Genetic Diversity of Switchgrass Populations in the Northeast

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INTRODUCTION

•Although a significant amount of genetic diversity exists within switchgrass (*Panicum virgatum*), little research has been conducted on the level of genetic diversity and local adaptation among different populations/ecotypes of switchgrass currently recommended for habitat restoration in the Northeast region of the US.

•Upland ecotypes (Fig 2) are commonly octaploids (2n=8x=72) and occasionally hexaploids (2n=6x=54) and are shorter, finer stemmed and more adapted to drier habitats (Lewandowski et al., 2003).

•Lowland ecotypes (Fig 2) are typically tetraploid (2n=4x=36), and are coarse-stemmed, tall growing and more robust than the upland ecotypes (Lewandowski et al., 2003).

OBJECTIVES

- The objectives of this study were to determine molecular and morphological differences within and between 14 different switchgrass populations.

MATERIALS AND METHODS

Plant Material

- Switchgrass seed from 14 populations (Fig. 1) were obtained from various sources.
- 'Carthage', 'Timber', 'Contract', 'Shelter' and 'High Tide' germplasm sources were obtained from the Natural Resources Conservation Service – USDA Plant Materials Center in Cape May NJ. Carthage, Timber, Contract and High Tide represented Northeast ecotypes.
- Standard cultivars developed in the Midwest and other germplasm sources from other countries included 'Caddo', 'Shawnee', 196 (PI 337553), Pav12, Turkey (PI 204907), 'Sunburst', 'Kanlow', 'Pathfinder', and 'Blackwell', obtained from the Plant Introduction (PI) collection curated by the Germplasm Resources Information Network (GRIN).
- Kanlow and Timber represented lowland ecotypes. All other populations expressed characteristics of upland ecotypes (Fig. 2).
- Seed of each population was germinated in Pro-Mix HP (K.C. Shafer, York, PA) in 12 x 15 inch flats.
- Individual plants were transplanted, grown under greenhouse conditions for approximately 8 weeks, and planted to a spaced-plant nursery in the spring of 2005 at the Rutgers University Plant Biology Research and Extension Farm at Adelphia, NJ (Fig. 2 and 3) for a total of 432 plants.

Figure 1. Switchgrass populations utilized in morphological and molecular marker analysis.

Populations

1) Caddo †
2) Shawnee
3) 196
4) Pav 12 ‡
5) Turkey
6) Sunburst
7) Kanlow
8) Shelter
9) High Tide
10) Pathfinder
11) Contract
12) Blackwell ‡
13) Timber
14) Carthage

† Numbers in table correspond to population numbers in *Structure* bar plots

‡ These populations did not yield enough DNA for molecular marker data analysis with *Structure*

Figure 2. Upland (left) and lowland (right) ecotypes of switchgrass.

Figure 3. *Panicum virgatum* 'Carthage'.

Morphological Markers

- Morphological measurements were taken on 12 individuals from each of the 14 switchgrass populations listed in Fig. 1 in 2005 and 2006.
- Measurements included plant height, panicle height, and flag leaf height, length and width.
- Measurements were taken approximately 1-2 weeks after anthesis.

Molecular Markers

- Leaf tissue was collected from 12 individuals from each population listed in Fig 1 for molecular marker analysis. DNA was isolated from leaf tissue using the Sigma® GenElute™ Plant Genomic DNA Miniprep kit (Sigma-Aldrich Co., St. Louis, MO).
- Publicly available switchgrass specific microsatellite (SSR) markers were utilized for the molecular marker analysis (Tobias et al, 2006).
- Thirty-two SSR primer pairs were tested. SSR markers were genotyped on all individuals using an ABI 3130 genetic analyzer. Fifteen primer pairs amplified polymorphic bands in our populations and these were used for molecular marker analysis.

Analysis

- Morphological and marker data was analyzed using the program *Structure* (Pritchard et al., 2000) which identifies clusters of related individuals from multilocus genotypes. The full data set was analyzed for all models from K=1 through 14.

RESULTS AND DISCUSSION

•Significant morphological (Fig 2) and molecular differences between switchgrass populations were observed.

2005 Morphological Data

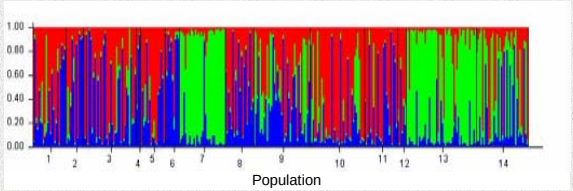
•*Structure* analysis of 2005 morphological data separated the populations into distinct groups (Fig 4). Kanlow'(7) and Timber'(13) grouped together based on morphological measurements. These two populations also looked phenotypically similar and represented the lowland ecotypes.

•Pathfinder(10), Contract(11), and Blackwell(12) were grouped together by morphological data.

•Morphological measurements also clustered Caddo(1), Shawnee(2), 196(3), Pav12(4), Turkey(5), and Shelter(8) .

•Morphological analysis in 2005 provided some delineation between upland and lowland ecotypes, but did not distinguish between Northeast and Midwest populations.

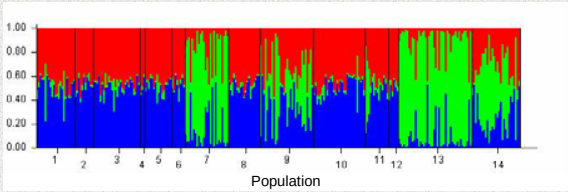
Figure 4. *Structure* bar plot at K=3 for 2005 morphological data. Populations are listed in Fig 1.



2006 Morphological Data

- Similar results were observed for *Structure* analysis of 2006 morphological data compared to 2005 morphological data.
- Kanlow(7) and Timber(13) again formed a distinct group based on morphology.
- Carthage(14) and High Tide(9) formed another cluster (Fig 5) in 2006.
- Upland and lowland ecotypes were grouped based on morphological data, but Northeast and Midwest populations were not completely distinguished using morphological data even though populations looked distinct.

Figure 5. *Structure* bar plot at K=3 for 2006 morphological data. Populations are listed in Fig 1.



•Molecular Marker Data

•*Structure* analysis of molecular marker data from 15 primer pairs also divided the populations into distinct groups (Fig 6).

•Kanlow(7), Timber(13), and Contract(11) composed one group, while Pathfinder(10) and Carthage(14) comprised a second cluster.

•Shawnee(2), 196(3), and Turkey(5) grouped together based on molecular marker data. Shelter(8) and High Tide(9) also formed a group.

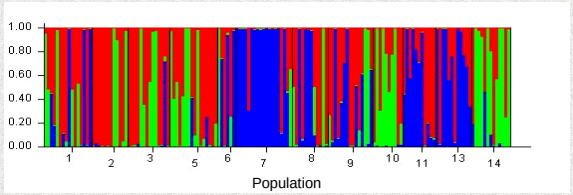
•Molecular marker analysis did distinguish Kanlow and Timber (lowland ecotypes), but Contract (upland ecotype) was also included in that grouping. This indicated that the analysis did differentiate between upland and lowland but it was not complete.

•The molecular marker analysis did not clearly delineate Midwest and Northeast populations.

•Molecular and morphological marker analysis did not produce similar results.

•Continued work with molecular markers is needed to further differentiate between switchgrass populations.

Figure 6. *Structure* bar plot at K=3 for molecular marker data. Populations are listed in Fig 1.



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