

Selection of pest and disease resistant, high-yielding shrub willows (*Salix* spp.) for biomass production from novel species hybrid pedigrees

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Fast-growing shrub willow (*Salix* spp.) is genetically diverse consisting of over 300 species and adaptable to many different site conditions. Our breeding program, begun in 1998, continues to exploit genetic diversity utilizing native, naturalized, and exotic accessions collected in the United States, Europe and Asia. Capitalizing on the ability of *Salix* to hybridize, over 700 intra- and inter-specific crosses have produced 270 new families using parents chosen for their yield, form, and resistance to pests and rust infection. Overall, exotics and exotic hybrids exhibit more resistance to feeding by imported and native willow beetle and less susceptibility to rust infection caused by *Melampsora* spp. when compared to native species. Novel inter-specific crosses using exotic species, such as *S. purpurea* (2x) x *S. miyabeana* (4x) and *S. viminalis* (2x) x *S. miyabeana* (4x) have produced putative triploid progeny, while progeny of *S. sachalinensis* (4x) x *S. miyabeana* (4x) are tetraploid. Using a stringent selection process the best individuals within the most productive families were tested in replicated small (4-30 plant) plot trials, followed by additional selection in replicated, 78-plant plot yield trials at multiple sites. In a selection trial begun in 2008 18 varieties performed better than the commercial variety, *S. sachalinensis* 'SX61', and eight new varieties ranked among the top 10 at the end of the first-year post-coppice on the Geneva, NY site. Replicated yield trials using 18 elite varieties were established in Tully, NY and Belleville, NY in 2005 and harvested in November 2008. Together, yield of the top five varieties averaged 12.15 and 10.51 tonnes ha⁻¹ yr⁻¹, respectively, after the first harvest. Seven varieties have been patented and are commercially available through Double A Willow, Fredonia, NY and many are currently in pre-commercial scale-up.

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Keywords: exotic, *Melampsora*, polyploidy, rust, *Salix miyabeana*, *Salix purpurea*, *Salix sachalinensis*, tetraploid, triploid, willow beetle

Preferred topic area:

1. Production Systems and Operations

Molecular Genetic Basis for Variation in Lignocellulosic Biomass Composition in Shrub Willow (*Salix* spp.) Bioenergy Crops

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Genetic improvement of fast-growing shrub willow (*Salix* spp.) requires a thorough understanding of the molecular basis for cell wall composition and molecular tools for early selection in breeding programs. By identifying the genes that are critical for regulating variation in biomass composition, we may be able to develop molecular markers for the early selection of genotypes with varied and improved biomass properties. Using wet chemical analysis and high-resolution thermogravimetric analysis, we have characterized significant variation in biomass composition, bark content, and stem density among commercial and pre-commercial genotypes of shrub willow produced through breeding. To gain an understanding of the molecular basis for difference in biomass composition, we studied genes encoding enzymes involved in lignin biosynthesis and selected carbohydrate active enzymes selected based on their functional characterization and conservation in *Populus trichocarpa* and *Arabidopsis thaliana*. Fragments of nearly 20 genes have been cloned from young stem cDNA of *Salix sachalinensis* and *S. miyabeana*. Expression profiling in willow stem apical tissue and developing stem tissue was performed for each isolated gene using probe-based quantitative real-time PCR. Two willow parental genotypes and six progeny within a hybrid family were selected for analysis and significant differences in expression among the individuals and between tissue types were observed for most of the genes. Patterns of gene expression were correlated with variation in the biomass composition of those genotypes to provide insights into the genetic regulation of lignocellulosic deposition in this important bioenergy crop.

Keywords: biomass composition, cell wall, cellulose synthase, glycosyltransferases, lignin, *Salix*

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Subject Area: Production Systems and Operations

Genetics, Pathology and Wood Science Abstracts

Prediction of Long-term Canker Disease Damage from the Responses of Juvenile Poplar Clones to Inoculation with *Septoria musiva*

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Stem cankers caused by the native fungal pathogen *Septoria musiva* severely limit production of hybrid poplars in eastern North America. A field experiment was conducted to determine whether short-term responses of poplar stems to inoculation with *S. musiva* was predictive of long-term canker disease damage. Stems of 27 poplar clones were inoculated during their first season of growth by removing the fourth or fifth fully expanded leaf and placing an agar plug colonized by an aggressive isolate of *S. musiva* over the resulting wound. Four months after inoculation, incidence of cankers, canker length, and percent of stem circumference affected (girdle) were recorded. Clones varied greatly in canker incidence (12-98%), mean canker length (11-53 mm) and mean girdle (14-94%). Logistic regression analysis was used to compare these responses to inoculation with canker disease damage categories assigned on the basis of information from longer-term field studies. Incidence, canker length, and girdle data all were informative, but girdle data most frequently correctly predicted assigned canker disease damage categories (24 of the 27 clones). Responses of 15 of these clones also were evaluated in a similar greenhouse experiment. Although responses in this greenhouse experiment again usually were predictive of long-term damage categories, the probability of correct prediction was lower for some clones. These results demonstrate the feasibility and potential benefit of screening juvenile poplar clones for responses to inoculation with *S. musiva* before undertaking field trials and release to growers.

Keywords: hybrid poplar, *Populus*, canker, *Septoria musiva*, disease, screen, resistance

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Subject Area: 1. Production Systems and Operations

Adaptation to climate change: Identification of climatically resilient hybrid poplar cultivars

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Woody biomass is a renewable resource that can serve as a feedstock to produce electricity and heat (bioenergy), as well as liquid fuels such as ethanol (biofuels) which in turn helps displace fossil fuel use. Sustainable woody biofeedstock production systems require a reliable supply of woody biomass. To be economically feasible, growth and yield of hybrid poplars needs to be accurately accounted for, and possible perturbations in biomass supply due to changes in climate must be accounted for to minimize risk in economic investments. However, there is limited understanding of the climatic sensitivity of short rotation woody crops such as hybrid poplars. The general objectives of this study is to identify climatically resilient hybrid poplar clones for woody biomass feedstock development. Specifically, tree-ring analysis methods (dendrochronology) were used to quantify the influence of climate on stem growth rates of hybrid poplars by measuring year-to-year changes in tree-ring width from different cultivars of hybrid poplar and relating annual growth patterns with past instrumental climate records (i.e., temperature and precipitation). Tree-ring analysis was conducted on a full-sib progeny plantation of different cultivars of hybrid poplars (*Populus x smithii* derived from different geographical variants of aspen parents: trembling aspen (*Populus tremuloides*) and bigtooth aspen (*Populus grandidentata*) located on Michigan State University property in the Sandhill Research Area. Climatic sensitivities that were identified in the hybrid poplar cultivars included: the degree of summer moisture stress, the timing of the start of the growing season, the timing of the end of the growing season, and the degree of winter harshness. Tree-ring analysis can thus contribute to retrospective screening of the sensitivity of hybrid poplar cultivars to climatic stressors.

Keywords: climate change, dendrochronology, hybrid poplar, tree improvement,

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Topic Area: Environmental and Landscape Processes and Benefits

Genetics, Pathology and Wood Science Abstracts

Inter-correlations of Physical and Chemicals Properties amongst Hybrid Poplars Grown in Wisconsin, Minnesota and Iowa

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ABSTRACT

Twenty-two different poplars were harvested across three sites (Arlington, WI; Iowa State University, Ames IA; Westport, MN) in November 2002 (7.5 years of age). The sample size was narrowed to 11 poplars from across all 3 sites in November 2003 and finally to 9 poplars from the Arlington, WI site in November 2004. There were many interesting correlations between physical fiber properties and outcome parameters relevant to papermaking. Similarly, there were also interesting correlations between chemical properties of the wood meals and outcome parameters relevant to the synthesis of bio-fuels and other bio-products. Two examples are shown in Figures 1&2 below. These results indicate that clonal selection could be an invaluable tool in many sectors of our emerging renewable economy.

Keywords: hybrid poplar, lignin content, S:G ratio, microfibril angle, pulping

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Subject Area: Pulp and paper manufacture, Bio-fuels, Bio-chemicals

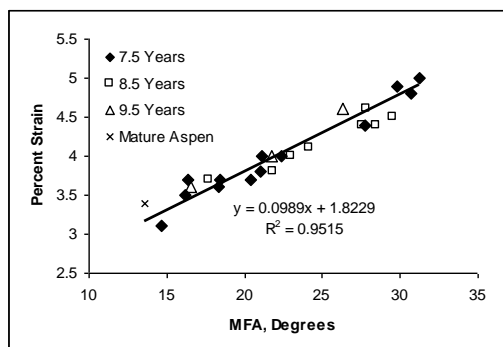


Figure 1: Microfibril angle (MFA) versus strain to the point of sheet rupture for a wide variety of poplars.

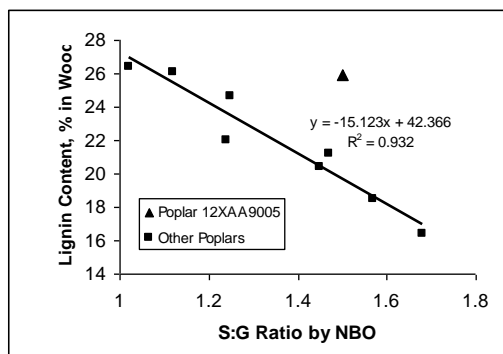


Figure 2: Lignin content versus syringyl to guaiacyl (S:G) ratio for poplars harvested from Arlington, WI.

Hot water extraction of shrub willow and sugar maple chips reduces ash content, increases energy content

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Shrub willows are being developed as a short rotation woody crop (SRWC) that can be grown on marginal agricultural land and combined with woody biomass from forests for the production of bioenergy, biofuels and bioproducts. The bark content in willow biomass is higher than other woody biomass because of its small diameter, which can increase the ash content and lower the melting point, thus increasing the potential for clinker formation and slagging. This study focuses on the changes in ash and energy content of sugar maple and three varieties of shrub willow (SV1, 9871-31 and SX64) with varying bark content (0%, 33%, 66% and 100%) that occur when a hot water extraction process is employed. This process is being developed for biorefineries that will produce biofuels, bioproducts and other products for combined heat and power. The mass removed in the extraction process ranged from 19.7 to 23.7% of the initial dry weight. The process caused a reduction in ash content of about 50% across all the samples, but varied among the varieties of willow and the amount of bark in the samples. Alkali metals were significantly reduced by the hot water extraction process. The heat energy content of the willow and sugar maple increased about 5% after the hot water extraction process because of the disproportionate removal of hemicellulose that has a relatively low heating value. Thus the hot water extraction provides a fermentable sugar stream for biofuel and products and can also improve the characteristics of willow and sugar maple biomass for pellet production and use in combined heat and power applications. This study further demonstrates how woody biomass with higher bark content can be modified for combined heat and power by managing its ash content while simultaneously producing other valuable products.

Keywords: SRWC, hot water extraction, bark content, ash content, alkali metals

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Subject Area:3