2025 AOSA Rule Proposal #8

Anthocyanin Rule Proposal-Cereals

PURPOSE OF RULE PROPOSAL: The purpose of this proposal is to add a note, under Poaceae Grass Family I-Cereals in AOSA Rules for Testing Seeds Volume 4, clarifying the anthocyanin color that can be present in *Secale cereale* and *x Triticosecale* seedlings.

PRESENT RULE:

ABNORMAL SEEDLING DESCRIPTIONS

Shoot:

. (see also notes 1 and 2)

Seedling:

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. seedlings with badly thickened and shortened roots and shoots due to injury from chemical treatment (see note 3).

NOTES

1. Seedlings grown in the dark or in low intensity light will exhibit increased elongation of the coleoptile and in some cases the mesocotyl. In towels, there may be considerable twisting of the shoot.

2. Splitting of the coleoptile tip occurs naturally as a result of expansion of the leaves inside and occurs after emergence and after the coleoptile ceases to elongate upon exposure to light.

3. Seedlings with badly thickened and shortened roots and shoots due to injury from chemical treatment are to be classified as abnormal. If such seedlings are difficult to evaluate on paper substrata, the interpretation should be based on the seedling performance in sand, or soil, or organic growing media.

PROPOSED RULE:

ABNORMAL SEEDLING DESCRIPTIONS

Shoot:

. (see also notes 1, 2, and 3)

Seedling:

.

. seedlings with badly thickened and shortened roots and shoots due to injury from chemical treatment (see note 4).

NOTES

1. Seedlings grown in the dark or in low intensity light will exhibit increased elongation of the coleoptile and in some cases the mesocotyl. In towels, there may be considerable twisting of the shoot.

2. Splitting of the coleoptile tip occurs naturally as a result of expansion of the leaves inside and occurs after emergence and after the coleoptile ceases to elongate upon exposure to light.

3. In *Secale cereale* and *xTriticosecale*, there may be a reddish to purplish color present in the coleoptile or leaves of the seedlings. Less frequently, this color can also be observed in other cereals. This color is due to the presence of anthocyanins and is to be evaluated as normal.

4. Seedlings with badly thickened and shortened roots and shoots due to injury from chemical treatment are to be classified as abnormal. If such seedlings are difficult to evaluate on paper substrata, the interpretation should be based on the seedling performance in sand, or soil, or organic growing media.

HARMONIZATION AND IMPACT STATEMENT:

The Federal Seed Act, Canada M&P, and ISTA Rules do not include notes on evaluating anthocyanins in seedling tissues.

SUPPORTING EVIDENCE:

While reviewing the results of the first Seedling Evaluation Survey (Poaceae, Grass Family-Cereals), it was evident there was confusion among analysts how to handle seedlings that had anthocyanins present. A significant number of analysts wrongly classified seedlings with anthocyanins as abnormal. Anthocyanins are water soluble pigments found in different types of plant tissues and can range in color from blue, purple, to red. In many grass species, seed coloration is partially due to anthocyanin pigmentation in the aleurone or pericarp. Under field conditions, anthocyanin development is usually the result of a combination of low temperature stress and high light intensity, producing the 'purpling' effect observed in turfgrasses leaves. In germination tests, first leaves of grasses, especially the tips, as well as coleoptiles, sometimes exhibit purplish-red coloration indicative of anthocyanin presence. As seedling development progresses, this purplish-red color disappears upon exposure to light and increased chlorophyl production. Among the cereals, anthocyanin development is most noticeable in first leaves and coleoptiles of *Secale cereale* and *x Triticosecale* seedlings.

Petrella, D.P., J.D. Metzger, J.J. Blakeslee, E.J. Nangle, and D.S. Gardner. 2016. Anthocyanin production using rough bluegrass treated with high intensity light. HortScience. 51(9) 1111-1120. doi:10.21273/HORTSCI10878-16.

Zykin, P.A., E.A. Andreeva, A.N. Lykholay, N.V. Tsvetkova, and A.V. Voylokov. 2018. Anthocyanin Composition and content in rye plants with different grain color. Molecules. 23(4):948. doi: 10.3390/molecules23040948. PMID: 29671758; PMCID: PMC6017340.

SUBMITTED BY:

Seedling Images Working Group: Heidi Jo Larson (Heidi.larson@sgs.com), Laura Donaldson (donaldson@indianacrop.org), David Johnston (david.m.johnstonrst@gmail.com), Kathy Mathiason (katherine.mathiason@sdstate.edu), Marija Topic (marijat@crookham.com), and Riad Baalbaki (rbaalbaki@cdfa.ca.gov).

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