

Meeting report

Is there gold at the top of the beanstalk?

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A report on the 3rd International Legume Genetics and Genomics Conference, Brisbane, Australia, 9-13 April 2006.

More than 230 delegates from 25 countries gathered in Australia recently for the latest annual conference on legume genetics, where they heard an optimistic forecast for the future of legumes as both an economic food commodity and a research topic, and talks on topical subjects ranging from noncoding RNAs, signal transduction, stem-cell research, and genomic profiling to pathogenic and symbiotic relationships. We have selected a small number of highlights in these areas from the many talks that were given at the meeting.

A valuable resource

Since the 1960s, the consumption of pulses has been going down, even in developing countries, giving ground to meat and grains. Concurrently, interest in legume research has waned and overall, plant-science graduates are scarce. But as Kofi Agblor (Saskatchewan Pulse Growers Association, Saskatoon, Canada) reminded the meeting, remarkable parallels exist between the recent economic history of mineral and energy resources and the status of legumes. In the 20 years before 2001, mining became marginally profitable and unfashionable, with declining investment in exploration, mines, and infrastructure. Since the beginning of this century, however, mineral and energy commodity prices have exploded. Today, everything in this field is at a premium, from geology graduates to tyres for mining vehicles. Similarly, economic and social forces are gathering that should again drive up legume cultivation and consumption, and give legume research a higher profile.

As Agblor pointed out, several strands are coming together to make legumes more attractive: protein security is precarious,

as poultry, meat, and seafood are variously threatened by bird flu, bovine spongiform encephalopathy (BSE), overfishing and heavy-metal contamination; rates of diabetes and obesity are sky-rocketing in developed countries as a result of diets containing too much fat and carbohydrate; and higher energy prices are increasing the cost of nitrogen fertilizers. The high protein and oil content of pulses and the ability to store protein easily in this form, combined with the ability of legumes to provide their own nitrogen, is set to make legumes attractive crops again. They may even have a future as a biofuel.

Nancy Terryn (Ghent University, Ghent, Belgium) reminded us, however, that there are barriers to exploiting the full potential of legumes, especially in developing countries. She argued that although low-level cultivation farming practices are environmentally friendly, the resulting poor yields may be unacceptable when it comes to feeding growing populations, as low-level cultivation requires up to three times the land area to produce the same crop biomass compared to conventional farming practices. And potential new crop legumes suitable for developing countries are not domesticated or improved for agricultural use.

RNA regulators

John Mattick (University of Queensland, Brisbane, Australia) presented evidence that multicellular organisms, including vascular plants, express a large repertoire of noncoding RNAs, and that their likely role is to regulate and direct the complex pathways of development in multicellular organisms. The function of some noncoding RNAs is gene silencing, and Bernie Carroll, also from the University of Queensland, reported elegant grafting experiments with *Arabidopsis* that showed the movement of RNA silencing from roots to shoots and identified several genes required for the translocation of RNA silencing. In this context,

Penny Smith (University of Western Australia, Crawly, Australia) showed that microRNAs (miRNAs) are prevalent in lupin phloem sap and that they may act as long-distance signal molecules. Florian Fruiger (CNRS, Gif-sur-Yvette, France) described the use of overexpression strategies that identified miRNAs that perturb root architecture in *Medicago*. Ralf Dietzgen (University of Queensland) described his group's work showing how durable virus resistance mediated by RNA interference (RNAi) can be achieved by targeting viral suppressors of RNA silencing and by simultaneously introducing target sequences from different viruses. Jiayu Wen (Australian National University, Canberra, Australia) showed that her *in silico* approach could identify over 2,000 non-coding RNA genes in *Medicago truncatula* and *Lotus japonicum*.

Dissecting pathways and gene families

One feature that makes legumes attractive as crops is their ability to fix atmospheric nitrogen by means of symbiotic bacteria - rhizobia and relatives - that live in nodules in the legume's roots. During root-nodule formation, rhizobia secrete lipochitooligosaccharide molecules called Nod factors; these recognize plant serine/threonine receptor kinases (NFR1 and NFR5) with LysM domains, and induce a series of downstream responses, initially in root hair cells, that lead to the expression of plant nodulation genes. Simona Radutoiu (University of Aarhus, Denmark) reported domain-swapping experiments that demonstrated the involvement of the receptor's LysM domains in the perception of Nod factors. Giles Oldroyd (John Innes Centre, Norwich, UK) described elegant work by his group showing that a gain-of-function mutation in the plant chimeric calcium/calmodulin-dependent protein kinase DMI3 activates the expression of plant nodulation genes independent of Nod factors. They showed that the gain of function requires the actions of two GRAS-family transcriptional regulators, NSP1 and NSP2. By the use of NSP2-GFP fusions, NSP2 was shown to relocate from the nuclear envelope/endoplasmic reticulum to the nucleus upon Nod factor elicitation. They found that DMI3 kinase activity was activated through the binding of calmodulin and calcium, and active DMI3 in turn regulated NSP2 activity to coordinate Nod-factor-induced gene expression. This indicates that NSP2 transduces calcium signals in plants.

The role of flavonoids in nodule initiation was explored by Gary Stacey (University of Missouri, Columbia, USA) and Ulrike Mathesius (Australian National University, Canberra, Australia), who both reported RNAi constructs, in soybeans and *Medicago*, respectively, that knocked down flavonoid pathway expression and abolished nodule initiation. Flavonoid complementation of the nodulation aberration was possible with *Medicago* but not with soybean, suggesting that there might be different flavonoid uptake systems, a situation similar to that reported by one of us (C.B.) for

Arabidopsis. Evidence for a role for flavonoids in affecting auxin levels was presented by Mathesius, while Stacey concluded that abolishing induction of the *Bradyrhizobium nod* gene in the root environment caused nodulation failure rather than flavonoids having a direct role in nodulation. In addition, Stacey described clever ways to visualize extracellular ATP, by using a luciferase as a reporter, and to discern its role in root-hair infection.

The control of petal shape and floral zygomorphy (bilateral symmetry) in *Lotus* was addressed by Da Luo (Shanghai Institute for Biological Sciences, Shanghai, China). He reported work by his group showing that a member of the TCP-box gene family, *CYC2* - whose function is mediated through an interaction with specific MYB genes - has a 'dorsalizing' activity in petal development (that is changing all the petals to that of dorsal petal identity). *CYC2* may interact with the gene *Kew1*, which controls lateral petal formation, as *CYC2 x Kew1* mutants only have ventral petal identities.

Steven Clark (University of Michigan, Ann Arbor, USA) described studies into how stem-cell populations are maintained in the apical meristem, the growing point of a plant shoot. These focused on teasing apart the contributions of individual members of the CLAVATA1 (CLV1), CLAVATA3 (CLV3), POLTERGEIST and REVOLUTA gene families in *Arabidopsis*. A key result was the characterization of the ability of different proteins containing CLV3/ESR domains (CLEs) to replace the secreted peptide, CLV3, function *in vivo*. The team showed that the different CLEs each possess a signal peptide, a variable domain and a conserved carboxy-terminal motif of approximately 15 amino acids, and that CLE functions are likely to be determined by their conserved carboxy-terminal domains. The next task will be to determine how active CLE peptides are cleaved from the variable domain, and which protease carries out this task.

Strategies for invasion

Soil nematodes invade legume roots and new insights into legume-nematode interactions were presented by David Bird (North Carolina State University, Raleigh, USA). Root-knot nematodes (RKNs) were found to induce cytoskeletal responses in legume root hairs similar to those elicited by *Rhizobium* Nod factors. Plants mutated in the Nod-factor receptors, Nfr1 and Nfr5, responded only very weakly to RKNs, suggesting that RKNs produce a Nod factor-like compound(s). In another case of molecular mimicry, soybean cyst nematodes were reported by Melissa Goellner Mitchum (University of Missouri, Columbia, USA) to contain CLV3-like elements in their genomes that may play a role in infection by aiding a successful interaction that does not trigger a defence response.

Karam Singh (CSIRO, Wembley, Australia) presented work on the genetic resistance to bluegreen aphid or blue alfalfa

aphid identified in *M. truncatula* and backcrossed into susceptible cultivars. The presence of the resistance gene makes the resistant plant less attractive to aphid infestation. The resistance segregates as a single dominant gene, *AKR*. Helge Küster (Centre for Biotechnology, Bielefeld, Germany) has used 70-mer oligonucleotide microarrays of *Medicago* to examine differential gene expression during nodulation and the formation of root mycorrhiza, and has identified genes of interest including signal transduction and transcription regulators many of which were not previously known. Prem Bhalla (University of Melbourne, Parkville, Australia) used RNA from microdissected shoot apical meristems (SAMs) to probe microarrays and identify shoot apical meristem specific genes. To address possible under-representation of SAM transcripts on microarrays, SAM cDNA libraries were examined and more SAM-specific transcripts were identified. These included LRR-kinase-like kinases, four integrin-like kinases, auxin binding proteins and auxin efflux transporters.

It will be interesting to see the progress made by the next meeting in Mexico in 2008 and to find out whether legumes have indeed enjoyed the same renaissance as resource commodities are undergoing now.