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FINAL STUDY REPORT
(Los Lunas Plant Materials Center (LLPMC), New Mexico)

Arriba western wheatgrass (*Pascopyrum smithii (*Rydb.) A. Love) Morphological Evaluation of 8 Germplasm Sources

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ABSTRACT

Western wheatgrass (*Pascopyrum smithii* (Rydb.) A. Love) is an important perennial grass species for agriculture and conservation. The Los Lunas Plant Material Center released a single-origin accession 'Arriba' for its' superior seed production, uniformity, rate of emergence and forage production in 1973. The objective of this study was to conduct a morphological evaluation of 8 different germplasm sources to determine whether there has been a migration of traits from the original release of Arriba western wheatgrass. This study examined differences among germplasm sources of: plant height, tiller length and leaf blade coloration. Seed was obtained from the following companies: Pawnee Butte, Granite Seed, Southwest Seed, along with 4 seed lots from the Upper Colorado Environmental Plant Center (UCEPC). Each of the 8 germplasm sources were propagated in a greenhouse and transplanted into a field of rod rows consisting of 25 individual seedlings per seed source. Tiller length, plant height, and leaf blade coloration data were evaluated for each individual within the 8 rod rows just prior to any intraspecific competition for resources. It was observed that the leaf blade coloration of the individuals within the row and among the rows exhibited great variation. Results indicated there was no difference in plant height among the germplasm sources, however, tiller length among the Arriba seed lots was highly significant ($P < 0.0005$) having a direct effect on forage yield over time. Broad-sense heritability estimates for western wheatgrass are incomplete (Robins et al., 2012) and thereby determination of the relative importance of genetic versus environmental effects cannot be quantified. However, it is known that allele frequencies differ among western wheatgrass populations within species and cycles of selection within populations (Ray and Harms 1994) and DNA variation among single-origin accessions is greater than half-sib families (Larson et al., 2003). From genome analysis, *Pascopyrum smithii* was found to be a cross-pollinating allo-octoploid or a hybridization of 4 different genomes. Hence, populations are extremely heterogeneous composed of highly heterozygous individual genotypes and thereby significant phenotypic variation should be expected owed to ecotypic variation and homology.

INTRODUCTION

Western wheatgrass is a cross-pollinating octoploid (2n=8x=56), cool-season, native, perennial sod-forming grass that is tolerant to moderately saline and alkaline soils. Due to its expansive rhizome development it is ecologically important for soil stabilization and is palatable for all classes of livestock. It has no known allelopathy and no negative impact on wildlife habitat (Waldron et al., 2011). Its large, diverse natural range includes most of the western two-thirds of the United States including dry sagebrush deserts. Western wheatgrass is known to display considerable ecotypic variation (Asay and Jensen, 1996).

Due to its extensive adaptability and utility the USDA from 1957 to 1973 evaluated 40 accessions of western wheatgrass to identify a superior cultivar. Arriba is a single-origin accession collected in Flagler, CO. It was released in 1973 for its superior seed production, uniformity and rate of emergence

and forage production (U.S. Department of Agriculture, 1977). During the 15 year period of evaluation eight field-size seed increase plantings of Arriba were made in which off-type plants were rogued out. No description of "off-type" plants or quantities of individuals rogued from the fields was provided by the study author in 1977. The information provided in the study file suggests the plant breeders evaluated Arriba within a cross-pollination block to bulk up seed quantities. Arriba was not bred to isolate desirable genes utilizing a recurrent selection breeding process that entails reselecting and inter-mating multiple generations to isolate superior inbreds and thereby, Arriba was released with high heterogeneity. Seed from these fields were direct combined and processed on an air-screen cleaner.

This evaluation resulted from observations that plants grown from seed of Arriba, purchased from the UCEPC, had visibly different floral structure and stature than the native western wheatgrass found growing in close proximity to the UCEPC. The objective of this study was to determine if there had been any morphological variation among the Arriba seedlots that would indicate the characteristics of this release had changed through generations of seed production.

MATERIALS AND METHODS

Certified seed of Arriba from 4 seed producers along with seed from the National Plant Germplasm System (NPGS) were shipped to the Los Lunas Plant Materials Center in May 2015: UCEPC: SFD-02-6A, SFD-07-UC4, 2014, SFD-11-Blend, Pawnee Butte:014-7363, Granite Seed: AGSM-NBS-TC4-ARR-1, Southwest Seed:2014 1045 and NPGS: PI 578777. The seed obtained from the NPGS had been provided to the NPGS in 1977 by the New Mexico USDA-SCS following the release of Arriba. The germplasm sources were evaluated alongside seed obtained from the NPGS designated as the standard for determination of variance levels among the seven germplasm sources. All eight seed lots of Arriba were sown into 4-inch plug trays on June 3, 2015 by the PMC staff. All other activities that took place to conduct this study were performed by the manager in order to control any introduced biases. On July 31, 2015 all eight seed sources were transplanted into 164 ml Ray Leach "Cone-tainers" (Stuewe and Sons, Inc., Tangent, OR, USA) leaving one seedling per container for a total of 25 individual seedlings per seed source. On September 16, 2015 all 200 containers were planted in Field 12 in rod rows with a 24-inch inter-row spacing and 38-inches between row spacing. The field was flood irrigated as necessary and weeds kept under control through cultivation. On May 3, 2016 the tiller length of each of the 200 individuals was recorded, and on May 19th measurements of plant height were documented as well. It was observed that among and within the seedlots of Arriba there were obvious differences in coloration of the leaf blades of individuals; some had a bluish tint, others a greenish coloration (Figure 1).

Figure 1. Photo of all 8 seedlots of Arriba depicting differences in leaf blade coloration

RESULTS AND DISCUSSION

Measurements of plant height and tiller length were taken prior to intra- and interspecific competition for available resources (Table 1). Statistical analyses of a one-way ANOVA (SAS Institute Inc., 1999) indicated there was no difference in plant height among the germplasm sources, however, tiller length among the Arriba seed lots was highly significant ($P < 0.0005$).

Table 1. Fisher's least significant difference of means of morphological traits of 8 seed lots of Arriba western wheatgrass, USDA-NRCS Los Lunas, NM 2016.

	Trait	
Seed lot	Height[1/] ---cm---	Tiller length[2/] ---cm---
NPGS:PI 578777	91.31 a[3/]	67.56 a
Southwest Seed: 2014 1045	91.24 a	57.91 ab
Pawnee Butte: 014-7363	90.55 a	56.79 c
Granite Seed: AGSM-NBS-TC4-ARR-1	90.22 a	68.38 a
UCEP: SFD-11-Blend	89.26 a	56.69 c
UCEPC: SFD-02-6A	87.40 a	55.37 c
UCEPC: SFD-07-UC4	83.82 a	52.02 c
UCEPC: 2014	87.71 a	66.04 ab

1/ Height (5/19/16); 2/ Tiller length (5/3/16); 3/ Means in columns followed by the same letters are not significantly different at P<0.05.

Western wheatgrass is an allo-octoploid (2n=8x=56) species comprised of the SSHHNNXX genome combination (Dewey, 1975; Wang and Jensen 1994). Essentially this allopolyploid has multiple sets of chromosomes that are different in type and origin. Each letter represents a different genome. One advantage of western wheatgrass being an allo-octoploid is the resulting heterosis which is characterized by increased vigor and superior qualitative or quantitative traits (Chen, 2010). The two known genera of *Pascopyrum smithii* through DNA sequencing are; *Elymus* and *Pseudoroegneria* (Jones et al., 2000). Their research suggests the genomic designations be revised to StHNsXm due to *Leymus* non-contribution of chloroplast to *Pascopyrum* as originally believed. Allopolyploidy results from concurrent hybridization and mutations in chromosome numbers that differ proportionally to the divergence of the parental genome (Comai, 2005). Through testing of amplified fragment length polymorphism (AFLP) of genotypes of western wheatgrass, DNA variation both between and within accessions was concluded (Larson et al. 2003). This variation likely causes differential gene expression that is measureable and observable as witnessed in this study.

Waldron et. al., (2006) reported a significant population, environment, and a population x environment interaction for mean seed yields of the same western wheatgrass germplasms grown at different locations. This lends support of the hypothesis that plant vigor may be amplified through selective pressures from cultural practices that favor larger individuals throughout the process of seed production, thereby giving rise to a false assumption of misidentifying Arriba within a production nursery. Regardless of whether or not this phenomena exists regarding the expression of vegetation vigor of Arriba western wheatgrass; all germplasm will display heterosis to the extent of water and nutrient availability and length of growing season due to its allopolyploidy.

Until the genetic heritability of Arriba western wheatgrass is quantified, the phenotypic variation of the morphological traits observed in this study could have resulted from any one of the genetic mechanisms of mutation, migration, or selection between ecotypes or by endemic strain genetic variability (ecotypic variation).

CONCLUSION

In summary, Arriba western wheatgrass is a cross-pollinated single-origin accession that was not bred to isolate desirable genes utilizing a recurrent selection breeding process; for that reason this allo-octoploid maintains great heterozygosity. In addition, considerable ecotypic variation exists among western wheatgrass populations for important agronomic traits including forage yield and plant height; consequently uniformity among phenotypes of Arriba should not be expected due to its homology and ecotypic variation. These findings are important because of the ever increasing concern of climate change and the capacity for adaptation. This evaluation suggests the native grass allopolyploids released by the Natural Resource Conservation Service-Plant Materials Program will adapt to changing site conditions.

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