Rule Change Proposal 8

1. PURPOSE OF PROPOSAL:

The purpose of this proposal is to provide recommendations in AOSA Rules Section 6.9 for effective usage and storage periods, as well appropriate storage conditions, for various chemicals and solutions used in the germination test. Since the purpose of these chemicals is to promote germination, it is critical that the efficacy of these chemicals be maintained to be effective. These are recommendations and not requirements. These chemicals and solutions include potassium nitrate, ethephon, ethylene, calcium nitrate, and gibberellic acid.

2. PRESENT RULE:

6.9 Explanation of Table 6A

...

- **g. Potassium nitrate.** A two-tenths (0.2) percent solution of potassium nitrate (KNO₃) is used in moistening the substratum and is prepared by dissolving two grams of KNO₃ in 1000 ml of distilled water. The grade of KNO₃ shall meet A.C.S. specifications.
- **h. Ethephon.** A 0.0029% solution of ethephon [(2-chloroethyl) phosphonic acid] shall be used to moisten the substratum. This solution is prepared by mixing 0.6 ml of a stock solution containing two pounds (2 lbs.) active material per gallon in a propylene glycol base with 5,000 ml of distilled water. A solution that is five times the normal concentration (0.0029%) may be used for extremely dormant seeds, provided seeds are transferred to substratum moistened with water after one to three days.
- **i. Ethylene.** Five (5) ml of ethylene gas per cubic foot of germinator space are injected into a germinator in which peanut seeds in moist rolled towels have been placed. Following injection of ethylene, the germinator is kept closed until the first count (five days). If germinator door is opened for the purpose of checking or re-wetting the samples, another injection at the same rate may be made.
- **j. Calcium nitrate**. Three-tenths to six-tenths (0.3-0.6) percent solution of calcium nitrate [Ca(NO₃)₂] is used in moistening the substratum and is prepared by dissolving three to six grams of Ca(NO₃)₂ in 1000 ml of distilled water. If Ca(NO₃)₂·4H₂O (calcium nitrate tetrahydrate) is used instead of Ca(NO₃)₂, 4.3 to 8.6 grams of Ca(NO₃)₂·4H₂O is dissolved in 1000 ml of distilled water. The grade of calcium nitrate shall meet A.C.S. specifications.

...

m. Viability testing of ungerminated seed. — Any of the following methods or combination of methods, unless otherwise specified, may be used to determine the viability of ungerminated seed that remain at the end of the prescribed test period. The results are to be reported as percentage dormant or hard seed as determined by the specified method.

...

(4) **Germination promoting chemicals:** Gibberellic Acid (GA₃) — The germination substratum should be moistened with the recommended concentration, 200 ppm–500 ppm (0.02 - 0.05%) of GA₃ for most cases. Stronger solutions may be used for stronger cases of dormancy. The required GA₃ concentration may vary according to species, cultivar, and year, intensity of dormancy and state of after ripening (Gaspar, S.J. Fazekas and A. Petho. 1975. Seed Sci. & Technol. 3:555-563; Bekendam, J. and J. Bruinsma. 1965. Proc. Int. Seed Test. Assoc. 30:869-886). When the concentration is higher than 800 ppm (0.08%), the use of a buffer is recommended. A 500 ppm solution of GA₃ is prepared by dissolving 500 mg GA₃ in one liter of water.

3. PROPOSED RULE:

6.9 Explanation of Table 6A

•••

- g. Potassium nitrate. A two-tenths (0.2) percent solution of potassium nitrate (KNO₃) is used in moistening the substratum and is prepared by dissolving two grams of KNO₃ in 1000 ml of distilled water. The grade of KNO₃ shall meet A.C.S. specifications. The following are recommendations for proper storage. Store the tightly closed powder container in a dark dry location at room temperature. Check for aggregation/clumping of the powder as a sign of improper storage and deterioration When stored properly, the shelf life of the crystalline powder salt is more than 5 years. Store prepared aqueous solution in a tightly closed container in a dark location at room temperature. When stored properly, prepared solutions have a shelf life of up to 5 years.
- **h. Ethephon.** A 0.0029% solution of ethephon [(2-chloroethyl) phosphonic acid] shall be used to moisten the substratum. This solution is prepared by mixing 0.6 ml of a stock solution containing two pounds (2 lbs.) active material per gallon in a propylene glycol base with 5,000 ml of distilled water. A solution that is five times the normal concentration (0.0029%) may be used for extremely dormant seeds, provided seeds are transferred to substratum moistened with water after one to three days. The following are recommendations for proper storage. Store the tightly closed concentrate solution in a cool dry location. Do not use the concentrate solution after the manufacture's expiration date, as effectiveness is reduced. Once made, use the prepared solution within 24 hours of preparation.
- **i. Ethylene.** Five (5) ml of ethylene gas per cubic foot of germinator space are injected into a germinator in which peanut seeds in moist rolled towels have been placed. Following injection of ethylene, the germinator is kept closed until the first count (five days). If germinator door is opened for the purpose of checking or re-wetting the samples, another injection at the same rate may be made. Follow proper procedures for handling and storage of gas cylinders recommended by the manufacturer.
- **j. Calcium nitrate**. Three-tenths to six-tenths (0.3-0.6) percent solution of calcium nitrate [Ca(NO₃)₂] is used in moistening the substratum and is prepared by dissolving three to six grams of Ca(NO₃)₂ in 1000 ml of distilled water. If Ca(NO₃)₂·4H₂O (calcium nitrate tetrahydrate) is used instead of Ca(NO₃)₂, 4.3 to 8.6 grams of Ca(NO₃)₂·4H₂O is dissolved in 1000 ml of distilled water. The grade of calcium nitrate shall meet A.C.S. specifications. The following are recommendations for proper storage. Store the tightly closed powder container in a dark dry location at room temperature. Check for aggregation/clumping of the powder as a sign of improper storage and deterioration When stored

properly, the shelf life of the crystalline powder salt is more than 5 years. Store prepared aqueous solution in a tightly closed container in a dark location at room temperature. When stored properly, prepared solutions have a shelf life of up to 5 years.

•••

m. Viability testing of ungerminated seed.

...

(4) **Germination promoting chemicals:** Gibberellic Acid (GA₃) — The germination substratum should be moistened with the recommended concentration, 200 ppm–500 ppm (0.02 - 0.05%) of GA₃ for most cases. Stronger solutions may be used for stronger cases of dormancy. The required GA₃ concentration may vary according to species, cultivar, year, intensity of dormancy, and state of after ripening (Gaspar, S.J. Fazekas and A. Petho. 1975. Seed Sci. & Technol. 3:555-563; Bekendam, J. and J. Bruinsma. 1965. Proc. Int. Seed Test. Assoc. 30:869-886). When the concentration is higher than 800 ppm (0.08%), the use of a buffer is recommended. A 500 ppm solution of GA₃ is prepared by dissolving 500 mg GA₃ in one liter of water. The following are recommendations for proper storage. Store the tightly closed powder container in a dark, dry location at 2 to 5°C. Under appropriate storage conditions, the chemical is stable and can be stored for several years. Store prepared solution in a closed container in a dark location at 2 to 5°C. It is best to use prepared solutions within 2-5 days after preparation. Alternatively, stock concentrate solution and prepared solution can be stored frozen at -20°C for 1-2 years.

4. HARMONIZATION AND IMPACT STATEMENT:

ISTA Rules – N/A

FSA/Regs Section 201 – N/A

Canadian M&P - N/A

5. **SUPPORTING EVIDENCE:**

Proper storage and usage information were based on known and published chemical properties of the different forms of each chemical. References included PubChem and relevant SDS (MSDS).

6. **SUBMITTED BY:**

David M. Johnston – RST/CSA Germination and Purity Program Coordinator Seed Programs Louisiana Dept. of Agriculture and Forestry 5825 Florida Blvd. – Suite 3004 Baton Rouge, LA 70806

Phone: (225) 925-4733

Email: djohnston@ldaf.state.la.us

Riad Baalbaki, PhD – CSA Germination Senior Seed Botanist California Department of Food & Agriculture Plant Pest Diagnostics Branch 3294 Meadowview Road

Sacramento, CA 95832-1448

Phone: (916) 262-3292

Email: riad.baalbaki@cdfa.ca.gov

7. DATE SUBMITTED:

October 16, 2019 – Original version February 20, 2020 – Amended version