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1. Mycotoxic metabolites produced by *Fusarium* species associated with feed refusal disorders in Western Australia

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Sheep, grazing in six different locations in Western Australia, were reported to partially or completely refuse to consume annual *Medicago* pods, which were later found by Barbetti and Allen (2005) to be contaminated with a number of different *Fusarium* species. Subsequent rat toxigenicity studies by Barbetti and Allen (2006) on 16 of these Western Australian *Fusarium* species showed they can cause high mortality rates. *Fusarium* species from other parts of the world are known to produce deoxynivalenol (causes vomiting and feed refusal in animals) and/or diacetoxyscirpenol (also causes reduced feed intake in animals) as part of their array of toxigenic secondary metabolites. The aim of this study was to isolate and characterize the secondary metabolites produced by the 16 *Fusarium* species associated with sheep feed refusal disorders in Western Australia. Purification of toxigenic fungal extracts using a brine shrimp (*Artemia salina*) bioassay guided fractionation and HPLC, and identification by GC/MS and NMR spectroscopy, revealed the presence of a number of trichothecenes (3-acetyldeoxynivalenol, deoxynivalenol, nivalenol, monoacetoxyscirpenol, diacetoxyscirpenol, and scirpenol), enniatins (A1, B, and B1) and zearalenone. These compounds showed significant activity with *Artemia salina* at concentrations as low as 5μg/ml, and are likely responsible for the sheep feed refusal disorders observed previously and also for the toxicity observed in the earlier rat tests.

2. Incidence and new records of *Mycosphaerella* species within a *Eucalyptus globulus* plantation in Western Australia

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Eucalyptus globulus is the predominant exotic hardwood plantation species in Western Australian (WA), and is often planted adjacent to native eucalypt forests. The increase in number of *Mycosphaerella* species associated with Mycosphaerella leaf disease (MLD) in *E. globulus* plantations in WA in the past decade has raised concern about the possible movement of pathogens between the native forests and plantations. In order to determine whether the introduction of new *E. globulus* genetics into WA may have further exacerbated this situation, juvenile and adult foliage were taken from a genetics trial near Albany, WA consisting of 60 full-sib families and *Mycosphaerella* species identified using morphological and molecular tools. Eleven species of *Mycosphaerella* were identified from one plantation: *Mycosphaerella fori* (*Pseudocercospora fori*) and *Mycosphaerella ellipsoidea* are new records for Australia; *Mycosphaerella tasmaniensis* (*Passalora tasmaniensis*) and *Mycosphaerella suttoniae* (*Kirramyces epicoccoides*) are new records for WA; and *Mycosphaerella nubilosa*, *Mycosphaerella cryptica*, *Mycosphaerella marksii*, *Mycosphaerella molleriana*, *Mycosphaerella lateralis*, *Mycosphaerella aurantia* and *Mycosphaerella parva*, previously recorded for WA. These records increase the number of known *Mycosphaerella* species from eucalypts in WA from 10 to 13. The increase in the number, distribution and impact of *Mycosphaerella* species contributing to MLD in WA is of concern both to the potential productivity of the plantations and the biosecurity of native WA *Eucalyptus* species. Continued monitoring of the plantation estate is required to understand the dynamics of the host–pathogen interactions.

3. Responses of native plants to infection with indigenous and introduced viruses

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The Western Australian flora is under threat from plant pathogens, including viruses. No investigations have been made to establish the susceptibilities of native species to infection with introduced or indigenous viruses. In a series of glasshouse experiments in which 14 native plant species were sap- or aphid- inoculated with 3 indigenous and 16 introduced plant viruses, infection with a least one virus was identified in 13 species from 6 plant families. In *Kennedia coccinea* (coral vine) plants the most severe damage was caused by BCMV-13B (*Bean common mosaic virus*-isolate 13B), infected plants developing leaf mottle, leaf deformation, stunting and shoot tip or plant death. *Kennedia prostrata* (scarlet runner) plants infected with BCMV-13B and CMV-SN (*Cucumber mosaic virus*-isolate SN) developed systemic necrosis, plant stunting and shoot tip or plant death. *Hardenbergia comptoniana* (native wisteria) plants only

became infected by HarMV-SP-1 (*Hardenbergia mosaic virus*-isolate SP-1) probably because its waxy leaves inhibited inoculation with other viruses. *Anigozanthos manglesii* (red and green kangaroo paw) plants became infected with CMV-SN and TSWV-Crb-1 (*Tomato spotted wilt virus*-isolate Crb-1) resulting in necrotic leaf symptoms, plant stunting and death of some plants. In this species, PVX-XK (*Potato virus X*-isolate XK) infection caused systemic necrotic streaking similar to symptoms induced by *Alternaria alternata* (inkspot) disease. In plants of *Kennedia prostrata* with BCMV-13B and *Gastrolobium bilobum* (heart leaf poison) with HarMV-SP-1, virus infection caused 96 and 97% losses in shoot dry weight respectively. In *Solanum symonii* (kangaroo apple) plants infected with AMV-EW/CMV-SN together or TSWV-Crb-1 alone, shoot dry weight was reduced by 60% and 99% respectively. Fresh weight per fruit was significantly reduced by infection with AMV-EW/CMV-SN or CMV-SN, and TSWV-Crb-1 infected plants produced no fruits. These results show that virus infection can cause severe damage to Australian native species, potentially impairing plant fitness and decreasing both competitive and reproductive ability.

4. Are the predatory mites, *Typhlodromips montdorensis*, *Neoseiulus cucumeris* and *Hypoaspis mile*, compatible with spinosad for the management of resistant western flower thrips, *Frankliniella occidentalis*?

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In Australia, Western flower thrips (WFT), *Frankliniella occidentalis* Pergande (Thysanoptera: Thripidae) have developed resistance to a range of insecticides including the biopesticide spinosad. To control resistant WFT, a combination of spinosad sprays and releases of predatory mites (Acari) may be required as part of an IPM program in strawberry. The present study assessed the effect of applying twice the recommended rate of spinosad (0.096 g ai/L) to the phytoseids *Typhlodromips montdorensis* (Schicha) (Phytoseiidae) and *Neoseiulus cucumeris* (Oudemans) (Phytoseiidae) which feed on the larvae, and *Hypoaspis mile* (Berlese) (Laelapidae) which feeds on the pupae. Spinosad was very toxic to *T. montdorensis*, *N. cucumeris* and *H. miles*, causing 100 percent mortality. Spinosad residues aged two, 24, and 48 hours also appeared to be very toxic to *T. montdorensis*, *N. cucumeris* and *H. miles* causing 96-100 percent mortality. Comparative toxicity indicated that spinosad residues aged 48 to 168 hours were less toxic to *N. cucumeris* followed by *T. montdorensis* and *H. miles*. The residual threshold for *T. montdorensis, N. cucumeris,* and *H. miles* was calculated as 146.76, 127.85, and 162.45 h (LT_{25}) respectively. A glasshouse trial showed that when mites were released on WFT-infested plants following six, five and seven days after a spinosad application, *T. montdorensis*, *N. cucumeris,* and *H. miles* significantly reduced WFT numbers compared to the control. *T. montdorensis* appeared to be the most successful in reducing WFT numbers followed by *N. cucumeris* and *H. miles*. The results indicate that predatory mites' releases can be successfully integrated with spinosad applications if required.

5. Genetic dissection of the *Medicago truncatula/Phytophthora medicaginis* pathosystem

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The oomycete genus *Phytophthora* is one of the most important groups of microbial plant pathogens worldwide. There are several important species of *Phytophthora* causing disease within the agriculture and horticulture industries in Australia, where farmers are bearing the cost of over $300 million annually in lost production and control measures. *P. medicaginis* causes a $3.5 million annual loss to the New South Wales chickpea (*Cicer arietinum*) industry alone. Other important Australian legume crops affected by *Phytophthora* are lucerne (*Medicago sativa*), soybean (*Glycine max*), lupin (*Lupinus alba*), clover (*Trifolium sp.*), cowpea (*Vigna unguiculata*) and adzuki bean (*Vigna angularis*).

Understanding of the host/pathogen interaction through the application of molecular techniques has the potential for discovering novel resistance genes. Once cloned these could reveal gene functions and provide a better understanding of mechanisms of pathogenesis and resistance. Via molecular mapping of resistance genes in the model legume *Medicago truncatula,* the aim of this project is to understand the molecular basis for host/pathogen interaction and to discover and genetically characterise novel plant resistance genes.

6. Host-pathogen interactions in the Mustard-White rust pathosystem: Protein expression profiling

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Albugo candida (white rust) is a serious disease of cruciferous vegetable and oilseed crops. Leaf and inflorescence infection causes yield losses up to 60% or more in India, and losses of up to 20% in Australia. Presently, canola-quality *B. juncea* is being developed to extend *Brassica* oilseed production to the lower rainfall areas of the southern Australian wheatbelt. Unfortunately, the varieties of *B. juncea* available in Australia appear highly susceptible to white rust. To understand the biochemistry of the host-pathogen interaction a proteomic study was undertaken to analyse changes in protein expression levels at different time points after inoculation during the interaction of *A. candida* with resistant and susceptible cultivars respectively. Some key regulators contributing to host resistance towards *A. candida* were determined that can be used to design genetic markers to screen for resistance available in *B. juncea* germplasm. Transcriptional expression changes shown by some enzymes are reflecting on how the fungus manipulates the host plant to get access to its resources/nutrient reserves. This gives a new insight into the understanding of mechanisms of host-pathogen interaction in this pathosystem.

7. Movement of pathogens between horticultural crops and endemic trees in the Kimberley

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Horticulture is Australia's second largest and fastest growing sector of agriculture, the majority of products being grown for the domestic market. Shoot blights, cankers and associated stem end rot of fruits caused by botryosphaeriaceous fungi, are amongst the most common diseases affecting tropical fruit production. Researchers in northern Australia (Queensland, Western Australia and the Northern Territory) have routinely isolated botryosphaeriaceous fungi from cankers and decayed fruit of tropical horticultural trees. Using molecular phylogenetic techniques fifteen botryosphaeriaceous species were identified as pathogens from cankers or endophytes of healthy tissue of tropical fruit trees in Australia; *Schizophyllium commune*, *Neofusicoccum mangiferum*, *N. ribis*, *N. parvum*, *Botryosphaeria dothidea*, *Macrophomina phaseolina*, *Neoscytalidium novaehollandiae*, *N. dimidiatum*, *Pseudofusicoccum kimberleyense*, *P. ardesiacum*, *P. adansoniae*, *L. theobromae*, *L. pseudotheobromae* and *L. parva*. *Lasiodiplodia pseudotheobromae*, *L. parva* and *P. kimberleyense* were identified from all regions (Western Australia, Queensland and the Northern Territory). Five species were shared between the native ecosystem and horticultural ecosystem. The pathogenicity between ten isolates to Kensington pride mangoes was determined.

8. Magic Mushrooms – The white *Amanitas* of Perth

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The framework of mycological taxonomy depends upon macroscopic and microscopic characteristics. For many taxa these techniques are inadequate. The use of genetic techniques in combination with the traditional methods can distinguish between previously indistinguishable collections. The aim of this research is to establish species parameters of a cohort of white *Amanitas* using this polyphasic approach. The newly described species are all growing in the same remnant of disturbed land on the periphery of a large university campus. Eucalypts and introduced pines are the dominant tree species over this area. Using the traditional macroscopic characteristics the field collections formed five groups. It was difficult to assign some collections using only field characteristics. When microscopic features (spore and basidia dimensions, presence and abundance of clamps, and subhymenial organisation) were examined two groups were amalgamated. Using ITS4 and ITS5 primers RFLP profiles were generated for all groups' collections. Initial results support the four groups.