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Vigor Testing Workshop

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VIGOR TESTING WORKSHOP

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DEFINITIONS OF SEED VIGOR

AOSA Definition

"Seed vigor comprises those seed properties which determine the potential for rapid, uniform emergence, and development of normal seedlings under a wide range of field conditions" (1980).

ISTA Definition

"Seed vigor is the sum of those properties which determine the potential level of activity and performance of the seed or seed lot during germination and seedling emergence.

"Seed which perform well are termed 'high vigor seed,' while those which perform poorly are called 'low vigor seed.'

"The aspects of performance which may show variations associated with differences in seed vigor include: (1) biochemical processes and reactions during germination such as enzyme reactions and respiratory activity, (2) rate and uniformity of seed germination and seedling growth, (3) rate and uniformity of seedling emergence and growth in the field, (4) emergence ability of seedlings under unfavorable environmental conditions.

"The effects of vigor level may persist to influence mature plant growth, crop uniformity, and yield.

"Many factors induce variations in the level of seed vigor, but the principal known causes are: (1) Genetic constitution, (2) Environment and nutrition of the mother plant, (3) Stage of maturity at harvest, (4) Seed size, weight or specific gravity, (5) Mechanical integrity, (6) Deterioration and aging, (7) Pathogens.

"Seed dormancy may obscure vigor potential of a seed lot in a laboratory, but it should not be regarded as a component of vigor if seedling emergence is unaffected in field sowings."

The principles adopted for a vigor test by ISTA are also of interest.

"A vigor test must be reproducible and the results must be proved to be correlated with a field performance characteristic such as seedling emergence under environmental stress.

"Vigor tests may be direct or indirect. Direct tests are those in which an environmental stress expected in the field is reporduced in teh laboratory and the percentage and rate of seedling emergence is recorded, e.g., Hilter test, cold test. Indirect tests are those in which other characteristics of the seed which have proved to be correlated with an aspect of field perfomance are measured, e.g., respiration rate, topographical tetrazolium reaction, conductivity test.

"No single method will satisfy all requirements and a method or combination of methods should be chosen to suit the crop or the environment into which it will be sown."

SEE ALSO GRAPHICAL REPRESENTATIONS BELOW:

CHECKLIST FOR SEED VIGOR TESTING

- NEED FOR INFORMATION ON VIGOR
- PURPOSE OF TESTING
- SUITABILITY OF TEST(S)
- REQUIRED LEVEL OF PRECISION (Equipment and Methods)
- LEVEL OF PRECISION ATTAINABLE
- EXPERIENCE AND/OR EXPERTISE WITH TEST(S) SELECTED
- INTERPRETATION CRITERIA
- CALIBRATION AND MEANING OF RESULTS
- DECISION-MAKING PROCEDURES.

ACCELERATED AGING TEST

Crops: Bean, Corn, Cotton, Pea, Peanut, Pepper, Wheat

Estimate seed longevity in warehouse storage. Predict life span under range storage conditions. Predict seed performance other than storability, such as stand establishment.

Principles:

1. Deteriorates seeds at high temp. (40-45°C) and high RH (>90%) for short period (48-96 hr).

2. Enhance deterioration so that germ decline is proportional to initial physiological potential of seeds.

   Ex: High Vigor Seeds - small decline germ
   Low Vigor Seeds - large decline germ

3. Germ response after AA relates to field performance under wide range of environmental conditions and to relative storability.

Equipment and Supplies:

1. Single sample inner chamber 'tray-method' provides standardized AA results.

2. Single sample inner chamber placed into outer chamber constant temp. of 40-45°C (± 0.1°C). See Table 1.

3. High RH in outer chamber prevents water evaporation in inner chamber.

4. Prevent water collecting at top-inside of outer chamber (condensation) and dripping on lid of inner chamber boxes. This causes condensation on inside of box increasing seed moisture, molds and rate of germ decline.

5. Inner chamber (tray) method uses plastic germ box 11.0 x 11.0 x 3.5 cm with lid.

6. Inside box placed on 10.0 x 10.0 x 3.0 cm wire-mesh screen.

7. 40 ml water added to each box.

8. Seeds should be single layer above water surface.

Procedures:

1. 40 ml water added to each box. Do not splash water on screen.

2. Seed samples determined on weight basis and placed on screen one layer deep. Larger seeds may require two boxes.

3. Secure lid on each box and place inner chamber into incubator. Do not open incubator during aging period.

4. After aging, remove seeds and plant according to standard germ test. Plant immediately after AA.

5. Check moisture content of seeds at the end of aging to ensure that procedures were proper.

Interpretation:

1. Evaluate according to AOSA Rule. Normal seedlings are considered vigorous.

2. Compare AA results with standard germ.

Observation:

1. AA is an excellent vigor test. Needs refinements in techniques, equipment, and time-temp. interactions for many seed kinds other than soybean seeds.

2. Soybean results show that sample size must be determined on a seed weight basis rather than seed number.

3. Control temperature of outer chamber precisely. Minor fluctuations influence germ results.

4. Do not compare results of treated with non-treated seed lots.

5. Seed lots should be at similar seed moisture before aging. Differences of 1-2% is OK at seed moisture less than 13%.

COLD TEST

Crops: Field Corn, Sweet Corn

Assessment of field performance

Evaluate fungicide efficacy

Genetic screening - cold, wet soil

Evaluate deterioration related to storage, freezing, immaturity, injury

Evaluate effects of mechanical damage on germination in cold, wet soil

Select seed lots for early spring planting

Basis for adjusting planting rates.

Principles:

1. Measures ability of seeds to germinate under high soil moisture, low soil temp. and microbial activity.

2. Simulates adverse field conditions and represents low germination that might be obtained under such conditions.

3. Standard germination represents highest germination.

4. Therefore, field germination will be between these extremes.

5. If cold test germination is near standard germination, the seed lot will germinate well over wide range of conditions.

6. Cold test germination is affected by heredity, mechanical injury, treatment and physiological condition. The test measures the combined effects of all factors.

Equipment and Supplies:

1. Plastic crisper (shoe box) - seeds actually planted in germination medium with natural microflora from field with temperature and moisture carefully controlled.

2. "Rolled towel" and "Tray" methods are better for high volume testing.

Rolled Towel Procedure

1. Two paper towels soaked in cold water (10°C) and plant 50 seeds. Cover seeds with thin layer sand-soil mixture (1 sand: 1 field soil).

2. Cover with one soaked - chilled paper towel, and roll as in standard germ test.

3. Place roll upright in plastic bucket and incubate at 10°C for 7 days.

4. Place at 25°C for germination and evaluate after 4 days just as in standard germination test.

Tray Method

1. Food service tray 45 x 66 cm (18 x 26 in.) used as container.

2. One sheet of 40 x 61 x 0.5 cm (16 x 24 x 0.25 in.) kimpak placed on tray and soaked in 1,100 ml chilled water (10^0c).

3. Two replicates of 100 seeds each (200 seeds) placed on top of kimpak. Press seeds into kimpak to stabilize.

4. Add excess day sand - soil mixture spread over seeds.

5. A levelling board to produce a level 0.4 cm (1/8 in.) below lip of tray is used to level soil.

6. Sand-soil equilibrated at 70% saturation.

7. Space trays at 7.5 cm (3 in.) intervals in enclosed food service cart with plexiglass back.

8. Place cart into 10^0C chamber for 7 days.

9. Transfer to 25^0C chamber for 4-day grow-out. Illuminate chamber for 8 hr. daily.

10. Evaluate seedlings as in kimpak germination test.

COOL GERMINATION TEST

Crop: Cotton

Principles:

1. A soil based cold test is too severe at 10^0C for cotton. Also, soil microorganisms retard cotton seedling growth under cold, wet conditions.

2. Cool germination test less severe to separate vigorous from less vigorous seeds. Low vigor seeds from warm season crop (cotton) will experience decreased growth rate and germination under cool conditions.

3. Determines it cotton seed lot is suitable for planting in cool soil. Lots with high percent of vigorous seedlings can be planted under a wide range of field conditions. Lots producing few vigorous seedlings should be planted only under very favorable field conditions.

Equipment and Supplies:

1. Same as for standard germination test.

2. Maintain constant 18°C and adequate humidity to prevent drying (18^0C is essential).

Procedure:

1. 200 seeds from each lot. Four reps of 50 seeds each on two moist towels.

2. Cover with two additional towels.

3. Roll towels and set upright (on end) in wire mesh basket.

4. One count made on 7th day for both acid and machine delinted seeds.

5. Normal seedlings with combined hypocotyl and root length of 4 cm (1 1/2 in.) or longer are counted.

6. Root-hypocotyl measurement is made from tip of radicle to point of cotyledon attachment.

7. Treat seeds with recommended fungicide. Only conditioned seeds should be tested. Not accurate on gin-run seeds.

Interpretation:

1. Normal seedlings according to AOSA Rules.
2. Evaluate root - hypocotyl - cotyledons - epicotyl.

CONDUCTIVITY TEST

Crops: Wrinkled Seeded Garden Peas, Soybeans, Field Corn

Principles:

1. Measurement of electrolytes leaking form plant tissues. All membranes lose integrity as seeds dry and mature.
2. During imbibition, membrane integrity is re-established. Vigorous seeds re-establish faster with less leakage than low vigor seeds.
3. Leakage of electrolytes as nutrients stimulate microorganism activity and secondary infection.

Equipment and Supplies:

1. Conductivity meter:
   - Dip cell must have a cell constant of 1.0.
   - Calibrate by a 0.01 M Kcl solution (0.745 g Kcl dried at 150°C for hr) dissolve in deionized water to make 1 liter.
   - In this solution, the meter should read 1,273 µmhos per cm at 20°C or 1,408 µmhos/cm at 25°C
   - Clean dip cell repeatedly with deionized water
2. Water:
   - Deionized or distilled not to exceed 5 µmhos/cm conductivity and sorted at 25°C (± 1°C) at least 24 hr. before use.
3. Glass or plastic beakers:
   - Base diameter influence results and should be 6-8 cm to provide adequate depth of water to immerse seed dip cell.
4. Incubator:
   - Germinator operating at a constant temp. of 25°C.

5. Seed moisture tester or oven:

   - Operate at 105^{0}C

Procedure:

1. Seed Moisture:

   - Determine seed moisture on fresh wt. basis.
   - Low (<11%) or high (>17%) should be adjusted.
   - Adjust low moisture at high RH/10^{0}C for 2 days. Desiccate high moisture seeds over CaCl for 7 days at 20^{0}C.

2. Removal of Seed Treatment:

   May not be necessary:

   - 150 seeds in flask with 100 ml methanol; shake gently for 2 minutes; rinse followed by second rinse with 100 ml methanol, and third rinse with 50 ml methanol.
   - Dry seeds in hood for 20 min. or at room temperature for 2 hr.
   - Prior to soaking and after weighing, each set of 25 seeds rinsed once with 20 ml distilled water.

3. Conductivity test:

   - 4 reps of 25 seed (uninjured) are weighed to 2 decimal places.
   - Place in 200 ml beakers and add 75 ml of deionized water.
   - Stir gently, cover breaker and place at 25^{0}C for 24 hr.
   - Measure conductivity immediately after removal from 25^{0}C.
   - Shake beaker with seeds gently for 10 - 15 seconds and immerse dip cell into the solution without filtration.
   - Rinse dip cell twice in deionized water and blot dry with filter paper.

- Conductivity per gram of seed wt. is:

$$\frac{\text{Conductivity (}\mu\text{ mhos) for each beaker}}{\text{wt. of 25 seeds}} = \mu\text{ mhos/cm/g}$$

- Report % of injured seeds

Interpretations:

- Measure conductivity immediately since temp. affects conductivity.
- Evaluate 8-12 beakers at a time. Others remain in 25^0C.
- Cracked seeds give cloudy appearance; initial moisture may be too low or injured seeds were not removed.
- High μ mhos/g = low vigor such as > 150 μ mhos/cm/g for soybean
- Not good for early planting or may be totally unfit.
- May lose vigor rapidly in storage.

Observations:

Factors affecting test:

- Purity of water and cleanliness of equipment
- Sample uniformity
- Soaking time and water temperature
- Initial seed moisture
- Temperature of steep water during test.

Alternate Procedure:

- Individual seed evaluation available with commercial machines which monitor leakage form individual seeds.
- 100 seeds placed into soaking try with 100 individual cells soak for 24 hours at 25^0C.

- Multi-head electrode (100 electrodes) fits over 100 cells of soaking tray cells and measures current in each cell.

- A partition selector setting for specific crop permits prediction of germination. Partition setting is a theoretical "breakpoint' which places each seed into live or dead category based on seed leakage in μ amps.

- Results in μ amps rather than predicted germination.

- Conductivity expressed in μ mhos but significant correlation exists between μ amps and μ mhos.

- Weigh the 100 seeds since size is not considered by the machine.

SEEDLING VIGOR CLASSIFICATION

Crops: Cotton, Garden Beans, Peanuts, Soybeans

Principles:

1. Four morphological sites for vigor evaluation: root - hypocotyl - cotyledons - epicotyl
2. Germination - each site develops promptly and free of defects.
3. Classify normal seedlings into strong or weak categories

Equipment and Supplies:

1. Same as for standard germination test.

Procedure:

1. Germ as prescribed by AOSA Rules
2. 8 reps of 50 seeds each
3. 2 moist towels on bottom - plant seeds
4. 2 moist towels on top
5. Orient for unobstructed growth of root, hypocotyl, epicotyl
6. Constant 25°C - dark or light
7. Light promotes rapid epicotyl growth
8. Maintain substrate moisture
9. Treat peanuts, cotton, garden bean seeds
10. Evaluate normal seedlings
11. Classify normal seedlings into strong or weak

Interpretation:

1. Weak normal seedlings subject to stress
2. More precise field emergence potential
3. Accuracy of interpretations of seedlings.