

PHYTOGEN

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Were you aware that? – Information about ASPS activities

Thanks to all the contributors for being prepared to give their time to compile the many informative articles in this issue of Phytogen

APSP OFFICE BEARERS - 2009

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A message from the editor

Dear Fellow ASPS Members,

Well here at last is the November issue of Phytogen coming with my abject apologies to all those contributors who worked against the clock and had their articles in on time. Some unexpected delays and technical difficulties have contributed to the late completion of this issue. Nevertheless it is full of a broad range of interesting and informative articles many from new contributors.

In this issue you will find familiar and new sections. "State of Affairs" from the ACT provides us with "the new look ANU" neatly dovetailed with contributions from CSIRO Plant Industry (Black Mountain) -- thanks to Peter Solomon for organizing this excellent contribution. Our "New PhD" section has been continued by Dr Alice Hayward from Christine Beveridge's group and as a first for "Discipline Highlights" Oula Ghannoum and David Tissue have prepared an account entitled *Eucalyptus Growth in Past and Future Climates.* There is a feature on The Australian Plant Phenomics Facility and thanks to Helen Irving "Twigs and Branches" is back.

The "Education Section" is new. I was inspired by receiving materials from Geoff Burrows & John Harper illustrating their Online Teaching modules and have combined these with reviews of two books by Teri O'Brien focused on teaching plant anatomy. I hope that we can continue this section as I am sure you would all agree that teaching our science in imaginative ways is a crucial component of promoting plant science.

Thank you all for your efforts and Seasons Greetings.

Tina Offler

A message from the president

I am looking forward to the meeting of ComBio2009 in Christchurch. It is nine years since our last combined meeting in New Zealand, which was in Wellington in 2000. This year there are two concurrent plant biology themes, and a third on agriculture and horticulture. At present over 850 people have registered, 80 of them being from our Society.

FASTS, the Federation of Australian Scientific and Technological Societies, to which our society subscribes, recently released a policy statement on Women in Science. I represented the Society at the FASTS workshop at Parliament House (see photo). The report can be accessed at <u>http://www.fasts.org</u>, by scrolling down their home page. This presents startling statistics on the advancement of women in relation to men in universities and research organisations. Although women make up 50 % of the total at PhD and postdoctoral level, there is a steady decline in relativity so that at the most senior levels women make up only 10% of the total. Actions are being taken to redress this worrying trend.

Rana Munns, (6/11/09)



ASPS Secretary's report for 2009

Taking over this role for the society is not straightforward as there is no manual or help menu, so apologies to those who have sought help and had slow response times. Fortunately, the fact that ComBio is happening in December this year gave us more time to sort out our awards. This year, while we had several worthy applicants, the Peter Goldacre Award will be given to Ben Trevaskis (CSIRO, Division of Plant Industry), for his contribution to understanding the molecular basis of flowering in cereals and the integration of vernalisation and long day pathways. His lecture is on Tuesday 8th December at 9.45am and follows the award of best paper in FPB to Joanne Tilbrook. On Sunday afternoon, there is an education symposium during which Amanda Able (The University of Adelaide) will enlighten us with some of her methods that have been recognised as this year's winner of the teaching award. While I never met JG Wood, we are fortunate to have his commemorative lecture delivered by Barry Osmond. As Barry put it: 'Joe Wood's work with Atriplex guided my Hons, MSc and PhD research, so it will be a signal honour to commemorate aspects of his work on the 50th anniversary of his death (8th December 1959)'. The society has also made two awards from the RN Robertson fund to assist Foteini Hassiotou (University of Western Australia) and Bianca Kyriacou (Flinders University) travel to other laboratories to broaden the research for their PhD projects. It is through our membership in ASPS and donations to the Robertson fund that we are able to support students attending ComBio and give awards to facilitate or recognise young researchers.

On a sadder note, the executive has decided to stop work on producing a second edition of Plants in Action. Brian Atwell, Susanne Schmidt, Mark Tester and Paula Jameson had taken on the task of revising the text and soliciting updated sections and new material. Despite a considerable investment of their time, the reality is that the costs of producing a full colour textbook have escalated beyond our means. On behalf of the society, I would like to thank them for their efforts. A discussion of how we can proceed will occur at the council meeting and AGM and we welcome your views and feedback to help guide our decisions.

John Evans

State of Affairs -- ACT

The Australian National University

A New Look for Plant Sciences at ANU

A new Research School of Biology and Division of Plant Sciences

Plant Sciences research at ANU is undergoing a structural reorganisation as a result of the formation of the new **Research School of Biology (RSB) (**<u>http://biology.anu.edu.au/</u>). Formerly, Plant Science researchers have been located in the Research School of Biological Sciences, the Department of Biochemistry and Molecular Biology (BaMBi) and the Department of Botany and Zoology (BoZo). However, a new model for ANU organization will bring these three groups of researchers together into a single new School. This means that the former departments and schools will cease to exist in 2010 and a new **Plant Sciences Division** within **RSB** will be the focal point for Plant Sciences research activities. Professor Murray Badger will be the new Head of the Division.

The Plant Sciences Division encompasses research which is focussed on four broad areas.

- Photosynthesis, photobioenergetics and plant energy biology
- Plant Environmental biology, functional ecology and global change
- Plant-microbe interactions
- Plant genomics, development and bioinformatics

Within these four areas the research addresses a wide spectrum of topics of fundamental importance to explaining plant function and performance in both natural and agricultural environments. Understanding how plants perform in response to global climate change, both now and in the future, is an important theme, particularly with respect to changing environmental stress factors. The research is distinctively integrative, using a wide range of approaches, spanning from genomic and molecular technologies to studies of plant biochemistry, physiology, ecology and evolution.

Plant Sciences Research Leaders

The research lab leaders of the new Plant Sciences Division and their general research interests are as follows. Highlights on some of the research undertaken by the lab leaders are listed below.

Owen Atkin	The importance of plant respiration in determining the scale and
	magnitude of future global environmental change
Murray Badger	Photosynthetic functional genomics of plants, algae and cyanobacteria
Marilyn Ball	Ecophysiology of salinity, freezing and high temperature tolerance
Fred Chow	Chloroplast thylakoid structure and function
Roderick Dewar	Application of statistical mechanics to several problems in environmental
	biology and beyond.
Michael Djordjevic	Plant development and stem cell biology, bioactive molecules and biofuel
	production
John Evans	Physiology of photosynthesis; interactions with nitrogen

Graham Farquhar	Coordination of CO_2 fixation and transpiration in plants - biophysics of
	CO_2 and water exchange between plants-soil and atmosphere
Adrienne Hardham	Cell biology of plant pathogenesis by fungi and oomycetes
Warwick Hillier	Oxygenic photosynthesis
David Jones	Fungal disease resistance
Josette Masle	Root:shoot communication, stress sensing and plant development under abiotic stresses
Ulrike Mathesius	Communication in plant-microbial symbioses
Anthony Millar	MicroRNA control of gene expression involved in Arabidopsis
-	development
Barry Pogson	Carotenoids, photosynthesis and drought
Dean Price	Analysis of photosynthetic CO ₂ acquisition by cyanobacteria
John Rathjen	Signal transduction mechanisms in plants
Michael Roderick	Interactions between water, carbon, energy and the environment
Iain Searle	Epigenetics and reproductive biology of plants
Peter Solomon	Functional genomics of plant-pathogen interactions
Susanne von Caemmerer	Photosynthetic CO ₂ fixation and water loss of leaves
Georg Weiller	Computational biology
Spencer Whitney	The photosynthetic CO ₂ -fixing enzyme, Rubisco
Richard Williamson	Cell wall biology
Thomas Wydrzynski	Photosystem II and protein engineering

New collaborative grouping at ANU with a focus on plant-microbe interactions

In the first years of its establishment, the Research School of Biological Sciences (RSBS) at the Australian National University consisted of six departments spanning a wide range of studies in plant and animal biology. One of the six was the Department of Developmental and Cell Biology led by Professor Denis Carr. After operating with this structure for about 17 years, in 1987 the departments were disestablished and RSBS was reorganized into nine Groups. During this process, the Department of Developmental and Cell Biology, under the leadership of Professor Brian Gunning and without change of constituent staff, became the Plant Cell Biology Group (PCB). Twenty-two years later, as part of the formation of the new RSB as described by Murray Badger above, on the 31st December this year, PCB will cease to exist and PCB staff will join other plant scientists in a new Division of Plant Sciences.

These changes of organizational structure within the RSB provide an opportunity for the formation of new, more informal collaborative groupings within each of the three intra-RSB Divisions. Seven lab leaders in the RSB will thus form a new Plant-Microbe Interactions Theme within RSB. Four of the seven senior staff, Prof. Adrienne Hardham, Dr David Jones, Dr Peter Solomon and Dr John Rathjen are currently in PCB. We will be joined by Dr Michael Djordjevic (ex-Genome Interactions Group of RSBS), Dr Ulrike Mathesius (ex-BaMBi) and Dr Celeste Linde (ex-BoZo). Together, research within the Plant-Microbe Interactions Theme will focus on the molecular biology underlying interactions of fungal and oomycete pathogens with resistant and susceptible plants, evolutionary and population biology of fungal phytopathogens and the interactions between Rhizobia and their plant hosts. Our research programs are described in a little more detail below.

Professor Adrienne Hardham - Infection of plants by fungal and oomycete pathogens

The aim of our research is to elucidate the cellular and molecular basis of the infection of plants by fungi and oomycetes, with a particular focus on rust fungi and oomycetes in the genus *Phytophthora*. As part of their pathogenicity strategies, rust fungi and *Phytophthora* species secrete effector proteins that enable plant infection by facilitating host penetration, suppressing plant defence and orchestrating changes in host cell organization and metabolism. Our research is directed towards identifying and determining the function of key pathogen effectors. We employ a fully integrated approach, using the latest techniques in molecular genetics and genomics, transcriptomics, proteomics and advanced microscopy. For more details of current projects, please go to http://www.rsbs.anu.edu.au/Profiles/Adrienne_Hardham/.

Dr. David Jones - Plant disease resistance

The main focus of our research is to determine the molecular mechanisms underlying fungal effector recognition in resistant plants and to exploit this knowledge to improve breeding for plant disease resistance. This includes studying the mechanism of entry of fungal effector proteins into plant cells in collaboration with Adrienne Hardham, and Jeff Ellis and Peter Dodds of CSIRO Plant Industry. The work on effector uptake focuses specifically on the uptake of rust effectors but the work on effector recognition also focuses on the recognition of extracellular effectors in the tomato-leaf mould (*Cladosporium fulvum*) and the tomato-fusarium wilt (*Fusarium oxysporum f. sp lycopersici*) pathosystems. We use a range of advanced molecular and microscopic techniques including the production and imaging of fluorescent fusion proteins to study effector/resistance protein localisation and interaction. For more information go to http://www.rsbs.anu.edu.au/Profiles/David_Jones/.

Dr. Peter Solomon - Functional Genomics of Plant-Pathogen Interactions

My lab is focused on understanding how fungal pathogens are able to cause disease on wheat. The main pathogen of interest is *Stagonospora nodorum* which is responsible for approximately \$108 million in yield

losses in Australia. It has been commonly thought that pathogens such as *S. nodorum* simply secreted a battery of lytic enzymes that lysed the plant cells preceding fungal growth, providing the nutritional requirements for the pathogen. Recent studies by my lab and collaborators (Richard Oliver, Richard Lipscombe, Tim Friesen) have now shown that the disease is much more complex than originally thought. We have shown that the fungal pathogen secretes effector proteins that are taken up by the host cells and interact in a gene-for-gene manner to cause disease. The mode-of-action of these effectors remains unknown and projects in my lab are now ongoing using proteomics, metabolomics and cell biology to better

John Rathjen, Celeste Linde, Peter Solomon, David Jones and Adrienne Hardham (left to right)

understand the nature of these proteins. My lab also has a strong interest in understanding the mechanisms of fungal sporulation. S. nodorum is a polycyclic pathogen meaning that it must go through several successive rounds of sporulation to cause yield losses. Modern functional genomics techniques have been used to dissect the sporulation process identifying key metabolites and genes that are required for the fungus to sporulate and cause disease. Studies are now underway trying to exploit this information and develop efficient anti-fungal strategies. Projects are available in any of the above areas Further information project and others. and opportunities are available at http://www.rsbs.anu.edu.au/ResearchGroups/PCB/profiles/Peter_Solomon/index.php

Dr. John Rathjen - Signal transduction mechanisms in plants

My lab works on all aspects of plant immunity, studying pathogenesis of the bacterium *Pseudomonas syringae* on plants. We study the earliest events of plant-microbe interaction, notably how the plant detects conserved bacterial molecules known as PAMPs, and the signal transduction events that transform these recognition events into immunity. We have made seminal discoveries on how the bacteria seek to shut down plant immunity using their compliment of virulence effector molecules, and how in turn the plant has evolved sophisticated molecular mechanisms to trap these effectors and activate defenses. My laboratory is a leader in the area of plant signal transduction, with an emphasis on protein biochemistry. Recently we have started an exciting new collaboration with CSIRO and the University of Sydney to use next-generation sequencing to characterize the genomes of wheat stripe rust, a severe pathogen on wheat crops worldwide. We will use these data to identify rust effector proteins to describe their roles in pathogenesis of this important fungal pathogen.

http://www.rsbs.anu.edu.au/Profiles/John_Rathjen/

Dr. Ulrike Mathesius - Manipulation of root development by soil microbes

Our research is aimed at understanding how plant symbionts (nitrogen fixing bacteria) and parasites (root knot nematodes) induce the development of new root organs, root nodules and galls, respectively. We found that microbes manipulate the plant auxin balance by interfering with auxin transport, accumulation and response, which is required to alter root development. This is similar to what happens during the formation of the evolutionarily older lateral roots, except that organogenesis is triggered by an external signal. Using a combination of genomics, proteomics, transport assays and microscopy of tagged proteins in the root, we are currently investigating the molecular mechanisms that are involved in the auxin transport and response changes in response to the further microorganisms. For details please see http://www.anu.edu.au/bambi/people/academic/mathesius.php.

Dr. Celeste Linde - Evolution of plant pathogens

Pathogens evolve. For example, they evolve to adapt to new deployment strategies such as resistance genes or fungicides or to new hosts facilitating host shifts. We use population genetics to understand the dynamics of pathogen evolution in both agricultural and natural systems. Various factors affect pathogen population structure eg. gene flow (migration), mating/reproduction system, genetic drift, selection, mutation. Using a combination of various molecular and phenotypic markers we investigate mechanisms involved in evolution of virulence, host shifts, population expansions and contractions, co-evolution and speciation.

Dr. Michael Djordjevic

Organogenesis of lateral organs in legume roots

A major research focus in my laboratory is to understanding how lateral root and nodule organogenesis is controlled in *Medicago*. The control of lateral organs in roots is important to better understand how

legumes can be manipulated to better adapt to the Australian environment. This is important as legumes underpin major agricultural productivity systems in Australia. As a model system we have studied an *in vitro* system for generating roots or somatic embryos and applied proteomic, transcriptomic and quantitative real time PCR approaches to identify key organ-specific genes: transcription factors, microRNAs, peptide signalling molecules and other genes that are likely to play key developmental roles. We want to understand how these genes control root formation or in vitro somatic embryogenesis in Medicago truncatula. We are also applying similar strategies to identify specific genes involved lateral root or root nodule formation and we aim to determine the function of these genes.

Biofuels research and characterisation of new bioactive molecules from plants

We are exploring the use of microalgal biomass as a source of renewable energy. We study the model algae, Chlamydomonas, to study and optimise the pathways responsible for the accumulation of oils suitable for biofuels production.

In collaboration with Professor Parish at the John Curtin Medical School we are determining the mode of action of plant bioactive molecules (Nod factors and other unknown molecules) that also modulate the process of blood vessel formation in mammals. These molecules have therapeutic potential in wound cardiovascular details healing, disease and cancer. For more see: http://biology.anu.edu.au/Labs/Djordjevic/ or http://www.rsbs.anu.edu.au/ResearchGroups/GIG/index.php

Mathesius lab: From Left to right: Anton Wasson (PhD student), Karsten Oelkers (PhD student), Chooi Hua Goh (Lab Technician), Cassandra Harris (Lab

Technician), Ulrike Mathesius (ARC Research Fellow), Britta Winterberg (Post Doctoral Fellow), Samira Hassan (Lab Technician).



Dr. Michael Djordjevic

Carotenoids, photosynthesis and drought - Professor Barry Pogson

Regulating pigment composition and plant architecture

Fruits and vegetables are an essential dietary requirement for humans and provide carotenoid-derived micronutrients, which promote health benefits including antioxidant activity, precursors for vitamin A biosynthesis, and prevention of macular degeneration of the eye. Animals do not produce carotenoids, so they have to gain them from their diet. Not all colours are due to carotenoids, so not all plants provide an adequate source of carotenoids, especially staple foods such as rice. Consequently, Vitamin A deficiency is one of the leading causes of death and blindness in developing countries.

Prof. Barry Pogson gave Dr Chris Cazzonelli, a member of the Pogson lab in the ARC Centre of Excellence in Plant Energy Biology, the challenge of finding what limits the production of carotenoids in plants and it wasn't long before he had identified a chromatin-modifying gene, SET DOMAIN GROUP 8 (SDG8). He found that it regulates carotenoid composition as well as flowering time, seed set, germination, root development and shoot branching. Dr Cazzonelli says, "Loss of function of SDG8 limits production of a carotenoid called lutein, which prevents age-related macular degeneration of the human eye. Furthermore, we have found preliminary evidence of novel hormonal-like roles for carotenoids in the differentiation of plant stem cells into roots, leaves and flowers." The essential roles of carotenoids, as well as the chromatin-modifying nature of SDG8, have opened a new door towards understanding epigenetic regulatory mechanisms that control plant development.

Drought tolerant plant gene discovered

An international group of plant scientists, led by Dr Gonzalo Estavillo and Professor Barry Pogson at The Australian National University have discovered a subtle mutation in Arabidopsis, a small, rapid growing plant, which may have important and far reaching implications for establishing drought resistance throughout the plant kingdom.

One potential the group is currently exploring is the application of the mutation to food crops such as rice or wheat, and the researchers will now begin to introduce the mutant characteristics into the elite wheat cultivars currently used in agriculture industry.

"The ultimate aim of the project is to develop wheat lines with improved drought tolerance and water use," explained Dr Estavillo. "The next step will be to identify wheat mutant plants lacking SAL1 genes identified by molecular biology procedures. We expect that these mutants should remain green, turgid and photosynthetically active, producing more leaves, flowers and seeds during mild to moderate water deficit."

Estavillo points out that with most climate models predicting that the vast wheat growing areas of southern Australia will become drastically drier over the next fifty years the prospect of drought resistant wheat offers much promise for ensuring long term food supply and economic wellbeing. This has been recognised by the Grains Research and Development Corporation, which recently provided further funding for Dr Estavillo and Professor Pogson to identify genetic variants of the SAL1 gene in wheat, in conjunction with CSIRO Plant Industry.

The SAL1 mutation also has the advantage of facilitating less controversial solutions to the enhancement of food crops. Because the basis of the mutation is a missing gene it would also be potentially possible to create drought tolerance in a plant like wheat without employing transgenic methods.

Functional Ecology Group, Research School of Biology - Professor Marilyn Ball and Dr Owen Atkin

In the Functional Ecology Group, we study the ecophysiology of plants, with emphasis on understanding the evolution and adaptation of plants to environmental factors, and how such adaptations relate to the structure and function of vegetation along complex environmental gradients. A combination of laboratory and field-based studies are used to quantify the fundamental physical and physiological processes underpinning plant responses to multiple environmental factors. We seek to integrate:

1. Stress physiology: understanding how attributes associated with increasing stress tolerance relate to inter-specific differences in growth and morphology of plants along complex environmental gradients.

2. Whole plant bioenergetics: understanding how respiration and carbon balance vary in response to environmental stresses, and how they relate to inter-specific variation in growth and structure of plants along complex environmental gradients.

3. Functional plant morphology: understanding the integration of transport processes with the structure and function of vegetation along complex environmental gradients.

Overview of Marilyn Ball's research

The Australian flora provides a unique opportunity for study of the biodiversity of adaptation to temperature stress because closely related species within single genera (e.g. Eucalyptus, Acacia, Poa) cover large areas with diverse environments. For example, populations of the snow gum (Eucalyptus pauciflora) are distributed naturally along altitudinal temperature gradients from sea level to the alpine tree line. Studies in other systems have found that the occurrence of freeze/thaw-induced embolism increases with xylem conduit diameter, with the implication that increasing resistance to freeze/thaw-induced embolism comes at the expense of hydraulic capacity. We are using the snow gum as a model system to explore how variation in hydraulic architecture within a species relates to freezing tolerance and the gas exchange characteristics of leaves along an altitudinal gradient.

The long-lived leaves of temperate evergreens must affect a compromise between summer tolerance of drought and high temperatures and winter tolerance of freezing. Both extremes involve tolerance of water stress due to dehydration of tissues and embolism of conduits. Changes in hydraulic architecture could reduce the incidence and severity of embolism, but at the expense of water transport to leaves. Similarly, changes in the display and properties of leaves could minimise the expenditure of water and reduce exposure to temperature extremes, but at the expense of light interception and carbon gain. We are studying relationships between structure and function in native Australian plants along complex gradients in temperature and rainfall with the goal of understanding relative costs and benefits to plant performance of different strategies of temperature tolerance. These trade-offs have implications for the morphology and function of leaves that might constrain carbon gain and affect the capacity of evergreen plants to respond to climate warming.

Another major focus of work is the functional diversity of salt tolerance in mangroves. Growth in saline environments requires a complex balance between carbon gain in relation to water loss and associated ion uptake. In mangroves, water use by leaves becomes increasingly conservative with increase in salinity and with increase in the salt tolerance of the species. We are studying how the water use characteristics of mangrove leaves growing along complex gradients in salinity and aridity relate to their morphology and the hydraulic structure of stems, and to the transport of ions in the xylem sap. We are using energy dispersive x-ray analysis of tissues visualised by cryo-scanning electron microscopy for quantitative elemental mapping of key ions in relation to water transport through root, stem and leaf tissues. The goal is to gain a better understanding of how uptake and storage of ions varies with salinity, and with mangrove species differing in salt tolerance. This fundamental work is being integrated with ecophysiological studies to link physiological processes with morphological constraints on leaf function in mangroves.

Overview of Owen Atkin's research

Climate-mediated changes in leaf respiration (R) are now accepted as an important component of the biosphere's response to global climate change. Plant R releases near ten times more CO_2 into the atmosphere than does the burning of fossil fuels. Therefore, variation in plant R has the potential to affect the extent to which atmospheric CO_2 will be sequestered by the terrestrial biosphere. Moreover, respiration produces the energy and carbon intermediates necessary for biosynthesis and cellular maintenance and consequently affects the functioning of individual plants and ecosystems. It is crucial, therefore, that we improve our understanding of the impacts of climate and environmental gradients on plant R. Because of this, our aim is to quantify the climate dependence of plant R, and determine the impact of variations in respiration on rates of net carbon uptake in a range of contrasting ecosystems over wide spatial and temporal scales.

Our research is assessing the impact of nutrient gradients, variations in water availability and changes in atmospheric CO_2 concentration on plant respiration. As part of this work, we are combining laboratory and field observations to establish if there are systematic patterns among contrasting plant species in how plant respiration responds to environmental gradients. These studies point to preservation of mitochondrial respiration under water stress to be vital in helping plants survive periods of drought and enable rapid recovery of productivity when soil moisture increases. Our work has also revealed that seasonal adjustments in the temperature response curves of R play a crucial role in determining the viability of tree growth in low productivity forest ecosystems. Moreover, we have shown the impact of thermal history on scaling relationships used to predict R being highly predictable. This finding enabled us to quantitatively incorporate thermal acclimation of R into the same coupled global climate-vegetation model used by the Hadley Centre in the UK. We are now working with modellers at the Hadley Centre/Met Office in the UK to further improve the representation of plant R in global climate-vegetation models.

Finally, we are investigating the physiological and biochemical basis of thermal acclimation of respiration to long-term changes in temperature. Our work points to temperature-mediated changes in enzymatic capacity playing a crucial role in thermal acclimation of plant R . We have used confocal imaging of GFP mitochondria and electron microscopy to visualize the impacts of cold acclimation on mitochondrial density, size and location within leaf cells; this work has highlighted the heterogeneity of mitochondrial populations in leaves and the extent to which long-term changes in growth temperature have differing effects on mitochondrial structure of contrasting cell types. More recently, using a novel development of the 18O discrimination method, we have also shown that activity via the alternative pathway increases transiently during the early stages of cold acclimation, but then decreases following development of leaves in the cold.

CSIRO Plant Industry (Black Mountain)

Genomics and Plant Development (compiled by Dr. Frank Gubler)

The Genomics and Plant Development Program, under the direction of Dr Frank Gubler, uses a broad range of capabilities, including molecular biology, genetics, genomics, bioinformatics, physiology and cell biology to tackle important questions in plant development and gene regulation. Studies in model plants

like Arabidopsis and rice inform new research extending to crop species of agricultural significance to Australia, primarily cereals (wheat and barley), canola and cotton. The core activities are focused on applying new technologies to deliver increased yield (apomixis, heterosis, and understanding plant reproductive biology), new mechanisms of gene control for biotechnology (small RNAs and epigenetic regulation), developing tools and traits for cotton improvement (fibre quantity and quality, marker assisted breeding), and bioinformatics and microscopy to support the above.

Apomixis and Heterosis.

Led by Dr Anna Koltunow, this group which is split between Canberra and Adelaide uses both model plants like Arabidopsis and the weedy *Heiracium* to study various aspects of seed development to understand the process of

Dr. Frank Gubler

apomixis whereby clonal or asexual seed production can occur with or without the process of pollination. Comparisons between apomictic and non-apomictic *Heracium* plants, for example, and detailed gene expression analysis in different cell types surrounding the egg cell are starting to unravel the molecular mechanisms for apomictic seed production and it is hoped that this could be applied to producing apomictic crop plants, particularly to fix the genetic gains that can be made through hybrids in many crops. Hybrid vigour (heterosis), whereby the F1 between two different inbred plants can have greater agronomic performance (more and bigger seeds, leaves etc) could have important applications in many crops but is limited by the high cost of hybrid seed production. The group led by Dr Liz Dennis is studying the molecular changes in gene expression in reciprocal hybrids between different Arabidopsis ecotypes and comparing them to their parents. This involves using deep sequencing, microarrays, small RNA and methylation analysis on a genome wide scale to define the genes that generate the hybrid vigour, opening up the possibility of generating hybrids in the future without the need for crossing.

Reproductive Biology

This is an area that targets aspects of cereal reproductive biology from flowering (timing), fertilization, spike structure, through to grain dormancy in wheat and barley. These traits largely impact on yield and are very important for protecting Australia's food security.

Small RNA and Epigenetics

The realisation that the large non-coding parts of plant and animal genomes were not "junk DNA" but had important regulatory functions in development and responses to biotic and abiotic stresses has sparked a global interest in their roles and potential uses in gene regulation. The CSIRO group under Dr Jean Finnegan, Dr Ming-Bo Wang and Dr Chris Helliwell are carrying out both basic and applied research to understand how the small RNAs (microRNAs, siRNAs etc), long non-coding RNAs are involved in regulating aspects of flowering, disease and other stress responses as well as designing tools that could be used by biotechnologists to use these systems to control specific genes or developmental pathways. They are also involved in understanding the role of both DNA methylation and chromatin modification in plant development using both ChIP (Chromatin immunoprecipitation) analysis and global methylation profiling particularly to study the cold regulated induction of flowering known as vernalization.



Cotton Biotechnology

Australia is an important exporter of cotton fibre to the textile industries of Asia and has moved largely to using GM varieties with insect and herbicide tolerance. CSIRO is now the sole provider of germplasm to the Australian industry and has an active and successful conventional and GM breeding program based in Narrabri, NSW with the Canberra labs providing all the molecular support for tracking transgenes through breeding populations and in providing quality control on breeding material prior to hand over of seed to a seed producer for bulk-up and sale. These commercial activities are supported by a trait and tool development pipeline to produce the next generation of traits that affect yield and fibre quality and for the discovery and application of markers to assist with breeding difficult traits like fungal and viral disease resistance in cotton. Developments and technologies in other parts of the Program are flowing through to cotton that provides a clear pathway to commercialisation. Of particular focus is the development and maturation of the cotton fibre on the seed and the group under Dr Danny Llewellyn are using printed cDNA and commercial Affymetrix arrays to probe global gene expression changes in a range of different cotton germplasm and mutants to identify transcription factors involved in the initiation of the cotton fibre (an elongated epidermal cell of the seed) as well as during the elongation and secondary cell wall thickening stages that determine the length, strength and fineness of fibres, all important commercial attributes for yarn and textile production. Silencing and over-expression of specific MYB and Homeodomain transcription factors in transgenic cotton, for example, is helping to understand the regulatory networks controlling fibre initiation and elongation and identifying new targets (other transcription factors and biochemical pathways) for future analysis.

Bioinformatics

With the explosion in data production from advances in chip technologies, including microarrays, ChIPon-chip, genome wide methylation analysis, tiling arrays, high throughput sequencing, high throughput SNP genotyping etc, the role of bioinformatic analysis has become central to many areas of modern plant science and many wet-lab scientists are having to become conversant with bioinformatic tools. Under Dr Jen Taylor CSIRO PI has developed a world-class bioinformatics team who work closely with biologists to develop pipelines for all sorts of microarray analysis, high throughput sequencing analysis of transcript abundance and small RNA abundance and the discovery of novel microRNAs, as well as for the analysis of genome wide methylation patterns following bisulfite treatment of DNA, genome assembly and SNP detection, genome browsers for displaying genomic data, mutant analysis and gene cloning, and metagenomics of environmental samples for microbial population studies.

Microscopy

This is a core capability across the site and central to all aspects of cell biology and gene function analysis and is led by Dr Rosemary White. A state-of the art facility provides both conventional microscopy with confocal and FTIR microscopy and environmental SEM and advanced image analysis to support all of the activities in cell biology including conventional structural analysis, particularly during reproductive development, localisation of proteins using GFP tagged reported constructs in transgenic plants, immuno-localisation of transcripts and proteins and jointly through the ANU laser capture microdissection for a variety of applications in gene expression analysis and cDNA library production from single cell-type and small tissue samples.

High Performance Crops for Australia (compiled by Dr Richard **Richards**)

Drought (compiled by Dr Richard Richards)

Lack of water limits crop growth and yield worldwide. Research to understand physiological factors that alter crop water use, water use efficiency and harvest index to plant improvement and crop agronomy is

active at CSIRO Canberra. Research is exploring the extent of genetic variation for both above and below-ground factors influencing yield under drought, the nature of the genetic control, specific genes or chromosomal regions that underpins this variation and the specific effect if this variation on growth and yield in water limited environments. Our aim is to introduce new levels of variation for important physiological traits into new wheat cultivars for Australia. We are currently researching traits involved with grain sterility, hormonal regulation of growth, the development and growth of leaves and roots, the efficiency of the exchange of CO₂ for H₂O and the partitioning of carbon to roots, stems and grain.

Cereal Growth (compiled by Dr Peter Chandler)

We have projects aimed at improving the early emergence and growth of wheat. Several new semi-dwarfing loci have been mapped, characterised and introduced into standard varieties. These loci have the advantage that dwarfism is not seen at early growth stages, so emergence and early vigour is better than in varieties containing Rht1 or Rht2. Lines with long coleoptiles and high early vigour have also been isolated, and the traits introduced into standard wheat varieties. Many 'overgrowth' mutants have been isolated following mutagenesis of severely dwarfed wheat. These are lines which still retain the original dwarfing mutation, but which have improved growth. Another trait of interest is reduced tillering (branching), and near-isogenic lines for the mutant tin (tiller inhibition) gene have been developed for assessing the agronomic potential of this trait as well as for detailed mapping, and eventually to identification of the gene responsible.

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Dr. Peter Chandler

Soil nutrients and toxicities (compiled by Dr Manny Delhaize, Dr Peter Ryan)

The objectives of our group at CSIRO "Soil nutrients and toxicities" are to improve plant production on hostile soils and especially soils that are acidic and deficient in phosphorus. We are increasingly interested in the broader influences of root-soil interactions and how root exudates, in particular, can affect wider aspects of plant growth by improving nutrition and providing protection from stresses.

Acid soils pose an impediment to sustainable food production globally but especially in many third world

countries. Aluminium and manganese toxicities represent the main limitation to plant production on acid soils and we have been identifying mechanisms that allow some plants to tolerate these stresses better than others. Our previous work demonstrated that wheat has at least two mechanisms of aluminium resistance which rely on malate and citrate efflux from their roots. These processes are controlled by members of different gene families suggesting that convergent evolution has been occurring for this trait in wheat. We identified the genes conferring this resistance, TaALMT1 and TaMATE1, and







demonstrated that they encode membrane proteins that facilitate organic anion transport. Studies from our group and others have now shown that similar genes confer aluminium resistance in a wide range of species in the same manner. We have also isolated genes essential for manganese tolerance in plants. These also encode membrane proteins that transport manganese ions, this time from the MTP family. Further work on this topic will establish the value of these genes to agriculture through their use as molecular markers and through transgenic approaches. Our collaborators for different aspects of this work have included Yoko Yamamoto' lab (University of Okayama); Steve Tyerman (Adelaide Univ), Harsh Raman (NSW DPI, Wagga Wagga), and Leon Kochian (Cornell Univ).

The group has a long-standing interest in phosphorus nutrition but this is developing into a major focus for the next few years. The group is particularly interested in the mechanisms by which some plants are able to extract more phosphorus from the soil than others. We developed a screen to assess the natural variation of phosphorus-use efficiency (PUE) in wheat and used this procedure to identify QTLs for PUE in two mapping populations. We found that phosphorus efficiency in wheat is a complex trait controlled by several genetic loci. We are trying to identify molecular markers for PUE for breeding purposes and hope to target key root traits to increase PUE with genetic engineering.

CSIRO Root architecture and rhizosphere processes team (Dr. Michelle Watt)

Dr Michelle Watt leads a team of researchers focused on increasing the productivity of wheat in conservation farming systems by improving the root systems and their associations with the soil biology. Wheat is the world's most important temperate crop and global demand will exceed current annual yield growth by 2020, with concurrent declines in agricultural land and water. Because roots are below ground and inaccessible, there are many discoveries yet to be made for crop improvement. The research activities in Dr Watt's team include (1) using non-invasive micro and macro imaging methods to understand how roots grow and interact with the soil in the field, (2) applying that knowledge to select for beneficial traits such as greater root vigour to access deep water, and root exudates to promote a beneficial rhizosphere microflora, and (3) testing novel wheat germplasm with beneficial traits in the field to validate for greater productivity, and eventual delivery to farmers. At CSIRO, we work with breeders and geneticists to develop new wheats and identify the genes associated with the traits for molecular breeding, agronomists for field validations, and microscopists and imaging experts to understand and track roots in field soils. Recently the team has started to apply the newly-sequenced grass model Brachypodium to the research- this grass is a miniature of wheat (the Arabidopsis for wheat) that will give us access to genes more quickly and allow mature root system studies in controlled conditions for the first time. Dr Watt's team has included Australian and international students and post doctoral fellows, and we have projects with the Grains Research and Development Corporation in Australia, the USDA-ARS in California, the Lawrence Berkeley Laboratories in California, and with the Indian Centre for Agricultural Research through the Australian Centre of International Agricultural Research.

Yield Potential, Photosynthesis and Phenomics (compiled by Dr Robert Furbank)

Bob Furbank and his team work in the newly commissioned High Resolution Plant Phenomics Centre at the CSIRO Phytotron. The High Res Centre is the Canberra node of the Australian Plant Phenomics Facility and operates in collaboration with the ANU and University of Adelaide. The Phenomics group are using non-invasive measurements such as chlorophyll fluorescence, digital growth imaging, infrared thermography and hyperspectral reflection to measure performance and yield of cereals, dicot crops and model species such as *Brachypodium* and *Arabidopsis* in controlled environments and the field. Recent major projects include participation in the C_4 rice initiative and an international collaboration with the USDA to use *Brachypodium* as a model to identify genes controlling key traits for yield and abiotic stress tolerance.

Genetic Engineering for Plant Improvement - Dr Jeff Ellis

Lr34 resistance - Dr Evans Lagudah

Durable disease resistance that defends a plant against multiple threats from within is highly desirable in commercial crops like wheat. Improved control of fungal rust diseases in cereal crops is critical not only to the financial survival of Australian farmers, but to food security worldwide. CSIRO Plant Industry's Dr Evans Lagudah and his team, in collaboration with the University of Zurich and the International Maize and Wheat Improvement Center (CIMMYT), have identified the wheat gene which provides resistance against leaf rust, stripe rust and powdery mildew. The gene, called Lr34, is the first of its kind to be identified in a commercial crop rather than a model plant. Usually one gene protects against only one disease, but tests conducted after identifying Lr34 gene sequence showed that it has provided partial but constant protection against leaf rust for over 80 years. The identification of Lr34 is intriguing because it has established itself as a durable and long-lasting defence mechanism found in wheat crops from around the world. Understanding the molecular nature of this type of resistance has important

implications for long term control of rust diseases. The wheat genome is large and complicated making specific gene identification difficult. The team successfully identified the location of Lr34 using the latest genetic techniques. Although the approximate location of the gene was previously known, Dr Lagudah and his collaborators have now been able to refine the location of Lr34 on the complicated wheat genome. This enabled them to obtain the exact sequence of this gene. After identifying the Lr34 gene sequence, the research team searched wheat lines from around the world and found Lr34 in the earliest cross-bred varieties first released in Italy at the beginning of last century. In Australia, Lr34 has been found in south-east Australian wheats but not in commercial cultivars released in South Australia or Western Australia. Future research will focus on how Lr34 works to confer resistance to disease. In the meantime, current research findings have been shared with WA and SA scientists and are being used towards incorporating Lr34 resistance into wheat varieties in those States. This work was supported in Australia by the Grains Research and Development Corporation.



Dr. Evans Lagudah

Dr. Chris Howard - Bulahdelah Bypass Orchid Conservation Project

The Bulahdelah Bypass Orchid Conservation Project aims to address issues for the survival, sustainability and translocation of three listed threatened orchid species *-Cryptostylis hunteriana*, Rhizanthella slateri and Corybas dowlingii (Fig. 1) - that are present within and adjacent to the proposed Pacific Highway Upgrade on the foot-slopes of Alum Mountain at Bulahdelah. The research is being funded by the NSW Roads and Traffic Authority and is being undertaken at the CPBR.

This research involves

- an examination of the floristic characteristics associated with each orchid species
- investigation of the pollination syndromes and species involved for each orchid species
- the location, hand pollination and collection of seeds of the three threatened orchid species at Bulahdelah
- the isolation, identification and establishment of the nature of the mycorrhizal relationships with each orchid species for seed germination

Using these data, alternative sites on Alum Mountain will be located for the initial translocation of plants directly affected by the construction of the Bulahdelah Bypass (Fig. 2). In addition, translocations will be performed with any *in vitro* propagated plants of all three species.

Dr. Joe Miller - The plant genus Acacia

The plant genus *Acacia*, with over 1,000 taxa, consists of shrubs and trees that are dominant throughout Australia. The CPBR has a major research project to develop a molecular phylogeny of *Acacia* to clarify questions of taxonomic circumscription, relationships, distribution, nomenclature, and ecology. The data sets and resulting phylogeny facilitate investigation of questions relating to morphological evolution, patterns of diversification, and biogeography.

In 2008 seed from over 300 *Acacia* species was germinated and plants are now growing in glasshouses and more are set to be germinated in the coming months. The seedlings are being measured for several characteristics, particularly the sequence of leaf development from pinnate to bipinnate leaves. DNA has been extracted from these, and from living plants at the ANBG, to be used in the DNA sequencing project. Over the past year we have sequenced six DNA regions for over 300 *Acacia* species and the work will progress to sample all available species. The analysis of this large dataset indicates new insights into the evolution of *Acacia*.

In conjunction with this research, a collaborative network of CSIRO and University scientists has been formed to integrate *Acacia* evolutionary studies with investigations of organisms that interact in some way with Acacia. These organisms include insects such as thrips and weevils, as well as rusts and rhizobia. This work will result in comparative phylogenetic studies of the interacting organisms.

Mr. Brendan Lepschi - The Australian Plant Name Index and the Australian Plant Census – plant names in the digital age

Keeping track of taxonomic and nomenclatural changes for any group of organisms can be a challenge. Greater understanding of evolutionary relationships, increasingly possible through new data sources and more rapid data analysis, almost inevitably results in taxonomic adjustments, accompanied by changes in nomenclature as well.

Australian botanists are fortunate to have a comprehensive listing of all the scientific names known to have been used for flowering plants in the Australian taxonomic literature. The *Australian Plant Name Index* (APNI) was initiated in 1973 by one of Australia's most prominent botanists, Dr Nancy Burbidge, as a precursor to the *Flora of Australia* project. Compiled by Arthur Chapman, the *Australian Plant Name Index* was published in 1991 as a four-volume work covering all relevant names published prior to 1989. The project was subsequently transferred into an electronic format to allow for corrections and continued updating. APNI is now managed by the Centre for Plant Biodiversity Research, a joint venture between the Australian National Botanic Gardens and CSIRO Plant Industry in Canberra.

In its current form, APNI is an on-line database of scientific names for plants used in the Australian taxonomic literature. Primarily focused on vascular plants, it provides detailed bibliographic information (the place of first publication and any relevant subsequent usages), authorship, information on the type specimen/s and some commentary. It does not provide an opinion on taxonomic concepts or indicate whether a name is currently accepted by the botanical community or considered to be a synonym.

There is an identified need for an information source to provide authoritative, reliable information on the currently accepted names for Australian plants. This was highlighted in 2003 when Commonwealth agencies attempted to align legislative schedules for the Environment Protection and Biodiversity Conservation (EPBC) Act with lists used by the States in their own legislation. Lack of congruence between taxonomic concepts and nomenclature between State and Commonwealth listings caused significant problems with these schedules.

In 2004 the Council of Heads of Australasian Herbaria (CHAH), representing all State and Commonwealth herbaria, agreed to produce a database presenting an agreed national view of the

scientific names used for Australian plants. This project, the *Australian Plant Census* (APC), will provide a current name for every vascular plant recognised to occur in Australia and its Territories, building on nomenclatural data already held in the comprehensive APNI database. It will also include all known synonyms (including phrase names and misapplications) and provide information on authors, references to the taxonomic and nomenclatural concepts adopted, geographic distributions, and relevant comments and notes. This information represents a consensus view of the herbaria involved and in some cases will be a compromise between conflicting scientific opinions.

Unlike previous published lists, APC is maintained as a dynamic database, constantly updated as new information is published. APC is accessible during its development, although it should be noted that agreed names are not available for all taxa as not all families have yet been treated. Completed families are indicated on the APC website (click on the "list of families" link). Some large groups such as the Proteaceae and Mimosaceae and Poaceae have been completed, but many major groups remain to be tackled by the project, including the Asteraceae, Fabaceae, non-eucalypt Myrtaceae, and Orchidaceae.

The APC is available at <u>http://www.anbg.gov.au/chah/apc/index.html</u> and the census team genuinely welcomes corrections or feedback on the project– please feel free to contact us via <u>cpbr-info@anbg.gov.au</u>



AoB PLANTS

AoB PLANTS - a new open access journal for plant biologists

Authors are turning in increasing numbers to open access journals to publish their work. The attractions of doing so are several. They include having greater control over copyright, the appeal and flexibility of the latest publishing technologies and, above all, having papers made available without charge worldwide and thus freely available to anyone who wishes to read them as soon as they are published. The newly launched journal AoB PLANTS offers these and other attractive features. It covers all aspects of plant biology, is owned and managed by plant scientists on a not-for-profit basis and is published by Oxford University Press. AoB PLANTS publishes 'Research Articles', 'Points of View', 'Reviews', 'Mini-reviews' and 'Technical Articles'. Submitted papers are evaluated against published minimum criteria for acceptability using a double-blind refereeing system. Papers will appear online within 3-5 days of acceptance and benefit from a full typesetting and proofing service. For an introductory period, there will be NO CHARGE to publish in AoB PLANTS. This creates the ideal opportunity for authors to try the new journal and enjoy the benefits of open access publishing at no cost. For further information contact Mike Jackson, Chief Editor AoB PLANTS, E-mail: mike.jackson@bristol.ac.uk or visit the web site http://aobpla.oxfordjournals.org/.

From Our New PhDs

Our recently completed PhDs who are the future of plant science and our society are encouraged to provide highlights of the research that earned them their new degree. Below is an account by Dr Alice Hayward. Please send me accounts of your research highlights if you have recently completed your PhD. Tina Offler

Interactions between strigolactone and auxin signalling in shoot branching

Alice Hayward

Diversity in plant architecture is largely generated by the post-embryonic regulation of meristem initiation and activity. Axillary meristems located in the axils of leaves give rise to axillary buds, which can either arrest or continue active growth to form axillary branches. A key regulator in this process is the hormone auxin, produced in shoot apical tissues and transported downwards in the polar auxin transport stream to inhibit bud-growth. A second hormone, cytokinin, acts antagonistically to auxin to promote branching. More recently, strigolactones, previously implicated in parasitic weed germination and mycorrhizal associations, were identified as a third class of hormone involved in branching inhibition.

The strigolactone biosynthetic pathway offers a novel target for the manipulation of plant architecture and yield as well as the germination of parasitic weed species that are detrimental to agriculture. My PhD research aimed to improve our understanding of the regulatory network controlling branching in plants. In particular I investigated the transcriptional regulation of strigolactone synthesis genes as a potential



Branching phenotypes of reciprocally grafted WT, *max4* mutant and *bdl* mutant plants (shoot/rootstock; Hayward *et al.*, 2009). WT roots can rescue branching in *max4* mutant shoots while *max4* and *bdl* roots can not, suggesting perturbed strigolactone production.

point of functional interaction between auxin and strigolactone signalling in branching control.

Using quantitative real-time PCR, the transcript abundance of the Arabidopsis strigolactone synthesis genes *MAX3* (MORE *AXILLARY GROWTH3*) and *MAX4* was found to be positively auxin regulated. This regulation was dependent on the AXR1/TIR1 auxin signalling pathway, which targets Aux/IAA transcriptional repressors for proteasomal degradation.

In particular, correct degradation of the Aux/IAA protein BDL (BODENLOS) was found to be necessary for optimal *MAX3* and *MAX4* expression. Gain-of-function mutants for *bdl* have significantly reduced *MAX4* and *MAX4* expression and increased branching. Previously, grafting studies revealed that the strigolactone branching inhibitor regulated by MAX3 and MAX4 is upwardly mobile. In this study, strigolactone application experiments and reciprocal grafting between max4, bdl and WT plants revealed that the reduction in MAX3 and MAX4 transcripts in bdl mutants is associated with perturbed strigolactone production and the bushy phenotype (see figure). Therefore it was proposed that auxin modulates branching, in part, by positively regulating strigolactone synthesis.

As previously shown for strigolactone synthesis genes in pea and rice, *MAX3* and *MAX4* expression was found to be feedback up-regulated in strigolactone pathway mutants of Arabidopsis. qRT-PCR analysis of *MAX3* and *MAX4* expression in double mutants for *max* genes and the auxin response mutant *axr1* revealed that this feedback regulation is *AXR1*-dependent in the shoot. Interestingly, *max* mutant shoots have increased amounts of auxin travelling in their polar auxin transport streams. Therefore, increased auxin level and/or signalling in conditions of reduced strigolactone signalling likely contributes to feedback regulation of the strigolactone pathway.

Comparative bioinformatic analysis of the MAX3 and MAX4 promoters and the promoters of orthologous genes in pea and rice revealed a number of conserved, putative *cis*-acting regulatory elements that could mediate auxin regulation as well as cross-talk with additional branching cues. In addition to analysing MAX3 and MAX4 transcription, the *max4* mutant in Arabidopsis was complemented with the orthologous gene from pea, confirming their orthology in branching regulation. Overall, my PhD research, combined with results from previous studies, reveals that auxin, cytokinin and strigolactone may control aspects of the biosynthesis, activity, level or distribution of each other via interlocking regulatory loops.

My PhD research was undertaken with A/Prof Christine Beveridge, Dr Fiona Filardo and Prof Peter Gresshoff at the ARC Centre of Excellence for Integrative Legume Research at the University of Queensland in collaboration with Prof Ottoline Leyser and her laboratory at the University of York, UK. I was fortunate enough to carry out a significant portion of my PhD research in York and I would strongly recommend international collaboration and travel where possible to other PhD candidates. This research was funded by an Australian Postgraduate Award, a UQ Graduate School Research Travel Award and a Travelling Fellowship from the Company of Biologists (Development). My PhD results are described in greater detail in the following research paper. Having recently completed my PhD I am now pursuing research into genetic diversity within Brassica species with Dr Jacqueline Batley at the University of Queensland.

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Messages from our SustainingMembers



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Discipline Highlights

Environment & Ecophysiology: Global Change

Eucalypt growth in past and future climates

Oula Ghannoum and David Tissue

Centre for Plants and the Environment, University of Western Sydney, Hawkesbury campus, Richmond, NSW 2753, Australia

Do eucalypt trees operate fundamentally differently today than they did prior to the industrial revolution? Will they operate differently a century from now? These questions provide the motivation for an ARCfunded project conducted by Professor David Tissue, Dr. Oula Ghannoum and their colleagues at the University of Western Sydney.

Background

Research into the response of plants to rising atmospheric concentrations of carbon dioxide ($[CO_2]$) has mainly focussed on the effects of elevated $[CO_2]$ (*e.g.*, doubling of current the current ambient $[CO_2]$). However, current ambient $[CO_2]$ (~390 ppm) represents a 35% increase above the atmospheric $[CO_2]$ which prevailed 200 years ago at the outset of the Industrial Revolution - more than 20% of this increase has occurred in the last 50 years.

The pre-industrial $[CO_2]$ of ~ 280 ppm had persisted during the last 10,000 years. In addition, atmospheric $[CO_2]$ has oscillated between 180 and 300 ppm for the last 1-3 million years. Therefore, the rise in atmospheric $[CO_2]$ since the early 1800s represents a significant change in $[CO_2]$ experienced by C_3 plants (Sage and Cowling 1999). Consequently, studying the response of plants to the recent rise in atmospheric $[CO_2]$ can help us understand their response to a future rise in $[CO_2]$.

Another drawback of many studies on the effects of atmospheric $[CO_2]$ is that they only investigate the direct responses to atmospheric $[CO_2]$ and ignore the fact that CO_2 is an important warming, greenhouse gas. Atmospheric $[CO_2]$ is predicted to approach 600 ppm during the 21st century (Solomon *et al.* 2007). Air temperature has increased about 1°C in the past century and is anticipated to increase a further 2-6°C during the course of this century (Solomon *et al.* 2007). Therefore, it is important to investigate the interactive effects of elevated $[CO_2]$ and high temperature rather than simply the focussing on rising atmospheric $[CO_2]$ in isolation.

The relative scarcity in studies addressing the sub-ambient $[CO_2]$ and the elevated $[CO_2]$ x high temperature aspects may reflect the technical and logistic difficulties associated with these experiments. Therefore, the Centre for Plants and the Environment at the University of Western Sydney (UWS) has set out to establish the research infrastructure required to undertake such important experiments.

This infrastructure includes temperature- and $[CO_2]$ -controlled glasshouses and whole-tree chambers focussing on *Eucalyptus* species. Considerable cost, time and efforts have been invested by UWS researchers in order to optimise each of these research facilities.

In this article, an overview of the inner workings of the glasshouse facility will be presented as well as a summary of results obtained so far. Future articles will cover other exciting research facilities operating at the Centre for Plants and the Environment.

Temperature- and [CO₂]-controlled glasshouse studies

Aim. The long-term goal of these studies is to provide a mechanistic basis for predicting eucalypt tree response to changes in $[CO_2]$ and associated climate change. The research represents the link between controlled environment studies and field-based experiments conducted at UWS.

Glasshouse. The glasshouse facility (shown in picture below) consists of six, naturally-lit glasshouse rooms $(3 \times 5 \times 5 \text{ m}^3, \text{ w x l x h})$ that are computer-controlled to maintain specific air temperature and atmospheric [CO₂].



Temperature control. Air temperature was controlled using a pair of air conditioners and gas heaters connected to each room and interfaced with a central computer. A software programme (PlantVisor, CAREL, Padova, Italy) operated (on-off) both the air conditioner and heater to achieve a temperature set-point.

Three compartments simulated the daily temperature of a 30-year average of a local (Richmond, NSW) day for the months of November to May (i.e. ambient temperature treatment). Three compartments simulated a constant 4°C step increase in temperature relative to the ambient temperature treatment. Temperature was maintained at constant values during the night and midday periods; and it was stepped up and down in two 4°C steps before and after the midday period. The average temperatures for the ambient and high temperature treatments were 26/18 and 30/22°C (day/night), respectively.

 $[CO_2]$ control. Within each temperature treatment, plants were grown at sub-ambient $[CO_2]$ (target 280 ppm, ambient $[CO_2]$ (target 400 ppm), and elevated $[CO_2]$ (target 640 ppm). Sub-ambient $[CO_2]$ was achieved by continuously passing compartment air over trays filled with calcium hydroxide (Schaefer Kalk GmbH & Co KG, Diez, Germany) within metal boxes fitted with fans (Thermoline Scientific, Sydney, Australia); calcium hydroxide was stirred daily and exchanged twice a week. Elevated $[CO_2]$ was achieved by injecting CO_2 gas (Food grade, AirLiquide, Australia) from pressurized cylinders through solenoid valves connected to a CO_2 monitor/controller (Lambda T, ADC BioScientific Ltd., Hoddesdon, Herts, UK). CO_2 was first passed through a Purafil® column to eliminate possible ethylene contamination.

 $[CO_2]$ was continuously monitored in all growth compartments by logging the voltage output of the CO₂ monitors/controllers using a data logger (DL2e, Delta-T Devices Ltd, Cambridge, UK). The CO₂ monitors/controllers were calibrated at regular intervals with pure N₂ and two CO₂ calibration gases (406 ± 12 and 714 ± 16 ppm) (AirLiquide, Australia). The average day-time $[CO_2]$ during the experimental period for the sub-ambient, ambient and elevated treatments was 290, 400, and 650 ppm, respectively.

Plant culture: Soil was collected from the A horizon of an experimental site located on the grounds of UWS, Richmond, NSW, Australia. The soil is a loamy-sand with low organic matter content, fertility and low water holding capacity. The soil was air dried, and 9 kg was added to 10 L cylindrical pots.

Seeds of Sydney blue gum (*Eucalyptus saligna*) and red ironbark (*Eucalyptus sideroxylon*) were germinated in a nursery at ambient $[CO_2]$. A month later, seedlings were transplanted into the pots, which were then transferred to the various glasshouse rooms. Plants were watered daily. Pots were routinely moved within the glasshouse rooms. Pots and their respective temperature and $[CO_2]$ treatments were swapped between glasshouse rooms on two occasions during the experimental period.



Physiological measurements: Once established, tree seedlings

were used for various growth, gas exchange and biochemical measurements. So far, one paper has been published using this experimental setup (Ghannoum *et al.* 2009), and the main results are summarised below.

Interactive effects of high temperature with sub-ambient and elevated $[CO_2]$ on growth of eucalypt seedlings

To investigate if *Eucalyptus* species have responded to industrial-age climate change, and how they may respond to a future climate, we measured the growth of fast- (*E. saligna*) and slow-growing (*E. sideroxylon*) seedlings exposed to pre-industrial (290), current (400) or projected (650 μ l L⁻¹) CO₂ concentration ([CO₂]) and to current or projected (current + 4°C) temperature. To evaluate maximum potential treatment responses, plants were grown with non-limiting soil moisture.

We hypothesise that: (i) eucalypts will exhibit a greater growth response to the transition in atmospheric $[CO_2]$ from sub-ambient to ambient $[CO_2]$ compared to the transition from ambient to elevated $[CO_2]$; (ii) the growth response to increasing $[CO_2]$ will be greater for eucalypts grown at high temperature compared to counterparts grown at ambient temperature; and (iii) the faster-growing *E. saligna* will be more responsive to elevated $[CO_2]$ and high temperature than the slower-growing *E. sideroxylon*.

Contrary to our hypotheses, we found that: *(i)* the growth response of eucalypt tree seedlings to increasing $[CO_2]$ was not greater in response to the transition from sub-ambient to ambient $[CO_2]$ compared to the transition from ambient to elevated $[CO_2]$; *(ii)* high temperature enhanced the growth response to atmospheric $[CO_2]$ for the transition between sub-ambient to ambient $[CO_2]$ only; *(iii)* the growth response to increasing $[CO_2]$ was greater in the slower-growing *E. sideroxylon* compared to the faster-growing *E. saligna*; and *(iv) E. sideroxylon* responded more strongly to elevated $[CO_2]$ than to high temperature, while *E. saligna* responded similarly to elevated $[CO_2]$ and high temperature.

These results suggest that fast- and slow-growing *Eucalyptus* species may not have responded to the change from pre-industrial to present ambient conditions in terms of growth, but that they possess great potential to respond to predicted increases in atmospheric $[CO_2]$ and temperature.

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A Message from Sapphire Bioscience



"Twigs and Branches 2009"

Nitrogen use imprinted in the genes

Nitrogen along with phosphorous are major nutrients limiting plant growth but who would think that it affects base selection of RNA. However, Acquisti *et al.* (2009 *Mol Biol Evol* **26**, 953-6) have shown that plants have adapted to ecological nitrogen limitations as crop plants have higher nitrogen contents in their transcribed RNA compared to undomesticated plants. Excitingly, this phenomenon is carried through to the proteome where crop plants and nitrogen fixing plants have proteins containing more amino acids with nitrogen rich side chains than undomesticated plants.

Using molecular mimicry to control photosynthesis

The bacterial pathogen that causes citrus canker, *Xanthomonas axonopodis* pv. *citri*, contains a plant natriuretic peptide – like (XacPNP) gene that is unique amongst bacteria (Nembaware *et al.* 2004 *BMC Evol Biol* **4**, 10). This bacterial protein causes increases of stomatal conductance, transpiration and photosynthetic rates and enhances the efficiency of light use during photosynthetic CO₂ fixation (Gottig *et al.* 2008 *Proc Natl Acad Sci* **105**, 18631-6.). It is speculated that expression of XacPNP allows the pathogen to create a favourable environment within the host for bacterial growth.

Danger ! Beware !

Plants recognise molecular patterns imprinting "DANGER" which activates a cascade of defence responses to contain the damage. This is not fiction! Boller and Felix (2009 *Ann Rev Plant Biol* **60**, 379-406) describe the different types of molecular patterns that specialised plant receptors recognise and so activate stereotypic defence responses. They cogently argue (and have great supporting diagrams) that the innate immune response is due to an overarching ability of plants to recognise danger signals from those produced by attacking microbes to endogenous signals indicating cellular damage.

Genetic technologies to find relevant genes

Having problems coming to terms with all the different types of genetic screens? The recent review by Papdi *et al.* (2009 *Funct Plant Biol* **36**, 696-720) could be the answer. They describe the strategies underpinning many of the techniques successfully used such as forward and reverse genetics, insertion and activation mutagenesis, TILLING, promoter trapping, and gene tagging.

Energy switches

Plant growth is very dependent on harvesting energy (photosynthesis) and using fixed energy stores effectively. Energy levels are reduced by events such as prolonged darkness and other stresses that interfere with photosynthesis. Baena-Gonzalez & Sheen (2008 *Trends Plant Sci* **13**, 474-82) describe how members of the SnRK (Sn related kinase) family energy and sense nutrient signals and integrate them with circadian clock and light signals to regulate the transcriptome where changes occur in >1000 genes.

Helen Irving



The Australian Phenomics Facility

The Australian Plant Phenomics Facility – new opportunities for plant scientists worldwide by Mark Tester¹, Robert Furbank² and Helli Meinecke¹

¹ The Plant Accelerator, University of Adelaide, SA

² High Resolution Plant Phenomics Centre, CSIRO Plant Industry, Canberra, ACT

Improvement of crops must be accelerated - crops that are higher quality, more disease resistant and productive in marginal conditions. To achieve this quickly requires a new capability in plant phenomics. Australia has traditionally excelled in plant physiology and crop genetics and breeding, but only limited efforts had been made to bring together these fields of expertise.

To address this weakness, the <u>Australian Plant Phenomics Facility (APPF)</u>, a new cross-institutional facility has been established. This involves two quite different, but highly complementary, research centres - *The Plant Accelerator* at The University of Adelaide, and the *High Resolution Plant Phenomics Centre (HRPPC)* at CSIRO Plant Industry and The Australian National University in Canberra.

The objective is for the two nodes to provide state-of-the-art capabilities for plant phenotyping, with controlled environments and field-based plant growth monitoring, using high throughput robotics, automated imaging and computing technologies. These are integrated with the ongoing adaptation and application of emerging phenomics measurement technologies. The APPF headquarters is based at The Plant Accelerator.

The scheme is funded by the Federal Government, together with local Government and host institution support. So far, over \$50m has gone into the Facility. This large investment was initiated after a review of scientific infrastructure needs in Australia identified a requirement for more focused plant research infrastructure and collaboration across scientific disciplines. It found that existing organisations have highly variable, and generally low quality, infrastructure dedicated to the growth of experimental plants within conventional glasshouses and plant growth cabinets. There are pockets of high-level capability, but they are distributed around the country in a series of unconnected plant breeding and research programs. The Australian Plant Phenomics Facility provides the opportunity to significantly increase plant science research in Australia, providing the chance to bring together genetics and physiology to open new areas of research and accelerate progress in established areas. Importantly, the facilities are also available for use by international researchers and for-profit companies.

Why Phenomics?

The need to increase global food production has never been more critical. Current UN forecasts estimate world output will need to double by 2050. With most arable land already being farmed and climate change threatening existing agriculture, the challenge is massive.

Australia faces its own unique issues with long periods of drought and increasing salinity undermining farm productivity.

Substantial government and industry investments in recent years have enabled Australia to make numerous advances in plant genomics and modern breeding technologies. Globally the stage has been reached where every crop plant genome is likely to have been sequenced within 10 years.

But science has hit a bottleneck in its ability to understand and relate the performance of particular plants with their genetic make-up. Progress on translating the huge database of genome knowledge into improved agricultural products has fallen behind. We are able to study genes and manipulate them, but the study of processes at the level of the whole plant has not been able to keep pace with these advances. As a result we are falling behind in our ability to measure the effects and consequences of those manipulations. This is termed the 'phenotyping bottleneck'. Relieving this bottleneck could significantly accelerate plant scientific advances and their application, as described in the recent article on plant phenomics by Elizabeth Finkel (2009: Science 325: 380-381). The opportunity to relieve this bottleneck is available due to rapid advances in imaging technology, robotic handling and cheap, powerful computers.

What is Phenomics?

Phenomics is the field of study concerned with the characterisation of phenotypes as a whole (the "phenome"). The suffix "-omics" is used by a wide array of other large-scale quantitative biology fields, and commonly involves high throughput technologies.

A phenotype is any observable characteristic or trait of an organism such as its morphology, development, biochemical or physiological properties, or behaviour.

Phenotypes result from the expression of an organism's genes as well as the influence of environmental factors and possible interactions between the two.

Phenomics enables researchers to understand and relate the performance of particular plants with their genetic make-up, resulting in the ability to accelerate progress in improving crops – generating crops that are more productive, disease tolerant and viable on marginal soils.

Phenomics improves our genetic understanding of yield, and increases a breeder's confidence in selecting new and improved breeding lines. It is an emerging area of plant science, which we hope will

~

1st International Plant Phenomics Symposium

The Canberra node of the APPF organised the 1st International Plant Phenomics Symposium, which was held on 22-24 April 2009. The symposium was a timely meeting of plant biologists focused on using plant phenomics and functional genomics to address crop productivity.

The Symposium was attended by 128 registrants (including 46 international participants). Registrants came from all the major phenomics centres, including the Director of the Scottish Crops Research Institute, the Director of Phytosphere and the Deputy Head of the Forschungszentrum Jülich, Germany, as well as researchers from the University of Sheffield, New York University, University of Dundee, Washington State University and Stanford University. There were also presentations and representation by commercial groups, including Monsanto, Bayer Crop Science and BASF / Crop Design.

The geographic spread of plant scientists, from Australian and international groups underscored the level of global interest in plant phenomics as a source of techniques for the better understanding of plant growth, performance and yield at the whole plant and population level. It was evident from presentations by extant and prospective plant phenomics centres in the UK, Europe and America that the levels of investment taking place are very high.

Nonetheless, the Australian Plant Phenomics Facility is acknowledged to be at the cutting edge in many ways, and to be taking a leadership role in encouraging collaboration.

The Symposium was structured to reflect scientific questions the technology addresses, with sessions on biotic stress (including fungal pathogens), abiotic stress (including screening for drought tolerance), growth and yield and ecosystem dynamics and climate change. Papers covered the range of techniques used in phenotyping – including visible and hyperspectral imaging for growth and disease analysis, chlorophyll fluorescence imaging, Infra-red thermography screening, and the use of radar reflectance to study soil and roots and others.

Looking to the future, it was agreed at the Symposium that a stronger vehicle for international collaboration should be established – and the <u>International Plant Phenomics Initiative</u> was launched. The Initiative will, over the coming months, develop an agenda and confirm priorities and actions at a meeting later in 2009. The agenda will likely include exchanging protocols, validating systems, exchanging staff for technical education and developing collaborative funding bids. The Initiative is being led by the Forschungszentrum Jülich and the Australian Plant Phenomics Facility.

About the APPF

Symposium.

The APPF has two nodes, *The Plant Accelerator* at the University of Adelaide and the *High Resolution Plant Phenomics Centre (HRPPC)* at CSIRO Plant Industry and the Australian National University in Canberra.

The Plant Accelerator – Adelaide

<u>The Plant Accelerator</u> is headquarters of the APPF and is located at the University of Adelaide's Waite Campus. The Plant Accelerator is a purpose built plant phenomics facility, which offers a range of greenhouses, high-throughput imaging stations (Smarthouses), growth rooms, laboratories and seed storage space. Its double glazed UV permeable acrylic skin provides good insulation properties and allows for UV penetration. The facility offers:

- A range of controlled growth environments in new, high quality facilities.
- Over 1 km of conveyors delivering radio-tagged plants automatically to state-of-the art imaging stations controlled by high capacity computing equipment.
- Equipment to image shoots in visible, near-infrared and far infrared spectra.
- Equipment for fluorescence imaging of shoots.
- Equipment to allow near-infrared imaging of pots to obtain a measure of water content in the soil.
- Automatic programmable watering to weight of plants on the conveyor system.
- Dedicated bioinformatics support to help manage and analyse data.

The Plant Accelerator will open its doors in January 2010 and will be available for Australian and overseas researchers undertaking public-good research as well as commercial organisations. View our brochure





Artists impression of The Plant Accelerator (top) and stages of construction (bottom)



HRPPC – Canberra

The <u>High Resolution Plant Phenomics Centre (HRPPC)</u> is located in Canberra at CSIRO Plant Industry and the Australian National University. This Centre focuses on "deep phenotyping" (delving into metabolism and physiological processes within the plant), in both controlled environments and in the field, and Reverse Phenomics (dissecting traits to discover their mechanistic basis).

Next generation research tools are being developed and applied to probe plant function and performance at medium and high throughput, under growth cabinet conditions and at managed field sites.

Recent advances in robotics, imaging and computing are used in applying these technologies and scaling them from the single plant to the ecosystem level. Two levels of service are provided in the HRPPC.

First, projects can be housed in the "Research Hotel" environment where screening systems can be developed using facility staff and resources then deployed in the facility and in the user's home institution.

Second, users' material can be screened for specific attributes using one or more of the modules housed at CSIRO or the ANU. This node of the facility focuses on flexibility from cereals to dicots and woody perennials at all stages of development.

The HRPPC opened in July 2009 and welcomes Australian and overseas researchers undertaking publicgood research as well as commercial organisations.

For further information about both facilities please visit http://www.plantphenomics.org.au/



Illustrations of the renovated phytotron (top), the interior of the new centre (bottom left) and a thermal image of field plots as an example of the field module (bottom right).

CONFERENCES 2010

International Conference on Plant Vascular Biology 2010

July 24-28, 2010, Ohio State University, Columbus, Ohio, USA

Co-Chairs: Biao Ding (Ohio State University, Columbus, Ohio, USA) David Hannapel (Iowa State University, Ames, Iowa, USA)

Plant Vascular Biology (PVB) includes studies on the biogenesis, structure and function of transport systems in plants, under conditions of normal plant growth and development as well as of plant interactions with pathogens. The transport systems cover broadly the xylem, phloem, plasmodesmata and vascular cell membranes. The PVB concept has emerged in recent years to emphasize the integrative nature of the transport systems and approaches to investigate them. PVB 2010 will bring together junior and senior researchers working on various aspects of PVB (structure, biogenesis, signaling, proteomics, genomics, nutrition, biomass, plant-biotic interactions, etc) from around the globe to share the latest findings, develop collaborations and identify new directions of research. The conference will also strive to bridge basic and applied aspects of PVB.

For more information please visit the conference homepage (http://www.ced.osu.edu/pvb2010conference/index.html).

11th International Symposium on the Genetics of Industrial Microorganisms (GIM 2010)

June 28 - July 1, 2010, Melbourne Convention and Exhibition Centre.

With the emphasis on the contributions of genetics coupled to microbiology we hope we can provide further developments in our field. Already contributions in our field are impacting our daily lives, including our health and well-being. Our society faces many more needs that our technologies may help with. These include new products from microbes, renewable and clean energy, bioremediation, microbial models and beneficial microbes.

The Conference was last held in Prague in 2006 and attracted over 800 delegates. The Conference will bring together the world's leading academics, agricultural specialists, food/information/energy technologists, health specialists, molecular biologists and pharmaceutical researchers.



CONFERENCE UPDATE: "PLASMODESMATA 2010"

Registration and abstract submission are now open for "Plasmodesmata 2010"

Come join us for an exciting week of intercellular communication, virus movement and intra- and inter-cellular gene silencing, while enjoying the unique atmosphere and magnificent views of the newly renovated Quarantine station, right on Sydney Harbour.

When: March 21-26, 2010Where: Quarantine Station, Manly, NSW

Earlybird registration deadline: **15th December 2009** Full registration deadline: **21st January 2010**

Abstract submission deadline: 21st January 2010

For more information please visit: http://www.bio.usyd.edu.au/pd2010/index.html

or email the Local Organising Committee at: pd2010@bio.usyd.edu.au

ROBYN OVERALL, CHAIR LOCAL ORGANIZING COMMITTEE

Functional Plant Biology

Editor-in-Chief: Dr Rana Munns Assistant Editor: Dr Yvonne Cheng

This has been an active year for FPB with an increase in the submission of high quality papers, particularly from Australia.

The Best Paper award for an early-career scientist was won by Joanne Tilbrook for her paper "Cell death in grape berries: varietal differences linked to xylem pressure and berry weight loss" by J Tilbrook and SD Tyerman, FPB *35*, 173-184 (2008). Joanne has been invited to present this work in a symposium at ComBio2009.

A special double issue on Plant Phenomics is just published, including papers on root methodologies, C4 genes into rice, thermal imaging for stomatal conductance, and hyperspectral methods for analysis at the leaf level. These papers are free on-line for 3 months.

The Evolution Series is well under way. These will all be free on-line for life. This is a virtual special issue that can be accessed as <u>http://www.publish.csiro.au/nid/103/aid/13408.htm</u>. Coming articles include the evolution of flowering, halophytes, C4 and CAM metabolism.

Special issues presenting Research Fronts on drought, salinity, acid soils and effector biology are in progress. Ideas for other topics are welcome.

Rana Munns Editor-in-Chief, FPB www.publish.csiro.au/journals/fpb

Education Section

Online Learning

Supermarket Botany – a fresh approach

Geoff Burrows and John Harper Charles Sturt University, Wagga Wagga gburrows@csu.edu.au & jharper@csu.edu.au



The use of Supermarket Botany is a popular approach to teaching plant structure and plant life cycles. It uses a student's existing knowledge of everyday food items to explore the differences between:

- fruit and vegetables,
- roots, stems and leaves, and
- flowers (with ovaries and ovules) and fruits (with seeds).

Although designed with students in mind, feedback indicates the website generates a high level of interest from the general public as they learn about common household products, while developing a new appreciation for plants.

We aimed to produce a resource that was botanically accurate, with a reasonable level of detail and was presented in an engaging format. Please see:

http://www.csu.edu.au/research/grahamcentre/education/

The web site is divided into two main areas:

- a tutorial that explains the differences between roots, stems and leaves, and also examines the differences between vegetative and reproductive tissues, and
- a test (called 'The Challenge') that allows students to apply the knowledge gained in the tutorial.

In 'The Challenge' students select an item from 'The Shelf' and are then required to select whether its major component is root, stem, leaf, flower, fruit or seed. We have done extensive surveys and have identified the common Supermarket Botany misconceptions. Thus we are able to customise the incorrect answer responses to give hints as to the correct answer. Once the correct answer is selected students go to the 'Why?' page, where high quality images provide supporting evidence.

Quantitative testing indicates the web application has similar learning outcomes to a traditional laboratory-based session, although it is designed to support, not replace, hands-on learning. Student responses include "I understood more in 15 min (using Supermarket Botany) than 2 hours of textbook reading."



The Virtual Floral Formula

Geoff Burrows Charles Sturt University, Wagga Wagga

gburrows@csu.edu.au

The study of flower structure is a standard component of almost all first year university botany and biology subjects. A knowledge of flower structure is important in understanding:

- how flowers facilitate the exchange of genetic information,
- different pollination syndromes,
- the development of fruits and seeds and the classification of fruit types,
- plant breeding and
- how to identify plants using keys and Floras.

While important, floral structure is often studied in a single laboratory session, based on a small number of specimens, the choice of which may be constrained by seasonal factors. Students may be examined on this material several weeks after the class, with no effective ways of reviewing the information.

I have designed an interactive website called 'The Virtual Floral Formula'. See:

http://www.csu.edu.au/herbarium/ and click on 'The Virtual Floral Formula'.

Users can select from 12 flowers, and for each flower scroll through a selection of images that show all the main structural features. They can then progressively assemble a floral formula for the flower from 13 drop-down menus (Fig. 1). When satisfied with their answer the 'Submit' button is clicked, after which the answer is compared and contrasted with the official answer and any differences are highlighted (Fig. 2). The images are now described and/or labelled (Fig. 2), allowing for any floral formulae discrepancies to be investigated.

As best as I can tell this is a unique resource, with nothing like it on the web for independent learning about flower structure and floral formulae. I suspect this application could be usefully used as a supporting resource in many first year university biology or botany subjects.

I would welcome comments or suggestions.



Book Reviews

Plant Anatomy Books, 2008

Dr Teri O'Brien

Research Associate La Trobe University, Botany Department Adjunct Associate, Monash University Former Reader in Botany, Monash University

Review of:

"Teaching Plant Anatomy Through Creative Laboratory Exercises" 2008. Peterson, R.L., Peterson, C.A., and L. Melville National Research Council of Canada. ISBN 978-0-660-19798-2.

This is comfortably the best illustrated modern text on how to go about teaching Plant Anatomy and a good deal of Plant Morphology at the same time. A very large number of the wonderful color illustrations are of fresh material, often backed up with excellent interpretative diagrams. Special topics are handled well by the use of the Box Format, a separate page illustrating a method or a theory or a tissue type, viz., polarizing microscopy (Box 3), tissue printing (Box 4), xylem (Box 10). All photomicrographs give the staining schedule used and are of an impressively high standard throughout.

With one exception, which I regard as a major mistake, most of my criticisms of this work are very minor and constitute nit-picking, designed really to invite the authors to improve something in the next edition. The serious error comes early on p.3., where it is stated "adjusting the condenser will achieve maximum contrast". This is simply not correct, and is a practice all too often used by inexperienced microscopists.

You should adjust the condenser as soon as you have a focused image of the specimen and to do that you use the field stop to get the light imaged close to Köhler conditions, with the condenser diaphragm left open. Now as you change objectives, you need only adjust the diameter of the field iris, or alter the contrast by the setting of the condenser diagram.

Condenser contrast comes from the setting of the condenser diaphragm and is only a minor issue in strongly stained specimens like many of those in this book, but it is critical in unstained ones of low inherent contrast. And no way should the condenser be defocused from the Köhler state to "increase contrast": this badly degrades the image and introduces a lot of false contrast and irritating phase contrast into the image. A simple and correct method to set up a microscope is given in O'Brien,T.P., and M.E.McCully 1981, The Study of Plant Structure: Principles and Selected Methods, now well and truly out of print but in many libraries around the world.

A list of minor issues follows:

1. The depth of field of a binoc m'scope is way bigger than that of a compound one at the same magnification, and I think the first paragraph therefore has an error in it. The degraded imaged you see at high magnification microscopes in binoculars is due, as I understand it, to the bad effects of scattered light impinging on a low numerical aperture, which goes with the increased depth of focus.

2. p.3. My understanding of the eyepiece adjustment is to compensate for the unequal vision in two eyes of the same investigator.

3. Cleaning a lens with dried aqueous stuff on it: spit is easily the best and safest.

- 4. Fig 20 looks like chloroplasts, not chromoplasts.
- 5. Box 3: most modern Polaroid sunglasses have curved lenses, not ideal for getting good extinction.

6. Don't forget leucoanthocyanins that are colorless, top of p.27

7. Pectic substances include all forms of pectins, the esters, the Ca salts as well as free pectic acids. Only the last is stained by TBO metachromatically. You can quantitatively stop metachromatic pink staining due to pectic acids by pre-treating a section with Ca chloride solution before T BO staining

8. CW label on Box 6 should go farther into the cell wall: presently it stops at the cell membrane.

9. Caption to Fig.89: they have helical thickening, not spiral thickening: a helix is not the same thing as a spiral! Re Box 11: Labels on the fibres and parenchyma cells would be good, and I have never seen a scalariform element with bands that have a free end. The ones I am familiar with all have that "free end" joined to one of the complete gyres.

10. Box 13: Safranin is a lignin stain and can give confusing results, but acid fuchsin is OK for a free space marker for about 20 minutes after which ultrafiltration begins to slow the transpiration path through the mesophyll, but you must mount sections in oil to view it. Caption to 104: unclear, you need to say what color the fluorescence of PTS is!

11. Say that all of Figs 114-116 are unstained. Fig 130: cells are not fibres and are lignified bundle sheath cells. Fig 153: there appears to be both TBO and Sudan in this image (aqua lignin in tracheary elements).

12. p.133: metachromicity! Metachromasia please! P.134: visible dust is never on the iris diaphragm unless it is a fibre hanging on the edge!

13. p.139: neutral red only works for vacuoles that contain a binding agent, such as polyphenols. And many years ago I.W. Bailey pointed out there were two types of vacuoles in pine cambium.

These 13 minor comments should not be taken as detracting from the usefulness of this work and it is a great credit to the authors that they have done it so well.

Review of: "Plant Structure, A Colour Guide" 2nd Edition, 2008. Bowes, B.G., and J.D.Mauseth CSIRO and Manson Publishing. ISBN: 978-0-643-09570-0.

The first confusing thing about this work is that it is called a 2nd Edition when in fact it is a revised Edition, since it has a different title from the 1st Edition "A Colour Atlas of Plant Structure ", 1996. This is followed rapidly by the issues raised with the notion of a Colour Atlas or a Colour Guide for a book on Plant Morphology and Plant Anatomy. While natural colours of plant parts, especially flowers and pigmented stems, give some indication of their function, the colours in this work seen in stained sections are never explained.

So while the photographs of whole plants and their parts are excellent, the interpretation of the photomicrographs of slides requires that the reader knows in advance that the majority of these sections seem to have been stained with safranin-fast green. In that combination, lignin and some polyphenolics in vacuoles stain red, unlignified cell walls and cyrtoplasts stain green, e.g., Fig. 380. But Fig. 378 shows mucilage-rich ray cells stained bright red. Since mucilage is rarely lignified and the other stain appears to be hematoxlin, the absence of data about the red stain is very unhelpful.

The presence of light micrographs of glutaraldehyde-osmium tetroxide fixed tissues (e.g., Figs 249 & 251) shows the expected improvement in the quality of fixation of the cytoplasm, but the absence of information about the staining renders the green of Fig. 249 and the deep blue of Fig. 251 uninterpretable. Nowhere is this worse than in Figs. 254 and 255. Fig. 254 shows unlignified parenchyma and cells of the phloem stained green, but Fig 255 beneath it shows all cell walls of the phloem stained bright red! This implies it is lignified, which is untrue for most phloem other than fibres in the majority of vascular plants. Presumably this is a section badly stained with safranin.

The authors fail to mention the very clear example of a passage cell in the endodermis in Fig. 255.

This absence of staining information is to be deplored and was heavily criticized by O'Brien and McCully in their 1981 book, The Study of Plant Structure: Principles and Selected Methods. This kind of issue should not be resurfacing in a modern book.

There is poor control of the background colour in almost every slide of material fixed by old techniques and stained by old style stains. There is no excuse for this lapse in control of background intensity and colour neutrality in modern photomicrography, no matter how old the slides are. Compare the backgrounds in the very dark versions in Figs 371, 372 and 387 which interfere a lot in stain interpretation, with the perfectly acceptable backgrounds in Figs 369 and 370.

Thus, I cannot support the enthusiastic review by Gasson, P., 2009, Bot. J. Linn. Soc 161, 103-104, but I do not want to be too hard on this work. The range of material covered is very impressive. Many of the photographs of whole plants and their parts are excellent, and in some cases, stunning (e.g., Figs 467, 476, 501 and 503).

If the purchaser wants a picture book with good photographs, and does not care about the quality of the photomicrographs, this work is useful. But if you want to learn plant anatomy in the 21stC, this is definitely not recommended.

Dr Teri O'Brien, Ph.D and JF(Harvard), D.Sc.(Monash)

A Message from QIAGEN

The Rotor-Gene® Q by QIAGEN

The unique centrifugal rotary design of the Rotor-Gene[®] Q makes it the most precise, versatile real-time PCR cycler available. Each tube spins in a chamber of moving air, keeping all samples at precisely the same temperature during rapid thermal cycling. Detection is similarly uniform. When each tube aligns with the detection optics, the sample is illuminated and the fluorescent signal rapidly collected from a single, short optical pathway. This thermal and optical uniformity results in sensitive, precise, fast real-time PCR analysis. It also eliminates sample-to-sample variations and edge effects. These are unavoidable in traditional block-based instruments due to temperature gradients across the block and multiple, complex optical pathways. The rotary design delivers: well-to-well variation below $\pm 0.01^{\circ}$ C (20 times less than block cyclers), uniform detection eliminating the need for ROX reference dye, and fast ramping and negligible equilibration times for short run times. With up to 6 channels spanning UV to infrared wavelengths, the cycler delivers the widest optical range currently available. The Rotor-Gene Q supports multiple PCR tube formats to suit a range of needs. As well as tubes, Rotor-DiscsTM, circular plates of vertically-oriented reaction wells, are available for accelerated setup and higher throughput (96 wells with 4 reference wells).

For more information Ph. 1800 243 800, email orders-au@qiagen.com





- *ASPS Website.* The ASPS website has been thoroughly revamped and is being continuously upgraded.
 - Membership dues can now be paid on line.
 - You can advertise jobs, PhD scholarships, conferences, books by contacting Kiran Sarfaraz via advertise@asps.org.au. To cover the costs involved, the society has introduced a small charge of \$30 for members and \$70 for non-members FOR EMPLOYMENT ADS ONLY. Advertising conferences and books (edited by society members or containing chapters written by society members) are FREE OF CHARGE.
- **RN Robertson Travelling Fellowship.** This named Fellowship recognises and celebrates the sustained contribution made by RN Robertson (Sir Bob) in nurturing young plant scientists in Australia spanning across four decades from the 1950's. The Australian Society of Plant Scientists is indebted to Hank Greenway and Joe Wiskich who generated and championed the early development of the RN Roberston Travelling Fellowship.
- **Student Travel Funds.** Funds are set aside each year to sponsor student travel to our annual conference (this year in New Zealand), and contribute to their professional development in plant science. Support will vary from year to year depending on the Society finances, location of meeting and number of applications. The Treasurer will apply a formula in calculating individual entitlements and takes these factors into account. Applicants must be financial members of ASPS and presenting a paper or poster at the ComBio meeting.
- Postgraduate Section. We are proud to announce that student members who have recently completed their PhD and had their thesis passed can submit a summary that features in Phytogen. The editors feel that this is an important opportunity for our postgraduate students to showcase their research. Such successful student members are advised that the summary can be accompanied by a key image in suitable format and that they should submit their items to the editor of Phytogen at any time for inclusion in the next issue.
- Society Funding for Workshops and Conferences. The society has a total of \$10,000 available each year to provide seeding money and sponsorship for up to four conferences organised by members. The amount available to assist each conference will be about \$2,500. For more details see the website: http://www.asps.org.au and take the link to conferences.
- Corresponding and Life Memberships. Life Membership recognises an outstanding and sustained contribution to the Society by a long standing ASPS member who, through their professional activities, has substantially enhanced the international profile of Australian plant science research. Corresponding Members are high profile overseas colleagues who have contributed substantially to plant science research within Australia. If you know of a deserving recipient for Life or Corresponding Membership, please consider putting a nomination forward. The procedure to follow is outlined on the ASPS website (see: http://www.asps.org.au and click on "About ASPS" where there is also a list of Life and Corresponding members).