



Annual Newsletter (n°4) August 2012



ENERGYPOPLAR (Enhancing Poplar Traits for Energy Applications) is an EC Seventh Framework Programme project aimed at further improving poplar trees as an energy crop. The work is directed to understand and improve traits such as yield and wood properties coupled to Bioethanol production. The project also addresses environmental and economical sustainability questions.

WITH WORLD ENERGY demand in 2050 set to increase by 84 per cent on the 2007 level, and liquid fuel demand and CO₂ emissions set to double from 2010 levels. In the same time we need to limit climate change's impact on the global temperature, the Intergovernmental Panel on Climate Change (IPCC) predicts a 50 per cent reduction of CO₂ emissions is needed to limit the temperature rise to acceptable levels. So, the search for greener alternatives to fossil fuels has never been more urgent. One very interesting possibility here is to further develop fast growing trees such as Poplar into even more efficient BioFuel crops.

Poplar is a Second Generation Bio fuels crop. One barrier Liquid BioFuels has been the perceived conflict between food crops – another ever-increasing demand – with the land needed to produce biofuels. However, species like poplars thrive in wide-ranging habitats, including marginal land not suitable for agricultural crops, and could prove appealing to farmers for reduced input costs and optimised land management. For this reason one of ENERGYPOPLAR's major aims is poplar's development as a bioenergy short rotation coppice crop (SRC) for large-scale European deployment on marginal land.

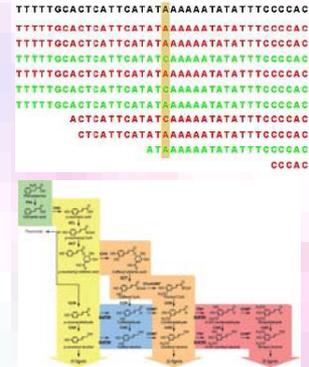
The EnergyPoplar project has now been successfully completed. The aims of the ENERGYPOPLAR project was to further develop poplar as a bio energy crop, specifically for sugar based fuels, short rotation coppice crop, suitable for large-scale deployment in Europe in areas unlikely to be used for food agricultural production.

Selected results and future possibilities

New breeding possibilities using improvements in using state-of-the-art genome analysis

We have developed a new breeding possibility which identifies and uses rare alleles present in the larger population of a certain species. We have named it "Breeding with Rare Defective Alleles (BRDA).

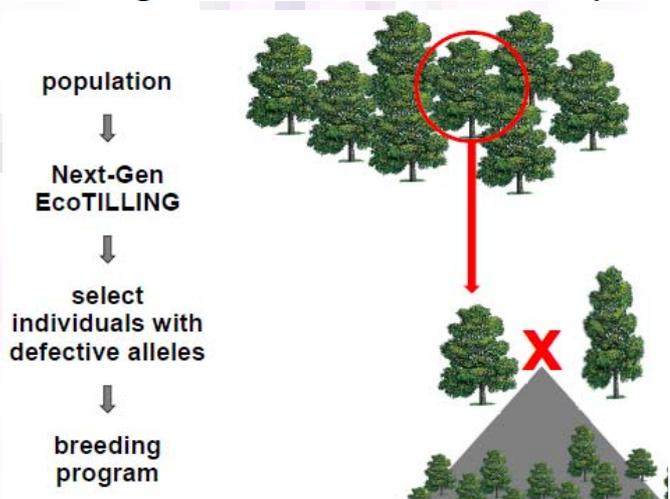
We used high throughput sequencing to identify rare alleles for genes conferring interesting recessive traits. Screening a population of 768 *P. nigra* accessions, we have identified a partially defective allele for the HCT gene, one of the lignin biosynthesis genes. The allele frequency was ~ 0.03. One *P. nigra* accession was homozygous for the defective allele. We have then shown that the tree homozygous for this recessive allele has an altered lignin structure corresponding to the expected lignin modification. This breeding strategy was called Breeding with Rare Defective Alleles (BRDA).



Further research could consist of targeted identification of defective alleles in promising genes in a genetically wide and diverse germplasm. This becomes very cheap as NG sequencing costs reduce. The BRDA strategy will speed up breeding, hence productivity and quality. The potential is therefore large.

This shows the possibility, with basis in the progress of sequencing and genotyping technology, to find and use natural occurring variants of specific genes in breeding strategies. We were able to identify gene variations that only occurred once in a population of 768 trees. Growing evidence suggests that rare functional variants, which are usually missed by genome-wide association scans, play an important role in determining the phenotype. This analysis package can be expanded to any sized tree or plant population, so the size of the population will be determining in which genes useful mutations can be identified. These mutations can then be introduced into breeding projects. We have conducted crosses with some of these mutants and variants in order to create homozygous trees with them. These trees are currently growing and the result will take a few years.

Breeding with Rare Defective Alleles (BRDA)

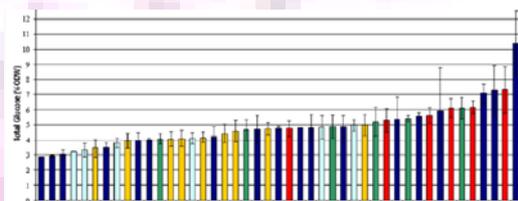


Improved growth and process ability

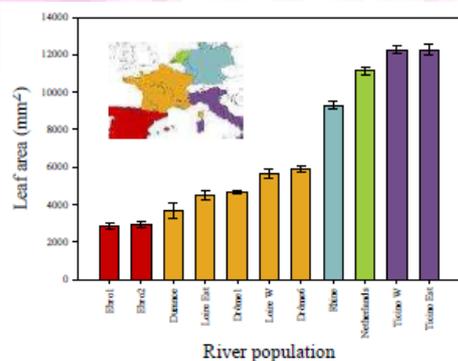
During the project we have analysed and investigated different factors coupled to improving Poplar as a 2nd generation BioFuel crop. These include growth, process ability of the wood to ethanol, water use efficiency and quantities of specific types of wood that have better process ability, e.g. tension wood. These data have generally been coupled to genetical analysis, both forward genetics in tree populations and reverse genetics using transgenic trees. Thus this has produced both knowledge and new target genes for future modifications and breeding.

- Using this information and knowledge we have during the end of the project used this data to initiate new crosses in the practical breeding of Poplars in France.
- We have also used this information to initiate collaborations to test identified genes in transgenic trees to directly see how they could be used.
- This information and knowledge has been and will be published, increasing the general knowledge.

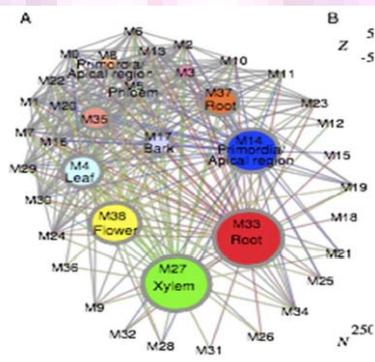
Some examples of data produced and used in the project:



Large variation in process ability of wood from different *Populus* genotypes *P. nigra*: approx 300% 'range. Data used for further breeding.



Variation in leaf morphology between clones adapted to different parts of Europe. Data used for genetic population analysis



Gene expression network used for molecular analysis and selection of targets genes for modifying wood and lignin properties.

Environment and economics

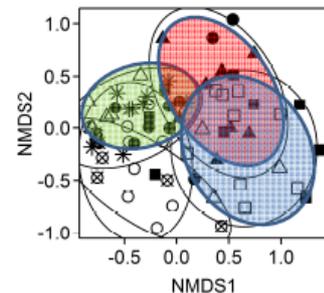
Because the economical and environmental parts of growing an Energy crop is essential to sustainable use, we have studied Environmental and Economical issues. Below some examples from these studies are presented.

Mycorrhizia. Poplar is a water demanding species. In symbiotic associations, mycorrhizal fungi ensheath poplar roots with a mantle, improve plant water use efficiency and rescue wood production when water is limiting and the soil is dry. The mechanistic basis of this beneficial influence of soil biota on poplar yield has been studied.

Due to the importance of mycorrhizia we also have developed an **Ectomycorrhizal Diversity Diagnosis tool** with which one can screen the mycorrhizia population using large scale sequencing techniques and thereby get a figure of the diversity of mycorrhizia present.

This technique was used in the project to make preliminary tests of the diversity of mycorrhizia coupled to the plantations of different poplar clones, species and GM poplar variants. These data will be published when fully analyzed.

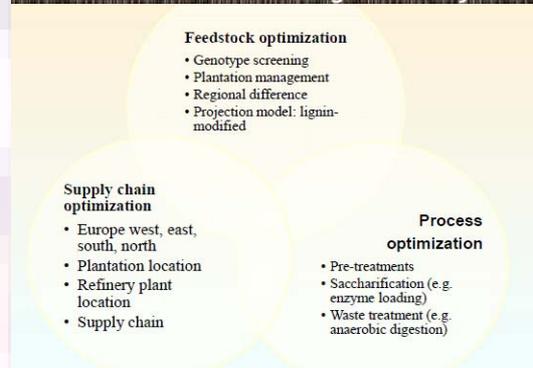
This method could be part of investigating the variability and importance of Ectomycorrhizal Diversity. If Ectomycorrhizal Diversity variation is found of importance, this method could be part of environmental risk assessments when needed.



Life cycle analysis. We have conducted LCA (Life cycle analysis) on the use of short rotation poplar and on the subsequent produced ethanol using different scenarios of development of factors such as nutrient use during Poplar growth, growth speed and wood process ability. Results from this analysis will be published and made available to all.

Data and results produced using this LCA will be useful: in decision making for biofuel project developers and for developers of dedicated bioenergy crops from Poplar, for Knowledge-based policy development by policymakers/regulators as well as for use in and contribution to the public debate regarding 2nd generation liquid biofuels such as Poplars.

LCA & Techno-Economic Modelling research objectives



The project is now finishing up and a Public report of the results will be published beginning of 2013.



For further information please visit: www.energypoplar.eu