# Rule Change Proposal No. 6

### **PURPOSE OF PROPOSAL**

Specify a pre-germination tetrazolium test as the method to determine the dormancy of the seed for *Andropogon gerardii* (Big Bluestem) in Table 3.

### PRESENT RULE

			First	Final	
		Temp.	Count	Count	
Kind of Seed	Substrata	°C	Days	Days	Additional Directions
Andropogon gerardii	P,TS	20-30	7	14	Light; Ungerminated seeds: see
big bluestem					sec. 4.2e and 4.9k.

### PROPOSED RULE

			First		
		Temp.	Count	Count	
Kind of Seed	Substrata	$^{\circ}\mathbf{C}$	Days	Days	Additional Directions
Andropogon gerardii	P,TS	20-30	7	14	Light (see Sec. 4.8s). Dormant:
big bluestem					Pre-germination TZ on
					200 seeds (see Sec. 4.8s)

## 4.7 Calculation of Percentage Germination

e. For *Andropogon gerardii* (Big Bluestem) report results of 400 seed germination as percent germination. If the percent viable seed from the 200 seed tetrazolium (TZ) test is greater than the 400 seed germination percentage, subtract the germination result from TZ result and report the difference as percent dormant seed.

4.8 s. **Big Bluestem** (*Andropogon gerardii*). - Two test methods as prescribed in Table 3 shall be used on each sample. Conduct a 200 seed pre-germination tetrazolium (TZ) test as prescribed in 4.9 k. (2c), record result as percent viable seed. Place 400 seed on blotters moistened with water and germinate for 14 days at 20-30° C in light, record result as percent germination. Refer to Sec. 4.7e for calculation and reporting results.

### **HARMONIZATION**

The International Seed Testing Association and Federal Seed Act (FSA) methods are not in harmony with each other and are not in harmony with the present AOSA method or this proposed method. The prechill and KNO<sub>3</sub> requirements listed in the FSA were removed from the AOSA Rules in October of 2002. The ISTA method does specify prechill, KNO<sub>3</sub> and atest duration of 28 days. Canadian Method and Procedures do not specify methods for this species.

### SUPPORTING EVIDENCE

Big Bluestem is a native indeterminate flowering warm-season grass which produces varying fruit sizes (seed units), some of which are commonly dormant at harvest. This dormancy

dissipates over one to three years, resulting in slow establishment of field plantings (Coukos 1944 and Byers 1973). Warm-season grasses are normally planted into warm soil conditions (20-25°C), compared to cool-season grasses, which can be planted in fall (dormant) or spring seedings. Most warm-season grasses are sold on a Pure Live Seed (PLS) basis, which includes germinated and dormant seeds. Therefore, breaking dormancy in the laboratory is not important in the pricing of these seeds, nor does it necessarily represent the expected field emergence potential of the seed lot. Tetrazolium is widely used for checking the viability of Big Bluestem seed lots and also is commonly used to check the viability of ungerminated seeds. Listing tetrazolium staining as the method for determining dormancy of Big Bluestem promotes testing standardization by reducing variation caused by allowing several viability determination options.

The proposed change is supported by the Native Seed Working Group's study of nine seed lots during the Summer/Fall of 2002. The Native Seed Working Group is composed of 14 different laboratories involved in testing native seeds; seven of these laboratories expressed interested in conducting these native grass studies. Three laboratories received seed from nine seed lots, the experimental design and data forms; one laboratory returned results (Mid-West Seed Service, Inc.). Testing was conducted using four true replicates of 100 seeds grouped into four separate blocks, each block containing only one of each seed lot and treatment combinations. Responses of nine Big Bluestem seed lots were evaluated across three test methods: 1) 400 seed TZ plus a 400 seed 14 day germination (400 pre-germ TZ), 2) 200 seed TZ plus a 400 seed 14 day germination (200 pre-germ TZ), and 3) 400 seed 14 day germination with TZ on remaining ungerminated seed (post-germ TZ). Data was collected, submitted and statistically analyzed by Amanda Patin, Mid-West Seed Services, Inc.

One laboratory returned results in this study with respective germination test responses for 36 observations are presented in Table 1. Viability of the nine seed lots ranged between 50-96%, 45-96% and 48-86% for the 400 pre-germ TZ, 200 pre-germ TZ and post-germ TZ, respectively. The Big Bluestem seed lots are representative of what laboratories could expect to see submitted to their laboratories.

**Table 1**. Total mean viable response from nine seed lots for 400 pre-germ TZ, 200 pre-germ TZ and post-germ TZ. N=36.

Mean Percentages Viable Seed						
Seed lot	400 pre-germ TZ	200 pre-germ TZ	Post-germ TZ			
1	81	83	59			
2	74	77	60			
3	76	79	63			
4	96	96	86			
5	83	94	72			
6	58	45	48			
7	63	71	68			
8	50	48	51			
9	76	78	58			
LSD	9.032	7.724	10.90			

Data in Table 2 demonstrates the comparability of the three methods averaged across the nine seed lots. The total viability for 400 pre-germ TZ and 200 pre-germ TZ are not statistically different; however the total viability obtained from the post-germ TZ is 11 and 12 % lower than the 400 pre-germ TZ and 200 pre-germ TZ, respectively.

**Table 2**. Mean germination, dormant and total viable seed averaged across nine seed lots for three methods. N=36.

	<u>Mean Percentages</u>					
Method	7d germ	14d germ	Dormant	Total Viable		
400 pre-germ TZ	35	55	18	73		
200 pre-germ TZ	35	55	19	74		
Post-germ TZ	36	53	9	62		
LSD	2.294	3.032	3.457	2.658		

### Literature Cited

Byer, K.L. 1973. Evaluation of methods of reducing seed dormancy in switchgrass, indiangrass, and big bluestem. Thesis South Dakota State University. 40p.

Coukos, D.J. 1944. Seed dormancy and germination in some native grasses. J. Amer. Soc. Agron. 36:337-345.

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