

Molecular evidence for genetic structure and diversity in jointed goatgrass (*Aegilops cylindrica*): what now?

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Abstract:

Polymorphic microsatellites were used to analyze accessions of jointed goatgrass (*Aegilops cylindrica*) in both its native and introduced range. Ninety-six individuals representing 51 counties in 12 western U.S. states and 230 individuals from 28 Eurasian countries were screened using fragment analysis. Results imply that genetic structure exists at a small scale within U.S. states and Eurasian countries rather than between these regions. This suggests the presence of gene flow among U.S. states and Eurasian countries in both ranges and multiple introductions of the species into the U.S. Cluster analysis using UPGMA of dice similarity matrix support the finding of multiple introductions into the U.S. This study is part of a larger project to evaluate the risk of transgenic introgression from drought tolerant wheat to jointed goatgrass. Genetic diversity implies that the risk will vary by genotype due to possible phenotypic diversity and variation in selection responses. Our results will help to clarify the processes driving the evolution of invasive species, while also contributing knowledge towards improving weed management practices.



Jointed goatgrass acting as the pollen donor in a plot containing wheat x jointed goatgrass hybrids (summer 2008).

Materials & Methods:

Ninety-six individuals from 12 western U.S. states were compared to 230 accessions representing the native range of jointed goatgrass in Eurasia.

U.S. accessions were obtained from field collections held by academic institutions. 145 of the Eurasian accessions were obtained from the USDA National Small Grains collections and the remaining 85 accessions were obtained from the Institute of Plant Genetics and Crops Plant Research Gatersleben, Germany. Plants were grown in the greenhouse and tissue for each accession independently collected, frozen, lyophilized, and ground for DNA extraction.

Six microsatellite primer pairs that amplified polymorphic alleles in U.S. jointed goatgrass accessions were chosen from previous screenings: gwm190, gwm301, gwm383 (Roder et al. 1998; Gandhi et al. 2005) and xcf48, xcf72, xcf79 (Guyomarc'h et al. 2002).

Amplified products were separated on an ABI 3130xl sequencer using labeled primers. Fragments produced by all primers were scored as presence/absence data using GeneMapper v 4.0. Cluster analysis was done using NTSys software (Applied Biostatistics Inc.; Rohlf 2000). Genetic distance was determined with the Dice-Coefficient and UPGMA. AMOVA tables were generated using ARLEQUIN (Schneider et al. 2000).

Results:

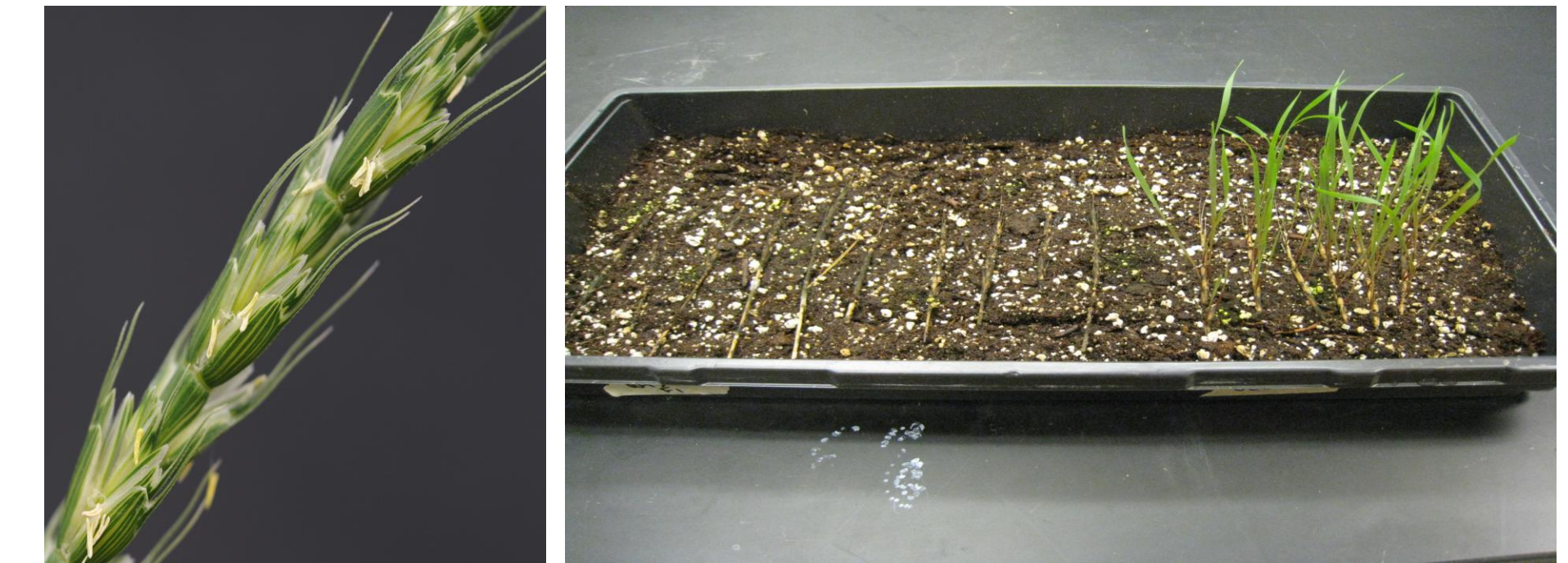
Eighty-eight fragments across six loci were amplified by the six microsatellite primer pairs. AMOVA results identify genetic structure in both the introduced and native range. In the U.S., 71.51% of variance is within states (Table 1). Similarly, in Eurasia, 87.30% of variance is within countries (Table 2).

These results show that the most variance, in both ranges, is within the smallest geographic scale tested in this study. For the introduced range this result suggests that either gene flow or introduction of multiple similar genotypes across the western U.S. has occurred. For Eurasia, the AMOVA reveals significant gene flow among countries. When the accessions were grouped by range, the AMOVA illustrated that 88.42% of the species variance is shared between the native and introduced range, with only 11.58% unique to either range (Table 3). Only three alleles were private to the U.S. indicating that the majority of the unique diversity is located in the native range.

Cluster analysis using UPGMA of Dice similarity matrix placed U.S. accessions into 19 groups interspersed across the Eurasian samples, supporting multiple introductions of jointed goatgrass into the U.S. (Figure 1). U.S. accessions did not cluster together by state, supporting the AMOVA results of variance within states.



U.S. accessions in field for seed increase (summer 2008).



Wheat x jointed goatgrass spike flowering (left). Germination of F_1BC_1 seeds compared to that of jointed goatgrass (right).

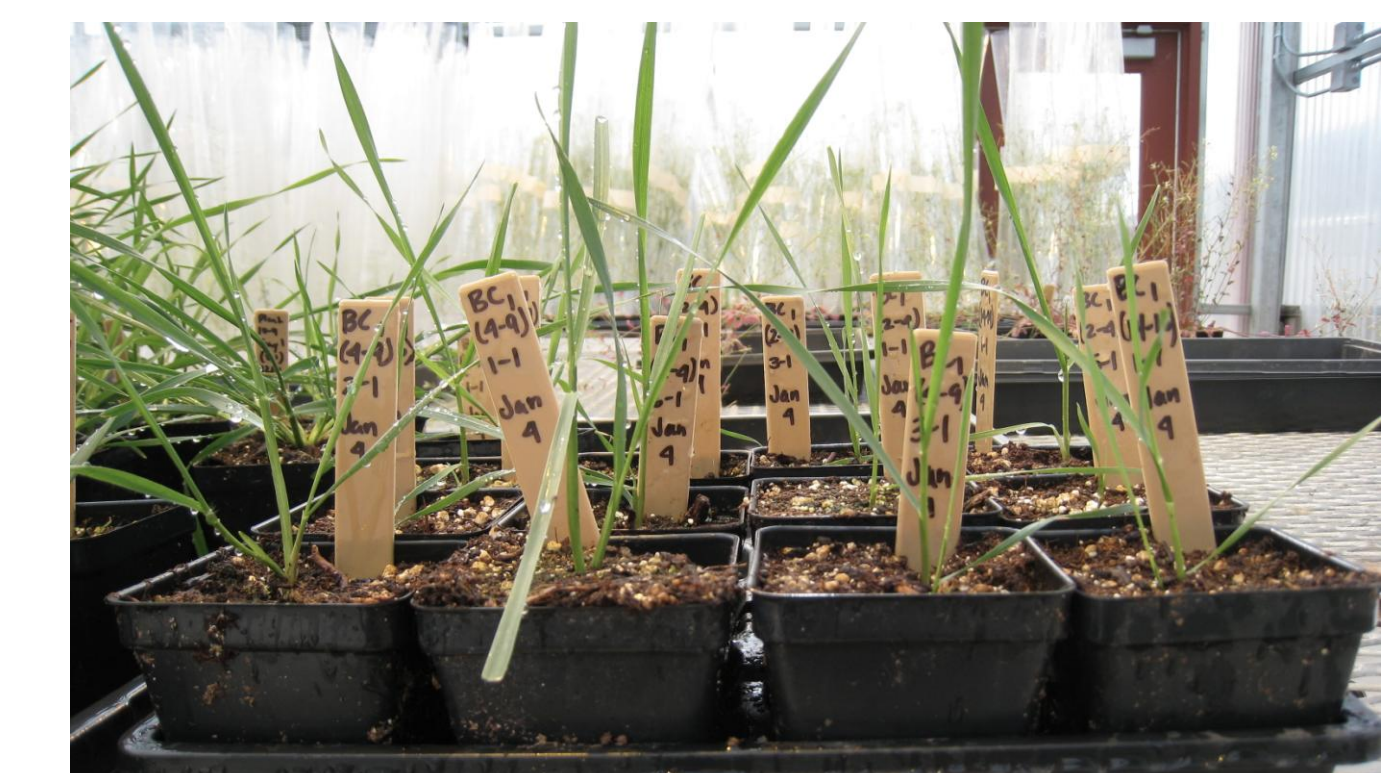
Implications:

Genetic diversity among accessions of jointed goatgrass in the U.S. suggests that genetically determined phenotypic diversity may exist. In the fall of 2008, field experiments began at two locations in Colorado to be evaluated in the summer of 2009. Thirty U.S. accessions representing 12 western states along with two wheat cultivars and wheat x jointed goatgrass hybrids will be evaluated under wet and dry treatments. In addition, the amount of gene flow from different U.S. jointed goatgrass accessions to wheat will be assessed.

The objective of the present and future studies is to evaluate the risk associated with commercialization of transgenic drought tolerant wheat. The results presented here suggest that the risk may vary among genotypes. Phenotypic diversity would imply that the selection response may vary in intensity according to the present level of drought tolerance expressed by each genotype. Germination, cytological, and further genetic studies will be pursued to closely examine this risk.



Newly planted wheat x jointed goatgrass hybrids (fall 2008) for evaluation of backcrossing to jointed goatgrass in the field.



F_1BC_1 plants germinated from spikes collected from field hybrids pollinated by jointed goatgrass (summer 2008).

Analysis of molecular variance:

Table 1: U.S. by state and regions

Population defined	Percentage of variation
Between regions	0.48
Between states	28.00
Within states	71.51

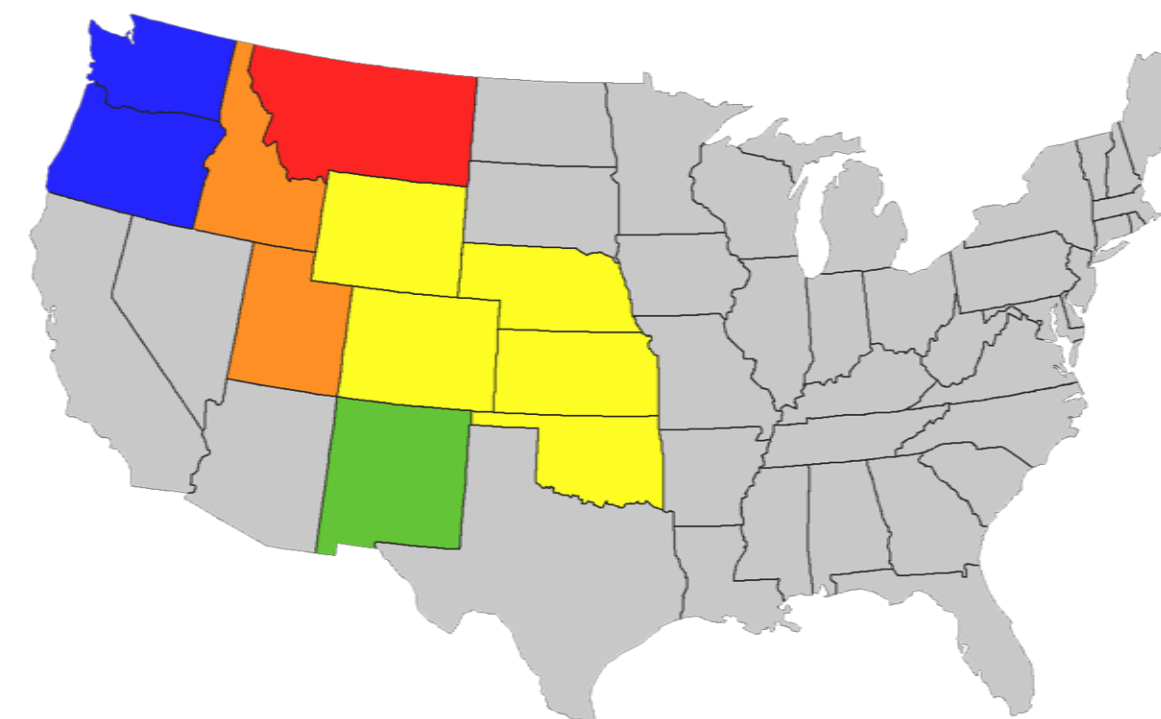


Table 2: Eurasia by country

Population defined	Percentage of variation
Between countries	12.70
Within countries	87.30



Table 3: Introduced versus native range

Population defined	Percentage of variation
Between ranges	11.58
Within ranges	88.42



Figure 1: UPGMA of Dice similarity matrix of Eurasian and U.S. accessions of jointed goatgrass.