The CILR launched its innovative one-week teacher professional development program in July.

“STEP IN LABS!” (Science Teachers Education Partnership IN Legumes And Biotechnology Studies!) was developed in partnership with Education Queensland as part of the Queensland Government’s Spotlight on Science initiative.

The program consisted of an extensive series of lectures and workshops designed to educate high school teachers on the cutting-edge techniques used in plant science.

Eleven teachers from Queensland secondary schools as far away as Goondiwindi and Thuringowa (near Townsville) took part in the program.

The CILR’s Director Professor Peter Gresshoff said the program aimed to show teachers the sophistication of plant biotechnology research.

“We hope the teachers will be inspired by this experience and take their enthusiasm back to the classroom.”

“We would like to see more students undertaking tertiary studies in plant biology,” Professor Gresshoff said.

The teachers themselves were enthusiastic and willing to learn more about extended experimental investigations, an area they felt required new training. In addition, they were hoping to gain some experienced contacts in scientific research. Many of the teachers were also excited at the prospects of taking some of the experimental workshops back to their classrooms, in particular those involving nitrogen fixation and nodulation.

The CILR intends to follow up this program with some of our Post-Docs visiting the teachers in their respective schools, and talking to the students about plant science and the importance of legumes. This is another area where the teachers felt education was required.

We would like to thank all of our Post-Doc researchers, lab assistants, and PhD and honours students for the time and effort they spent giving lectures and setting up workshops in order for the teachers to gain a thorough understanding of the research conducted by the CILR.
Pod news

Legumes and Lollypops a Big Hit!

During July the Centre took part in the University of Queensland’s Experience Science Program for Year 11 and 12 school students. More than 100 students from a number of schools throughout S.E. Queensland participated in a two-hour plant science workshop “DIY Plant Genetic Engineering - The Backyard Scientist’s Toolkit” put together by the Cooperative Research Centre for Sugar Industry Innovation through Biotechnology (CRC SIIB) and the CILR.

Students extracted DNA from bananas, subcultured orchid seedlings aseptically, stained fungal hyphae and observed them microscopically, and investigated nodulation. CILR researchers Yu-Hsiang Lin and Cuc Nyugen led a nodulation workshop designed to demonstrate how single point mutations can create both non-nodulating mutants and supernodulating mutants.

Students from Macgregor High School said they greatly enjoyed the experience. “We are keen to learn more about plant science, and tertiary courses offered in the areas of plant science and biotechnology”.

“The best part of an experience like this is that you get to deal with actual scientists in a real university”, said John Paul College students. Of course, the free lollypops handed out at all the various workshops didn’t go astray!

L-R: Students measuring soybean shoot length; Viewing nodules under a microscope; Cuc quizzing students using CILR’s new postcard quiz.

Stress and Somatic Embryogenesis

In a recent paper published in Plant Cell Reporter (July 2006), CILR scientists Dr Kim Nolan, Nasir Saeed and Assoc. Prof. Ray Rose, from the Newcastle Node, have investigated the stress kinase gene MtSK1 in relation to somatic embryogenesis in Medicago truncatula.

*M. truncatula* can be regenerated by somatic embryogenesis by the use of a suitable genotype and an auxin plus cytokinin. The stress response induced by explant wounding and culture is increasingly recognised as an important component of somatic embryo induction. MtSK1 was cloned and investigated, and found to be a member of the SnRK2 sub-group of the SnRK group of plant kinases. These kinases play a role in stress responses of plants. MtSK1 expression in induced by wounding in the cultured tissue independent of auxin or cytokinin. The likely role of MtSK1 in stress-induced signalling will facilitate the relating of stress-response pathways to auxin and cytokinin-induced signalling and in the understanding of the molecular mechanisms involved in the induction of somatic embryogenesis in *M. truncatula*.

Director’s report

We tend to focus on the scientific achievements of the Centre… publication, patents, seminars, grants. But there are other benefits that we have achieved, namely in the field of human resource development and capacity building. Achievements in this area have long-lasting effects caused by an odd multiplier effect.

As I look back over my career as a plant scientist, I most remember the people with whom I have worked. Yes, colleagues and collaborators are important but most significant are the younger people, the undergraduates, the Honours students, the technical staff, the office staff, the postgraduate students and the postdoctoral research staff to whom I look back with a sense of achievement.

Many started as total novices; tolerance, teaching and a lot of scribbles and arrows on spare paper were needed. Many had no clue of practical molecular biology or plant biology, their genetic knowledge was based on prokaryotic haploids, and often they spoke rather poor English. But all had a willingness to learn, an urge to succeed and underneath it all, an excellent level of intelligence. Many came as rough diamonds, some came as stars. In the end they all succeeded. They all passed through my academic life, developed a varying levels of friendship and all moved on to a fascinating array of occupations. Some moved continents, settled in new countries and gave their children a new life. Some returned to their country and brought improved education, and science awareness.

Many of them are in academia; a Deputy Vice-Chancellor, Dean of Science, professsors, senior lecturers, research officers, head technicians, and research group leaders. Many of them became internationally recognised scientists both in plant molecular genetics as well as animal biology (medicine, entomology). Some took novel routes towards their occupations as public servants, communicators, patenting officers, quarantine administrators. Others went into commercial science rising to the level of Director of International Marketing in one key Australian instrumentation manufacturer and supplier.

I am proud of them all and thank them for their patience in being supervised by me and their efforts of pushing science forward. As an academic my legacy is both that of knowledge as well as people. This capacity building has an immense multiplier effect as these people move on, educate others, facilitate new things which are all beyond my capabilities. Yes, I have two wonderful sons, but I also have a large academic family of whom I am proud. I guess it is a matter of genes and memes that we all pass on (c.f., The Selfish Gene by Richard Dawkins, for reference to ‘memes’).

As an ancient Chinese proverb states: If I am interested in the next year, I grow rice; if I am interested in the next decade I grow trees; if I am interested in the next century, I grow people. How true, especially when we consider the need for plant scientists in a future that sees demands for sustainable production of food, biomaterials and fuel.

Peter M. Gresshoff
Director, CILR.
August 2006

Hospital Technology in Plant Science Labs: Soybean plants on the Drip!

If you have been wandering about the intravascular drip arrangements seen around the Brisbane node, it is no more than Yu-Hsiang Lin (see Pod People p.8) supplying soybean plants with leaf-extracts from various sources. The method involves cutting the petiole, connecting a snug-fitting tubing which is attached to a Feeding Reservoir (nothing more than an Eppendorf tube) supported by a pole (no more than a food skewer), then removing the air bubble with a small hypodermic needle and a subsequent period of pant growth to create a bioassay. Yu-Hsiang got great results from this approach analysing the leaf-derived NARK factor controlling Autoregulation of Nodulation.

Soybean plant receiving petiole feeding (a blue food dye is used for visualisation. Photo: Y-H.Lin)
The formation and maintenance of nodules on legume roots by rhizobia, is a process strictly controlled by the plant. While rhizobia contribute fixed nitrogen to the plant, the plant, in turn, provides large amounts of photosynthate to the rhizobia. There are multiple levels of control over the initiation of a nodule, and we have been interested in the plant signals that initiate nodules and control nodule numbers on the root system. We focused on the plant hormone auxin as a potential regulator, because it is known to be necessary for controlling cell divisions and organ differentiation.

We have sought to answer the following question: how does the plant control where auxin accumulates to regulate meristem formation? Three recent papers from our lab have given us some clues about how the plant regulates auxin transport and accumulation.

First on the scene was Giel van Noorden, who came from The Netherlands to undertake his PhD with Prof. Barry Rolfe and Dr Uli Mathesius on the question of how shoot control of nodule numbers is regulated. It is from the shoot that the so-called auto-regulation signal is sent to the root to prevent the initiation of excessive numbers of nodules. Most of the auxin in the plant is synthesized in the young leaves and transported actively down to the root. Therefore, Giel tested whether long distance auxin transport might be part of the autoregulation process. In collaboration with Dr Christine Beveridge from Queensland University, Giel devised and optimised a system to measure long distance auxin transport in whole seedlings. Giel’s efforts resulted in a major breakthrough when he discovered that rhizobia inoculated at the root tip somehow manage to tell the shoot to down-regulate the supply of auxin to the root. Since the down-regulation of auxin coincided with the timing of autoregulation, it might be a mechanism to inhibit nodule formation by starving them of auxin and possibly carbohydrate supply. Giel used an autoregulation mutant in Medicago truncatula, sunn (super numerary nodules), kindly provided by Dr Julia Frugoli from Clemson University in the US, to show that this mutant is unable to down-regulate auxin transport after inoculation. Instead it appears to constitutively oversupply the root with auxin, which might be a factor in allowing excessive nodules to form on the roots. This was confirmed by direct auxin measurements thanks to the collaboration of Dr John Ross and Prof. Jim Reid in Hobart! We are now keen to find out how the autoregulation gene, a leucine-rich repeat receptor like kinase (in soybean termed NARK for nodule auto-regulation receptor kinase), regulates the loading of auxin from the shoot to the root. This is an important question because auxin is a global regulator of organ formation in general, including lateral root formation, which is important for establishing root architecture.

While auto-regulation is one mechanism of regulating nodule numbers, it is not the only one the plant has up its sleeves. Ethylene is another plant hormone important in development. Ethylene is a regulator of defense responses as well as being able to regulate auxin transport, although this process is poorly understood in legumes. Joko Prayitno, who is just finishing his PhD thesis on the regulation of nodule numbers by ethylene, again co-supervised by Barry and Uli, found that ethylene is a second regulator of auxin transport which acts independently of the autoregulation mechanism. Ethylene acts locally in the root, as Joko established from grafting experiments. Joko used an ethylene insensitive mutant of M. truncatula, called sickle, for the sickle-shaped root that forms as a result of dense clusters of nodules. Joko found that sickle is different from sunn in that it doesn’t oversupply the root with auxin. However, it was also insensitive to long distance auxin transport inhibition by rhizobia, suggesting that an ethylene dependent root control of long distance auxin transport exists.

If long distance auxin transport might control nodule numbers, does auxin transport also control the initiation of nodules after the first inoculation? To answer this question we developed a method to measure local auxin transport at the site of inoculation of the root with rhizobia. We knew from previous experiments that rhizobia locally and transiently inhibit auxin transport, followed by auxin accumulation to start a nodule. Using both supernodulating mutants, Giel and Joko showed that the long distance regulation of auxin transport is independent from short distance auxin transport regulation. Both mutants showed local auxin transport inhibition and auxin accumulation at the site of the first nodule formation, even though they had defective long distance auxin transport. However, both mutants end up with more auxin at the nodule initiation site, and this might stimulate more nodules to form initially in these mutants. In the sickle mutant, this
While ethylene might be a negative regulator of auxin accumulation at the nodule initiation site, the skl mutant still showed the early and transient local auxin transport inhibition that we think is important for establishing a nodule primordium. So what regulates this early auxin transport inhibition? Previous studies suggested that plant flavonoids could act as internal auxin transport inhibitors. Flavonoids might bind to a protein that interacts with auxin transport proteins. Flavonoids are induced by rhizobia in about the same location that the auxin transport changes occur. Despite this correlative evidence, it was previously not possible to test genetically if flavonoids are required for auxin transport inhibition. No flavonoid deficient mutants are available in legumes, as most genes of the flavonoid biosynthetic pathway are encoded in multigene families. This was a sticking point until Anton Wasson came along to try his luck at an Honours project to silence the flavonoid pathway in the model legume Medicago truncatula.

Anton designed an RNAi construct to silence the first gene of the flavonoid pathway. We were fortunate to have Dr Flavia Pellerone, a post doc with expertise in plant transformation and plant molecular biology, who turned our lab into a transgenic factory for gene knockouts using RNA interference. Soon plants producing transgenic “hairy roots” that lacked flavonoids were created. When these plants were inoculated with rhizobia they didn’t form nodules, as expected. Measuring auxin transport in these roots showed that the flavonoid-deficient roots had higher transport rates of auxin (as expected because flavonoids inhibit auxin transport), and most importantly, inoculation with rhizobia did not result in the inhibition of local auxin transport. Hurray!!

We now think that the plant locally regulates auxin transport inhibition using flavonoids. This is followed by increased auxin transport and accumulation, which are regulated partly by local ethylene signalling. On top of this local regulatory system, long distance control of auxin loading from the shoot to the root is regulated by the sunn gene, and might be important for determining how many nodules the plant allows on its roots. Being able to control where the auxin goes and how much of it is transported might give us the ability to regulate plant architecture in general.

References:


Surprising GM Food Attitudes

Earlier in May, CILR PhD student Lucy Carter travelled to Cairns to present at Biotechnology 2020, a public forum organized by Biotechnology Australia. Lucy’s presentation discussed general ethical issues associated with the use of novel gene technology in food products. Some of the issues covered related to the use of mandatory labelling regimes, potential applications of new biotechnologies, and cultural concerns associated with the use of novel technologies. Some popular myths surrounding new biotechnologies were also busted. The audience consisted of various stakeholders including professionals, semi-professionals, and the lay public.

An opportunity was presented to speakers to ask one question of the public following their presentations. The question posed by Lucy was, “Knowing that all genetically modified products sold in Australia and New Zealand require mandatory labelling, do you intend to purchase products that contain genetically modified ingredients?” The audience response was not surprising. 56% of respondents answered “yes”, while 19% answered “no” and 25% answered, “not sure”. Some discussion afterwards revealed that of those that answered in the negative, the reasons put forward were not related to a concern about potential harm to human health as commonly asserted in the literature. In general, respondents who said they would not purchase GM products simply intended to support local manufacturers whose products were grown and sold locally, irrespective of the process used. The current generation of GM products was thought to be largely beneficial to multinational agribusiness, not consumers.

These responses have no statistical power and contain some methodological bias in that attendees of such events are often self-selecting. However, these responses are of interest in that they provide a deeper understanding of other, less often discussed concerns about GM technology. Human health concerns are often touted as the primary reason the public will not purchase (or consume) GM products. This may be so in the UK as an unfortunate consequence of the Bovine Spongiform Encephalitis (BSE or ‘mad cow disease’) health crisis in the 1990s. For Australians however, concerns about GM products appear to be less health-related and more linked to economic and political issues.

Soul Kitchen

“The difficulty lies, not in the new ideas, but in escaping from the old ones, which ramify...into every corner of our minds.”

John Maynard Keynes (1935).

“Prioritise......

Everyday do something on your urgent list and something on your admin list. Also do something for yourself, something you enjoy and that keeps you entertained. Something that ensures the passion for your work is retained.”

Dr Christine Wells (2006)

Coming up in 2006 . . .

August
20-25 8th International Congress of Plant Molecular Biology
Adelaide, South Australia
27-30 Genomics in the Barossa
Barossa Valley, South Australia

September
24-28 ComBio
Brisbane, Queensland

Later...
January 2007 Plant and Animal Genomics Conference
San Diego, CA
Bio-ethics Update

The 11th conference of the Australasian Bioethics Association was held at Queensland University of Technology, 5-8 July 2006. Bioethics has typically focused on ethical issues in biomedicine, though its net has occasionally been cast more widely to include problems in the biological sciences more generally.

Professor John Mattick addressed ethics and ideologies in biology and medicine. He reviewed the spectacular developments that have taken place in genetic understanding, especially over the last ten years, which has taken us to the point where individual genomes sequencing may be a part of the medical diagnostic repertoire within the next five years. This would revolutionise genetic discovery, genetic diagnostics, therapeutics and personalised medicine. However Professor Mattick believes that there is a darker side that needs to be addressed. The rapid developments in fields of genetic technology have generated public concern. He singled out GM of plant species, as well human GM, as cases of this concern. An emerging obdurate ideological fundamentalism is opposed in principle to any genetic manipulation of organisms. Mattick argued that there is an urgent need to promote better understanding of science and technology and its benefits, to ensure that prejudice and ignorance do not prevent us from harvesting the benefits these technologies present.

Mattick suggested that the challenge is perhaps easier in connection with medicine than it is in other areas of fruitful genetic research, because when it comes to the crunch, concern with health always trumps. This perhaps is illustrated by the fact that there is far less objection to the use of GM in connection with medicine and pharmaceuticals than there is with food production and agriculture.

An emerging conviction is that, where misgivings about the applications of genetic technologies arise, the onus of proof is shifting towards opponents of these technologies to demonstrate the likelihood of significant harm before these technologies can be constrained. Perhaps a cautious precautionary approach was more easily justified in the early stages of the development of these technologies, but as their benefits become increasingly clear, and the supposed hazards turn out often to be more apparent than real, workers in the various fields of genetic inquiry seem increasingly irritated by exaggerated concerns expressed by opponents of these technologies. That of course isn’t to say that anything goes. There is a clear sense that there are accepted boundaries not to be transgressed.

Dr Stephen Coleman, from the School of Humanities and Social Sciences at the University of New South Wales read a paper dealing with public concern about GM of crops such as canola, and the question of food safety. Dr Coleman discussed the appropriate role of liberal democratic governments, like those in Australia and New Zealand, in the regulation of the sale and advertising of food, especially in connection with dealing with the way that values and public opinion should be addressed and balanced against scientific fact.

A problem arises when the public perception of risk is based on poorly understood, or exaggerated concerns. This problematic situation perhaps can only be addressed by a gradual process of public education. Food regulation typically considers risks more carefully than benefits, and a great deal of work remains to be done to address the problem of squaring public policy, always (of course) sensitive to public opinion, with the sound empirical understanding of scientific facts and the genuinely rational assessment of risks. Lucy Carter, a CILR-supported PhD student (currently completing a thesis on ethical issues in relation to GM), presented a paper exploring indigenous world-views and their capacity to illuminate ethical issues raised by novel gene technologies. (A report of Lucy’s participation in a GM food forum appears on page 6.)

Although, understandably, biomedical issues continue to dominate the bioethical agenda, it is clear that emerging ethical issues associated with genetic technologies have resulted in issues relating to the plant biosciences becoming less peripheral to bioethical inquiry than they once were.
Pod people

First Class Honours for CILR Student

CILR honours student, Yu-Hsiang Lin, was recently awarded first class Honours for his thesis titled “A bioassay for an analysis of the soybean SDI signalling molecule”. Yu-Hsiang’s research project involved a novel method of feeding soybeans with a ‘drip’, similar to those used in hospitals for human patients (see p.3). Yu-Hsiang’s Petiole Feeding Assembly (Gravity System) was developed to feed extracts from both wild-type (Bragg) and AON (Autoregulation Of Nodulation) deficient mutant (nts1007) leaves into the supernodulating mutant nts1007. nts plants have mutations in the GmNARK gene, discovered by CILR researcher Assoc. Prof. B. Carroll and Prof. Peter Gresshoff in 2003. nts1007 plants fed with wild-type leaf extracts showed a significant reduction in nodule numbers, similar to the nodule number of wild-type plants. nts1007 plants fed with nts1007 (mutant) leaf extracts showed supernodulation. Therefore Yu-Hsiang was able to demonstrate that an inhibitory substance exists in leaf extracts, consistent with other work completed by CILR scientists.

Yu-Hsiang was born in Taiwan and moved to Australia in 1996. He completed his undergraduate study at UQ, graduating with a Bachelor of Biotechnology with first class Honours in July this year.

Yu-Hsiang said he greatly enjoyed completing Honours within the CILR. In particular he appreciated conducting novel research within a real research environment and working with real scientists, something he hadn’t experienced during his undergraduate coursework studies. “As the youngest member of the lab, it was really nice to have the guidance of more experienced scientists, especially when things fail, as they so often do in research”. Yu-Hsiang also commented on the level of camaraderie between lab members, and especially enjoyed the progression of friendship towards activities outside the laboratory environment.

Next on the agenda for Yu-Hsiang is furthering his studies towards a PhD, either in Australia or overseas. However before he embarks on the next phase of his life, Yu-Hsiang has some advice for students either currently completing Honours within the CILR, or considering joining the CILR. “Commitment is the most important aspect of Honours. You need skills and knowledge, but you also need commitment in order to gain the most out of your honours research year”.

New Education and Outreach Manager

Sadly, we must bid farewell to Dr Stephanie Williams from the CILR. Stephanie has been the Centre’s Education and Outreach Manager since mid-2005, and during her time here has organised a number of workshops, established our new website, and worked extremely hard to increase publicity regarding legumes and their importance to society. Stephanie is leaving us to take up a new position with Research Australia, based in Sydney.

Our new Education and Outreach Manager is former CILR Honours student Lisette Pregelj. Lisette graduated with first class Honours from the Centre in 2004, after completing her thesis titled “Investigation of the Transcription of the Phenylpropanoid Pathway in Soybeans”. Since then, Lisette has gained her Masters in Technology and Innovation Management from UQ and travelled throughout Europe and Malaysia. She is excited to be re-joining the Centre, and looks forward to continuing Stephanie’s work.