Inheritance of leaf variegations in *Miscanthus*

Darren Touchell, Jeremy Smith and Thomas Ranney

NC State University, Dept. of Hort. Science, Mountain Horticultural Crops Research and Extension Center, 455 Research Drive, Fletcher, NC 28732-9244

darren_touchell@ncsu.edu

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Significance to Industry: Miscanthus is a grass native to Asia and has become a popular ornamental plant in the United States. It is a valuable nursery crop and also has significant potential as a bio-fuel crop. In some areas of the United States, and particularly in Western North Carolina, *Miscanthus* has naturalized along roads side verges and disturbed areas raising environmental concerns.

Miscanthus contains 14 species of which *M. sinensis* is most common in the nursery industry. Currently, there are over 100 named varieties of *M. sinesis* (1), which are noted for their variegations, different foliage characteristics, compactness and inflorescence color. Vertical and horizontal leaf variegations are the most valued characteristics. Understanding the genetics and heritability of these desirable characteristics will help in developing strategies to simultaneously breed for ornamental traits and non-invasiveness.

Nature of Work: Leaf variegations arise from mutations that make chloroplast formation unstable. While these mutations are most often found in the chloroplast genome they can also occur in the nuclear genome (3). Mutations in the nuclear genome can be distinguished from those in the chloroplast genome by the inheritance pattern. Specifically, chloroplast mutations are typically inherited maternally, while mutations in the nuclear genome show Mendelian inheritance (2). In the present study we report on the mode of inheritance for horizontal and vertical leaf variegations in *Miscanthus sinensis*.

M. sinensis ‘Strictus’ (ST) and *M. sinensis* ‘Variegatus’ (VAR) were selected as two cultivars representing horizontal and vertical leaf variegations, respectively. Reciprocal di-hybrid F_1 and F_2 populations were produced to study inheritance of the two variegations. Backcrosses (BC) population to each parent were also produced. Further backcrosses with *M. sinensis* ‘Zebrinus’(ZEB), *M. sinensis* ‘Little Zebra’ (LZ) and *M. sinensis* ‘Superstripe’ (SS) were conducted to determine if horizontal variegations in these cultivars was attributed to the same gene. Similarly, backcrosses with *M. sinensis* ‘Silberfeil’ (SILB) were conducted to determine if vertical variegations could be attributed to the same gene. All crosses were conducted during the summer of 2006. Hybridizations were performed by placing the two flower heads in a paper bag and shaking for 30 s. Seed was collected after 30 d and germinated on moist filter paper. Germinated seed was transferred to pinebark substrate supplemented with 0.75 kg/m^3 micronutrients (Micromax) and 1.5 kg/m^3 lime. Phenotypes were scored during the spring/summer of 2007. It was hypothesized that horizontal and vertical variegations were independent traits and inherited in a Mendelian recessive manner. Chi-square analysis was used to analyze departures from expected rations on segregating families.

Results and Discussion: Reciprocal F_1 crosses yielded progeny that were all non-variegated and green, with the exception of 3 horizontally variegated plants and 1 vertically variegated plant, which were attributed to infrequent self-pollination (Table 1). This suggests that both horizontal and vertical variegations were not maternally inherited characteristics arising from mutations in the chloroplast genome.

In the F_2 progeny, a lower than expected number of horizontally variegated plants (19 observed vs. 31.3 expected) and combined horizontally and vertically variegated plants (3 observed vs. 10.4 expected) were observed, therefore having a poor fit to the expected 9:3:3:1 ratio. The chi-square test for independence of linkage ($\chi^2 = 1.24$, $P = 0.2$) suggested no evidence of linkage between the horizontal and vertical variegations. However, segregation ratios for BC populations strongly fit the expected 1:1 ratio for variegated:non-variegated phenotypes for both horizontal and vertical variegations (Table 1). Further, phenotypic expression was erratic and seemed to be influenced by seasonal and/or other factors not considered in this study. The lack of uniformity in expression may have influenced data collection and it is possible that further variegations will be expressed as plants mature. The combination of these data and observations suggest that simple recessive inheritance is currently the most likely model for both horizontal and vertical leaf variegations in *M. sinensis*.

Progeny from crosses with other horizontally and vertically variegated cultivars provide good fits to a 1 variegated: 1 green ratio, strongly suggesting that the genes responsible for variegations remain the same across cultivars. However, the intensity of variegations often varied within each cross. Therefore it is likely there are other genes that interact with the phenotypic expression of variegations.

Horizontal and vertical variegations in *Miscanthus sinensis* are likely to be inherited in a simple recessive Mendelian manner. However, the expression of variegations (e.g., phenology, intensity, etc.) appears to by modified by other factors. Further studies are currently being conducted to develop variegated, non-invasive ornamental cultivars.

Literature Cited:

1. Darke, R. 2007. The encyclopedia of grasses for living landscapes. Timber Press.Portland.
2. Fawole, I. 2001. Maternal inheritance of plant variegation in cowpea, *Vigna unguiculata* (L.) Walp. Theoretical and Applied Genetics. 102: 459-462.
3. Sakamoto, W. 2003. Leaf-variegated mutations and their responsible genes in *Arabidopsis thaliana*. Genes and Genetic Systems. 78: 1-9.

Table 1. Segregation analysis for variegated foliage traits in *Miscanthus sinensis*

		Progeny (No. of plants)						
Cross[Z]	Population	Green	Horizontal	Vertical	H+V	Expected Ratio	χ^2	*P*
ST x VAR	F_1	75	3	0	0	1:0:0:0	_	_
VAR x ST	F_1	70	0	1	0	1:0:0:0	_	_
F_1 x F_1	F_2	111	19	33	3	9:3:3:1	14.57	.05
ST x F_1	BC_H	16	14	0	0	1:1:0:0	0.17	0.5
VAR x F_1	BC_V	33	0	39	0	1:0:1:0	0.51	0.5
ZEB x F_1	BC_{ZEB}	21	20	0	0	1:1:0:0	0.05	0.5
LZ x F_1	BC_{LZ}	12	15	0	0	1:1:0:0	0.37	0.5
SS x F_1	BC_{SS}	30	26	0	0	1:1:0:0	0.3	0.5
SILB x F_1	BC_{SILB}	32	0	34	0	1:0:1:0	0.08	0.5

[Z]ST = 'Strictus', VAR = 'Varigatus', ZEV = 'Zebrinus', LZ = 'Little Zebra', SS = 'Superstripe', SILB = 'Silberfiel'