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Abstract to be submitted for conference session *Tree Biotechnology: molecular understanding of tree growth and development and applications for tree improvement*

## Investigating the molecular and physiological basis of drought tolerance in *Populus*

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*Key words: Populus, biomass productivity, transcriptome profiling, drought adaptation*

## **Background**

Considering the increasing demand for both, food production and production of bioenergy feedstock, energy crop production systems should preferentially utilise perennial crops grown on marginal sites [1]. Ongoing climate change, especially increasing drought events, put biomass productivity at an increasing risk [2]. Consequently, there is a need to develop new germplasm of perennial biomass crops characterised by high productivity and the ability to maintain productivity under water limited growth conditions. To this end, it is pivotal to understand the underlying physiological and molecular mechanisms of drought stress responses of bioenergy crops, and to investigate the variation of drought stress responses among differentially adapted genotypes of bioenergy crops. Because of its adaptation to a broad range of habitats, its fast growth and available molecular and genomic resources, the genus *Populus* is considered as a model system for woody biomass plants [3]. Here, we report on a study of drought stress adaptation in selected genotypes of *Populus nigra*. Our main hypothesis is that *Populus* genotypes from environments differing in precipitation and mean annual temperature harbour different physiological and molecular mechanisms to cope with limited water availability, which can be exploited to produce improved germplasm for sustained biomass production.

## **Methods**

A drought stress experiment was conducted in the greenhouse facilities at INRA Nancy (Champenux, France) in early summer under ambient light conditions. Plants of three genotypes of *Populus nigra* ("Italy", "France", "Spain") originating from habitats characterized by increasing water limitation were exposed to well-watered conditions or moderate drought for five weeks. Plants were adjusted to the respective level of water availability three times per day on an automated watering system. Measurements of height and diameter growth as well as stomatal conductance and parameters of photosynthetic gas exchange were carried out twice a week.

In order to study the development of the drought effect on the biochemical and transcriptional level over time, we collected mature leaves, fine roots, and developing

secondary xylem of plants of genotype “Italy” once a week. At the end of the experiment, trees of all three genotypes were sampled for biomass determination, and mature leaves, fine roots, and developing secondary xylem were collected for biochemical analyses and transcriptome profiling. Transcriptomics were carried out by mRNA-sequencing (50 bp single-end sequencing with 6x-multiplexing on an Illumina Hi-Seq2000 instrument). Quality-filtered sequence reads were aligned to the *Populus trichocarpa* transcriptome v3.0 [4], and analysis of differential expression was carried out using the Bioconductor package DESeq2 [5].

## **Results and Conclusions**

We found constitutive differences in biomass productivity among genotypes, and a significant reduction of biomass accumulation under drought, irrespective of genotype. Height and diameter growth rates differed among genotypes and were reduced early after the onset the drought treatment. Measurements of photosynthetic gas exchange revealed genotype effects on transpiration rates per unit leaf area, net CO<sub>2</sub> assimilation rates, and stomatal conductance to water vapor. Drought provoked an increase of intrinsic water use efficiency, with some genotype effects. The genotypes differed constitutively in wood anatomical traits, and some traits were also affected by drought.

To link differences in phenotypic traits with changes in the underlying transcriptome, samples of developing xylem, fine roots, and leaves were collected for transcriptome profiling by RNA-Seq (four biological replicates per organ, per treatment, and per genotype). The RNA-seq data of the time-course experiment (genotype “Italy”) revealed a strong treatment effect on all three analysed tissues. In both, fine roots and developing xylem, a considerable number of genes displayed a treatment effect that changed over time (treatment-x-time interaction). Transcriptome profiles obtained from plants of all three genotypes sampled at the end of the experiment revealed a strong drought-effect in mature leaves and fine roots, while the transcriptome of the developing xylem was more resilient. In addition, very strong genotype effects were detected, while only a small number of genes showed genotype-x-treatment-interactions. These pronounced genotype effects on the transcriptional level support our hypothesis of a molecular basis of the observed genotypic variation in biomass productivity.

## Competing interests

The author declares that they have no competing interests.

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