

Rule Change Proposal No. 9

PURPOSE OF PROPOSAL

Specify a pre-germination tetrazolium test as the method to determine the dormancy of the seed for *Sorghastrum nutans* (Indiangrass) in Table 3.

PRESENT RULE

<u>Kind of Seed</u>	<u>Substrata</u>	<u>Temperature °C</u>	<u>First Count Days</u>	<u>Final Count Days</u>	<u>Additional Directions</u>
<i>Sorghastrum nutans</i> indiangrass	P,TS	20-30	7	14	Light; Ungerminated seeds: see sec. 4.2e and 4.9k.

PROPOSED RULE

<u>Kind of Seed</u>	<u>Substrata</u>	<u>Temperature °C</u>	<u>First Count Days</u>	<u>Final Count Days</u>	<u>Additional Directions</u>
<i>Sorghastrum nutans</i> indiangrass	P,TS	20-30	7	14	Light (see Sec. 4.8s). Dormant: Pre – germination TZ on 200 seeds (see Sec. 4.8s).

4.7 Calculation of Percentage Germination

e. For *Sorghastrum nutans* (Indiangrass) 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. **Indiangrass** (*Sorghastrum nutans*). - 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.

Note: if proposals 6, 7, and 8 are adopted all four species will appear in sections 4.7e and 4.8s.

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 KNO₃ requirements listed in the FSA were removed from the AOSA Rules in October of 2002. The ISTA method does specify prechill, KNO₃ and test duration of 28 days. Canadian Method and Procedures do not specify methods for this species.

SUPPORTING EVIDENCE

Indiangrass 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 Indiangrass seed lots and also is commonly used to check the viability of ungerminated seeds. Listing tetrazolium staining as the method for determining dormancy of Indiangrass 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. Four laboratories agreed to participate on this species and received seed from nine seed lots, the experimental design and data forms; two laboratories returned data (Nebraska Crop Improvement Association and 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 Indiangrass 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. Two laboratories returned results in this study and their respective germination test responses for 72 observations are presented in Table 1. Viability of the nine seed lots ranged between 67-91%, 69-89% and 62-88% for the 400 pre-germ TZ, 200 pre-germ TZ and post-germ TZ, respectively. These Indiangrass seed lots are representative of what laboratories could expect to see submitted to their laboratories.

Table 1. Total mean viability response from nine seed lots for 400 pre-germ TZ, 200 pre-germ TZ and post-germ TZ. N=72.

Seed lot	Mean Percentage Viable		
	400 pre-germ TZ	200 pre-germ TZ	Post- germ TZ
1	67	69	62
2	74	70	66
3	87	88	78
4	89	85	84
5	91	89	88
6	88	88	80
7	82	86	72
8	75	83	86
9	79	78	71
LSD	3.996	5.256	5.369

Data in Table 2 shows the comparability of the three methods averaged across the nine seed lots and two laboratories. 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 5 and 6 % lower than the 400 pre-germ TZ and 200 pre-germ TZ, respectively.

Table 2. Percent mean germination, dormant and total viable seed for three methods averaged across nine seed lots and two laboratories. N=72.

Method	Mean Percentages			
	7 d germ	14 d germ	Dormant	Total Viable
400 pre-germ TZ	43	53	28	81
200 pre-germ TZ	43	53	29	82
Post-germ TZ	43	53	23	76
LSD	1.820	1.837	2.125	1.405

In Table 3 the results of both laboratories are comparable with the 400 pre-germ TZ and 200 pre-germ TZ methods; while the post-germ TZ laboratory results were significantly different between the two laboratories.

Table 3. Total mean viable response for two laboratories averaged across nine seed lots for 400 pre-germ TZ, 200 pre-germ TZ and post-germ TZ. N=72.

Lab	Mean Percentage Viable Seed		
	400 pre-germ TZ	200 pre-germ TZ	Post-germ TZ
1	81	82	74
2	82	82	79
LSD	1.884	2.477	2.531

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.

SUBMITTED BY: Mid-West Seed Services, Inc. Please submit questions to: Tim Gutormson, RST, Mid-West Seed Services, Inc., 236 32nd Ave., Brookings, SD, 57006, ph 605-692-7611, timg@mwseed.com.

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