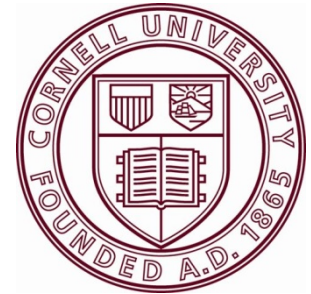


# *Breeding Triploid Hybrids of Shrub Willow with Improved Yield and Biomass Composition*

Larry Smart, Associate Professor  
Cornell University, Dept. of Horticulture  
New York State Agricultural Experiment Station  
Geneva, New York, USA



lbs33@cornell.edu  
[www.hort.cornell.edu/smart/](http://www.hort.cornell.edu/smart/)

 WILLOWPEDIA <http://willow.cals.cornell.edu>



# Willow biomass can be a feedstock for biopower, heat, and liquid biofuels



Lyonsdale Biomass Power Plant, Lyons Falls, NY  
Photo: Catalyst Renewables



VeraSun Station, Brookings, SD



ACT Bioenergy Wood Boiler

...each with its own optimal biomass composition

# Shrub Willow

## Breeding Goals:

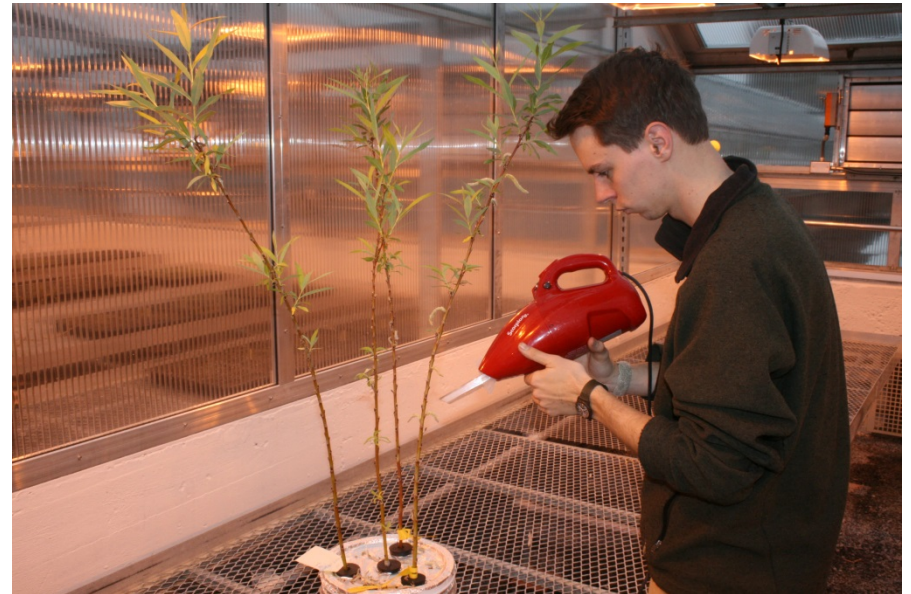
- Yield, yield, yield
- Pest/disease resistance
- Density/composition
- Form: harvesting/cuttings
- Stress tolerance for marginal sites





# Breeding Approach:

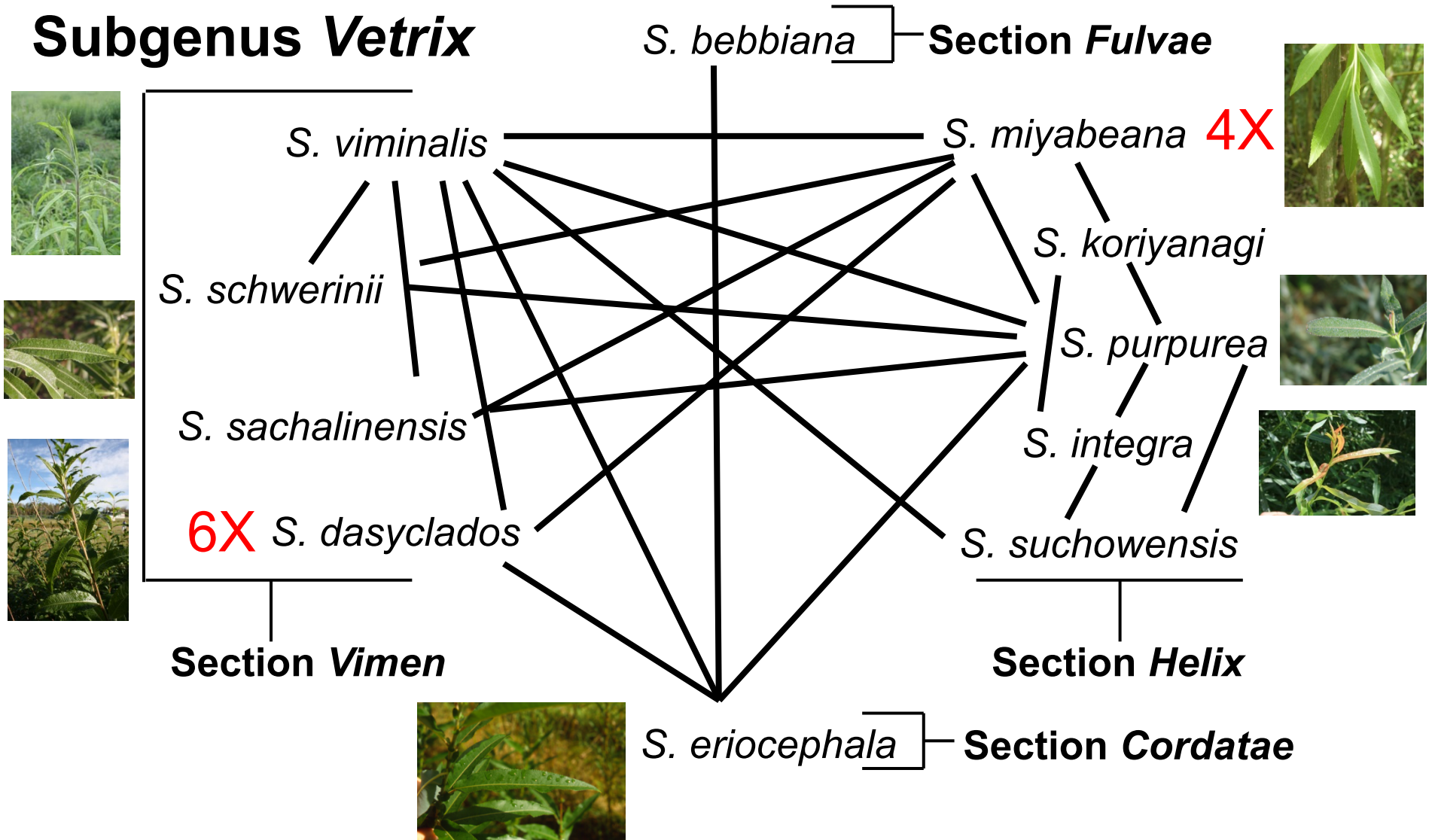
- Capture hybrid vigor and combine traits through controlled pollination and species hybridization



# Breeding Strategy:

*Exploit the diversity of Salix through hybridization*

## Subgenus *Vetrix*



*Compilation of results from US, Canadian, and European breeding programs*

# Inter-specific Hybridizations



**~10,000  
seedling  
progeny  
produced  
in last 5  
years at  
Cornell**

## **F<sub>1</sub> hybrids**

*S. integra* x *S. purpurea*  
*S. cordata* x *S. eriocephala*  
*S. purpurea* x *S. eriocephala*  
*S. purpurea* x *S. viminalis*  
*S. purpurea* x *S. sachalinensis*  
*S. purpurea* x *S. gilgiana*  
*S. purpurea* x *S. suchowensis*  
*S. koriyanagi* x *S. purpurea*  
*S. koriyanagi* x *S. integra*  
*S. koriyanagi* x *S. miyabeana*  
*S. viminalis* x *S. miyabeana*  
*S. viminalis* x *S. eriocephala*  
*S. x dasyclados* x *S. miyabeana*  
*S. x dasyclados* x *S. eriocephala*  
*S. x dasyclados* x *S. viminalis*  
*S. eriocephala* x *S. purpurea*  
*S. matsudana* x *S. alba*  
*S. sachalinensis* x *S. eriocephala*  
*S. sericea* x *S. purpurea*  
*S. sericea* x *S. eriocephala*  
*S. sericea* x *S. sachalinensis*  
*S. discolor* x *S. cinerea*  
*S. discolor* x *S. eriocephala*

*S. miyabeana* x *S. suchowensis*  
*S. miyabeana* x *S. dasyclados*  
*S. integra* x *S. suchowensis*  
*S. alberti* x *S. purpurea*  
*S. alberti* x *S. miyabeana*  
*S. alberti* x *S. viminalis*

## **Multi-species hybrids**

*S. viminalis* x (*S. purpurea* x *S. miyabeana*)  
*S. viminalis* x (*S. viminalis* x *S. miyabeana*)  
*S. purpurea* x (*S. purpurea* x *S. miyabeana*)  
*S. purpurea* x (*S. viminalis* x *S. miyabeana*)  
(*S. viminalis* x *S. schwerinii*) x *S. cinerea*  
*S. miyabeana* x (*S. purpurea* x *S. miyabeana*)  
(*S. kori* x *S. pur*) x *S. miyabeana*  
(*S. kori* x *S. pur*) x *S. suchowensis*  
(*S. kori* x *S. pur*) x (*S. kori* x *S. pur*)  
(*S. pur* x *S. sach*) x (*S. pur* x *S. sach*)  
(*S. suchowensis* x *S. viminalis*) x *S. purpurea*  
(*S. suchowensis* x *S. viminalis*) x *S. miya*  
*S. miya* x (*S. vim* x (*S. schwerinii* x *S. vim*))  
(*S. alberti* x *S. leocopithecica*) x *S. miya*  
(*S. alberti* x (*S. integra* x *S. such*)) x *S. miya*  
(*S. alberti* x (*S. integra* x *S. such*)) x *S. pur*  
(*S. integra* x *S. suchowensis*) x *S. purpurea*  
(*S. integra* x *S. suchowensis*) x *S. such*  
(*S. integra* x *S. suchowensis*) x *S. purpurea*  
*S. purpurea* x ((*S. kori* x *S. pur*) x *S. such*)  
*S. vim* x (*S. vim* x (*S. schw* x *S. vim*)) x *S. miya*



# Selection and Scale-up Strategy

**Controlled pollinations**  
- start seeds in gr chamber  
- transplant to greenhouse

↓  
**Plant seedlings  
in field**  
*1,000's*

**Family Screening Trial**  
Single-plant plots in  
family rows

*2-3 years* ↓ **Select, propagate**  
*60-80*

**Selection Trial**  
Single site, replicated,  
multi-plant plots

→ **Select 12-15**  
*2-4 years*

**Yield  
Trials**



# Selection and Scale-up Strategy

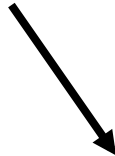
**Controlled pollinations**  
- start seeds in chamber  
- transplant to greenhouse

Plant seedlings  
in field



**Family Nursery Beds**  
Single-plant plots in  
family rows

Propagate all  
in families



**2013 Selection Trial**  
284 clones, 4 reps  
3-plant plots

**2014 Selection/QTL Trial**  
1085 clones, 4 reps  
3-plant plots





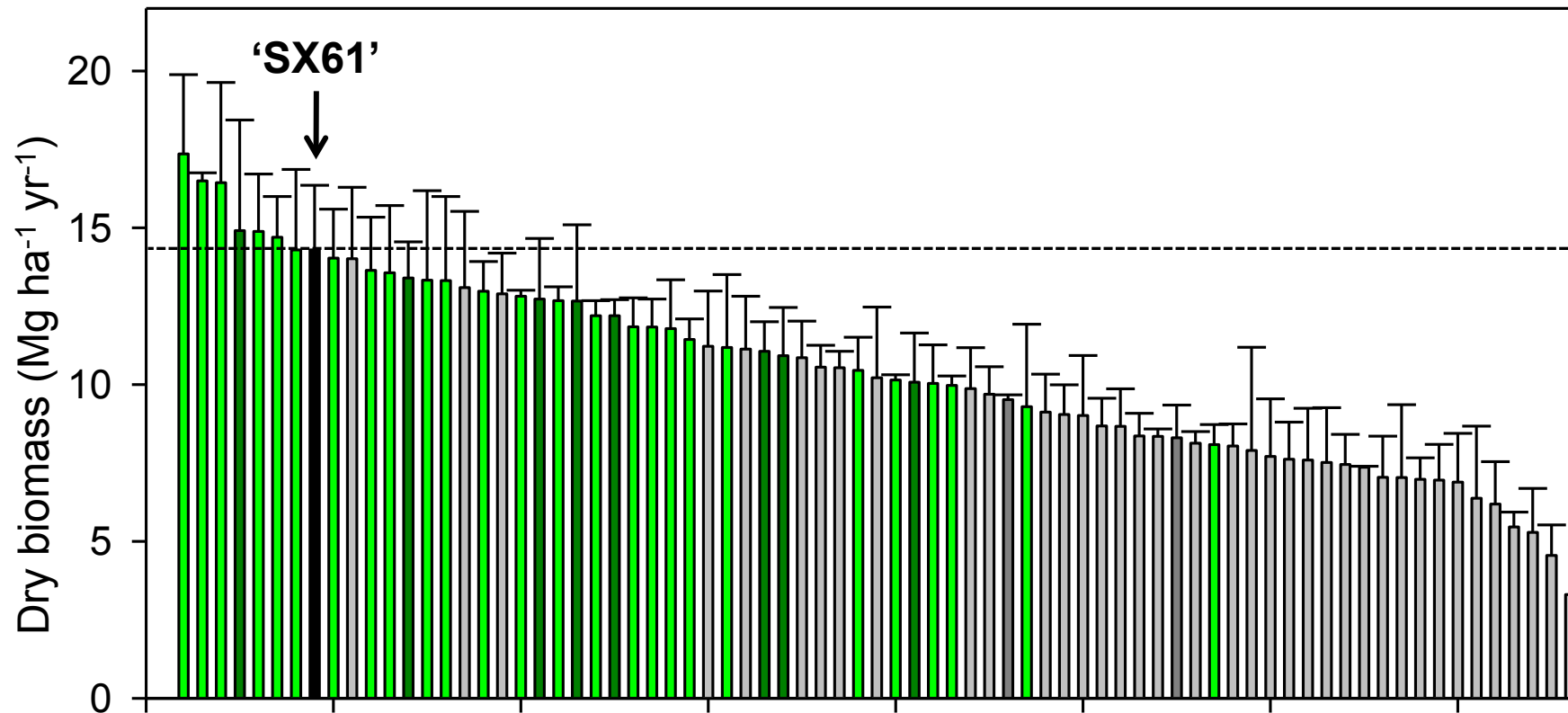
# 2008 Genetic Selection Trial - Geneva, NY

- 24-plant plots, 3 replicates, 75 clones
- Biomass harvested from middle 8 plants in Dec. 2011



# 2008 Genetic Selection Trial - Geneva, NY

- 6 new genotypes ranked higher than 'SX61'
- Top genotype produced 21% greater yield



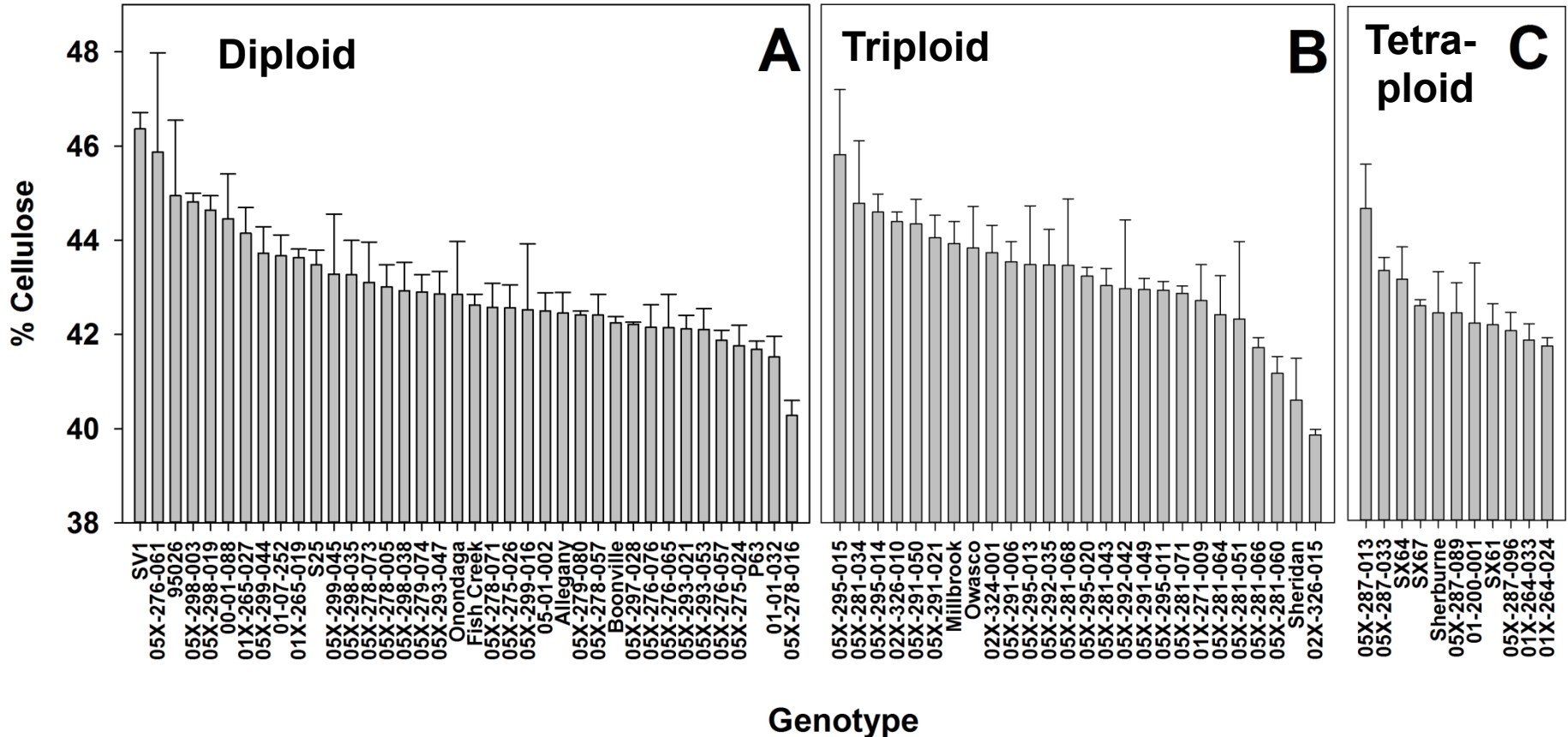
- DIPLOIDS = 8.3 dry Mg ha<sup>-1</sup> yr<sup>-1</sup> (n=39)
- TRIPLOIDS = 12.7 dry Mg ha<sup>-1</sup> yr<sup>-1</sup> (n=26)
- TETRAPLOIDS = 12.5 dry Mg ha<sup>-1</sup> yr<sup>-1</sup> (n=9)
- PENTAPLOIDS (n=2)



# 2008 Genetic Selection Trial - Geneva, NY

## *Third-year cellulose content*

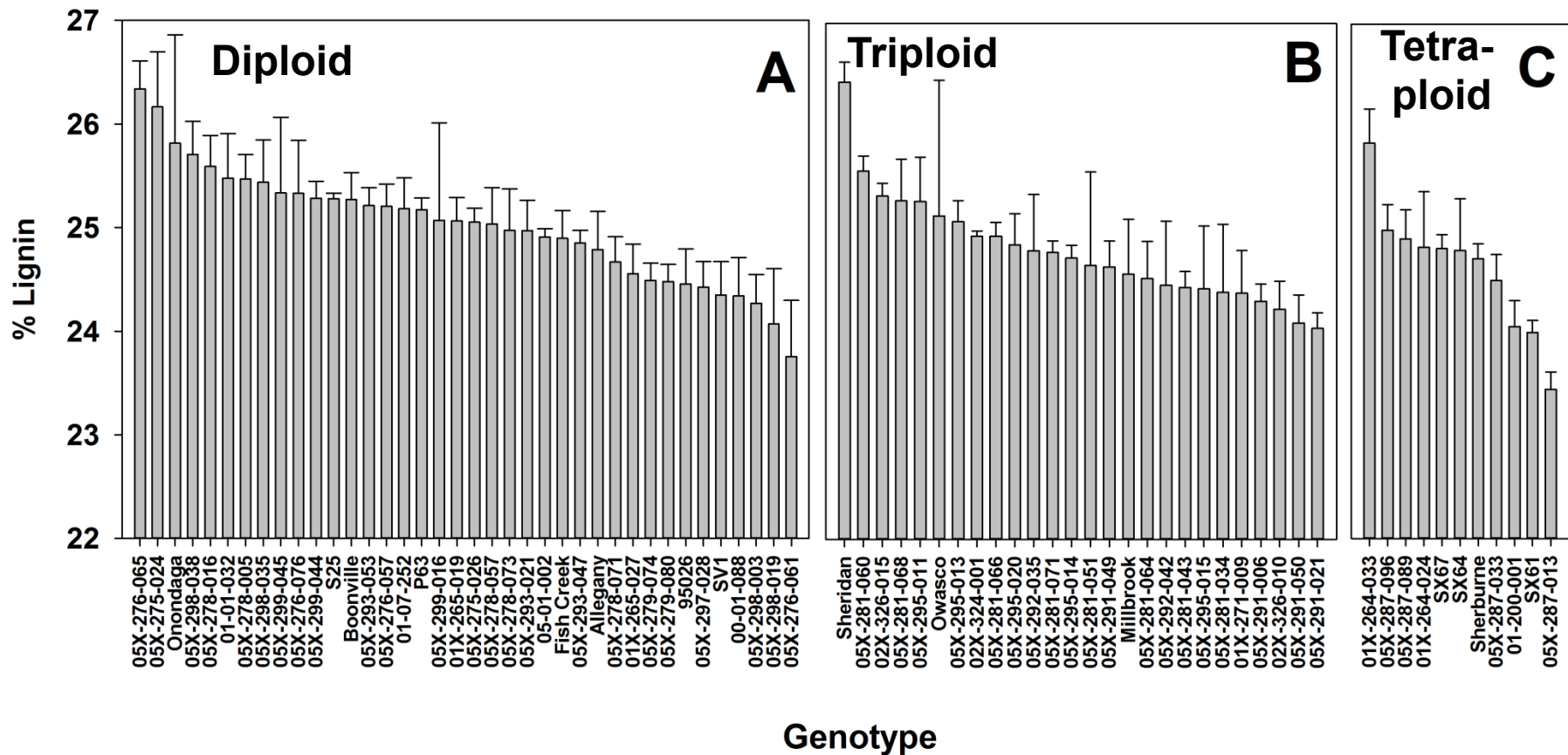
- positively correlated with yield
- strongly negatively correlated with lignin and ash
- not significantly different by ploidy



# 2008 Genetic Selection Trial - Geneva, NY

## *Third-year lignin content*

- negatively correlated with yield and height
- negatively correlated with cellulose, positively with ash
- significantly lower in triploids and tetraploids

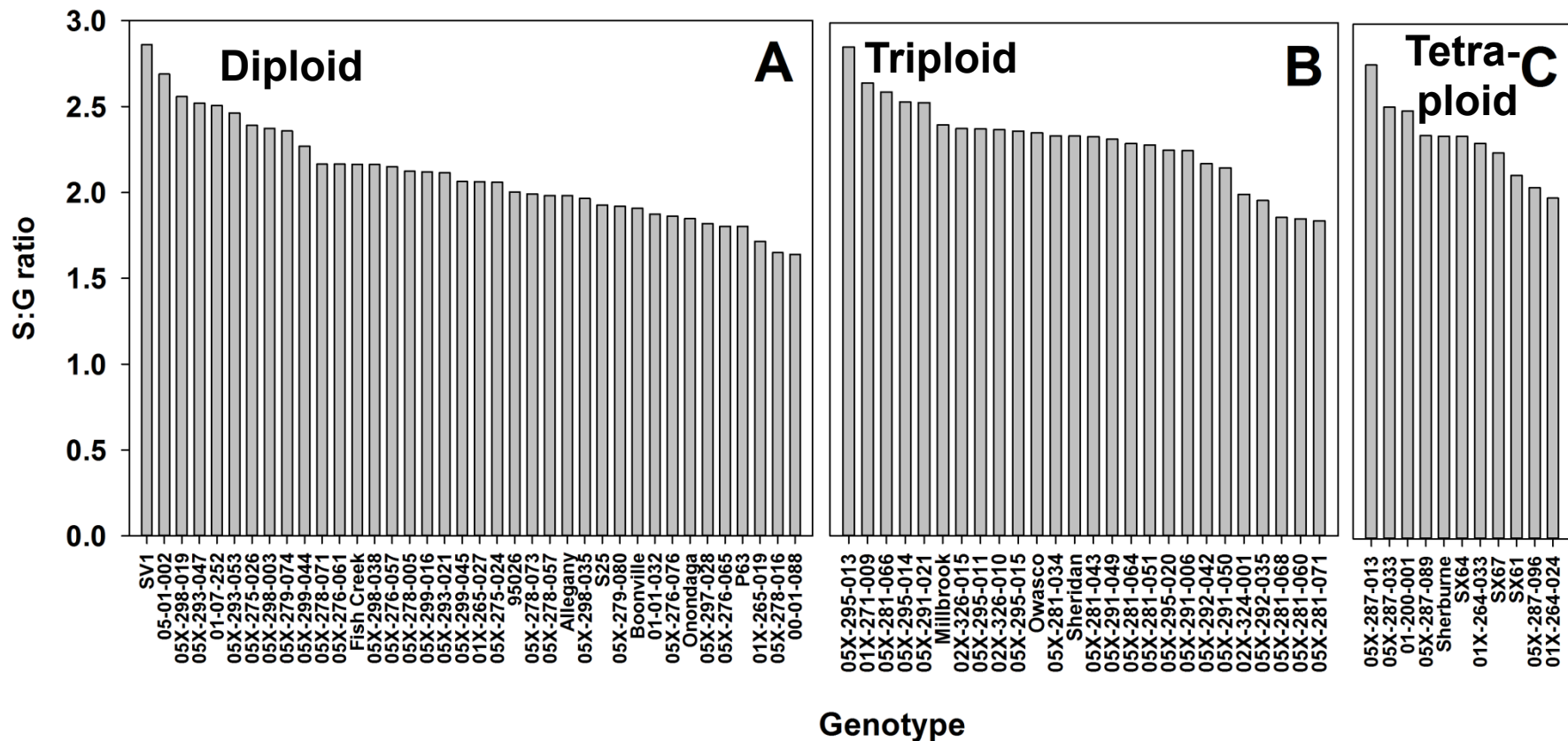




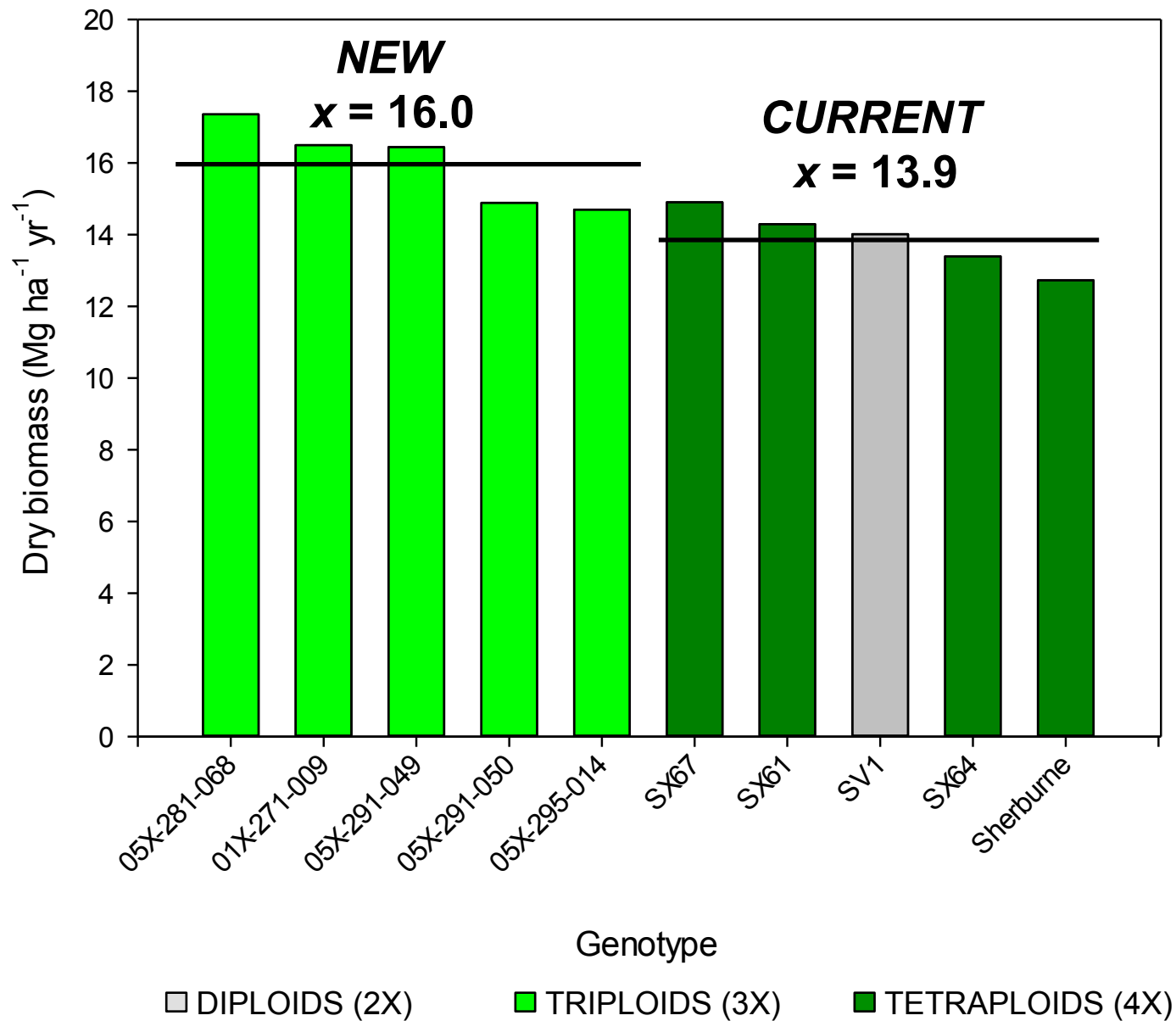
# 2008 Genetic Selection Trial - Geneva, NY

## Third-year lignin S:G ratio

- positively correlated with yield, height, density, cellulose
  - negatively correlated with lignin and ash
  - only one rep analyzed from year 3 due to cost

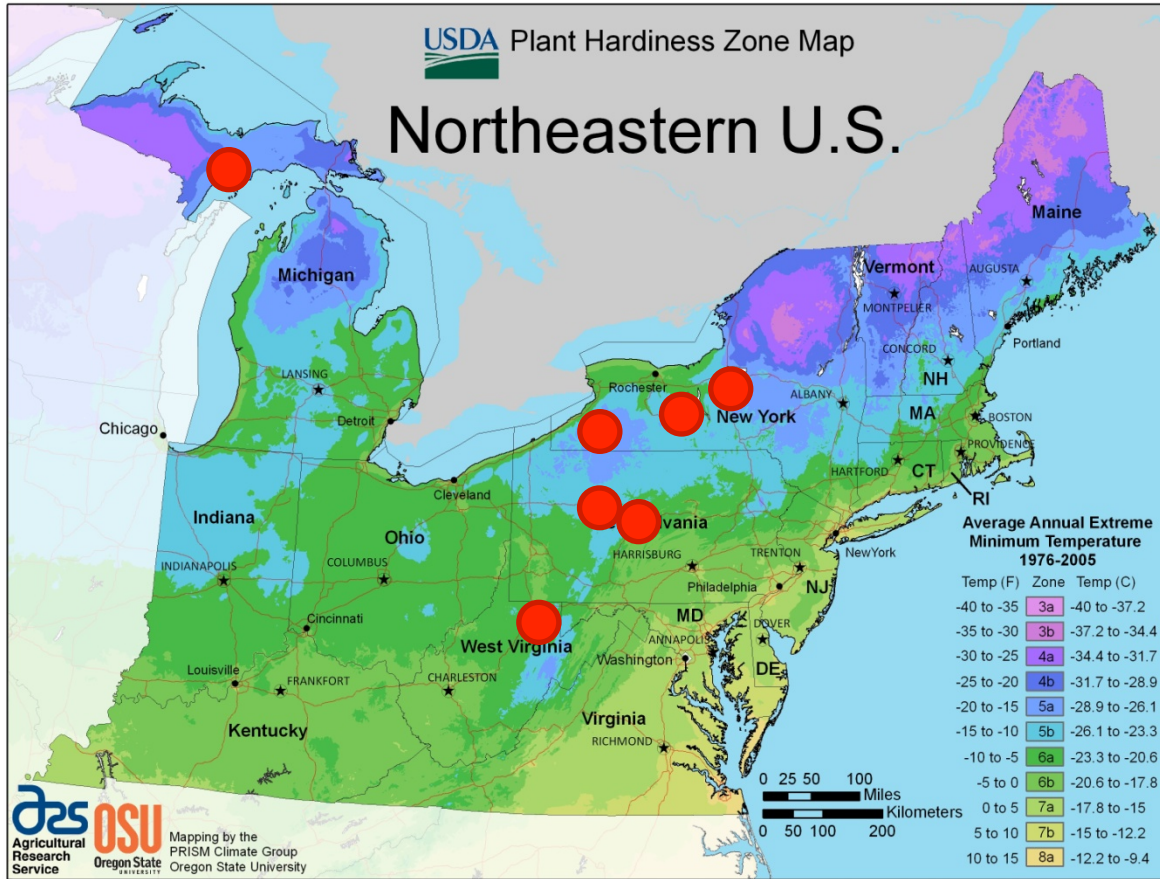


# Mean of Top Five New vs. Current Cultivars = 15% increase





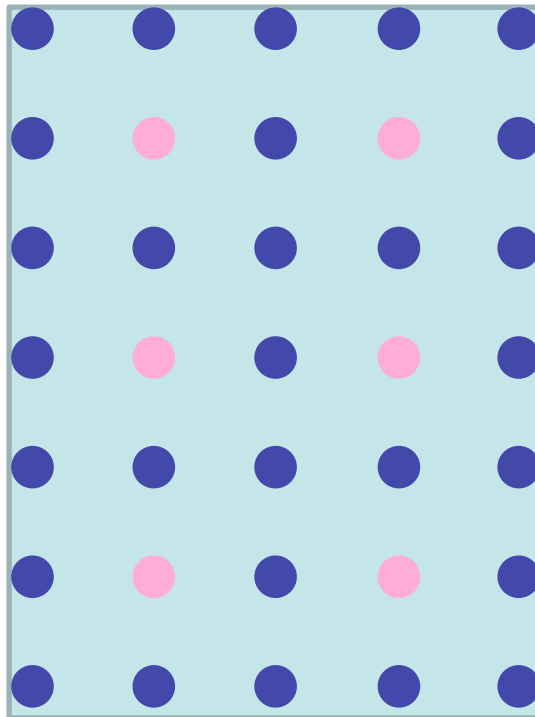
# NEWBio Regional Trials



- Yield Trials (24 cultivars, 48 plant plots)
  - two sites are reclaimed mine land
  - one site – paired amended/unamended

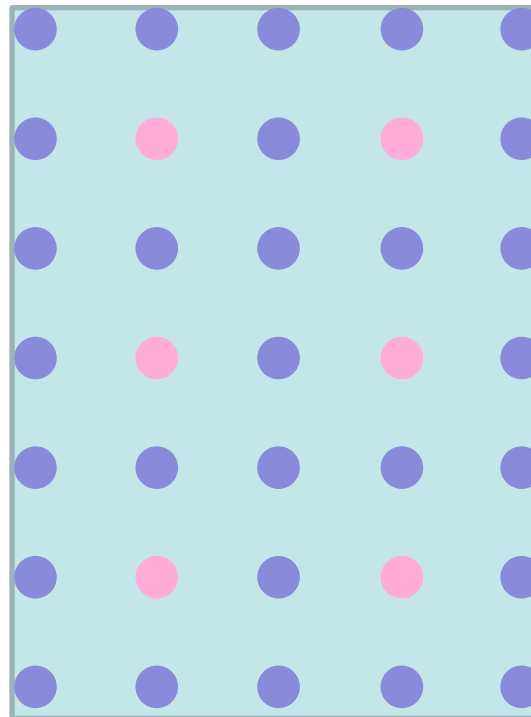
# Long-term Triploid Breeding Strategy: *Population improvement of diploids and tetraploids* *Crossing blocks, half-sib seed collected in the field*

**Foundation**



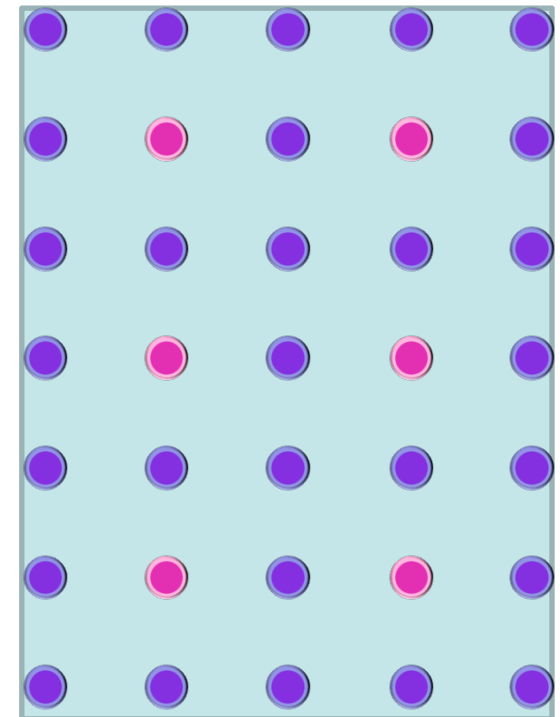
♀, ♂ =  
unimproved  
tetraploids

**Unimproved**



♀ = unimproved  
tetraploids  
♂ = unimproved  
diploids

**Improved**



♀ = improved  
tetraploids  
♂ = improved  
diploids



## ***Conclusions...***

- We can capture hybrid vigor in willow through hybridization of diverse species.
- Natural variation in ploidy among *Salix* spp. can be exploited to produce triploid progeny.
- Biomass composition traits vary significantly among diverse genotypes and are correlated with yield and height; some traits differ by ploidy.
- Most promising commercial cultivars are triploid and are essentially sterile.

# Thanks to...

**Michelle Serapiglia**  
**Fred Gouker**  
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**Art Stipanovic (ESF)**  
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**Ray Miller (MSU)**  
**Steve DiFazio (WVU)**  
**Tim Volk (ESF)**  
**Armen Kemanian (PSU)**  
**Marvin Hall (PSU)**

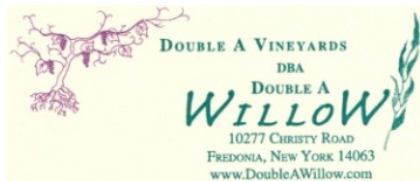


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