

Meetings

Fagaceae trees as models to integrate ecology, evolution and genomics

IUFRO Conference, Genetics of Fagaceae and Nothofagaceae, in Bordeaux, France, October 2012

The Fagaceae family includes over 1000 tree and shrub species such as the familiar oaks (genus *Quercus*), beeches (*Fagus*) and chestnuts (*Castanea*), which are widespread in the northern hemisphere. However, the family also includes related genera such as *Castanopsis* and *Lithocarpus* that comprise hundreds of species in Asia, whereas the southern beeches belong to a closely related family (Nothofagaceae) distributed in South America and Australasia. Fagaceae species have high ecological value, as illustrated by the fact that the Chestnut Blight Disease, caused by a pathogenic fungus, is considered to have produced the greatest ecological disaster in the history of the United States (Wheeler & Sederoff, 2009). They also have major economical importance for their wood, bark (cork), fruits, and ecosystem services. Their value has driven much applied research in forestry including genetics and breeding, but it became clear during the recent IUFRO (International Union of Forest Research Organizations) Genetics of Fagaceae and Nothofagaceae Conference (<http://colloque4.inra.fr/iufro2012>), that these species are increasingly being used as models to integrate ecology and evolution.

'... the biological species concept popularized by Ernst Mayr ought to rather be named the 'puritanical' species concept.'

The conference brought together over 100 participants from Europe, the Americas and Asia and summarized research efforts on all aspects of the genetics and evolution of these species. A major issue in such a species-rich group is to clarify processes affecting species diversification, including ecological speciation. Studies on adaptation to past, current and future conditions (e.g. as a consequence of ongoing climate change and the emergence of new pests) were also numerous. These issues justify current investments made to decipher the genomes of a few representative Fagaceae species.

Species diversification and ecological speciation in Fagaceae

Guido Grimm (Swedish Museum of Natural History, Stockholm, Sweden) highlighted that the lineage made up of the ecologically constrained beeches (genus *Fagus*, 10 species) contrasts with its highly diverse sister lineage, which includes oaks (genus *Quercus*, c. 600 species). Possible causes for differences in speciation rates were further discussed by Paul Manos (Duke University, Durham, NC, USA). He showed that while acorn-like fruits have evolved independently several times, they were not directly associated to increased diversification rates. To better understand key innovations, a lot has to be learned about the Fagaceae in the tropical and subtropical areas where the highest species richness occurs (i.e. South East Asia and Mexico).

Presentations in this meeting demonstrated that substantial genome-wide colinearity exists among species and even genera. This surprisingly high level of conserved gene order in an evolutionarily deep order (Fagales) further extends at the gene sequence level to woody plants in general. For instance, the unigene sequences and gene models of *Quercus* and *Castanea* species reported at this conference consistently showed highest sequence similarity with genes from woody plants such as *Prunus persica*, *Populus trichocarpa* and *Vitis vinifera*. These observations suggest that due to long generation times, woody plants share a slow evolutionary clock (Petit & Hampe, 2006), thereby facilitating comparisons among them.

This high level of conserved colinearity also helps explain observed levels of hybridization. In oaks, high hybridization frequency and difficulties in delimiting species has resulted in them being shortlisted among the 'botanical horror stories' (Rieseberg *et al.*, 2006). Oaks infamous reputation started with Darwin's *Origin of Species*. In Chapter 2 on doubtful species he writes: "Look at the common oak, how closely it has been studied...[yet] in this country the highest botanical authorities and practical men can be quoted to show that the sessile and pedunculated oaks are either good and distinct species or mere varieties." However, contrary to what Darwin implied, Fagaceae species can generally be fairly well delimited with modern molecular approaches, as shown in several talks and posters at this Conference. Nevertheless, hybridization and introgression are clearly widespread with both Fagaceae and Nothofagaceae. As provocatively put by Chuck Cannon (Texas Tech University, Lubbock, TX, USA), the biological species concept popularized by Ernst Mayr ought to rather be named the 'puritanical' species concept. Cannon presented a model allowing fuzzy mating behaviour. He showed that tree species coexistence is greatly enhanced when hybridization is possible. Being choosy must somehow be both less critical and more problematic in these long-lived and highly fecund organisms that are locked in space.

Another route towards understanding speciation is to determine the genetic basis of interspecific incompatibility. For instance, Laure Villate and Catherine Bodénès (INRA, Bordeaux, France) showed, using a high density SNP-array and high density linkage maps, that segregation distortion can be mapped along the chromosomes and co-localized with loci that are interspecific differentiation outliers in several oak species, suggesting that some sexual barriers are evolutionarily conserved. Oliver Brendel's (INRA, Nancy, France) presentation instead focused on the mechanisms underlying species differences in water use efficiency in *Quercus robur* and *Q. petraea* with him reporting striking differences between them in the diurnal dynamics of stomata opening. This ecological difference is considered to help keep these broadly sympatric species apart. In the near future, it is envisaged that the genetic basis of the traits that have triggered ecological divergence and those involved in sexual incompatibility barriers will be elucidated, which should help understand the dynamics of ecological speciation.

Adaptation to biotic and abiotic environments

The role that diversification and local adaptation play in community assembly is a fundamental question at the interface of ecology and evolution, as explained by Jeannine Cavender-Bares (University of Minnesota, Saint Paul, MN, USA). Focusing on the adaptive radiation of the American oaks, she found both stasis and parallel adaptive evolution in the history of this group, with important consequences for biogeography and community assembly.

The rapidity of ongoing climate change and the pace with which new diseases emerge are raising questions with regard to the short-term response of long-lived forest trees. Adaptation of oaks, beeches and chestnuts to these new conditions represent an important research front that was discussed in Bordeaux. Rapid adaptation is facilitated by long distance gene flow (Kremer *et al.*, 2012), and several examples of the efficacy of pollen dispersal were reported at the conference. Victoria Sork (UCLA, Los Angeles, CA, USA) showed that in valley oak, adaptation to different environmental conditions within the species range in California must have constrained historical patterns of pollen gene flow, even at presumably neutral markers (Sork *et al.*, 2010). In other words, there would be 'isolation by adaptation' (and not merely by distance) at the range wide scale in this species. This geographic genetic structure was illustrated with an association between climate variables and genome-wide structure of transcriptome data.

Using common garden experiments has a long tradition in forest genetics. Such tests detect geographic clines that are interpreted as resulting from adaptation to some environmental factor; for example many such clines have been described for bud burst, an important adaptive trait. Jean-Paul Soularue and Antoine Kremer (INRA, Bordeaux, France) described a model of assortative mating caused by phenological differences (Soularue & Kremer, 2012). Their model, which includes long distance pollen flow, shows that assortative mating can create geographic patterns of genetic differentiation for bud burst that are independent of direct selection on the timing of bud burst.

Using a different type of modelling approach, Sylvie Oddou-Muratorio (INRA, Avignon, France) reported that rapid evolution could take place along an altitudinal gradient in the European beech. The strongest response was found at the highest altitudinal level as a consequence of the much stronger mortality taking place there, a useful reminder that adaptation comes at a cost.

Many studies on the tolerance to pathogenic fungi or to herbivorous insects have focused on the American and European chestnuts. However, a major advance was made on pedunculate oak, as François Buscot (Helmholtz Centre for Environmental Research, Halle, Germany) reported that his group had established a unique experimental platform to study plant–microbe interactions *in vitro* (Herrmann & Buscot, 2008). *Quercus robur* has an endogenous rhythmic growth, expressed under optimal culture conditions. The project investigated how the species coordinates its physiological and molecular responses during interactions with beneficial and detrimental organisms considering the rhythmic shoot and root growth fluctuations. It was fascinating to realize that a tree that can live hundreds of years and weigh over 20 tons (Fig. 1) can be cloned and investigated on a Petri dish, just like the model plant *Arabidopsis thaliana*.

Fagaceae enter the genomics era ...

In Fagaceae, although no complete assembled genome is available to date, a number of steps forward in genomic resource development have recently been achieved. These data are accessible via electronic and physical repositories and constitute a knowledge foundation to better understand the structure, function and evolution of the genome of these important species.

A first step was to establish an atlas of expressed genes with full length coding sequences from transcriptome sequences obtained from different tissues or developmental stages (Soler *et al.*, 2007, 2008; Ueno *et al.*, 2010; Barakat *et al.*, 2012; Spieß *et al.*, 2012; Torales *et al.*, 2012). The next step is to sequence the genome of Fagaceae species (haploid genome size: 700–900 Mb) for research to continue to grow and flourish. This has turned into reality with the launch in the United States of the Chinese chestnut (presented



Fig. 1 Mixed beech and sessile oak forest in the National Forest of Sillégné, Moselle, France. Photograph courtesy of Alexis Ducouso.

by John Carlson, Penn State University, PA, USA) and in France of the pedunculate oak (presented by Christophe Plomion, INRA Bordeaux, France) genome projects. By combining different sequencing technologies with deep sequencing coverage and libraries of different insert size allowing long range assembly, the genome of these two species are now being sequenced and assembled. The inherent challenges in assembling the complex genomes of highly heterozygous, undomesticated plants were raised during the Conference. Improved bioinformatics tools are urgently needed to ensure that we realize the great promise and opportunities that exist for use of genomics tools with woody plants and other out breeding species.

Together these developments set the stage for a new era in Fagaceae ecological genomics. We foresee numerous applications for forest management and prediction of response to climate change or pest and disease outbreaks that should help maintain healthy forests with tremendous economic, ecological, and social value in the future.

**Rémy J. Petit^{1,2*}, John Carlson³, Alexandru L. Curtu⁴,
Marie-Laure Loustau^{1,2}, Christophe Plomion^{1,2},
Antonio González-Rodríguez⁵, Victoria Sork⁶ and
Alexis Ducousso^{1,2}**

¹INRA, UMR 1202 BIOGECO, Cestas, France;

²Univ. Bordeaux, UMR 1202 BIOGECO, Talence, France;

³Department of Horticulture and The Huck Institutes for Life Sciences, The Pennsylvania State University, University Park, PA, USA;

⁴Department of Forest Sciences, University of Transilvania, Brasov, Romania;

⁵Centro de Investigaciones en Ecosistemas, Universidad Nacional Autónoma de México, Morelia, México;

⁶Department of Ecology and Evolutionary Biology and Institute of the Environment, University of California Los Angeles, Los Angeles, CA, USA

(*Author for correspondence: tel +33 557122837; email petit@pierroton.inra.fr)

References

- Barakat A, Staton M, Cheng C-H, Park J, Buang N, Yassin M, Ficklin S, Yeh C-C, Hebard F, Baier K *et al.* 2012. Chestnut resistance to the blight disease: insights from transcriptome analysis. *BMC Plant Biology* 12: 38.
- Herrmann S, Buscot F. 2008. Why and how using micropropagated trees rather than germinations for controlled synthesis of ectomycorrhizal associations. In: Varma A, ed. *Mycorrhiza*. Berlin, Germany: Springer-Verlag, 439–466.
- Kremer A, Ronce O, Robledo-Arnuncio JJ, Guillaume F, Bohrer G, Nathan R, Bridle JR, Gomulkiewicz R, Klein EK, Ritland K *et al.* 2012. Long-distance gene flow and adaptation of forest trees to rapid climate change. *Ecology Letters* 15: 378–392.
- Petit RJ, Hampe A. 2006. Some evolutionary consequences of being a tree. *Annual Review of Ecology, Evolution, and Systematics* 37: 187–214.
- Rieseberg LH, Wood TE, Baack EJ. 2006. The nature of plant species. *Nature* 440: 524–527.
- Soler M, Serra O, Molinas M, Garcia-Berthou E, Caritat A, Figueras M. 2008. Seasonal variation in transcript abundance in cork tissue analyzed by real time RT-PCR. *Tree Physiology* 28: 743–751.
- Soler M, Serra O, Molinas M, Huguet G, Fluch S, Figueras M. 2007. A genomic approach to suberin biosynthesis and cork differentiation. *Plant Physiology* 144: 419–431.
- Sork VL, Davis FW, Westfall R, Flint A, Ikegami M, Wang HF, Grivet D. 2010. Gene movement and genetic association with regional climate gradients in California valley oak (*Quercus lobata* Née) in the face of climate change. *Molecular Ecology* 19: 3806–3823.
- Soularie JP, Kremer A. 2012. Assortative mating and gene flow generate clinal phenological variation in trees. *BMC Evolutionary Biology* 12: 79.
- Spieß N, Oufir M, Matusíková I, Stierschneider M, Kopecky D, Homolka A, Burg K, Fluch S, Hausman J-F, Wilhelm E. 2012. Ecophysiological and transcriptomic responses of oak (*Quercus robur*) to long-term drought exposure and rewetting. *Environmental and Experimental Botany* 77: 117–126.
- Torales S, Rivarola M, Pomponio M, Fernandez P, Acuna C, Marchelli P, Gonzalez S, Azpilicueta M, Hopp H, Gallo L *et al.* 2012. Transcriptome survey of Patagonian southern beech *Nothofagus nervosa* (= *N. alpina*): assembly, annotation and molecular marker discovery. *BMC Genomics* 13: 291.
- Ueno S, Le Provost G, Léger V, Klopp C, Noirot C, Frigerio J-M, Salin F, Salse J, Abrouk M, Murat F *et al.* 2010. Bioinformatic analysis of ESTs collected by Sanger and pyrosequencing methods for a keystone forest tree species: oak. *BMC Genomics* 11: 650.
- Wheeler N, Sederoff R. 2009. Role of genomics in the potential restoration of the American chestnut. *Tree Genetics & Genomes* 5: 181–187.

Key words: biotic interactions, bud burst, hybridization, modelling, phenology, phylogeny, speciation, whole genome sequencing.