



evoltree

EUROPEAN PROJECT ON
FOREST BIODIVERSITY



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evoltree EXTERNAL NEWSLETTER

evoltree is an interdisciplinary network of scientists in Europe. This Network involves 25 research groups from 15 European countries that are working together to identify and study genes of adaptive significance in order to evaluate the contribution they make to the evolution of tree species and tree communities. Research will also be carried out on organisms that interact with trees such as insects and mycorrhizal fungi.

www.evoltree.eu



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A. INTRODUCTION

Message of the coordinator

by *Antoine Kremer*, *evoltree* coordinator



ANTOINE KREMER is Director of the Joint Research Unit BIOGECO at INRA Bordeaux and Coordinator of EVOLTREE. His main research interest is the evolution of genetic diversity „from genes to phenotypes“.

antoine.kremer@pierroton.inra.fr

Forest Biodiversity comes to the gene level... This issue of the newsletter provides several examples where diversity in natural populations of trees, fungi and insects is described and inventoried at the gene sequence level (articles by Stephen Cavers, Marc Buée, Kirsten Evertz, Carole Kerdelhué, Delphine Grivet). This was impossible a few years ago. But the examples shown in this newsletter illustrate one of the major achievement of *evoltree*, which is the production of a very large number of gene sequences in a variety of trees and associated organisms. Thanks to the availability of gene sequences it is now possible to monitor biological diversity at the most refined level where it is expressed. The next step is to disentangle within the massive data that are now accessible, the component of diversity that has either functional, ecological or also economical relevance. This is a very challenging issue, that was also among the goals of *evoltree*, but will need more intensive research using different complementary approaches, including the comparison with more traditional methods as progeny, provenance testing, or reciprocal transplantation experiments. An illustration of such an experiment installed in the ISS is given in the newsletter (article by Reiner Finkeldey). In the meantime, the existing resources that were developed may lead to



School children sampling acorns from under the oak stand at ISS Punkaharju, Finland (Photo by E. Beuker, Metla).

other practical applications as genetic delineation of provenance regions, fingerprinting of species or populations, and certification. Other applications may be found once the achievements and outputs produced within *evoltree* are discussed and shared with the stakeholders and end users.

B. PROGRESS IN INTEGRATION ACTIVITIES

School children help out at ISS Punkaharju

by *Egbert Beuker*



EGBERT BEUKER is forest scientist at the Finnish Forest Research Institute, Punkaharju Research Unit. He is interested in genetic adaptation in forest trees and tree breeding (e.g. aspen).

egbert.beuker@metla.fi

Every year during spring or early summer several primary school classes from around Punkaharju visit the forest areas of the Punkaharju Research Unit of the Finnish Forest Research Institute Metla. These forest areas are part of the *evoltree* Intensive Study Site (ISS) located in Punkaharju. The main attractions in the area are the arboretum, the tallest tree of Finland - a European larch *Larix decidua* of about 47 m of height, and the nature conservation area. The children's visit is often combined with an attendance to the Finnish Forest Museum, Lusto, just next door the Punkaharju Research Unit. The guides of the Research Unit organize activities for the children



Position of Punkaharju Research Unit, Finland. Source: wikimedia.org

such as tree planting in the arboretum. Each child gets its own tree, which will be geo-referenced for later visits and recoveries.

For some reason, last year, class 4b of the Savonlinna University Practice School and their teacher, Teija Piisilä, had to postpone their visit until autumn. Of course there is a lot to explore in the forest also in autumn but, unfortunately, there was no possibility to arrange any tree planting that late in the year.

However, at that time, the process of collecting acorns from a small oak (*Quercus robur*) stand was taking place. The collection was part of ISS transfer experiment which was about to start, led by Reiner Finkeldey within one of evoltree's Jointly Executed Research Activities (JERA 4). So the children were asked to help in the collection. First, they were given a short and very simple explanation about evoltree and the reason for the sampling. Helping with real research work as well as knowing that the acorns were going to be sent to Germany, to germinate, grow into small trees and be distributed all over Europe, made this quite exciting for the children. Under the supervision of our technician, Tarja Salminen, hundreds of acorns were collected in a short time. After the external quality of our acorns was checked by our staff, they were delivered to Germany for germination where the small oaks are now struggling to survive until they will be planted next spring.

Of course the children had the possibility to keep some acorns for themselves, so that they may even be growing their own trees and one day they may wish to start their own research.

Updates on databases: evoltree data curation and integration activities

by Arouna Woukeu



AROUNA WOUKEU is researcher in the Multimedia Group at the University of Southampton. His research interest lies in integration of bioinformatic resources, Technology Enhanced Learning and Digital Libraries.
aw1@ecs.soton.ac.uk

Within evoltree (IA1.1), a number of database applications are being developed to curate and share various resources between evoltree partners and the scientific community. Among these databases are genetic and QTL mapping databases (CMap and SalicaceaeMap), and the SSR and SNP databases.

SSR and SNP databases in Southampton (P22)

The evoltree SSR and SNP databases have been operational for some time and based at the University of Southampton School of Biological Sciences (see this address: www.evoltree.soton.ac.uk).

The SSR database is currently populated with data from several species including *Populus trichocarpa*, *Populus deltoides*, *Quercus robur*, *Quercus petraea*, *Quercus rubra*, *Quercus myrsinifolia*, *Castanea sativa*, *Castanea crenata*, *Castanopsis cuspidata*, *Fagus sylvatica*, *Fraxinus excelsior*, etc.. Users can log onto the web site and upload data, search and browse the content of the database, and view statistics. A way to automatically link these databases with genetic maps databases is being devised.

Data Upload

There are two simple steps involved in uploading SSR or SNP data into the database.

First, users need to download the appropriate data submission template from the web site, and format their data set accordingly.

Then they need to save their data set spreadsheet into csv format and follow the online instructions to upload the data onto the database. Users can browse/search the database to ensure that their data have been uploaded. To simplify and avoid duplication in the login process, evoltree users can now automatically get redirected to the SSR/SNP databases from the main evoltree portal without the need for additional login.

CMap and SalicaceaeMap databases (P22)

The evoltree comparative mapping database (CMap) for Salicaceae and Pinaceae, and the genotypic and phenotypic database (SalicaceaeMap) are also accessible online (see this address: www.evoltree.soton.ac.uk).

The CMap application allows users to update map data and view comparisons of genetic and physical maps. Instead, SalicaceaeMap (previously called PopulusMap), currently being tested, is aimed at storing and managing raw molecular (genotypes) and phenotypic (phenotypes) data which can be used for QTL detection and genetic map construction. SalicaceaeMap also stores information about corresponding pedigrees, and is linked to CMap. Both these databases are accessible from the main evoltree portal (www.evoltree.eu).

C. RESEARCH HIGHLIGHTS

Population structure and adaptive variation of Scots pine in Scotland (IA4)

by W. Wachowiak & Stephen Cavers



STEPHEN CAVERS is a molecular ecologist at Centre for Ecology and Hydrology (UK). He is interested in genetic diversity, gene flow and adaptation.

scav@ceh.ac.uk



WITOLD WACHOWIAK is molecular geneticist at the Institute of Dendrology, PAN (Poland). His main research interest is in evolutionary processes in plants including genetic diversity and adaptive variation in forest trees.

witoldw@rose.man.poznan.pl

Scots pine (*Pinus sylvestris* L.) is the most widely distributed conifer species in the world. In Scotland, where it is the national tree, the species reaches the most north-westerly part of its distribution and is a keystone component of the remnant Caledonian forests. Less than one hundred fragments of the ancient native pine woods remain in Scotland, scattered across diverse sites which are highly differentiated in annual rainfall and temperature, growing season length and soil type. To conserve what remains, forecast the likely responses of remnant populations to environmental change and develop science-based management strategies, it is vital to assess genetic structure of these populations and gain a better understanding of the process of local adaptation. The Scottish pine woods have unique biological characteristics, but also cultural, recreational and potentially economic value. There is a clear justification for a more detailed examination of the Scottish range of the species to create more effective strategies for the management of the remaining natural resources. To achieve this will require contributions from different scientific fields, bringing together ecology, forestry and molecular biology to search for the signatures of evolution in natural populations.

A progeny trial of Scottish *P. sylvestris*, including samples from 21 populations across an environmental gradient and including all of the designated UK seed zones for the species, has been initiated at the Centre for Ecology and Hydrology, Edinburgh, UK. The study aims to establish patterns of variation in quantitative traits and mating system for the purposes of genetic resource conservation

in fragmented Scots Pine populations. That initiative is supported by the **evoltree** integration activity package that supports mobility (IA4). The outputs of the trial were extended by applying molecular population genetics and candidate gene-based approaches to address questions about the environmental and evolutionary factors influencing patterns of DNA sequence variation (SNPs) in local populations. Nucleotide diversity patterns were studied across many genes including, for example, some potentially involved in control of dehydrative stress and wood formation. DNA sequence information was compared between populations which had previously shown differentiation at quantitative traits. Preliminary results indicate the influence of demography and selection on nucleotide diversity in Scottish populations and their unique features relative to mainland European populations.

The support of the **evoltree** mobility grant also provided an excellent opportunity for interaction and integration of the work activities between the partners involved and has promoted technology transfer, especially in relation to SNPs methodology, and design, maintenance and measurement of physiological and growth parameters in experimental trials. This successful application of genomic approaches provides immediate guidance



Natural Scots pine stand in Ben Eighe, Scotland (Photo by Stephen Cavers).

on new directions for future research and should help to develop a sound science base for conservation of *P. sylvestris* in the UK.

Late Holocene spread of Norway spruce and beech in southern Scandinavia – combining modelling, ancient DNA and paleoecology

by Richard Bradshaw



RICHARD BRADSHAW, University of Liverpool, is interested in the long-term history of trees in Europe. He studies macrofossils and ancient DNA.

richard.bradshaw@liverpool.ac.uk

Understanding how tree species have spread during the Holocene has long been a source of fascination for many researchers in the fields of botany, paleoecology and more recently genetics. Spreading patterns and especially spreading rates of tree species have become a heavily debated topic because earlier estimates, based on palaeoecological data alone, were artificially rapid as small outlying populations were often undetected in the paleoecological record. More realistic estimates can be obtained by combining paleoecological and genetic research methods and data in the way that we are working within **evoltree**. These estimates of spreading rate are important during a period of forecasted rapid climate change as they can give us a clue as to how threatened the different species are and how the effect of climate change can best be mitigated.

In a recent project we combined the different scientific backgrounds from **evoltree** partners to work on the long-term history of beech (*Fagus sylvatica*) and Norway spruce (*Picea abies*), two of the most widespread and commercially important species in central and northern Europe respectively through the study of sub-fossil pollen, macro fossils and ancient DNA. In the latest stage of this project this information was taken as a basis for modelling the changing distributions of beech and Norway spruce in southern Scandinavia during recent millennia and for gaining insights into the controls of the spreading process using data-model comparison. We found that the spread of Norway spruce during the last 4,000 years gave a good match between the observed data and a diffusive spread at a rate of 250 metres per year, with a moderate long-distance dispersal component. It showed that Norway spruce had some

difficulty in penetrating a compact moving front of beech. The best estimate for the spreading rate of beech in turn was 100 metres per year, although the model gave a less good fit to the data than for Norway spruce. This suggests that factors other than dispersal were also limiting the spread of beech, which is consistent with the reported need for local disturbance prior to stand-scale expansion of beech populations. The areas of Europe with suitable climates for Norway spruce and beech are likely to alter, creating the risk that the two species might not spread with sufficient speed to track the suitable habitats. The better understanding of the two species migration patterns can now be incorporated in the different climate change scenarios to forecast these potential risks more accurately.

High-throughput molecular tools highlight fungal community structure in an extremely complex subterranean world

by Marc Buée, Marlis Reich and Francis Martin



MARC BUEE is member of the „Tree-Microbe Interactions“ Unit, INRA Nancy. His main research activities are environmental factor influence on fungal community structure and functional diversity study of ectomycorrhizal fungi. buee@nancy.inra.fr



MARLIS REICH is supported by a Marie Curie PhD scholarship, within the framework of the TraceAM programme. She is interested in Mycorrhizal fungi focusing on *Cenococum* sp, *Glomus* sp, and *Laccaria* sp. reich@nancy.inra.fr



FRANCIS MARTIN is leader of UMR INRA, Nancy. Involved in the 'Tree-Microbe Interactions' Unit, which focuses on functional genomics of the ectomycorrhizal interaction.

fmartin@nancy.inra.fr

During the thirties, Lourens G. M. Bass-Becking formulated two general laws concerning microorganisms diversity, summed up in the phrase “Everything is everywhere ... the environment selects”. It is certain that fungi are ubiquitous and these organisms play key roles, in particular in decomposition and recycling of nutrients or plant nutrition in territorial ecosystems. Fungi are the largest component of biodiversity on earth



Picea abies (Photo by Sascha Rösner | fotouristen.de).

after insects, with a number of species estimated to 1,5 million, of which only 5-10% have been described so far. In temperate and boreal forests also, this diversity is very important, particularly in contrast to the small number of tree species. During the last fifteen years, PCR-based molecular methods and DNA sequencing have been routinely used to identify fungi and these molecular ecology studies have provided an interesting insight into the complexity of these species, particularly for ectomycorrhizal fungi, and in the processes that structure these fungal communities. Such approaches have been used to determine how the spatial and temporal variations of fungal populations and communities are affected by biotic and abiotic factors, including host tree species influence. With improvements in molecular genotyping techniques, appropriate DNA databases and powerful in silico capacities of data analysis, innovating high-throughput molecular diagnostic tools, such as phylochips or massively parallel sequencing (pyrosequencing), can be used to investigate this huge biodiversity and cataloguing soil fungi on the larger scale imposed by field studies of a very heterogeneous subterranean world.

Methods

For these reasons, we have developed and validated the first large-scale ribosomal ITS phylochip to study the impact of host trees and seasonal changes on

fungal communities in six different forest treatments (conifer and hardwood plantations). For the design and development of the NimbleGen phylochip, 16,987 fungal ITS-sequences were used to create 84,891 species-specific oligonucleotides for 10,318 fungal species. Oligonucleotides were spotted in four replicates on the phylochip. Results of phylochip analysis were validated with results of cloning/Sanger-sequencing and were compared to first 454 pyrosequencing results.

The fungal community structure was evaluated using fungal ITS region fragments obtained from eight soil samples in each of six host-tree plantations. Fungal ITS amplicons were labelled and used for Nimblegen oligo-phylochip hybridization. Moreover, fungal ITS-1 amplicons were amplified for 454 pyrosequencing, generating between 100 to 250 bp sequences. A total of 185,806 PCR amplicons was obtained after sequencing and 180,213 were retained after trimming. The operational taxonomic units (OTUs) were defined at 97% sequence similarity, a commonly recognized level for comparative analysis. Rarefaction was used to compare fungal richness of the different soil samples. Rarefaction curves obtained with 25,700 (Douglas fir) to 35,600 (spruce) sequences showed that sequencing effort should be increased to reach the asymptotes of curves. A maximum of 1,000 OTUs was estimated in the oak soil samples. Multiple data sets were compared with MEGAN software to provide taxonomic affiliation of DNA reads.



Lymantria monacha moth (Photo by Andrzej Oleksa).

Multivariate statistical analyses were performed on the relative proportions of the whole fungal species obtained with the two technologies.

Results

The comparison of the six treatments showed a strong host effect on the community structure of terrestrial forest fungi, independently of approaches. These high-throughput methods could be applied to study the impact of tree species as drivers of terrestrial fungal diversity in the context of global warming and plant spatial dynamic.

Genetic diversity in *Lymantria* spp. populations in relation to their host trees

by Kirsten Evertz



KIRSTEN EVERTZ is a PhD student from the Max Planck Institute for chemical ecology. Her interests are in ecology and animal-plant-interaction. In particular she studies the genetic diversity of *Lymantria* spp. in relation to their host trees.
kevertz@ice.mpg.de

Lymantria dispar (gypsy moth) and *Lymantria monacha* (nun moth) (Lepidoptera: Lymantriidae) are important forest insect pest species in North America and Europe. Both *Lymantria* species are polyphagous and have to cope with a wide spectrum of different chemical defenses by different host trees. There is anecdotal evidence for genotypes that are specialized for deciduous leaves vs. conifer needles but firm evidence for differentiation is lacking. The focus lies in digestive physiology of *Lymantria dispar* and *Lymantria monacha* feeding on different food sources and on which genes are putatively involved in



Larvae of *Lymantria dispar* moth (Photo by Kirsten Evertz).

adaptation to different host plants in *Lymantria dispar* (more the generalist) and *Lymantria monacha* (more the specialist)? To identify candidate genes involved in detoxification, we have conducted feeding assays where we compare groups of larvae that have been fed on different food sources (oak, spruce, artificial diet). RNA was extracted from larval midguts to determine differences in gene expression, using cDNA-AFLP method. After cutting differently expressed fragments, those were sequenced and then compared with cDNA libraries from both species. Genes related to digestive activities were found. Within the framework of [evoltree](#), we will focus on the digestive physiology of populations collected from deciduous trees vs. populations collected from conifers in several European field sites (the Intensive Study Sites established across Europe).

Genetic characterization of a phenologically aberrant population of the pine processionary moth

by Carole Kerdelhué



CAROLE KERDELHUÉ is member of BIOGECO at INRA Bordeaux and works in forest insect genetics. Her main interests are phylogeography and neutral genetic diversity. She started to develop programs on the genetic bases of adaptive traits.
carole@pierreton.inra.fr

The winter pine processionary moth (PPM) *Thaumetopoea pityocampa* is a phytophagous Lepidoptera associated with pines all around the Mediterranean Basin. Its larvae feed on pine needles and show a gregarious behavior, causing heavy damages due to defoliation. They develop mostly during winter, and build typical silk nests. Moreover, the larvae are highly urticating and can provoke strong allergic reactions.

All over its vast distribution range, the PPM shows some variability in phenology as an adaptation to the local climate and environment. In regions of low altitudes and latitudes, where summers are hot and dry, reproduction takes place in late summer or in fall, probably because both eggs and young larvae are susceptible to heat. In northern latitudes and higher altitudes, reproduction can occur as early as June. In all cases, once larval development is completed, the caterpillars leave their host tree to pupate in the soil between January and April, depending on climatic conditions, forming typical processions. An obligate diapause then takes place until adult emergence.



Typical silk nests of the pine processionary moth *Thaumetopoea pityocampa* on a pine tree (Photo by Helena Santos).

In 1997, an 'abnormal' population of the pine processionary moth was discovered in the Leiria National Forest in Portugal. It showed a shifted life cycle, with sexual reproduction in spring rather than late summer, larval development in summer rather than winter and processions in October rather than February (1). Interestingly, PPM individuals having the normal phenology were still found in the same forest. Monitoring of adult activity through trapping showed that periods of sexual reproduction did not overlap between the normal 'winter' population and the shifted 'summer' population, which suggested that gene flow was probably reduced or absent between the two sympatric groups. We started a fruitful collaboration with Manuela Branco (Instituto Superior de Agronomia, Universidade Técnica de Lisboa) and Helena Santos as a PhD student. Preliminary genetic analyses, based on neutral sequences, undoubtedly showed that individuals from the shifted 'summer' population belonged to the same species as the normal winter individuals. This confirmed that we did not deal with a rare, cryptic species of the same genus. Moreover, microsatellite analyses based on five loci strongly suggested that the 'summer' population was founded recently from a small number of individuals originating from the local 'winter' population (2).

We will take advantage of this unique situation to study the genetic bases of phenology in the pine processionary moth. Candidate genes involved either in biological clocks or in diapause termination have been selected and will be searched for in cDNA libraries that were developed during the course of **evoltree** for *Thaumetopoea pityocampa* on both the 'summer' and the 'winter' populations. Specific primers will be developed and the selected genes will be sequenced in natural populations to study their nucleotide diversity and to determine if this variability can be associated to differences in the insect phenology. New microsatellite loci, under construction within **evoltree**, will also be used to better characterize the genetic structure of the PPM and propose a more accurate scenario explaining the origin of the 'summer' population.

(1) Pimentel C, Calvao T, Santos M, et al. 2006. Establishment and expansion of a *Thaumetopoea pityocampa* (Den. & Schiff.) (Lep. Notodontidae) population with a shifted life cycle in a production pine forest, Central-Coastal Portugal. *Forest Ecology and Management*, 233: 108-115.

(2) Santos H, Rousselet J, Magnoux E, et al. 2007. Genetic isolation through time: allochronic differentiation of a phenologically atypical population of the pine processionary moth. *Proceedings of the Royal Society of London Series B*, 274: 935-941.



An adult pine processionary moth *Thaumetopoea pityocampa* (Photo by Helena Santos).

Update on population genomics research of Mediterranean conifers (JERA2, IA1.2)

by Santiago C. González-Martínez and Delphine Grivet



DELPHINE GRIVET is a member of the INIA-CIFOR in Madrid. Her main research interest is population genetics and genomics, phylogeography, gene flow and conservation genetics of temperate forest trees.
dgrivet@inia.es



SANTIAGO C. GONZALEZ-MARTINEZ is a member of the INIA-CIFOR in Madrid. He is interested in population genetics and evolutionary processes in plants, with a focus on Mediterranean trees and Iberian endemics.
santiago@inia.es

The Mediterranean basin is considered a global hotspot of plant diversity. Mediterranean forests are characterized by their spatial and temporal complexity and heterogeneity in terms of both their biotic (fauna and flora) and abiotic (geography, geology, geomorphology, pedology, bioclimate) components. Within this range, pines exhibit high genetic variability and/or phenotypic plasticity that explain their high colonizing ability and the central role they play in vegetation dynamics in the Mediterranean region. The fact that pines are able to live in such contrasted environments leads to speculation about the mechanisms involved in their adaptation, whether they are the same across all the species, and about the consequences of current and future climate changes in the Mediterranean zone on the adaptive potential of pines. In that perspective, several initiatives are underway to understand the molecular evolution underlying adaptation of Mediterranean pines within the framework of the **evoltree** project.

At the single-species level

One main objective is to sequence some target candidate genes involved in drought tolerance across the natural distribution range of two model Mediterranean pines: *Pinus pinaster* Ait. (in collaboration also with P1a, INRA) and *Pinus halepensis* Mill. To achieve this goal, molecular information (available from the New World pine species *Pinus taeda* L. and also newly developed within **evoltree**) was successfully transferred to these two pine species. Several candidate genes, including some from the well-known dhn (dehydrin) and asr (ABA/water

stress/ripening-induced) families, were analyzed for each species, and the information of these genes of adaptive relevance was combined with that of neutral markers (microsatellites) to look at the species demographic and evolutionary histories. In particular, in *P. halepensis* we have found a notable loss of diversity and fixation of derived polymorphisms during colonization of its western range, whereas in *P. pinaster* we found high levels of nucleotide variation in all its range.

In addition, genetic resources are being currently produced within **evoltree** in order to extend the genetic knowledge to two other target Mediterranean pines, namely *P. pinea* L. (notable for its near absence of organelle variation at the wide range scale) and *P. canariensis* Chr. Sm. Ex DC (a current Canary Islands endemic, formerly with a wide distribution in the European continent). In parallel, another initiative focuses on increasing the genetic resources available in *P. pinaster* and *P. halepensis* by transferring a set of 12 full-length genes related to water stress and initially developed in *Pinus pinea*.

At the multi-species level

The CRIEC (Candidate gene Resequencing In European conifers) sequencing initiative combines resources from **evoltree** with those from CNR, INIA, USDA, UC Davis and other American and European institutions, with the following main objectives: a) to test for transferability of ~300 expressed candidate genes (initially developed analyzing *Pinus taeda* ESTs) to a wide range of conifer species within the genus *Pinus* (*P. leucodermis*, *P. brutia*, *P. cembra*, *P. halepensis*, *P. nigra*, *P. pinea*, *P. canariensis*, *P. uncinata* and American southern yellow pines) and outside this genus (*Abies* sp., *Juniperus* sp., *Cupressus* sp., *Austrocedrus* sp., *Larix* sp., *Cedrus* sp., *Taxus* sp.) the genus *Pinus*, with a focus on European species; b) to discover SNPs and to provide first estimates of nucleotide diversity for a wide range of conifer species using ortholog expression candidate genes. Levels of transferability have been very promising so far (over 60 genes sequenced), as high as ca. 75% in some pines and from 4% to 43% for the other conifers.

From the various genetic resources produced within these different initiatives, the identification of candidate genes controlling traits of interest, as well as the evolution of these genes across conifer species, constitutes one of the ultimate goals of the work. To reach this goal, one strategy consists in searching for association of the genomic polymorphisms detected in contrasting natural populations with some adaptive traits (e.g. growth, phenology, response to drought or cold),

as measured in common garden experiments (to avoid any environmental effects). Then, the polymorphisms that are positively associated to adaptive traits can be screened in wide-range samples of natural populations. Another way to look for adaptive variation is to explore the correlation between the discovered polymorphisms and some environmental variables (e.g. precipitation, temperatures) at the species distribution scale. Altogether, the polymorphisms revealed by population genomic and association studies will allow the identification of the genes underlying ecologically important traits and the description of the consequences of naturally occurring variation at these loci on fitness.

Will tree seedlings adapt to changing climates? A new **evoltree** project aims to find answers

by *Reiner Finkeldey*



REINER FINKELDEY, University of Göttingen, is interested in population genetics of forest tree species. Further research focus lies within ecological consequences of anthropogenic influence and global change.
rfinkel@gwdg.de

Trees produce in irregular frequencies huge numbers of seeds, but only a few will survive until maturity. This offers unique opportunities for selection processes to increase the adaptation of tree populations, in particular during early stages of stand development. Within the framework of **evoltree** an experiment was set up to investigate the growth and survival of pine (*Pinus sylvestris*) and oak (*Quercus* spp.) seedlings, and to compare genetic structures of the adapted (surviving) with the non-adapted (dead) plants. The main long-term objective is to identify genetic variation which is responsible for the different survival abilities of tree seedlings growing in diverse climates.

Pine and oak seeds were harvested in, or close to, five of **evoltree**'s Intensively Studied Sites (ISS): Blizyn (Poland), Punkaharju (Finland), Solling (Germany), Valais (Switzerland), and Ventoux (Southern France). The seedlings are currently been raised in greenhouses at Göttingen University (Germany) and will be distributed to the same sites to be planted following a common design. A large number of seedlings representing each provenance (400 plants) is going to be planted at each trial site. At least three provenances will be represented for each species at each of the five sites. The experimental approach differs from the establishment of provenance trials since the focus is on the observation of

the seedling stage, which is particularly critical due to the strong selection pressure. Thus, different survival rates of seedlings from the various 'provenances' will be primarily investigated to gain additional insight on selection processes by comparing different provenances.

Before the seedlings are distributed to the different trial sites, a small amount of tissue is taken from each plant, which will be prepared for long-term storage in the repository center of **evoltree**, located in Seibersdorf, Austria. This will allow assessing, for the first time, the genetic structure, not only of the surviving individuals, but also of non-adapted, dead seedlings.

The trials will be monitored for at least two growing



A trial for **evoltree**'s seedling selection experiment. Top: preparing **evoltree**'s seedling selection experiment: oaks from Switzerland growing in the greenhouse in Göttingen (Photos by Reiner Finkeldey). Center: different growth and germination of oak seeds in **evoltree**'s seedling selection experiment. Provenances from left to right: Switzerland, Poland, France, Germany, Finland (Photo by Reiner Finkeldey). Bottom: preparation in a beech forest in Solling, Germany. Seedlings will be protected from damage by mice and other animals in steel cages.

seasons, in 2009 and 2010 and, depending on survival rates in different locations, even longer. The first results of the comparison in the genetic makeup of adapted and dead plants will be available during 2010.

COMMUNI-TREE - Tree community genetics: what is the first year of research telling us?

by Katalin Tuba and Ferenc Lakatos



KATALIN TUBA, a PhD student at the University of West-Hungary, is interested in interactions between insects and their host species. She focuses on insect associates of natural *Populus nigra* and its commercial clones.

tuba@emk.nyme.hu



FERENC LAKATOS, a forest entomologist at the University of West-Hungary, is interested in ecology and diversity of forest insects. His main research interests are in population genetics of insects associated with woody plants.

flakatos@emk.nyme.hu

COMMUNI-TREE, a subactivity of [evoltree](#), tries to determine and understand the influence of genetic diversity of trees on the diversity of associated organisms. Twelve partners throughout Europe are engaged in several projects on this topic. The main issues addressed in COMMUNI-TREE are the following:

(I) the relationship between tree genetic diversity and the diversity of associated organisms (i.e., insects



All stages of the a poplar feeding insect (eggs, larvae and adult beetle) can emerge at the same time (Photo by Ferenc Lakatos).

and mycorrhizas), (II) the identification of the genetic characteristics of trees influencing the diversity of associated organisms.

Katalin Tuba and Ferenc Lakatos, members of the University of West Hungary (UHW, Sopron, Hungary) are working on these aspects, focusing on the model tree species *Populus nigra* 'Italica' (Lombardy Black Poplar) and its associated insects.

The newsletter team (NT) approached Katalin for an interview about her research work.

NT: in establishing a pan-European project such as COMMUNI-TREE, you must have encountered problems in separating the influence of the environment from the role of the host tree genotype in determining the diversity of associated organisms. How did you achieve the necessary level of standardization across site in the 'Lombardy Black Poplar' project?

Katalin: as a very first step, we tried to see what species of insects feeding on trees were found by partners in all the project sites. The locations selected were in France, Germany, Hungary, Italy and Poland. We identified six main functional groups of insects, occurring with different frequency in all study sites: chewers, skeletonisers, leaf miners, sucking and sap feeding insects, gall makers and leaf rollers.

The next step was to compare different survey methods and to examine where we needed to select sample trees to avoid special local effects.

NT: How did you move forward?

Katalin: first we tested different survey methods. We marked some leaves, we observed their conditions in April and collected them later in September. At the same time, on adjacent *P. nigra* 'Italica' trees, we monitored unmarked leaves, selected by chance during the whole survey season. For what concerns the selection of sample trees, we chose pairs of *P. nigra* 'Italica' trees at seven sites each in West-Hungary. We also picked different locations among the study sites (e.g., urban environments, villages and fields, surrounded only by agricultural lands).

NT: What results did you obtain from this testing?

Katalin: the method based on unmarked leaves turned out to be more efficient in terms of efforts to be put into field work and information obtained. In addition, we observed it was difficult to get a really random selection of the leaves to be marked. Moreover, chewing, sucking and leaf mining insects tend to prefer young leaves.

Thus, these functional groups were potentially neglected when using the method based on marking leaves, as these are harvested late in the season. Finally, we found we would risk to damage pre-selected leaves and twigs when marking them, and sometimes they would even fall prematurely.

So, we concluded that the method based on marked leaves would be just suitable to assess insect damage by one generation of a particular insect species, or for a short-term parallel monitoring of damage caused by several species.

NT: What about the categories of sample trees that you determined? Did you find that the type of environment has a lot of influence on the types of associated insects?

Katalin: indeed, species composition varied substantially between the types of environment (i.e., urban, village and the field). Furthermore, the proportion of insects within respective functional groups varied. For example, monophagous leaf miners were found to be more abundant within the field, while polyphagous chewers preferred those trees located in villages.

NT: How did you make these results available, in order to contribute to the pan-European COMMUNI-TREE project?

Katalin: we developed some guidelines, based on our results, which contained several images showing types of damage procured and the insect species responsible, divided by functional groups. This was a very useful contribution, appreciated by the partners involved in the COMMUNI-TREE project.

NT: are there other PhD students, at the University of West Hungary, besides you, who are engaged in the COMMUNI-TREE project? Are you planning a follow-up to the ongoing initiative?

Katalin: we have three master students contributing to the COMMUNI-TREE-related activities. We are supervised by Prof. Ferenc Lakatos, at the Institute of Silviculture and Forest Protection, University of West Hungary. The common garden experiments that were established within the framework of COMMUNI-TREE have already yielded very promising results, therefore further investigations are planned for the future.

NT: Katalin, thank you very much for the interesting interview, and good luck in your forthcoming research work!



Carpets of wood anemone *Anemone nemorosa* are flowering in a beech *Fagus sylvatica* stand in early april, Germany (Photo by S. Rösner | fotouristen.de).

Mycorrhizas on silver fir – effects of long term adaptation and genetic differentiation of the hosting trees

by Kathrin Donges



KATHRIN DONGES is a member of the mycology group at the University of Marburg. Her research activities are ectomycorrhizal community genetics, ectomycorrhizal species diversity and population genetics. donges@staff.uni-marburg.de

A major focus of **evoltree** is on community genetics, i.e. the study of the influence that the genetic diversity of trees exerts on the diversity of associated organisms within the ecosystem. We assume that the communities of associated organisms vary among tree genotypes because these carry different information at relevant genes.

Common garden experiments have generated knowledge on these aspects within different ecosystems (1), but little is known about biogeographical effects, such as, for

example, the influence that long-term adaptation to the environment has on the organisms interacting with, and depending on trees.

An experiment was set up to examine these issues in the Black Forest (Germany), with funding from DFG (German Science Foundation) and **evoltree**. The research is focused on different silver fir (*Abies alba* Mill.) provenances and the objective is to look at the amounts and types of ectomycorrhizae (ECM) which colonize their roots, during one vegetative season. One fir provenance ('Nagold') is locally adapted, with origin from the Black Forest, whereas the other two ('Romania' and 'Macedonia') belong to the south eastern silver fir lineage, which evolved independently since the last ice age (2). We hypothesized that the 'foreign' provenances would host less species of the native mycorrhizae, presenting a limited overall degree of mycorrhization on their roots. Additional expectations were the following: (i) mycorrhization induces a better growth performance of host trees, and (ii) the silver fir provenances selected for the study show genetic differentiation due to their long-term isolation, with an influence on the spectrum of associated ECM.

Seedlings and saplings were established following a nested random-plot design on two adjacent area cleared of forest, in spring 2007. The young silver firs were harvested after four months and examined to determine the rate of mycorrhization and the number of ECM species found on their roots. In addition, individual growth performances and the genetic differentiation of the various tree provenances, were analyzed. A multivariate principal component analysis (PCA) was carried out to identify the most important variables determining the patterns observed.

The diversity of ECM species found on the different provenances was in general quite high, considering the short inoculation period and the age of the host trees. Mycorrhization rate was higher for the local provenance. Moreover, the total mycorrhization had a significant positive effect on host tree performance, as measured through parameters such as root and shoot weight (ANOVA), with local provenance presenting a superior performance.

The number of surviving individual of silver firs varied on the different plots, thus rarefaction was calculated (3) in order to assess the real ECM species diversity per provenance, and to determine potential effects of location and development stage (i. e. ontogenesis) on the degree of mycorrhization.

Mean species diversity was highest for the local fir provenance and lowest for the Romanian provenance (Figure 1). An unpaired t-test revealed slightly significant differences for just one factor (ontogenesis).

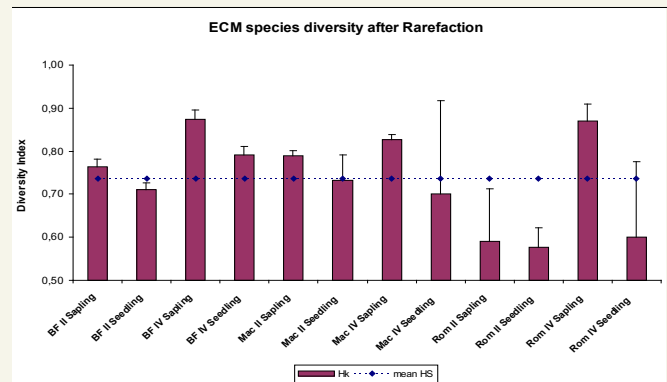


Figure 1: ECM species diversity after rarefaction of categories found in two plots (II and IV), with three provenances of silver fir (BF – Black Forest, Mac – Macedonia, Rom – Romania), at different stages of development (sapling – pre-germinated silver firs, seedlings – sown silver firs). The blue dotted line indicates the mean diversity index of ECM species.

The analysis of the genetic differentiation between silver fir populations was performed using five SSR markers (4,5). It revealed a strong separation of the local Black Forest provenance from the two south eastern provenances. This can be explained by the different post glacial history of the provenances.

The results of the PCA revealed a correlation between the genetic differentiation of tree populations and the associated ECM diversity.

These findings are interesting but further investigations would be needed to support these results, especially by removing the effect of factors such as location and development stage which are difficult to control.

(1) Whitham TG, Bailey JK, Schweitzer JA, et al. (2006) A framework for community and ecosystem genetics: from genes to ecosystems. *Nature* 7: 510-523.

(2) Liepelt S, Cheddadi R, de Beaulieu J-L, et al. (2008) Postglacial range expansion and its genetic imprints in *Abies alba* (Mill.) – a synthesis from paleobotanic and genetic data. *Rev Palaeobot Palyno*. doi:10.1016/j.revpalbo.2008.07.007

(3) Petit RJ, El Mousadik A, Pons O (1998) Identifying populations for conservation on the basis of genetic markers. *Conservation Biology* 12: 844-855.

(4) Cremer E, Liepelt S, Sebastiani F, et al. (2006) Identification and characterization of nuclear microsatellite loci in *Abies alba* Mill. *Mol Ecol Notes* 6: 374-376

(5) Hansen OK, Vendramin GG, Sebastiani F, et al. (2005) Development of microsatellite markers in *Abies nordmanniana* (Stev.) Spach and cross-species amplification in the *Abies* genus. *Mol Ecol Notes* 5: 784-787.

The genome of a smelly fungi: Understanding the ecology of black truffle (*Tuber melanosporum*)

In its February 2009 issue n. 323, the journal Science published an article about recent findings from research activities on truffle ecology and genomics. The author of the article, John Bohannon, summarizes information from different sources and interviews leading scientists in this area of research, including Francis Martin, [evoltree](#) member and leader of the black truffle genome project at INRA (French National Institute for Agricultural research) in Nancy. Besides scientific findings, Bohannon is undoubtedly intrigued by the culinary and cultural role of truffles in Europe.

The fruiting bodies of white (*Tuber magnatum*) and black truffles (*Tuber melanosporum*) are among the most expensive foods in the world. Thanks to their unmistakable taste and aroma, truffles are highly appreciated by gourmets, who are ready to pay up to thousands of Euro for a kilogram of the delicacy. Beyond the secrets of their appreciated taste and aroma, scientists are interested in the ecology and genome of these fungi. The full genome sequence of the black truffle was completed in 2008, while for the white truffle the task is still ongoing and should be completed soon.

The investigations recently concluded have revealed interesting characteristics of these valuable commodity. It was previously thought that truffles were almost clonal and therefore genetically very similar across their distribution range, with differences in their aromatic properties mainly determined by environmental conditions (e.g. soils, local climate). However, the recent findings show that the black truffle genome is highly variable. Researchers from the Institute of Plant Genetics in Perugia, Italy (Francesco Paolocci, Andrea Rubini, and Sergio Arcioni) and from INRA-Nancy (Claude Murat and Francis Martin) genotyped more than 200 black truffles from 13 regions across southern Europe and found that they form locally distinct varieties. These findings have immediate very useful applications as molecular markers could be used to detect the origin of truffles and help to contain 'counterfeiting'.

This is indeed an issue. Sometimes, species which look similar to the high value truffles, but are less aromatic and more abundant, such as the Chinese species *Tuber indicum*, are sold for a very high price, because they are mistaken for black truffles from other more famous regions. Molecular biologists have a chance to become strong allies of the truffle industry.

The researchers Martin and Murat, at INRA-Nancy, modelled the evolutionary history and phylogeography of truffle populations. They showed that *Tuber melanosporum* existed in southern Italy and Spain 12,000 years ago, and later spread northwards over the Alps and Pyrenees, colonizing Europe. *Tuber melanosporum* instead was restricted to the central part of Italy and did not cross the Alps. Therefore, it is questionable whether the white truffle will manage to withstand the pressure posed by climate change.

The findings from the genome project contributed to cast light on the sexual behaviors of truffles. The fungus consists of hyphae which are symbiotically associated with the roots of tree species, to form the so-called ectomycorrhizal symbiosis. At a specific time of the year, the fungus forms temporary reproductive organs, which are the parts we appreciate on our tables. These fruiting bodies are now known to be the result of a sexual encounter between two different fungi, while it was previously believed that truffles were self-mating.

In his interview, Francis Martin remarked how we understand little about truffle ecology. For instance, it is still challenging to understand why the soil surface, nearby trees which present association with black truffle, is completely different and looks like scorched. This is believed to be the result of chemical signals coming from the fungi. Therefore, Martin and his colleagues in Italy have recently started new work on gene expression



Emerging flowers and leaves from the rowan *Sorbus aucuparia*, Poland (Photo by S. Rösner | fotouristen.de).

patterns which might reveal the mechanisms behind these observations. Possibly, these investigations will reveal the nature of the chemical signals exchanged between hyphae and roots and will enable to identify the genes responsible for the development of the fruiting bodies and their particular flavour.

At the end of his article, Bohannon wonders whether Europeans would be ever able to better appreciate a genetically engineered truffle aroma coming from more easily cultivated organisms. The answer from the experts leaves no doubts that consumers will never accept genetically modified portobellos with the whiff of tartufo bianco d'Alba!

Source: Bohannon, J 2009: Rooting around the truffle genome. *SCIENCE* 323: 1006-1007. DOI: 10.1126/science.323.5917.1006

D. OUTPUTS OF MEETINGS AND TRAINING

3rd Annual Meeting of **evoltree**, February 2009

by *Silvia Fluch and Johanna Schmidt*



SYLVIA FLUCH is group leader at the Austrian Research Centers in Seibersdorf (Austria) and head of the plant resource centre. She is involved in research activities on functional genomics and utilisation of gene resources, with special interest in European tree species. silvia.fluch@arcs.ac.at



JOHANNA SCHMIDT is a software developer at the Austrian Research Centers, Seibersdorf (web applications and design). She is administering the EVOLTREE Portal and develops the e-lab application. johanna.schmidt@arcs.ac.at

The 3rd Annual Meeting of **evoltree** took place from February 2nd to 6th, 2009 in Baden, Austria.

Invited speakers from Europe and the United States presented the most recent developments in the different thematic areas covered within the **evoltree** framework:

- i) 'Genomic signatures of adaptive evolution in plants' by M. Tenaillon and 'Searching for selection in the Scots Pine Genome' by O. Savolainen,
- ii) 'Population genomics and complex trait variation in natural Populations' by T. M. Olds and 'Population genomics in hybrid zones of poplar' by C. Lexer,
- iii) 'Putting Canadian and US genomic research projects



Oak leaf from last autumn on the forest floor, where wood anemone *Anemone nemorosa* are flowering in early april, Germany (Photo by S. Rösner | fotouristen.de).

into the picture of European forestry research' by J. Bousquet and N. Wheeler and iv) 'Use of genomic tools to investigate past vegetation dynamics' by E. Willerslev and 'Global change Ecology: 10 years of forest ecosystem experiments tell us what trees will do' by G. Taylor.

Scientific sessions were followed by technical workshops that would gather all working units to allow planning of the joint activities to be implemented till the end of the project.

A visit was paid to the ARC (Austrian Research Centers) campus. The partners of the network had the possibility to visit the automated storage unit of the repository centre as well as the PICME labs. Moreover, the booths of Nexus and Whatman on site were well visited. Presentations on forestry related research issues investigated at ARC were given, covering different topics, from isotope analysis in wood, to forest soil microbial research, climate modelling and forest genomics.

After the visit, a social dinner kindly cosponsored by NEXUS Biosystems, the manufacturer of the storage units at the repository centre, was organized in a typical local tavern ("Heuriger"), where the participants enjoyed

typical Austrian home made meals in an environment characterized by a traditional atmosphere.

evoltree at the 19th session of the United Nations Food and Agriculture Organization's (FAO) Committee on Forestry (COFO)

by *Barbara Vinceti*



BARBARA VINCETI, a forest ecologist at Bioversity International, coordinates research and networking initiatives focused on conservation and sustainable use of forest genetic resources outside Europe. She leads the *evoltree* Dissemination activities. b.vinceti@cgiar.org

Between March 16th and 20th, 2009, over 550 participants from member states gathered at FAO in occasion of the 19th session of the United Nations Food and Agriculture Organization's (FAO) Committee on Forestry (COFO), held jointly with the first edition of the World Forest Week. The participants included representatives of countries, heads of forestry departments, UN agencies, and intergovernmental and non-governmental organizations, all brought together to identify emerging policy and technical issues, to seek solutions and advise FAO and others on appropriate action.

The Committee on Forestry (COFO) is the most important of the FAO Forestry Statutory Bodies. The Committee is convened every two years, at FAO headquarters in Rome. On Monday, 16 March 2009, Jan Heino, the Head of the Forestry Department at FAO, welcomed the participants and highlighted the need for cooperation among forest-related organizations. He reminded the need to address climate change and the challenges posed by the global economic crisis, and emphasized the need for institutional adaptation.

Among the issues discussed in plenary was the proposal put forward by the FAO's Commission on Genetic Resources for Food and Agriculture's (CGRFA) to prepare a State of the World's Forest Genetic Resources report to be complete by 2013. In the following discussion, participants generally supported developing the implementation of such an effort. In particular the Czech Republic, representing the EU, stressed broad participation in formulating the report.

The US representative recommended focusing on the most threatened species and genetic diversity, and actions to preserve them and noted that the State of

the World's Forest Genetic Resources report has to be grounded on countries' cooperation and should be undertaken in collaboration with relevant regional and global programmes.

For the first time, in conjunction with the COFO session, approximately twenty special events were held throughout the week as part of "World Forest Week." These events were intended to create a more informal dialogue and open participation, with delegates speaking in their personal capacity and not as state representatives. On March 17th, 2009, a special event on the event on the preparation of a report on the State of the World's Forest genetic Resources was organized. Reiner Finkeldey, from Göttingen University, was among the invited speakers and in his presentation he had a chance to emphasize the importance of and the threats to forest genetic diversity, our increasing but limited knowledge of genetic resources, and the critical need for international collaboration on the issue. In this context he presented the collaborative scientific efforts undertaken in ongoing initiatives related to forest genetic resources. The initiatives explicitly mentioned were EUFORGEN and *evoltree*. The full presentation given by Reiner Finkeldey (activity leader in *evoltree*) can be found on the *evoltree* website (www.evoltree.eu), in the Dissemination section.



Plenary session at FAO during the 19th session of the United Nations Food and Agriculture Organization's (FAO) Committee on Forestry (COFO) held jointly with the first edition of the World Forest Week (photo: B.Vinceti | Bioversity International)

In occasion of the COFO and World Forest Week, Bioversity International set up a booth in the main atrium at FAO to distribute publications and display posters of various ongoing projects (EUFORGEN, EUFGIS and *evoltree*). Further information on COFO and the World Forest Week can be found within the site maintained by IISD, at <http://www.iisd.ca/fo/cof19/>.

E. FORTHCOMING EVENTS AND ANNOUNCEMENTS

Training course on DNA sequence analysis and association genetics: 13th-17th July 2009, Valsain, Segovia (Spain).

The training initiative will start on Sunday 12 July with a pre-course on sequences assembly, post-processing and genomic resources exploitation, as part of CRIEC (Conifer Re-sequencing Initiative). Deadline for applications: 31st May 2009. The programme is presented below:

- 13-14th July: DNA sequence analysis (nucleotide diversity, linkage disequilibrium and recombination, demographical models and signatures of selection).
- 15th July: quantitative genetic analyses of common garden experiment (BLUP analysis).
- 16-17th July: association genetics (case-control studies, general linear models, mixed linear models, Bayesian methods and family-based approaches (QTD)).

Number of participants: the maximum number of participants is 20 (except for the precourse that is open to all CRIEC contributors).

Organization: this workshop is an intensive 5-day (approx 40 hr) short-workshop, with topics organized into 9 modules (morning/afternoon sessions) and a focus on practical exercises. Theoretical and practical expertise in the analysis of DNA sequences and the development of association studies will be complemented with evening talks by leading researchers in the field. Modules will include both lectures (PowerPoint slides) and practical exercises (computer-based exercises). Analysis of own data-sets are encouraged during the practicals.

Audience: Post-docs, graduate students and advanced undergraduates within the NoE [evoltree](#) (except for the pre-course that is open to all CRIEC contributors). For each module, basic concepts will be reviewed briefly prior to introducing more advanced content and the most commonly used methods will be applied in the



Leaf of *Fagus sylvatica*. (Photo by : S.Rösner | fotouristen.de)

practicals during the afternoon sessions. Resource / reference materials will be provided, using a combination of printed and online resources.

Contact: Ricardo Alía (alia@inia.es);

Santiago C. Gonzalez-Martinez (santiago@inia.es).

International conference on "Fagaceae Genomics": 10th - 13th November 2009 in Raleigh, North Carolina (USA).

An international conference on Genomics of Fagaceae is planned from 10th - 13th November 2009 in Raleigh, North Carolina, USA. The conference is co-organized by the NSF project on genomics of chestnut and the EU [evoltree](#) Network. A web page should be available soon. One of the main outcomes of the conference will be the sharing and exchange of genomic resources among the European and US groups. We will also discuss opportunities for cooperation between the two communities. We hope that there will be strong participation of European groups at the conference, and that various presentations will be made by [evoltree](#) partners.

Participation to the conference will be supported by dissemination activity SEA4. Therefore, if you (as an [evoltree](#) member) are interested in participating, you should announce your intention to Michele Morgante, who is responsible for SEA42.2. Then the budget for the participation will be allocated within WP4. Contact: Michele Morgante (michele.morgante@uniud.it).

E. PUBLICATIONS AND OTHER PRODUCTS

OLBRICH M. et al. (2008). Quantification of mRNAs and house-keeping gene selection for quantitative real-time RT-PCR normalization in European beech (*Fagus sylvatica* L.) during abiotic and biotic stress. *Z. Naturforsch.*, 63c: 574-582.

ABSTRACT: Analyses of different plant stressors are often based on gene expression studies. Quantitative Real-time (qRT-PCR) is the most sensitive method for the detection of low abundance transcripts. However, a critical point to note is the selection of housekeeping genes as an internal control. Many so-called 'housekeeping genes' are often affected by different stress factors and may not be suitable for use as an internal reference. We tested six housekeeping genes of European beech by qRT-PCR using the Sybr Green PCR kit. Specific primers were designed for 18S rRNA, actin, glyceraldehyde-3-phosphate dehydrogenase (GAPDH1, GAPDH2), tubulin, and ubiquitin-like

protein. Beech saplings were treated with increased concentrations of either ozone or CO₂. In parallel, the expression of these genes was analyzed upon pathogen infection with *Phytophthora citricola*. To test the applicability of these genes as internal controls under realistic outdoor conditions, sun and shade leaves of 60-year-old trees were used for comparison. The regulation of all genes was tested using a linear mixed-effect model of the R-system. Results from independent experiments showed that the only gene not affected by any treatment was actin. The expression of the other housekeeping genes varied more or less with the degree of stress applied. These results highlight the importance of undergoing an individual selection of internal control genes for different experimental conditions.

AYLOTT M.J. et al. (2008). Yield and spatial supply of bioenergy short rotation coppice and willow in the UK. New Phytologist. doi: 10.1111/j.1469-8137.2008.02396.x

ABSTRACT: Limited information on likely supply and spatial yield of bioenergy crops exists for the UK. Here, productivities are reported of poplar (*Populus* spp.) and willow (*Salix* spp.) grown as short-rotation coppice (SRC), using data from a large 49-site yield trial network. A partial least-squares regression technique was used to upscale actual field trial observations across England and Wales. Spatial productivity was then assessed under different land-use scenarios.

Mean modelled yields ranged between 4.9 and 10.7 oven-dry tonnes (odt) ha⁻¹ yr⁻¹. Yields were generally higher in willow than in poplar, reflecting the susceptibility of older poplar genotypes to rust and their tendency for single stem dominance. Replacing 10% of arable land, 20% of improved grassland and 100% of set-aside grassland in England and Wales with the three most productive genotypes would yield 13 Modt of biomass annually (supplying 7% of UK electricity production or 48% of UK combined heat and power (CHP) production).

Results show existing SRC genotypes have the immediate potential to be an important component of a mixed portfolio of renewables and that, in future, as new and improved genotypes become available, higher yields could extend this potential further.

TAYLOR G. et al. (2008). Future atmospheric CO₂ leads to delayed autumnal senescence Global Change Biology, 14, 264-275.

ABSTRACT: Growing seasons are getting longer, a phenomenon partially explained by increasing global

temperatures. Recent reports suggest that a strong correlation exists between warming and advances in spring phenology but that a weaker correlation is evident between warming and autumnal events implying that other factors may be influencing the timing of autumnal phenology. Using freely rooted, field-grown *Populus* in two Free Air CO₂ Enrichment Experiments (AspenFACE and PopFACE), we present evidence from two continents and over 2 years that increasing atmospheric CO₂ acts directly to delay autumnal leaf coloration and leaf fall. In an atmosphere enriched in CO₂ (by ca. 45% of the current atmospheric concentration to 550 PPM) the end of season decline in canopy normalized difference vegetation index (NDVI) – a commonly used global index for vegetation greenness – was significantly delayed, indicating a greener autumnal canopy, relative to that in ambient CO₂. This was supported by a significant delay in the decline of autumnal canopy leaf area index in elevated as compared with ambient CO₂, and a significantly smaller decline in end of season leaf chlorophyll content. Leaf level photosynthetic activity and carbon uptake in elevated CO₂ during the senescence period was also enhanced compared with ambient CO₂. The findings reveal a direct effect of rising atmospheric CO₂, independent of temperature in delaying autumnal senescence for *Populus*, an important deciduous forest tree with implications for forest productivity and adaptation to a future high CO₂ world.



Emerging flowers and leaves from ash tree *Fraxinus excelsior*, Germany (Photo by S. Rösner | fotouristen.de).

evoltree

GLOSSARY

AFLP: amplified fragment length polymorphism, a highly sensitive tool used in molecular biology to detect DNA polymorphisms.

alleles: A known variation (version) of a particular gene or genetic marker.

allopatric: When two different plant or animal species occur in different regions or in spatial isolation, they are allopatric <-> sympatric.

amplicon: Amplicons are the target pieces of DNA which are amplified during Polymerase Chain Reactions (PCR).

amplification: Natural or artificial duplication of DNA regions (=> amplicon).

ancient DNA: DNA from remote periods or times early in history.

arboretum: A place where trees, shrubs, and herbaceous plants are cultivated for scientific and educational purposes.

candidate genes: A candidate gene is a gene, located in a chromosome region suspected of being involved in the expression of a trait such as a disease, whose protein product suggests that it could be the gene in question. A candidate gene can also be identified by association with the phenotype and by linkage analysis to a region of the genome.

cDNA: Complementary DNA: DNA synthesized from a => mRNA template in a reaction catalyzed by the enzyme => reverse transcriptase. cDNA therefore comprises functional DNA regions (eukaryotes: exons and introns, prokaryotes: exons) which are expressed at a special physiological state. Continuous cDNA fragments are called => ESTs

chloroplast: A major component of a plastid in green plants and eukaryotic algae of any colour. It is involved in photosynthesis. Prokaryotic photosynthetic organisms do not have chloroplasts. Due to prokaryotic ancestors (=> endosymbiotic theory), chloroplasts exhibit their own DNA material, which is used for population genetics.

clone / cloning: Cloning is the process of producing genetically identical duplicates of organisms or DNA fragments.

conifers: Cone bearing trees. This is a class of the Gymnospermae which includes needle-leaved trees such as pines and cypresses. Their flowers are in cones, and male and flower cones are separate. The oldest (bristlecone pine) and the largest (sequoia) extant organisms belong to this Class. Their unique feature is the inheritance of cytoplasmic DNA (chloroplasts) via pollens.

consensus sequence: The nucleotides or amino acids most commonly found at each positions of the sequences of related molecules.

cpDNA: chloroplast DNA, see => Chloroplast

deciduous: plants (trees) which leaves are falling off or shed seasonally or at a certain stage of development in the life cycle.

dehydrative stress: Stress in plants caused by a lack of water or depletion in body fluids.

demography: The statistical study of populations with respect to size, density, and distribution.

detoxification: The reduction or loss of harmful substances.

diapause: A period of physiologically enforced dormancy between periods of activity.

DNA sequence: DNA sequencing is the process of determining the nucleotide order of a given DNA fragment.

ectomycorrhizal fungi (ECM): mutualistic fungi associated with the fine roots of trees in temperate forests.

expressed sequence tag (EST):

fitness: The ability of organisms to reproduce (e.g., level: high and low fitness)

gene expression: Gene expression is the process by which inheritable information from a gene, such as the DNA sequence, is made into a functional gene product, such as protein or RNA.

gene flow: The movement of genes within a population or among populations following genetic admixture. Gene flow

creates new combinations of genes or alleles in individuals that can be tested against the environment. This way it is one of the sources of variation in the process of natural selection.

genetic diversity: Genetic diversity is a level of biodiversity that refers to the total number of genetic characteristics in the genetic makeup of a species.

genotype: The genetic characteristic of an organism that can not be seen (<-> phenotype).

genotyping: The process of determining the Genotype of an organism/Individual.

haplotype: The particular combination of alleles in a linked group encoded by genes in close vicinity on the same chromosome or organellar DNA.

heterozygote: A diploid organism showing different alleles at one locus.

holocene: The geological epoch from 11,700 yrs until today.

homozygote: A diploid organism showing two identical alleles at one locus.

hybridization: Hybridization is the process of combining different varieties or species of organisms to create a hybrid.

in silico: Processes performed with the computer.

ITS: Internal transcribed spacer. Ribosomal DNA spacer region, frequently used for molecular species identification.

ISS: Intensive Study Sites. Large scale ecosystem plots, where trees and selected associated species are mapped, genotyped and phenotyped

JERA: Jointly executed research activities

JSP: Joint Scientific Project

mating system: ... is any of the ways in which animal societies are structured in relation to sexual behavior.

microsatellite markers: Microsatellites, or => Simple Sequence Repeats (SSRs), are polymorphic loci present in nuclear and organellar DNA that consist of repeating units of 1-6 base pairs in length. They are typically neutral, co-dominant and are used as molecular markers which have wide-ranging genetic applications (e.g. population genetics).

mitochondria: A cell organelle in eukaryotic cells, supplying cellular energy, therefore involved in signalling, cellular differentiation and growth, cell cycle and cell death. Due to prokaryotic ancestors (=> endosymbiotic theory), mitochondria exhibit their own DNA material, which is used for population genetics.

monophagous: When species only forage at one specific type of food.

multiple probe concept: Multiple oligonucleotide probes to detect target organisms at different or same phylogenetic levels.

neutral sequences: variations in DNA sequences which are neutral in terms of having no effect on the phenotype.

oligonucleotides: a short nucleic-acid (DNA) chain usually consisting of up to approximately 20 nucleotides.

orthologous: Homologous sequences are orthologous if they were separated by a speciation event: when a species diverges into two separate species, the divergent copies of a single gene in the resulting species are cited to be orthologous.

Palaeoecology: a scientific branch of ecology focussing on ancient environments, plants and animals.

paralogous: PCR: The polymerase chain reaction (PCR) is a technique widely used in molecular biology. It derives its name from one of its key components, a DNA polymerase used to amplify a piece of DNA by in vitro enzymatic replication.

phenological: Concerning the phenology: a branch of ecology focussing on periodic biological phenomena that are correlated with seasonal conditions.

phenotype: The visible or measurable (i.e., expressed) characteristics of an organism (see => genotype).

phenotyping: The process of determining the Phenotype of an organism/Individual.

phylochips: DNA microarrays consisting of rRNA-targeted oligonucleotide probes that were designed according to the "multiple probe concept".

phylogeny: An evolutionary tree showing the inferred relationships of descent and common ancestry of any given taxa.

phylogeography: Phylogeography is the study of the historical processes that may be responsible for the contemporary geographic distributions of individuals. This term was introduced to describe geographically structured genetic signals within and among species. Past events that can be inferred include population expansion, population bottlenecks, vicariance and migration.

physiology: A discipline of science which studies mechanical, physical, and biochemical functions of living organisms.

polyphagous: organisms which feed on many different food types.

primer pair: Pair of oligonucleotides, whose sequences are complementary to a special region within the template DNA (e.g. genomic DNA). Primer pairs are used for starting the amplification reaction of the => PCR.

progeny: the offspring.

pyrosequencing: A new technique used to sequence DNA using chemiluminescent enzymatic reactions. Here, DNA strands are sequenced and then synthesized enzymatically (the complementary strand).

quantitative traits: ecological characters of species (or other taxa) that can be quantified, e.g. number of seeds, spores or biomass.

rarefaction: a technique in ecology for standardization and comparison of species richness in differently sized samples.

RFLP: Restriction fragment length polymorphism, which is a variation in the DNA sequence of the genome that is detected and cut by restriction enzymes (DNA endonucleases that bind and cut specific palindrome DNA sequences). RFLP analysis is used for molecular approaches (e.g. population genetics) by analyzing the size of the resulting fragments by gel electrophoresis.

RNA: Ribonucleic acid. A molecule with a long chain of nucleotides. Similar to DNA but with the base uracil rather than thymine. RNA is central to the synthesis of proteins.

Sanger-sequencing: A DNA sequencing method developed by Frederick Sanger. Here, the PCR reactions are determined (by the use of dideoxynucleotide triphosphates (ddNTPs) as DNA chain terminators).

sequencing: DNA sequencing is the process of determining the nucleotide order of a given DNA fragment.

SNP: Single nucleotide polymorphism: DNA sequence variation occurring when a single nucleotide (A, T, C or G) of a shared sequence differs between individuals (of one or different species).

SSR: Simple Sequence Repeat, see => Microsatellite marker

sub-fossil: Remains of plants or animals are sub-fossil when they are not yet completely fossilized.

sympatric: When two different plant or animal species occur in the same region, they are sympatric <-> allopatric.

ubiquitous: Animal or plant species are ubiquitous when they occur under almost all environmental or habitat conditions.

link¹: <http://dendrome.ucdavis.edu/NealeLab/>

link²: http://www.evoltree.eu/index.php?option=com_content&task=view&id=238&Itemid=327

link³: http://www.evoltree.eu/index.php?option=com_content&task=view&id=194&Itemid=318

link⁴: <http://www.evoltree.eu/geonetwork>

link⁵: <http://www.europeanforestweek.org/home/en/>

link⁶: <http://dx.doi.org/10.1007/s11295-008-0141-5>

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EVOLTREE ACTIVITIES

EVOLTREE WILL LINK FOUR MAJOR DISCIPLINES (genomics, genetics, ecology and evolutionary studies) to improve understanding of forest ecosystems structures, dynamics and processes, by investigating their adaptive diversity, their role in structuring the diversity of associated species (insects and mycorrhizal fungi) and their evolution in response to environmental changes.

THE NETWORK IS FOSTERING INTEGRATION through interdisciplinary research. The genomic activities will be conducted within a 'laboratory without walls' where techniques will be integrated and applied to a wide range of trees and associated species, starting with model species. EVOLTREE will install and enhance the necessary integrated experimental infrastructures, information systems and bioinformatics resources for common use by the partners. Large data sets will be compiled and made accessible for the analysis of geographic and temporal distribution of genetic diversity.

EVOLTREE WILL SPREAD ITS KNOWLEDGE and expertise for the purpose of education, biodiversity monitoring, and conservation. The network will develop training capacities and facilitate mobility opportunities throughout Europe.

SYNERGIES WILL BE ACHIEVED through facilitating the implementation of large-scale genomic projects, by integrating different groups into multidisciplinary research teams, working in intensively studied plots.

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Bioversity International is associated with **evoltree** and responsible for its dissemination activities. Contact: Dr. Barbara Vinceti, Bioversity International, Rome, Italy.
E-mail: b.vinceti@cgiar.org



evoltree is coordinated by INRA. Project Coordinator, Dr. Antoine Kremer, INRA, Bordeaux, France.
E-mail: kremer@pierroton.inra.fr



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Junior Editorial Team of the **evoltree** newsletter (newsletter@evoltree.de):

Kathrin Donges (donges@mail.uni-marburg.de),
Lars Opgenoorth (Lars.Opgenoorth@staff.uni-marburg.de),
Sascha Rösner (mail@sascharoesner.de)