

NATIONAL TURFGRASS EVALUATION PROGRAM (NTEP)

ANCILLARY TRIAL – KENTUCKY BLUEGRASS DNA GENOTYPING

Report on NTEP Kentucky Bluegrass DNA Genotyping

Rutgers University - Josh Honig Ph.D., Stacy Bonos Ph.D., and William Meyer Ph.D.

Background

In Fall 2011, the NTEP Policy Committee decided to fund an ancillary study that uses new DNA genotyping techniques, developed by Rutgers University researchers, to classify Kentucky bluegrass entries. Therefore, each entry sponsored and submitted to the 2011 National Kentucky Bluegrass Test was charged an additional fee to conduct this analysis by Rutgers University. The results of this analysis were completed in spring 2015 and are therefore, published for the first time here (see the following sheets). The Rutgers University team of Dr. Josh Honig, Dr. Stacy Bonos and Dr. William Meyer will prepare and publish, in a refereed scientific journal, more detailed information on the procedures conducted and data collected. The following is a summary of the results from the NTEP entries only.

Procedures

One hundred fifty-six (156) cultivars and accessions of Kentucky bluegrass were seeded into in the turf greenhouse facility in New Brunswick, NJ. The 156 entries included the following: all 82 entries of the 2011 NTEP Kentucky Bluegrass test, 74 additional Kentucky bluegrass cultivars/accessions, and one natural *Poa annua* population collected at Rutgers Horticultural Research Farm II (North Brunswick, NJ). These entries were field planted in spring 2012.

In June 2013, all individual entries were visually inspected for apomixis in the field nursery during the flowering period of Kentucky bluegrass. Of the 156 original Kentucky bluegrass cultivars/accessions, 13 Kentucky bluegrass cultivars/accessions (12 from the added cultivars/accessions, plus the standard cultivar America) exhibited less than 50% apomixis in the field nursery. This low level of apomixis was assumed to be contamination in the affected entries, resulting in all 13 cultivars/accessions being removed from further analysis. The loss of the standard cultivar America was compensated for by including a number of America-type cultivars in the added group of cultivars/accessions.

Due to the findings of Bushman et al. (2013), DNA from eight (8) plants of each entry was collected to represent a cultivar/accession. Thirty-five (35) microsatellite markers from Honig et al. (2010) were used to DNA genotype eight plants from each of the 144 bluegrass cultivars, experimental selections, and accessions (1152 total samples).

Results and Summary

Some departures from the previously defined (Honig et al., 2012) bluegrass classification types include Julia cultivars/accessions being split into two groups in the current analysis, as well as a number of European cultivars/accessions/collections (mixed with some North American cultivars/accessions/collections) being interspersed between some of the traditional classification types.

The current microsatellite analysis of the 2011 NTEP Kentucky Bluegrass Test shows that the vast majority of all cultivars, experimental selections, and collections were uniquely identified (genetically differentiated) with the current set of microsatellite markers. Strong genetic support was found for the Midnight, Cynthia, Julia, BVMG, Limousine, Mid-Atlantic, America, Shamrock, P-105, and Compact bluegrass classification types. Additional entries or analyses may be needed to clarify the genetic relationships of the split Julia classification type and the remaining unclassified (predominantly European) entries.

Literature Cited

- Bushman, B. S., et al. 2013. Molecular markers highlight variation within and among Kentucky bluegrass varieties and accessions. *Crop Sci.* 53(5): 2245-2254.
- Honig, J. A., et al. 2010. Isolation and characterization of 88 polymorphic microsatellite Markers in Kentucky bluegrass (*Poa pratensis* L.). *Hort Sci.* 45(11): 1759-1763.
- Honig, J. A., et al. 2012. Classification of Kentucky bluegrass cultivars and accessions based on microsatellite markers. *Hort Sci.* 47(9): 1356-1366.

2011 NTEP KENTUCKY BLUEGRASS TEST - DNA GENOTYPING RESULTS FROM RUTGERS UNIVERSITY

Entry No.	Entry Name	Classification/Type	Entry No.	Entry Name	Classification/Type
1	BAR 12PP 612	Eurasian	42	Avid	P-105
2	BAR 8PP 504	Eurasian	43	Empire	America
3	America	Unreliable seed stock	44	Burl 3-51	Eurasian
4	A05-361	Mid-Atlantic	45	Burl 06-11	America
5	Cabernet	Mid-Atlantic	46	PST-K4-3	Eurasian
6	Award	Midnight	47	PST-K9-90	Eurasian
7	NuChicago	Midnight	48	PST-K10-106D	Julia
8	Kenblue	Eurasian	49	BAR PP 119327	Eurasian
9	J1770	Midnight	50	Barduke	BMVG
10	J-1136	Midnight	51	Baron	BMVG
11	Rush	Limousine	52	Barvette HGT	Eurasian
12	Sudden Impact	Midnight	53	BAR VV 112916	Eurasian
13	J-1853	Midnight	54	BAR PP 110358	Eurasian
14	Waterworks	America	55	BAR PP 119326	Eurasian
15	A05-329	America	56	BAR VV 118532	Eurasian
16	Oasis	America	57	Skye	Midnight
17	A98-363	America	58	Blue Coat	Eurasian
18	RAD-1492	Eurasian	59	AKB 2555	Shamrock
19	A04-38	Shamrock	60	A00-4199	Eurasian
20	A05-360	Mid-Atlantic	61	A06-26	America
21	PPH 9131	Eurasian	62	PICK TD8	America
22	DPPP 818	BMVG	63	SRX 5321	Shamrock
23	PP 10847	Limousine	64	PICK TD9	Eurasian
24	PICK 033	Mid-Atlantic	65	Blackjack	Eurasian
25	PICK MP07	Eurasian	66	Arrowhead	Shamrock
26	PICK 4340	America	67	PST-K9-99	Eurasian
27	SRX 446	Mid-Atlantic	68	Lunar	Eurasian
28	SRX 2758	Mid-Atlantic	69	PST-T10-18	Eurasian
29	SRX 4338	America	70	Endurance	Eurasian
30	LTP-08-6	America	71	PST-K9-97	Eurasian
31	A00-2882	Compact	72	Rubix	Eurasian
32	Keeneland	America	73	3733	Eurasian
33	A06-46	Eurasian	74	Dauntless	Shamrock
34	A05-TB-382	Mid-Atlantic	75	A05-999	Eurasian
35	H99-1653	Eurasian	76	A06-47	Eurasian
36	Bluenote	America	77	RAD-507	America
37	Legend	America	78	A04-36	Shamrock
38	Zinger	Shamrock	79	Shamrock	Shamrock
39	A03-1017	Midnight	80	A01-1106	Compact
40	A04-74	America	81	Midnight	Midnight
41	RAD-849	Eurasian	82	Thermal Blue	Eurasian

SORTED BY ENTRY NUMBER

2011 NTEP KENTUCKY BLUEGRASS TEST - DNA GENOTYPING RESULTS FROM RUTGERS UNIVERSITY

Entry No.	Entry Name	Classification/Type	Entry No.	Entry Name	Classification/Type
14	Waterworks	America	1	BAR 12PP 612	Eurasian
15	A05-329	America	2	BAR 8PP 504	Eurasian
16	Oasis	America	8	Kenblue	Eurasian
17	A98-363	America	18	RAD-1492	Eurasian
26	PICK 4340	America	21	PPH 9131	Eurasian
29	SRX 4338	America	25	PICK MP07	Eurasian
30	LTP-08-6	America	33	A06-46	Eurasian
32	Keeneland	America	35	H99-1653	Eurasian
36	Bluenote	America	41	RAD-849	Eurasian
37	Legend	America	44	Burl 3-51	Eurasian
40	A04-74	America	46	PST-K4-3	Eurasian
43	Empire	America	47	PST-K9-90	Eurasian
45	Burl 06-11	America	49	BAR PP 119327	Eurasian
61	A06-26	America	52	Barvette HGT	Eurasian
62	PICK TD8	America	53	BAR VV 112916	Eurasian
77	RAD-507	America	54	BAR PP 110358	Eurasian
			55	BAR PP 119326	Eurasian
22	DPPP 818	BMVG	56	BAR VV 118532	Eurasian
50	Barduke	BMVG	58	Blue Coat	Eurasian
51	Baron	BMVG	60	A00-4199	Eurasian
			64	PICK TD9	Eurasian
31	A00-2882	Compact	65	Blackjack	Eurasian
80	A01-1106	Compact	67	PST-K9-99	Eurasian
			68	Lunar	Eurasian
11	Rush	Limousine	69	PST-T10-18	Eurasian
23	PP 10847	Limousine	70	Endurance	Eurasian
			71	PST-K9-97	Eurasian
4	A05-361	Mid-Atlantic	72	Rubix	Eurasian
5	Cabernet	Mid-Atlantic	73	3733	Eurasian
20	A05-360	Mid-Atlantic	75	A05-999	Eurasian
24	PICK 033	Mid-Atlantic	76	A06-47	Eurasian
27	SRX 446	Mid-Atlantic	82	Thermal Blue	Eurasian
28	SRX 2758	Mid-Atlantic			
34	A05-TB-382	Mid-Atlantic	48	PST-K10-106D	Julia
6	Award	Midnight	19	A04-38	Shamrock
7	NuChicago	Midnight	38	Zinger	Shamrock
9	J1770	Midnight	59	AKB 2555	Shamrock
10	J-1136	Midnight	63	SRX 5321	Shamrock
12	Sudden Impact	Midnight	66	Arrowhead	Shamrock
13	J-1853	Midnight	74	Dauntless	Shamrock
39	A03-1017	Midnight	78	A04-36	Shamrock
57	Skye	Midnight	79	Shamrock	Shamrock
81	Midnight	Midnight			
			3	America	Unreliable seed stock
42	Avid	P-105			

SORTED BY CLASSIFICATION/TYPE