POPCAN:
Genetic improvement of poplar trees as a Canadian bioenergy feedstock

Carl Douglas and Shawn Mansfield
Project Leaders
Large Scale Applied Research Project Competition

2011-2014
POPCAN Research Team

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Climate change creates demand for renewable transportation fuels

2010 second warmest year on record

At the current 5% Federal ethanol supplementation target in Canada ~2 billion litres are required annually

Higher targets (10%) likely in the future

Current Canadian production is ~1.3 billion litres (agricultural feedstocks)
Biology of bioconversion of solar energy into biofuels

## Biomass crop productivity

<table>
<thead>
<tr>
<th>Crop</th>
<th>Total Yield (ton/ha/yr)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sorghum</td>
<td>40-55</td>
</tr>
<tr>
<td>Sugarcane</td>
<td>80-120</td>
</tr>
<tr>
<td>Switchgrass</td>
<td>14-18</td>
</tr>
<tr>
<td>Miscanthus</td>
<td>30-41</td>
</tr>
<tr>
<td>Corn Stover</td>
<td>3-5</td>
</tr>
<tr>
<td>Poplar</td>
<td>12-24</td>
</tr>
</tbody>
</table>

### Fast growing trees: poplar

- J. Eaton

### Perennial grasses: switchgrass

- S. Long
Comparison of forest tree plantation species

Rotation Length (years)

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Populus trichocarpa and *P. balsamifera* as Canadian biomass feedstocks

Poplars are highly productive over large Canadian regions and are well suited as potential biomass plantation species.
Afforestation potential for poplar

150,000 km² (28% of the eligible land base) within the Prairie Provinces estimated as suitable for poplar afforestation

AAFC Agroforestry Development Centre
Ethanl from wood

Size reduction → Pretreatment → Hydrolysis (sugar release) → Fermentation → Ethanol
Secondary cell walls as hydrocarbon feedstocks
Trait improvement/domestication

Biomass

Decrystallization

Hydrolysis
Water

Conc. H₂SO₄

Hydrolysis

Lignin Utilization

Ethanol
dehydration

Ethanol recovery

Acid Reconcentration

Acid/Sugar Separation

Water

Neutralization Tank

Purified Sugar Solution

Gypsum

Fermentor
Key traits for improvement

Biomass yield
• productivity, adaptation, pathogen susceptibility

Recalcitrance
• sugar release, cell wall structure, polymer biochemistry

Socio-economic challenges

Land use policy

Public acceptance
The Genome of Black Cottonwood, *Populus trichocarpa* (Torr. & Gray)


We report the draft genome of the black cottonwood tree, *Populus trichocarpa*. Integration of shotgun sequence assembly with genetic mapping enabled chromosome-scale reconstruction of the genome. More than 45,000 putative protein-coding genes were identified. Analysis of the assembled genome revealed a whole-genome duplication event; about 8000 pairs of duplicated genes from that event survived in the *Populus* genome. A second, older duplication event is indistinguishable coincident with the divergence of the *Populus* and *Arabidopsis* lineages. Nucleotide substitution, tandem gene duplication, and gene chromosomal rearrangement appear to proceed substantially more slowly in *Populus* than in *Arabidopsis*. *Populus* has more protein-coding genes than *Arabidopsis*, ranging on average from 1.4 to 2.6 putative *Populus* homologs for each *Arabidopsis* gene. However, the relative frequency of protein domains in the two genomes is similar. Overrepresented exceptions in *Populus* include genes associated with lignocellulosic wall biosynthesis, meristem development, disease resistance, and metabolite transport.

**Tuskan et al. 2006. Science 313, 1596**

**Nis-1 reference genome is one representation of the genome**

**Large scale variation in different individuals**

**Single Nucleotide Polymorphism - SNP**
Natural populations are reservoirs of genetic and phenotypic variation

*Populus trichocarpa*
Stein Valley BC, Canada
May 2010
Populus trichocarpa collections

BC Ministry of Forests

US Department of Energy
Energy Biosciences Center
UBC Totem Field replicated trial
BC Ministry of Forests collection of ~500 individuals
Agriculture Canada Balsam Poplar (AgCanBaP) collection

Bill Schroeder, Agroforestry Development Centre, Indian Head SK
• Analyze variation in wood chemistry and biomass traits in natural populations grown in common gardens

• Find variants ("SNPs") in all genes within the populations by large scale whole genome resequencing – >1000 poplar genomes

• Identify SNP variants that underlie traits of interest - "association genetics"

• Use SNP variants in marker-assisted breeding to accelerate feedstock improvement
POPCAN Partners

Data generation and analysis

- US DOE BioEnergy Sciences Center
- Joint Genomics Institute
- University of Pretoria, South Africa
- Texas Tech U
- Vancouver Michael Smith Genome Sciences Centre

Breeders and endusers

- Alberta Pacific Forest Industries Inc.
- Greenwood Resources Inc.
- Agriculture Agri-food Canada Agroforestry Development Centre (PFRA)
- BC Ministry of Forests, Lands, and Natural Resource Operations
Social science component - GELS

- Policy and Law on forestry plantations
- Economic models for biomass plantations
- Social acceptability of land use change
- Land use change model: biomass vs. agriculture

GELS team leader: Marty Luckert, Department of Resource Economics & Environmental Sociology, University of Alberta
“In the long view of recent human history, oil production will be a narrow window”


“It isn’t a matter of whether bioenergy and all other forms of carbon-neutral energy will be able to replace petroleum, but when, how, and to what extent.”
- Nick Carpita, President, American Society of Plant Biologists