Genetic Resources, Genomic Diversity, and Functional Trait Variation in the Perennial Triticeae

Steve Larson, Ivan Mott, Shaun Bushman, and Richard Wang
Functional working groups at FRRL

Biomass and Biofuels

- Altai, basin, giant, mammoth wildryes
- Intermediate / tall wheatgrasses

Low-Maintenance Turfgrasses

- Rhizomatous crested wheatgrass
- Thickspike wheatgrass

Irrigated Pasture Plants

- Roadcrest

Rangeland Ecology and Restoration

- Bluebunch wheatgrass
- Crested wheatgrass
- Basin wildrye
- Russian wildrye
Perennial Triticeae grasses are classified into nine genera comprised of six distinct genomes.
**Psathyrostachys and Leymus “wildryes” share the same Ns genome**

**Psathyrostachys**
- Eight diploid or autopolyploid species, which define Ns genome
- Native to arid regions of central Asia
  - Russian wildrye (*P. juncea*) is valuable forage in its native range and western North America

**Leymus**
- 50 allopolyploid species containing the Ns and Xm genomes
- Most abundant in eastern Asia
  - Important native forages such as *L. chinensis*
- 15 North American taxa
  - 10 native
  - 1 circumboreal (*L. mollis*)
  - 4 introduced, including cultivated Altai wildrye (*L. angustus*)

*Psathyrostachys* and *Leymus* species used for gene introgression in wheat
Phylogeny of *Leymus* chloroplast genomes

**EURASIAN**

*Leymus* taxa

**NORTH AMERICAN**

*Leymus* taxa

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**L. racemosus**

**L. cinereus** / **L. triticoides**
Basin wildrye

*(Leymus cinereus)*  

- Largest native grass of western North America  
- Where water and soil accumulate, croplands  
- Good early spring forage, grows coarse in summer  
- Elevated growing point susceptible to clipping  
- Large prolific spikes, seed shattering explosive  
- Poor seedling establishment  
- Five U.S. & Can. cultivars / 231 USDA accessions
DNA fingerprinting and flow cytometry analysis

basin wildrye ecotype

4x 8x
Western
Rocky Mountain
Great Basin

basin wildrye cultivars

Magnar
Trailhead, Tetra, and Washoe
Continental (Magnar x Trailhead)
Creeping wildrye (*Leymus triticoides*)

- Relatively short (up to 1.3 m) but strongly rhizomatous grass
- Once dominant species in Central Valley, where it is still cultivated as saline biomass crop
- Good clipping and grazing tolerance
- Non-shattering, but recalcitrant seed dormancy
- One cultivar, Rio / 19 USDA accessions
Bayesian cluster analysis of 600 genotypes reveals some hybridization between species and ecotypes.
Experimental creeping x basin wildrye hybrids

**Functionally important traits:** rhizomes, seed dormancy, salt-tolerance, seed-shattering, and biomass heterosis.

**Hypothesis:** The tall plant height, large stems, and large leaves from basin wildrye combined with the prolific rhizomes and tillers of creeping wildrye produce biomass heterosis.
EST library from perennial rhizome/tiller buds creeping x basin wildrye hybrids

- 12,281 Sanger EST unigene contigs (average 973 bp)
- 69 STS primer pairs for nine of then known lignin biosynthesis genes
- 1,798 SSR primer pairs
- 91% amplify Leymus (*NsXm*)
- 74% *Psathrostachys juncea* (*Ns*)
### Genetic maps for creeping x basin wildrye hybrids

<table>
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<th>LG</th>
<th>Total no. markers</th>
<th>Map Distance (cM)</th>
<th>Markers cM⁻¹</th>
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<td>360</td>
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Alignment of 88 mapped *Leymus* ESTs to a high-density *Hordeumum* EST map

Leymus linkage groups

Hordeum linkage groups

4Ns / 5Ns reciprocal translocation
Comparison of QTLs that controlling plant height, rhizomes, biomass heterosis, and other traits.
Russian wildrye
(*Psathyrostachys juncea*)

- One of most versatile forage grasses for dryland pastures (e.g. late summer through winter)
- 20 – 40 cm precipitation
- Stays greener and cures later with higher protein
- The *psathyros* “fragile” *stachys* “spike” can be problematic in seed production
- At least 9 U.S. & Can. Cultivars / 202 accessions
Transcriptome library of Russian wildrye

Russian wildrye
*Psathyrostachys juncea* (Ns)
- 270,000 reads (429 bp)
- 17,200 contigs (690 bp)
- 22,700 unmatched (369 bp)
- 40,000 isotigs

creeping x basin wildrye
*Leymus* (Ns X)
- 11,281 Sanger EST unigenes (1017 bp)
- 375 mapped ESTs

Unknown ancestor (X)
Psuedoroegneria
• 15-20 Diploid or autopolyploid species, which define \textit{St} genome
  – Mostly either Asian or Eurasian
  – One species, bluebunch, native but very widespread and abundant in western North America

\textit{Elymus}
• 150 allopolyploid species containing \textit{St} and \textit{H} genomes
• 39 native or naturalized species in North America
  – \textit{Elymus repens}

\textit{Pascopyrum}
• One allooctoploid (\textit{StHNSXm}) species, western wheatgrass, endemic to North America
Bluebunch wheatgrass
(*Pseudoroegneria spicata*)

- Most widespread, abundant native bunchgrass western U.S.
- 30 – 50 cm precipitation
- Very palatable, nutritious forage
- Does not tolerate heavy grazing, easily overgrazed
- Four cultivars / 184 USDA accesses
Bayesian cluster analysis of 565 genotypes identify 20 metapopulations of bluebunch wheatgrass

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<td>33.9 ± 1.5</td>
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Allotetraploid Snake River wheatgrass
(*Elymus wawawaiensis*)

- Very similar to diploid bluebunch wheatgrass, but contains both *St* and *H* genomes
- Two cultivars / 47 USDA accessions
- Secar has been most widely used native grass cultivar, because it was mistaken as BBWG
- Sensitive to clipping / overgrazing
Streambank wheatgrasses
(*Elymus lanceolatus*)

- Allotetraploid with **St** and **H** genomes
- Forage and low-maintenance turf types
- Better clipping and grazing tolerance
- Lower forage yield potential
- At least 4 cultivars
- 29 USDA accessions
Snake River x Streambank wheatgrass hybrids

- Rhizomes
- Turfiness
- Forage yield
EST libraries for the St and H genomes of *Pseudoroegneria* and *Elymus* “wheatgrasses”

- **Bluebunch wheatgrass**
  - *Pseudoroegneria (St)*
  - 8,780 Sanger EST unigenes (1017 bp)
  - 451 polymorphic SSRs

- **Snake River x Streambank**
  - *Elymus (St H)*
  - 7,212 Sanger EST unigenes
  - 133 polymorphic SSRs
### Genetic maps for Snake River x Streambank wheatgrass hybrids

<table>
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<th>Subgenome</th>
<th>AFLP markers</th>
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Synteny among rice, wheat, and *Elymus* wheatgrass identify homoeologous linkage groups

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Western wheatgrass
(*Pascopyrum smithii*)

- Only known species of allooctoploid (StHNSX) genus
- Most widespread and abundant native sod grass of Northern Great Plains
- Seed dormancy problems
Genetic resources from *Elymus* and *Leymus* have strong potential in western wheatgrass.

- **Streambank wheatgrass**
  - *Elymus lanceolatus* (St H)
  - 7,212 Sanger EST unigenes (1017 bp)
  - 133 polymorphic SSRs

- **Creeping wildye**
  - *Leymus tricoides* (Ns X)
  - 11,281 Santer EST unigenes (1017 bp)
  - 850 polymorphic SSR/STS

- **Psathyrostachys** (Ns)
  - 40,000 Roche EST isotigs (507 bp)

- **Unknown** (X)
  - 40,000 Roche EST isotigs (507 bp)

**Hordeum** (H) **Pseudoroegneria** (St)
- 8,780 Sanger EST unigenes (1017 bp)
- 451 polymorphic SSRs

**Psathyrostachys** (Ns) **Unknown** (X)
- 40,000 Roche EST isotigs (507 bp)

**Pascopyrum** (St H Ns X)
- 850 polymorphic SSR/STS
Genetic markers for seed dormancy in *Leymus* may have application in western wheatgrass.

Creeping wildrye (*Leymus triticoides*)

Basin wildrye (*Leymus cinereus*)
Intermediate and tall wheatgrasses share the E genome of genus *Thinopyrum*

**Thinopyrum**

- Ten diploid, autopolyplploid or allopolyploid species containing at least one E genome (set)
- Intermediate and tall wheatgrasses are cultivated and widely naturalized throughout North America
- Native to Mediterranean region and other parts of Eurasia
Intermediate wheatgrass
(Thinopyryum intermedium)

- Relatively drought tolerant hay and pasture grass, very good single-crop hay
- Many chromosome introgressions in wheat
- Perennial cereal breeding in U.S.
- At least 12 “active” cultivars released in U.S. / Can.
- 252 USDA accessions
Tall wheatgrass
(Thinopyrum ponticum)

- Used for hay, pasture, and conservation
- Very salt tolerant with high biomass potential
- Poor forage quality
- $10n=70$ autopolyploid (genetically recalcitrant)
- Six cultivars released in U.S. and Canada
- 18 USDA accessions
Transcriptome libraries for allohexaploid intermediate wheatgrass and its diploid ancestors

**Pseudoroegneria** (St)
- 8,780 Sanger EST unigenes (1017 bp)
- 451 polymorphic SSRs

**Thinopyrum elongatum** (Ee)
- 448,000 reads (422 bp)
- 28,800 contigs (662 bp)
- 44,400 unmatched (373 bp)
- 73,200 isotigs

**Thinopyrum bessarabicum** (J or E^b^)
- 364,000 reads (426 bp)
- 14,800 contigs (716 bp)
- 23,700 unmatched (348 bp)

**Intermediate wheatgrass**
**Thinopyrum intermedium** (E^e^E^b^St)
- 567,000 reads (432 bp)
- 27,700 contigs (646 bp)
- 55,100 unmatched (384 bp)
- 27,700 isotigs

**Tall wheatgrass**
**Thinopyrum ponticum** (E^e^E^e^E^e^E^e^E^e^)

- 4X
- 6X
- 8X
- 10X
## Development of genetic mapping families for intermediate wheatgrass forage varieties

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<th>High Yield</th>
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<td><strong>PI 401020</strong> (Turkey)</td>
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<td><strong>PI 383561</strong> (Turkey)</td>
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<td><strong>PI 440002</strong> (Karachay-Cherkess)</td>
<td><strong>15 + 21</strong></td>
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Crested wheatgrass

(\textit{Agropyron \textit{spp.}})

- Widely naturalized and recommended forage, withstands heavy grazing
- Two recognized taxa in U.S. (\textit{A. cristatum} and \textit{A. fragile})
- Diploid or autopolyploid forms of \textit{P} genome
- About 10 cultivars released in U.S. and Can.
- More than 700 USDA accessions
Crested wheatgrass (*Agropyron* spp.)

- CDII (Hycrest selection)
- Hycrest (I28 x tetraploid)
- I28 (doubled diploid)
- Nordan (4X)
- Kirk (4X)
- Hexaploid
- rhizomatous Roadcrest (4x)
Summary of genetic resources representing genomic diversity among perennial Triticeae

- **Hordeum (H)**
  - 8,780 Sanger EST unigenes (1017 bp)
  - 451 polymorphic SSRs

- **Pseudoroegneria (St)**
  - 7,212 Sanger EST unigenes
  - 133 polymorphic SSRs

- **Thinopyrum (E)**
  - 111,700 Roche EST isotigs (487 bp)

- **Psathyrostachys (Ns)**
  - 40,000 Roche EST isotigs (507 bp)
  - 850 polymorphic SSR/STS

- **Leymus (Ns X)**
  - 11,281 Santer EST unigenes (1017 bp)

- **Thinopyrum intermedium (EESl)**
  - 82,000 Roche EST isotigs (471 bp)

- **Unknown (X)**
  - 2X

- **Agropyron (P)**
  - 4X

- **Hordeum (H)**
  - 6X

- **Psathyrostachys (Ns)**
  - 8X

- **Pascopyrum (St H Ns X)**