can also produce misleading results unless effective protocols are strictly followed. Current analytical methods use molecular and microscopic methods, but should be chosen carefully as they are not always related to actual abundance or plant response in the field.

**Keywords.** mycorrhizal fungi, field, revegetation, inoculum, measurement, methods.

## **Some melanised root-associated fungi may increase organic carbon in soil**

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**Abstract.** Organic carbon makes a crucial contribution to soil function. Loss of organic carbon from soil places sustainable food production under serious threat. Current methods of soil management that aim to increase organic carbon in soil have unpredictable outcomes in part due to misunderstandings surrounding the long-term storage of organic compounds in soil. Melanised root-associated fungi have access to the centre of soil aggregates, an anaerobic environment within which the oxidation of organic compounds is greatly reduced. Further, melanised root-associated fungi possess a recalcitrant polyaromatic compound within their cell walls, melanin. Melanised root-associated fungi therefore have the potential to increase levels of organic carbon within soil aggregates, of which aromatic compounds make up a significant portion. A compartmental pot study was conducted in order to test whether melanised root-associated fungi isolated from native plants within the Sydney basin (NSW, Australia) could enhance organic carbon in soil. Over 14 weeks, 20 of 24 melanised root-associated fungi increased levels of carbon in an aggregated carbon-rich agricultural soil. Nine of these fungi concurrently increased concentrations of phenolic materials in soil. The role of melanised root-associated fungi in the enhancement of organic carbon in soil may be greatly underestimated.

**Keywords.** soil aggregates, soil carbon, polyphenolic, root endophytes, melanised fungi, carbon sequestration.

## Can inoculation with fungi improve the rehabilitation of a minesite?

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**Abstract.** Contemporary mine rehabilitation utilises spoil (rock removed from above and between coal seams) to fill the mine void. Stripped sub and/or top soils are spread atop the spoil and vegetation is seeded. However, in some cases the quality and quantity of replaced soils is inadequate for reasonable rehabilitation. Practitioners are seeking potential alternatives to topsoil or ameliorants for spoil. Pot trials suggest amendment of spoil with organic matter and fungi may result in a surface with reasonable qualities. At an experimental field site in the Hunter Valley of NSW, the inoculation of spoil with microbes (rhizobia, ectomycorrhizal fungi, arbuscular mycorrhizal fungi and dark septate endophytic fungi) and municipal waste compost is being tested. Treatment is primarily intended to improve the organic carbon content of spoil. Organic carbon contributes physical, chemical and biological benefits (binding particles; improving plant growth; increasing water, mineral and nutrient status; and providing energy for biological processes). Eighteen months following planting with Australian native plants (*Hakea sericea*, *Acacia parvipinnula*, *Corymbia maculata* and *Dodonea viscosa*) the effects of treatment are not yet clear. Total carbon is in the range of 2-5% and this includes coal dust and carbonates. More detailed analysis and further harvests are planned.

**Keywords.** spoil, municipal waste compost, inoculation, rehabilitation, Australian native plants.

**Parallel evolution of the fungal pathogen *Cryptococcus neoformans***

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**Abstract.** The selective pressure placed on an organism under environmental stress can lead to the proliferation of adaptive mutations. Pathogenic organisms encounter a hostile host environment during infection and long-term colonization in particular provides opportunity for adaptive mutations to spread through the population. The fungal pathogen *Cryptococcus neoformans* currently has an estimated global burden of over 600,000 deaths per annum making it of significant clinical relevance. A key feature of *C. neoformans* pathogenesis is the ability to establish dormant infections, as immunocompetent hosts are thought to acquire an asymptomatic or subclinical initial infection that remains latent until immune suppression, at which time the infection can escalate. Such a pattern presents an ideal background from which adaptive mutations can emerge. However identifying these key mutations in studies of the species is hampered by limited clinical samples and the large number of mutations observed between isolates across the 20 Mb genome. Serial isolates represent one potential way to overcome these issues by reducing the number of mutations between isolates, however the uncontrolled host environment leaves open the question of whether the mutations occurred within the host or the environment. To support a significant role within the host, we have focused on identifying mutations affecting the same gene in multiple independent isolates, relying on the idea that the similar pressures encountered by each of the isolates has selected for changed function of these gene products. To date this work has uncovered multiple genes under apparent parallel evolution and through their characterization we hope to reveal key components of the infection process of this pathogen.

## The role of sirtuins in silencing in *Cryptococcus neoformans*

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**Abstract.** The opportunistic fungus *Cryptococcus neoformans* is responsible for causing cryptococcosis, which is invariably fatal if left untreated. A range of known virulence factors enables the fungus to infect a human host, however regulation of these factors is poorly understood. Recent results suggest that the number of rDNA repeats in a single fungal strain is frequently altered during infection. In *C. neoformans* isolated from patients, there are consistently changes in the array length from initial infection isolate to a relapse isolate. In *Saccharomyces cerevisiae*, Sir2 controls both the maintenance and silencing of the rDNA array and based on this knowledge, sirtuins, and the role they play in silencing was investigated in *C. neoformans*. Five predicted sirtuin-encoding genes were identified in the *C. neoformans* genome. All five of the identified sirtuins were deleted from the genome. Initial findings after a battery of phenotypic tests indicated that the sirtuins are playing a variety of roles in the cell, including influencing virulence factor production and maintaining and silencing the rDNA array, which is of particular interest as these sirtuins may possibly be modulating virulence. The sirtuins also impact the ability of *C. neoformans* to survive in a non-dividing state, as the loss of the sirtuins resulted in cellular death far earlier compared with the wildtype. This suggests that sirtuins in *Cryptococcus* play a role in longevity in a similar fashion to that seen in *Saccharomyces*. Future work in this area will produce even more vital knowledge regarding the regulatory occurrence of gene silencing and the potential role that it may play in the virulence of *C. neoformans*.

**Keywords.** silencing, rDNA array, longevity.



# **An investigation of the ecology and bioactivity of endophytes from *Pittosporum angustifolium***

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**Abstract.** Knowledge of the ecology and antimicrobial activity of endophytes within Australian plants is currently limited. This project investigated the endophytes of *Pittosporum angustifolium*; an Australian plant known for its medicinal use by indigenous Australians and from which antimicrobial chemicals have been extracted (from the essential oils of its leaves and fruit). Plants were sampled from seven sites in SE Queensland. Endophytes were cultured from plant tissue onto potato dextrose agar. Sequencing of ITS-DNA regions of fungal endophytes and SSU-DNA regions of bacterial endophytes revealed a variety of different species isolated from plants sampled at the different sites - evidence against any host specific interactions. Primary screening of isolates occurred on Sensitest agar against ATCC type strains of *Staphylococcus aureus*, *Serratia marcescens*, *Escherichia coli* and *Candida albicans*. Primary screening resulted in strains of *Pseudocercospora fuligena* being identified as candidates for further investigation. Work is still ongoing to confirm the bioactivity of the *P. fuligena* isolate and to identify any bioactive compounds produced.

**Keywords.** endophyte, *Pittosporum angustifolium*, bioactivity.

# Investigation of the anti-microbial properties of endophytes from *Santalum lanceolatum*

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**Abstract.** *Santalum lanceolatum* is a native Australian plant which has a history of prior use by the indigenous Australian population to treat infections. This project looks at the fungal endophytes present in *Santalum lanceolatum* and whether bioactive compounds from the endophytic fungi may be able to be used as antimicrobials. At this stage two sites in the Darling Downs region have been sampled with both sites producing a wide variety of endophytes from the plant tissue when grown on potato dextrose agar. One site has already been screened on Sensitest agar against ATCC type strains of *Staphylococcus aureus*, *Serratia marcescens*, *Escherichia coli* and *Candida albicans*, with no observable inhibition as to the growth of these pathogens. If any bioactive compounds are found to be effective, then the molecular structure of the compound is to be identified along with the sequencing of the ITS-DNA of the endophytic fungi from which it came. Work on this project will be continuing throughout 2014.

**Keywords.** Endophyte, *Santalum lanceolatum*, bioactivity.