



# **Hawkesbury Institute for the Environment**

## **Summer Scholarship Research Program 2020**

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## **Project 49: Talking trees: dynamic stems with rain and dry during climate change**

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### **Project description**

Trees don't talk in the normal manner that we consider talking. However, in spite of wood being almost exclusively dead tissue, it exhibits vital and dynamic fluctuations linked to the movement of water through the xylem. This is a chance to help unravel the meaning of these fluctuations and what trees tell us through them, by measurements of stem diameter and water status to understand plant behaviour under climate change. Our goal is to help uncover how water and carbon transport and mechanical properties influence stem diameter fluctuations in Eucalyptus trees in a western Sydney woodland right next to the Hawkesbury campus of Western Sydney Uni.

Point or band dendrometers have been increasingly used to study these dynamic fluctuations, referred to as stem diameter variation (SDV; 1–10  $\mu\text{m}$  changes). SDVs occur as a result of environmental variation. Variation in the water content in stems, originating from reversible shrinkage and swelling of dead and living tissues, and irreversible growth, all contribute to SDVs. Thus, high-resolution SDVs are widely recognized as a useful drought stress indicator.

South-eastern Australia including Richmond, NSW has seen an increasing trend toward dry and hot summers. We will measure SDVs on mature Eucalyptus trees during summer, with both manual and automated data collection techniques. We will determine the magnitude of daytime SDVs, and seek to associate them with wet/dry and cool/hot cycles, as well as with higher atmospheric CO<sub>2</sub> concentration at the EucFACE facility.

### **Project Aims**

The behaviour of trees with respect to the environment and climate change will be measured and analysed using stem dendrometers. Our aims are:

- Gain experience with scientific equipment
- Measure shrink/swell behavior of trees.
- Relate the stem diameter variation (SDV) to soil water availability and its changes
- Learn how to summaries and report your findings to others

## **Project Methods**

This project uses tree dendrometers that record diurnal stem diameter variation as the basis for assessing how trees behave and 'talk' with their environment. We will select a set of study trees amongst those being manually monitored for long-term growth at EucFACE. The measurements involve re-starting an existing set of dendrometers with a few new ones that can be installed and monitored for diurnal stem diameter variation (SDV). After learning new techniques, it is also possible to monitor concurrent fluctuations in plant moisture status with water potential measurements on leaves and stems during wet and dry periods.

A full suite of relevant environmental measurements at EucFACE, including light, air temperature, air humidity, and soil moisture are available on-line through the HIEv system. These measurements can be used to determine the 'drying power' of air on a daily basis, and with heat waves during summer. As well, cycles of rain and dry can also be analysed.

## **Opportunity for Skill Development**

This project will introduce the student to the fields of plant physiology and ecology. The student will develop skills using several pieces of scientific equipment, as well as skills for proper data collection, organisation, hypothesis-testing and analysis.

- The student gains the ability to make and interpret scientific measurements
- The student will see how inclement environmental conditions affect tree behaviour, and also
- The student learns about the precision and accuracy of measurements.
- The student gets the opportunity to work with a few different electronic sensors
- The student learns how to analyse the data quantitatively, and present it appropriately

The student will learn to function independently after receiving proper guidance, learn how to work in a team, and learn how to contribute to a research team effort.

## **Students are required to have the following skills/meet the following pre-requisite(s) to apply**

- Student candidates should be willing to learn how to work with quantitative data, but no higher math's skills are needed.
- Student candidates should be willing to work in the field while following safe-work practices.
- He/she must be willing to implement Covid-19 hygiene practices and social-distancing when required.
- Work will sometimes involve brief inclement conditions (hot and/or sunny days during summer).

## **Project 50: Assessing biodiversity benefits in urban environments**

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### **Project description**

More than 4.2 billion people live in urban areas, representing ~3 percent of the Earth's land area. As the human population grows, cities around the globe will continue to expand their footprints and increase their demands for goods and services. Ecosystem services, such as temperature regulation and biodiversity support, are provided by urban forests in cities worldwide. However, in cities, habitat loss is caused by the replacement of natural habitat with tarmac, pavement and other impervious surfaces. These changes have a negative effect on the abundance and richness of invertebrate species by decreasing habitats, including floral resources and nesting sites. The Which Plant Where (WPW) project, funded by Hort Innovation Australia, aims to increase the knowledge base that will help make our cities more liveable not only now but under future climatic conditions. An essential part of having a liveable city is the maintenance (and expansion) of green spaces that provide many core benefits to society, including biodiversity support.

This summer scholarship project focuses specifically on how different plant attributes are related to the provision of ecosystem services on a local scale, focusing specifically on how different planting structure and complexity are related to abundance and diversity of pollinator and other insects on a local scale. The student joining our project will examine insect biodiversity, using a variety of measurements such as plant biometrics and insect sampling - with student interest driving additional or alternative data collection. A variety of measurement techniques (e.g. tree biometrics and insect surveys) will be used to obtain data on which vegetation traits (e.g. leaf area index, height, canopy density) are associated with the maximum provision of biodiversity benefits. The student will be introduced to the concepts of ecosystem services and will have the opportunity to learn about survey design, sampling strategies, data management and develop their skills in the relevant statistical techniques and software. This project also offers enormous opportunity benefits to the student via interactions with a team of post-docs and Ph.D. students contributing to related questions as a part of the wider WPW project.

### **Project Aims**

To understand the effect of structural complexity of vegetation on invertebrate abundance and diversity in an urban context.

### **Project Methods**

This project involves fieldwork, sampling, and data collection from the Which Plante Where Living Lab research facilities at Western Sydney University's Hawkesbury Campus. Additionally, fieldwork will potentially be conducted in other Living Lab sites in different locations within the greater Sydney region. All fieldwork will be supervised in conjunction with the primary supervisor and the associated Ph.D. student (Mahmuda Sharmin). Research conducted for this project is mostly undertaken outdoors, so a willingness to spend time in the field is important for this project.

Data collection for this project will include spot measurements of different biological and physical parameters (such as tree height, diameter at breast height) and insect sample collection, followed by species identification. This will contribute to a comprehensive evaluation of which tree species and vegetation characteristics are better to increase local biodiversity in an urban context. All these findings will be shared with industry partners and plant nurseries and will help to advocate the importance of having appropriate tree species in our cities to maximise the benefits they bring to urban populations.

### **Opportunity for Skill Development**

This project will provide a sound understanding of the fundamentals of research – from hypothesis development to experimental design and data collection – providing a solid framework for future participation in HDR study in the area of urban ecology. The student joining this project will have the opportunity to tailor their research skills development from measurements to data analysis and inference focussing on applied topics, such as insect identification and biodiversity assessment. In addition, they will have the opportunity to learn about a wide range of ongoing projects run by affiliated Ph.D. students and post-docs conducting research on related questions using the WPW framework, gaining experience of how to work as part of a wider research team. As part of the WPW team, they will be supported in connecting ecological theory to practice, developing research questions, hypotheses and predictions and, if interested, learning about writing for a general audience and the scientific community.

### **Students are required to have the following skills/meet the following pre-requisite(s) to apply**

A driving licence is desirable but not required.

## **Project 51: Effects of drought on plants and pollinators**

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Second Supervisor

### **Project description**

Native pollinators provide huge economic benefits to Australian agriculture, and ongoing work conducted at HIE is examining the importance of non-crop floral resources in sustaining pollinator populations outside of the relatively short flowering periods of the major crop plants. Unfortunately, native pollinators face a variety of threats that are contributing to declines in their abundance across the globe, ultimately reducing their ability to provide these important pollination services within agricultural and horticultural systems. Climate change is a major factor affecting pollinators at local, regional and global scales, via a number of direct and indirect routes. Direct effects of drought or increased temperatures (especially heatwaves) can influence the activity and resource requirements of pollinators, via impacts on their metabolism. Indirect effects such as shifts in the timing of flower production relative to the life cycle of pollinators, and changes in both the availability and quality of floral resources can also drive shifts in plant-pollinator interactions.

The affiliated larger experiment is a new project lead by the Community Interactions subgroup of the DRI-Grass facility, a long-term grassland drought manipulation experiment at Hawkesbury Campus. This project will take advantage of a field-experiment that has been established at HIE where, for the past year, mounted cameras have been recording the diversity and abundance of flowering plants under various drought treatments. The project will provide a unique opportunity to integrate field-collected data with computational image analysis and allows students to learn classic plant and pollinator survey methods all from the comfort of your own home. Subject to Covid-19 safety requirements, there will also be the opportunity to visit the field experiment and get involved in hands-on data collection.

### **Project Aims**

This project aims to determine the effect of drought on plant phenology including the diversity and abundance of flowering plants, and the timing and duration of flowering within a grassland community.

### **Project Methods**

This project will take advantage of a field-experiment that has been established at HIE where, for the past year, mounted cameras have been recording the diversity and abundance of flowering plants under various drought treatments. This project will provide a unique opportunity to integrate field-collected data with computational image analysis and allows students to learn classic plant and pollinator survey methods all from the comfort of your own home.

### **Opportunity for Skill Development**

Skill development from this project will provide the student with a sound understanding of the fundamentals of research – from hypothesis development to experimental design and data collection – providing a solid framework for future participation in HDR study in the area of climate change ecology, plant ecology and pollination ecology. The student joining this project will have opportunity to will learn how to collect data using image processing, analyse experimental data and communicate their findings through writing a scientific manuscript. Finally, as a member of the Community Interactions subgroup of DRI-Grass the student will be supported in connecting ecological theory to practice, developing research questions, hypotheses and predictions and, if interested, learning about writing both for general audiences and for the wider scientific community.

### **Students are required to have the following skills/meet the following pre-requisite(s) to apply**

The essential criteria for student participation in the Summer Scholarship Program are sufficient for skill pre-requisites. Bachelor of Science, or Bachelor of Natural Science students are most likely to benefit fully from this project, however students with an expressed interest in insects, plants, ecology, and/or climate change research who are enrolled in other programs are welcome to apply. Students should be willing and able to work from home on a computer able to handle image processing software (all free), with access to zoom video for meetings with supervisors.

## **Project 52: The effects of hive design and climate on stingless bee activity**

**Supervisor(s):** Mark Hall - [mark.hall@westernsydney.edu.au](mailto:mark.hall@westernsydney.edu.au)  
Principal Supervisor

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Second Supervisor

### **Project description**

Native Australian stingless bees (*Tetragonula* spp.) are often kept in managed hives, due to their utility for crop pollination and ease of handling. Yet little is known of the activity levels of the most commonly kept species (*Tetragonula carbonaria*) and how these may be mediated by climate conditions (e.g. temperature and relative humidity). Further, there are many types of commercial hive available for housing this species, and these differ in their dimensions and the materials used in their construction. Each of these may impact how colonies regulate thermal conditions within hives, which may then influence colony strength and activity levels. Reduced activity by colonies will decrease the utility of these bees in pollinating important commercial crops. Here, we will conduct a range of passive experiments, monitoring bee activity levels at four different hive types. These hives have a system installed that constantly records hive weight, temperature and relative humidity (both inside and outside the hive), and light intensity. We will conduct video recording and observation at hive entrances to record the number of foragers leaving and returning to hives to directly compare with climatic and hive weight measures. We may also couple this with audio recordings within the hive to determine if/how colonies signal changes in conditions. Comparisons will be made between the four hive types and across different times of the day to determine which factor(s) most influence forager activity and colony strength of Australian stingless bees.

This work will contribute to the wider work on stingless bee foraging efficiency and behaviour, funded by Hort Innovation. It will aid understanding of forager activity and help determine potentially suitable hive characteristics to aid the use of stingless bees for crop pollination. Results will be presented in a peer-reviewed manuscript, milestone reports for the project and presentations to the scientific community and general public.

### **Project Aims**

- To investigate if stingless bee activity levels are directly related to thermal conditions (temperature and relative humidity) within hives
- To investigate if different hive types differ in their thermal capacity and therefore modify activity levels
- To determine if activity levels differ over the course of the day in relation to temperature, relative humidity and light levels

### **Project Methods**

Surveys will be conducted at two field sites within close proximity to Hawkesbury campus, where native bees have been installed for an extensive set of experiments. Regular surveys at hive entrances will be conducted throughout the day at each hive on warm sunny days during peak activity periods (e.g. 9am to 4 pm). The number of bees leaving and returning to hives per 2-minute period will be recorded by the student. Audio and video recordings of hive entrances may also be required to capture more data. This will be replicated across multiple days and hive types. Hive weights, temperature and humidity data will be made available to the student for comparison with activity data.



### **Opportunity for Skill Development**

This project will enable the student to manage a scientific research question, from preparing the study, searching for literature, conducting experiments, performing analyses and writing both a final report and contributing to a scientific paper. They will gain knowledge of the behaviour of stingless bees, some of their ecology and their potential as crop pollinators. They will develop key skills required of further study (e.g. honours or masters degrees). A key component will be learning some basic statistical skills and the ability to interpret and discuss results.

### **Students are required to have the following skills/meet the following pre-requisite(s) to apply**

No additional skills required

## **Project 53: Do forest soils breathe more under eCO<sub>2</sub>?**

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Second Supervisor

### **Project description**

As atmospheric CO<sub>2</sub> concentrations have surpassed 400 ppm and continue to rise, there remains great uncertainty in how ecosystems will respond to predicted future climate scenarios. It is generally perceived that under non-limiting conditions, elevated atmospheric CO<sub>2</sub> (eCO<sub>2</sub>) stimulates photosynthesis and plant growth which could act as a sink for atmospheric CO<sub>2</sub>. Observations have also demonstrated that eCO<sub>2</sub> can increase below ground C allocation where C can either be respired back to the atmosphere via autotrophic or microbial pathways or can enter more stable soil C pools. Recent work from our flagship Eucalyptus Free Air CO<sub>2</sub> enrichment (EucFACE) forest experiment suggest that when nutrients are scarcely available the potential for offsetting increases in atmospheric CO<sub>2</sub> may be limited and that the amount of CO<sub>2</sub> released back to the atmosphere under eCO<sub>2</sub> may increase resulting in a positive feedback. However, the magnitude of this feedback is dependent on soil moisture dynamics. Further work at the research facility has demonstrated the P-fertilisation can overcome limits to tree growth at the site. However, the impacts of P-fertilisation on below-ground microbial processing and soil respiration are unknown. Preliminary work has demonstrated that microbial biomass increases following P-fertilisation, but uncertainties around whether this facilitates soil carbon accumulation, or increases the rate at which CO<sub>2</sub> is respired back to the atmosphere are not known. This project seeks to determine the long-term impacts of eCO<sub>2</sub> on rates of soil respiration by utilising a combination of high temporal data collected from automated soil respiration collars and in-field survey measurements at more spatially resolved scales to determine the long-term effects of eCO<sub>2</sub> on feedback responses in a nutrient limited Eucalyptus forest. Further, we aim to determine how soil respiration responds to P-fertilisation in this nutrient poor forest system. The project will expose students to in-field measurements at the unique, world-renowned EucFACE research facility, and importantly allow them to interact with the multidisciplinary research team undertaking a wide array of project within the site, exposing them to research expertise and methods that go beyond the scope of this project and will be offered the opportunity to participate in sampling campaigns that are undertaken by the broader soils community working at EucFACE. The student will be trained in methods of data acquisition, quality control and analysis and will offer the student opportunity to both collect in-field measurements as well as explore and analyse existing data collected from automatic respiration chambers.

### **Project Aims**

The main objective of this study is to build on existing soil respiration data base for the EucFACE facility to assess the long-term effects of eCO<sub>2</sub> on soil respiration. Importantly this project will also determine the effect of P-fertilisation on soil respiration within this ecosystem. This will provide important baseline data prior to the establishment of a P-intervention experiment that aims to determine the ecosystem response to the interactive effects of eCO<sub>2</sub> and P-fertilisation and will serve to improve model predictions of the response of forest systems to rising atmospheric CO<sub>2</sub> concentrations.

## **Project Methods**

Manual survey measurements of soil respiration will be made fortnightly, approximately every two weeks at permanent locations established within each of the experimental rings (8 per ring), using portable infrared gas analyzers (Li-8100-103, Licor Environmental, Lincoln, NE, USA). The student will be directly engaged in measurements and will work side by side with the academic researchers to make in-situ measurements using two identical units. The student will be directly engaged and be trained in downloading measurement data, quality checking and analysing the data using specialised software (SoilFlux Pro, Licor Environmental, Lincoln, NE, USA). Additionally, the student will use this software to determine daily soil respiration measurements from data collected from automated long-term chambers (Li-8100A, Licor Environmental, Lincoln, NE, USA) located within each of the experimental rings. The student will learn how to screen data for anomalies and learn skills in dealing with large data sets. The student will also learn skills associated with maintenance of scientific equipment. Throughout the student will be trained by the academic researchers, when face-to-face allows as well as remotely by using zoom meetings and live software training sessions.

## **Opportunity for Skill Development**

The student will be involved in using specialised equipment (Li8100 infa-red gas analyser) and software (Soil flux pro) for collection of in-situ, real-time soil respiration data. Key skills in data acquisition, quality control and organisation of experimental data will be developed. Additionally, the student will be guided to design a robust experimental protocol for collection of soil respiration data on the P-addition plots. Here skills will be developed around assessing sources of variation, quantifying field variation and determining appropriate number of replicates needed for field measurements.

## **Students are required to have the following skills/meet the following prerequisite(s) to apply**

Basic undergraduate training is sufficient. An eye for attention to detail and ability to work with spreadsheets is desirable.

## **Project 54: How do local landmarks influence the foraging behaviour of *Tetragonula carbonaria*, an Australian stingless bee species used as a commercial pollinator**

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Second Supervisor

### **Project description**

The stingless bee species *Tetragonula carbonaria* is increasingly being used as a managed pollinator species on crops in Australia such as macadamia. Stingless bees, much like honeybees, live in large eusocial colonies containing 10s of thousands of individuals. A subset of the colonies' members, the foragers, scour the surrounding landscape for pollen and nectar sources to feed the colony. While we rely on the bees performing these foraging trips in order to pollinate our crops, we still understand very little about how these bees are using landscape cues to orientate themselves in the landscape.

We aim to study the effect of landscape by providing the bees with prominent landscape features that we can shift and modify, and observing how these changes influence where the bees go when visiting spatially spread out arrays of food sources.

### **Project Aims**

The project aims to better understand the foraging behaviour of *Tetragonula carbonaria*, an Australian native stingless bee species that is becoming increasingly important as a managed pollinator of tropical crops such as macadamia. The project will address the following questions:

- How do stingless bee foragers use landmarks to navigate through spatially spread out forage sources?
- How long do landmark based flight paths persist once the landmarks have been removed?

### **Project Methods**

The student will take a leading role in conducting the field work for this experiment. 6 *T. carbonaria* colonies shall be tested one by one under the same conditions. The student shall perform the following steps with each colony in the experiment:

1. A stingless bee colony shall be placed in the middle of a large flat field with no available forage sources within 300m of the colony
2. A gravity feeder containing 50/50 sucrose/water solution shall be placed at the entrance of the colony, and the bees shall be trained to feed on it.
3. The gravity feeder shall be moved to a position 50m North from the colony, and placed next to a 3m x 3m x 3m tent that shall act as a mobile landmark. Newly arrived bees shall be marked at the feeder with a paint pen. There shall be 3 empty feeder's places East, South and West of the colony. All 4 feeder locations shall be filmed for the duration of the experiment to monitor bee activity levels.
4. Over 4 consecutive days, the landmark shall be shifted at random to each of the 3 other cardinal point locations. At this stage of the experiment all 4 feeders shall contain 50/50 sucrose/water solution.
5. Steps 1 – 4 shall be repeated for each of the 6 colonies.

### **Opportunity for Skill Development**

The student shall develop the skills required to conduct field based behavioural experiments with stingless bees. At the data extraction and analysis phase they shall learn how to use video analysis software to count the activity levels of stingless bees at the feeders. If they choose to progress to the final stages of the experiment, they will gain data analysis and manuscript writing skills. The goal of this project is to create a publishable dataset. The student will have the opportunity to become a co-author on this publication if they choose to be involved in all stages of the research process.

### **Students are required to have the following skills/meet the following prerequisite(s) to apply**

- The student must display a willingness to learn new skills
- The student must be prepared to work with insects
- The student must be willing to work long hours in the outdoors
- A knowledge of the biology of social insects is desirable but not essential.

## **Project 55: The thermal biology, energetics and nutritional ecology of wild koalas**

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Second Supervisor

### **Project description**

This project forms part of a broader project titled “Understanding and mapping how thermal and dietary constraints combine to restrict koala habitat and determine refugia”. That broader project will address knowledge gaps identified by the Koala Research Plan (NSW) associated with climate change and extreme weather events, namely habitat suitability under climate change, refugia and water supply. Droughts and heatwaves are recognised as important agents of mortality for koalas, and are expected to increase in frequency and severity under climate change. Maintaining an adequate intake of water is essential for koalas to thermoregulation under hot conditions, and koalas famously rely on water in leaves for most of their water intake.

The broader project comprises three components: 1) a field study focused on the thermal biology, energetics and nutritional ecology of wild koalas, which will monitor simultaneously the thermal and energetic physiology, feeding and nutritional ecology, and energy use of wild koalas through a range of weather conditions, to substantially improve our understanding of how biophysical and foliage parameters affect the ability of individual koalas to maintain adequate nutrition, hydration and thermoregulation. These factors ultimately limit the persistence of koala populations. No previous model of koala habitat suitability has recognised that hot temperatures can severely restrict the ability of koalas to feed, and thus to maintain water intake. We will quantify the magnitude of this effect, which fundamentally alters the current dominant paradigm of how heat affects koalas. The effects of temperature on feeding are magnified in the presence of leaf toxins, yet this also is not recognised in current models of koala bioenergetics. 2) Foliar water content is a vital parameter that affects the ability of koalas to persist in hot environments. Using satellite remote sensing with ground calibration, we will measure and map how leaf moisture varies across NSW through a range of climate and weather scenarios. 3) We will integrate the first two components into a new dynamic bioenergetic model for the koala. This model will be used to map the current and projected bioenergetic niches of koalas across NSW under future climate and weather scenarios, substantially improving the accuracy of koala habitat suitability predictions.

This summer scholarship project sits within the first component of the broader project, and will address koala behavioural responses to thermal conditions, and improve our understanding of the digestibility, and hence energetic value, of koala diets

## Project Aims

The overall project aims to understand current and future habitat suitability for koalas by i) investigating the behaviour and thermoregulatory physiology of wild koalas in response to extreme environmental conditions, ii) linking these detailed individual responses with measurements to determine nutritional and water constraints imposed by selected leaf food intake, and iii) integrating this new information about key functional relationships to develop more accurate and dynamic mechanistic models that can predict spatiotemporal variation in koala mortality risk under projected climate change scenarios. Although koalas are well-adapted to coping with exposure to varying environmental conditions, severe heat and drought conditions can lead to mortality and heat-induced water stress limits the extent of suitable habitat for koalas. Biophysical and correlational models (particularly those using weather extremes) both show that critical limits to thermal balance are fundamental to predicting habitat refugia. Despite these insights, confidence and precision of model outputs is limited by uncertainty about how intake and nutrient and water composition of foliage affects the capacity of koalas to resist heat stress, and how leaf moisture content varies at a fine scale over space (i.e. among trees) and time. A better understanding of these model-sensitive constraints might substantially alter our understanding of what is suitable koala habitat, both now and in the future.

By the time the summer scholarship commences, 6 koalas will have been captured in the Campbelltown region and fitted with GPS collars to allow the remote download of koala movement, accelerometry and shade/light exposure data. The koalas will also have been implanted with heart rate loggers and core body temperature sensors, and be fitted with acoustic recording devices to monitor the timing and duration of feeding bouts, however these data will only become available when koalas are recaptured after the summer scholarship project and the recording devices are retrieved. However, it is essential that we manually monitor the behaviour and movement of koalas in order to validate the data that is subsequently logged “on-board” the animals. This behavioural calibration will be the first aim of this summer scholarship.

We have recently demonstrated in Victoria that different eucalypts eaten by koalas can differ dramatically in their digestibility, and as a consequence, in the energy that koalas derive from these diets. The second aim of this study will involve the estimation of the digestibility of koala diets in this study.

## Project Methods

The first aim of the summer scholarship project is to observe wild koalas and record behavioural patterns including the timing and duration of feeding bouts, movement and other activities, and tree choice. These behavioural data will eventually be compared with data collected by devices carried by the koalas, allowing us to ensure that we are subsequently interpreting the recorded data appropriately. Some examples of how behavioural data can be linked to recorded digital data are as follows:

- Devices carried by the koala will record accelerometry data. With careful observation, accelerometry data matched to the time of observation should be able to be linked to particular behaviours, such as walking, climbing, feeding, and grooming.
- Devices carried by koalas will record acoustic signals. Again, acoustic data can be matched in time to behavioural observations, allowing us to identify bouts of feeding and bite rates from the acoustic recordings.
- Temperature and exposure to sunlight will be recorded by the GPS collar and can be downloaded remotely. We need to understand how these data correspond to microhabitat selection by koalas at different times of the day and under different thermal conditions.

The collected data and eventually, the more continuous data collected on-board, will be used to test hypotheses about how ambient temperature affects koala behaviour, including the timing and duration of meals and the choice of tree species and individuals for shelter and for feeding. One key hypothesis is that hot temperatures will reduce the ability of koalas to detoxify and thus tolerate plant toxins and to digest large meals, and that tree choice and feeding behaviour will alter accordingly.

The summer scholar will locate koalas using a VHF radio signal (manual radio tracking) and will make scheduled observations of koala behaviour. The fieldwork will be performed in conjunction with a PhD student and/or a postdoctoral researcher.

A second aim of the summer scholarship project is to improve our understanding of the digestibility of koala diets. This will be achieved by the collection of leaves from trees consumed by koalas and of faecal pellets collected from under these koalas' trees. Silica will be used as an indigestible marker of the extent of digestion. Silica concentrations will be determined precisely in both leaf and faeces using a colorimetric assay, and the increase in silica concentration in faeces relative to leaf can indicate the extent of digestion that has occurred.

### **Opportunity for Skill Development**

The student will obtain experience in field research techniques, including radio tracking, remote download of GPS data, collection of leaf and faecal samples, and techniques of behavioural observation, recording and data analysis. There is a possibility that the student will observe koala captures and handling. The student will also gain experience in the laboratory analysis of leaf chemical attributes.

### **Students are required to have the following skills/meet the following pre-requisite(s) to apply**

- Some demonstrated experience with fieldwork or a background which demonstrates that the student is comfortable working in the bush, sometimes under uncomfortable circumstances, including at night.
- The ability to take detailed and reliable notes and to manage and summaries data and samples.
- An ability to follow clear instructions and a demonstration in order to undertake laboratory chemical analysis of leaf properties



## **Project 56: Controlling disease induced losses to agriculture and forestry**

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### **Project description**

Both in agricultural and in forestry settings, death caused by the soil-borne pathogens lead to serious losses that threaten the sustainability of our forestry and agricultural industries. Losses in productivity in these sectors are especially important to consider as increasing frequency of climactic extremes are leading to a higher percentage of crops with stressed and immunocompromised plants, a situation that will result in ever decreasing productivity to these key industries in Australia. As such extremes are predicted to come more frequently in the coming decades, we must begin future-proofing these ecologically and economically vulnerable populations. Depending upon the crop studied, infected plants usually show symptoms such as fungal hyphae penetrating root surfaces, wilting and loss of vitality to the plant, and sometimes the production of mushrooms clusters aboveground. Once soil-borne pathogens are established, symptoms in the standing crops typically include crown thinning, reduced height growth and distress fruiting that eventually degrades into the full death of the host. Current prevention strategies of soil diseases largely rely on proper hygiene, but once the pathogen arrives it will progressively spread and cause the death of host crops across large areas. Therefore, we need other means by which to combat these pathogens. One method will be to identify how pathogens alter host plant physiology with different proteins and metabolites to promote disease progression so that we can eventually breed plants that are immune to these pathogenic chemicals. In order to do this effectively, we need to gain a better understanding of how pathogenic fungi are able to cause disease.

This summer project will advance our understanding of the 'tools' used by pathogens to induce plant death using two different tactics:

1. The student will get an opportunity to do some practical lab work (30-50% of the project) by characterising the role of three pathogenic proteins that may enable pathogens to cause cell death of plant tissues. The successful candidate will learn a variety of molecular biology techniques necessary to clone the genes that code for the proteins and then create genetically modified plants expressing the fungal proteins. The student will then characterise the impact that these fungal proteins have on root development, health, and biochemical activity.
2. The student will spend the remaining portion of the project doing computer-based analyses of a metabolomics data set of plant roots being colonized by a pathogen. The student will seek to identify how different pathogens use different secondary metabolites during the colonization of a disease susceptible host. This will teach the student a variety of advanced data analysis packages as well as familiarize the student with how to use statistical analysis to mine large data sets.

The outcome of this project will be a better understanding of the plant cellular processes impacted by the disease so that new crops can be chosen in future that are resistant to the pathogen and will equip the student with career-relevant skills.

### **Project Aims**

- Analyse a metabolomics data set and write up the results of the analyses with appropriate figures
- Clone three fungal genes with a putative role in host colonization
- Transform plant with cloned genes
- Phenotype transgenic plants and write up results with appropriate figures

### **Project Methods**

This lab-based component of this project (30-50% of the time) will use a range of classical molecular biology techniques including nucleic acid extraction, nucleic acid manipulation, Polymerase Chain Reaction (PCR), gene cloning, as well as plant phenotyping such as cell death analyses and growth characterisation. The student will be engaged in this process in a hands-on manner with supervision from the two nominated supervisors. The student will be directly involved in the planning and running of the experiments. With the mentoring of the supervisors, the goal will be to get the student comfortable enough with the techniques used so that they may also be able to work more independently by the end of the scholarship period.

The computer-based component of this project (50-70% of the time) will use a range of data analysis platforms for untargeted metabolomics as well as teach the student a number of different statistical analysis packages using the R programming language. As with the laboratory-based component of the project, the goal will be to get the student comfortable enough with the techniques used so that they may also be able to work more independently by the end of the scholarship period.

### **Opportunity for Skill Development**

Often within undergraduate courses, students gain the opportunity to do simple molecular biology techniques, but rarely do they have the opportunity to develop these skills beyond a very basic knowledge. The successful student who undertakes this project will have regular one-on-one mentoring to develop molecular biology techniques such as nucleic acid extraction, gene cloning, plant mutagenesis, as well as help with planning experiments and coaching on statistical data analysis. Further, students rarely get the opportunity at the undergraduate level to work with large data sets, nor to learn the statistical approaches needed to ask scientifically relevant questions of these resources. For the metabolomics component of this project, the student will get this opportunity to learn job-relevant skills that will help them in their future career endeavours.

### **Students are required to have the following skills/meet the following prerequisite(s) to apply**

The student will be required to have a background in microbiology with some past experience in a laboratory setting.

## **Project 57: Next Generation Sequencing and Bioinformatics Analyses of Bacterial Genomes from Insect Pests**

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### **Project description**

Bacteria have a single ring chromosome comprised of DNA that is mostly organised into units called genes. Most of these genes encode proteins and determine the types of functions that bacteria can perform. Some bacteria live inside the cells of insects (intracellular) and are highly beneficial to the insect due to the types of proteins they can produce, while others can be deleterious. It is important to understand the proteins these intracellular bacteria produce, as they affect the biology of their host and may be used either as novel, environmentally-friendly biological control agents for insect pests, or, conversely, as target to disrupt function in insects. Therefore, this research is to be seen of relevance to pest management in agriculture, forestry and natural environments.

The DNA of whole chromosomes can be sequenced using modern technologies. Therefore, it is possible to determine all the genes they contain and, consequently, all the proteins bacteria are able to make. Previous research has enabled us to produce large datasets of short DNA sequences using a “shotgun” sequencing method (part of what is widely called Next Generation Sequencing [NGS]). In this method, whole chromosomes are broken into small pieces, the small fragments are sequenced and then overlapping fragments are assembled using specialised bioinformatics programs to generate long contiguous DNA sequences. By having complete chromosome assemblies (and therefore the bacterial genomes), we can then compare the gene content and gene sequences found among closely related bacteria. No prior experience of NGS is required.

Psyllids are a diverse group of insects that can damage plants through defoliation and spread of plant pathogens. They feed on plant sap and, because of this restrictive diet, they have a strong association with intracellular bacteria that can synthesise food components missing in their diets. The bacterium, *Carsonella*, which helps the psyllid meet its nutritional requirements, is found in all psyllids, but all psyllids also appear to have other intracellular bacteria (secondary endosymbionts) that are specific to particular psyllid species or genera that may also provide their hosts with various benefits.

Markus Riegler’s research group at the HIE, Western Sydney University have been studying psyllids since 2012 when a severe outbreak of a previously undescribed species of lerp-forming *Cardiaspina* psyllid infested large numbers of the dominant eucalypt species, the greybox (*Eucalyptus moluccana*), in western Sydney’s Cumberland Plains Woodland. Our interest has been to study the psyllids and their association with bacterial endosymbionts (symbionts that live within host cells), with our main focus on Australian psyllids which feed on eucalypts, including many members of the genus *Cardiaspina*. Through comparative genomics, which is the sequencing and annotation of the genomes followed by comparison with other similar organisms, we can identify metabolic pathways, infer function and determine evolutionary relationships. Sequencing DNA from a psyllid generates data for the psyllid as well as its bacteria, including *Carsonella* and secondary endosymbionts. The laboratory aspect of the sequencing has been

done, and substantial sequencing data is available; the assembly, annotation and analysis is proceeding, but has yet to be completed. There is scope in this project to learn and perform all aspects of the bioinformatics pipeline – assembly, annotation and analysis – with the aim of gaining a better understanding of the impact of these bacterial endosymbionts on the biology of these psyllids, and may lead to strategies for managing outbreaks.

### **Project Aims**

The overall aim is to assemble and compare the genomes (here the single chromosomes of the bacteria) of the intracellular bacteria associated with *Cardiaspina* spp. associated with eucalypts. The specific aims are to:

1. Utilise genomic datasets derived from NGS to assemble the complete genomes of the secondary endosymbionts from these insect hosts; and,
2. Use a variety of open source bioinformatics tools to compare these genomes by identifying the genes they contain and comparing gene content.

### **Project Methods**

Next Generation Sequencing of genomic DNA from an insect provides DNA sequences of the insect host and its microbiota (microbes associated with the host). Genomic datasets have already been generated from DNA extracted from six *Cardiaspina* species. The genomes of the symbiont, *Carsonella*, from these host species have already been assembled. The next phase is to assemble the genomes of the secondary endosymbionts, including *Sodalis* and *Arsenophonus* bacteria from these datasets and compare their complements of genes using standard NGS technologies.

1. Assembly of secondary endosymbiont genomes. Assembling any organism's genome, including small bacterial genomes, can be difficult and labour intensive; however, it is a task made more manageable when reference genomes are available that are expected to be similar. Published reference genomes are available, making the assembly of *Sodalis* or *Arsenophonus* an achievable task.  
The work will use 'CLC genomics workbench'. This is a computer system with a graphical interface that performs bioinformatics processes including assembly of short sequences into longer contiguous sequences (contigs). The BLAST algorithm is used to identify contigs of interest, following which they are mapped to target contigs in CLC then a manual examination of the mapping quality is performed to verify the complete genome sequence.
2. Comparative genomics. Annotating genomes (that is, finding the genes and naming them based on similarity to other known genes) and comparing genomes to determine gene content is then performed using bioinformatics tools, many of which use the Linux command line to implement programs. The genomes of other intracellular bacteria have been assembled and compared in this way by the research group at HIE, and a robust pipeline for all methodologies has been produced that will be the guide for this project.

### **Opportunity for Skill Development**

This project will use NGS and bioinformatics analyses, technologies revolutionising medicine and biology. Although NGS data can now be acquired with ease, its downstream analysis is problematic, as data sets are growing in size, producing a 'data deluge' that requires analysis by specialised personnel. The student will receive hands-on training and the project provides an opportunity to gain experience and knowledge of NGS technologies. These are highly sought after and transferrable skills and can be applied to applications of NGS in other areas of biology or medicine. In addition, the project will enhance the student's competencies in molecular biology and bioinformatics through gaining further knowledge of genes and their functions.

**Students are required to have the following skills/meet the following prerequisite(s) to apply**

The project will consist entirely of computer-based analysis of biological data; therefore, attention to detail, a knack for problem-solving, and the ability and desire to spend time on detailed analysis is a must. No direct familiarity with Linux command line or any computer language is a prerequisite, but basic computer skills with Microsoft Word and Excel, and the aptitude to understand and the desire to learn the basics of command line is necessary. A student's experience may come from genetics and molecular biology, or from computer / data science.

## **Project 58: Functional traits, hydraulic resistance and recovery capacity in Eucalyptus**

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### **Project description**

Drought events associated with climate change are impacting ecosystems all over the world. Understanding drought vulnerability is important to prioritize conservation and management of species. Measuring drought vulnerability/drought tolerance has been a task for researcher in last few years showing, as a general trend, that climate-origin is associated with hydraulic vulnerability; a proxy of drought vulnerability. Nevertheless, there is a lot of variability linked possibly with different strategies to deal with drought. Finding gaps and factors from this variability will help to prioritize species conservation to a finer scale within ecosystems and biomes.

Eucalyptus genus consist of an excellent example to study this variability since is widely distributed across diverse climates in Australia, expressing differences in habit, height and morphological traits that should have an effect in the integrated strategy to drought. Additionally, Eucalyptus has diverse regeneration strategies that are usually studied under fire disturbances but not drought and should influence specially recovery processes. Since that in this project we focus in represent a gradient of climate of origin (related water variables as temperature and precipitation) from three regeneration strategies (seeder regenerators, lignotuber producers which re-sprout only from trunk-base and lignotuber and epicormic re-sprouters, known as combinational regeneration, which resprout from base and upper parts from trunk) to explore its variation in morphological traits, hydraulic vulnerability (capacity to maintain water transport under drought) and recovery. We are Including widely and restricted species since most of studies include wide distribution species in which, theatrically, could be more potential for adaptation.

The summer project will encompass the performance and physiological assessment of around 12 species of Eucalyptus during well-water conditions under nursery conditions. Growth and morphological traits are going to be measured as well as the hydraulic vulnerability of every species. This first part will allow to determine the effect of climate of origin and regeneration strategy under well-watered conditions and its influence on hydraulic vulnerability of stem and leaves. This first piece of the puzzle will be after connected with measurements of water control strategy under drought and subsequent recovery.

### **Project Aims**

- Understand drought vulnerability of Eucalyptus in terms of climate, functional and life history traits
- Characterize performance and morphological functional traits of Eucalyptus of diverse climate, distributions and regeneration strategies under well water conditions.
- Determine effect of climate and regeneration strategy on hydraulic vulnerability and its relation with particular functional traits.

## **Project Methods**

Eucalyptus plants will be around four months old starting the summer scholarship period under nursery and well water conditions with drip irrigation automatized system. The plants are going to be randomly located in blocks in a Polytunnel structure. Measurements of height, non-destructive leaf area (with a transparent grid), basal stem diameter and number of leaves will be measured in eight to ten plants per species every fortnight.

Additionally, four Eucalyptus plants per species are going to be harvested to measure hydraulic vulnerability by the optique technique in which a plants are let to dehydrate in a bench, while its water potential (proxy of water quantity) is measured with a psychrometer installed in the stem-exposed xylem; formation of bubbles are followed installing clamps with installed Raspberry Pi camera illuminated with LED which take photos during the dehydration. The images will be processed with the software Image J to determine hydraulic vulnerability curves in which water potential is related with apparition of embolism events (bubbles). The student will be trained to use this technique. In the harvested plants and once finalized he dehydration processed, wood density will be measured as volume/dry weight and leaves, stem and roots will be separated and dry oven for at least 24 h under 75°C to weight dry biomass of each and total plant biomass. Finally, ten leaves will be collected from around five plants to determine specific leaf area; for that leaves will be scanned to determine leaf area with Image J software and leaves will be oven dried to determine dry weight.

The student will collect data from a pool of all species and will analyse them to characterize species under well water conditions and its relation or influence in drought vulnerability.

## **Opportunity for Skill Development**

The student will gain skill and experience in understanding how an experiment is set, learning to measure plant performance in terms of different measures across the plants as well as being methodical in managing big quantities of plants in experiments. Also the student will learn to measure morphological traits as specific leaf area and wood density that are pretty useful in determine ecological function and strategy. The student will learn the impressive optic method to determine hydraulic vulnerability curves; this will approach him/her with novel tools in physiological research which we hope will motivate its curiosity and research expertise. Finally, the student will be guide to analysed the data and obtain graphics and statistics that led to make and support solid conclusions.

## **Students are required to have the following skills/meet the following pre-requisite(s) to apply**

First and most important thing is motivation and commitment with the experiment. Understanding of basic plant biology, good organisation and analytical skills. Accuracy in taking measurements.