CDFA Seed Lab Functions

- Regulatory Samples - Purity, Noxious Weed, and Germination
- Seed Investigations
- Service Samples - Identification, Purity, Germination, Seed Vigor
- Identifications for County Ag Commissioners (high-risk dog team interceptions), Border Inspection Stations, private labs
- Association of Official Seed Analysts (AOSA) - streamline national seed analysis rules and improve uniformity in analysis
- Training new seed analysts for the future
CDFA Seed Lab-sample dividing, purity, herbarium, germination
Purity analysis

• Pure seed unit (PSU) and required working weight for each crop
• Each sample approximately 2500 seed units by weight
• Each sample is separated into pure seed, other crop seed, weed seed, and inert matter (soil, sticks, chaff, broken seeds, plant parts, fungi)
• Test these 4 components for compliance using Federal Seed Act or AOSA tolerance tables
• Our lab routinely tests over 100 different crop species
Components of Seed Purity Separation:
Pure Seed, Other Crop Seed, Weed Seed, Inert Matter

Purity Components: Pure Seed, Other Crop Seed, Weed Seed, Inert Matter
Seed Mixtures

• Regulatory Samples - over 50 crop species mixtures per year
  • Examples - turfgrass mixtures, grain mixtures, forage mixes, erosion control mixes (commonly 2-8 species per sample, one 17-species sample this year)
• Seeds of all species must be completely separated by hand for purity analysis and each species tested separately for germination
• Preparation time for mixtures ranges from 3 to 20 hours per sample
Noxious Weed Seed Examinations

• Conducted on all regulatory samples of agricultural crop seeds
• Each sample approximately 25,000 seed units by weight according to Federal Seed Act and AOSA Rules
• Zero tolerance for Prohibited Noxious Weed Seeds (stop-sale order)
• Restricted Noxious Weed Seeds must be within tolerance of zero per sample (maximum of 2 seeds per 25,000) or listed on the label
California Noxious Weed Seeds

- Over 180 species are listed as noxious weeds in California Code of Regulations
  - Prohibited noxious weed seeds
  - Restricted noxious weed seeds
- No tolerance for noxious weed seeds in certified seed lots

Photo: Scotch thistle, a prohibited noxious weed seed that badly degrades rangeland; D. Meyer, CDFA Seed Lab
Giant hogweed, *Heracleum mantegazzianum*, a Federal noxious weed

- Old World species
- Established in the Pacific Northwest and eastern U.S.
- “A”-rated pest in California subject to exclusion and eradication
- Hazardous to humans and livestock
- Not currently known to occur California

Photo credits: Plant, L. Mehrhoff, Univ. of Connecticut, Bugwood.org; Fruits, J. Scher, USDA APHIS, Bugwood.org
Seed Investigations

• We provide unbiased analyses to help resolve seed disputes or problems reported by the Seed Control officer

• Can help protect seed sellers

• Examples:
  • Unusually low or variable germination results
  • Poison control referrals - poison hemlock plants in a vegetable garden near carrots planted from seed
  • Seed testing result - poison hemlock likely present in the environment and not from the carrot seed

Photos: Plant, Ohio State Weed Lab, bugwood.org; Seeds, Steve Hurst, bugwood.org
Seed Identification at CDFA

• Service Sample Seed Identifications (39 samples in fiscal year)
• Seeds from global trade found as contaminants and unable to be identified by the private labs (92%)
• Can’t sell the seed lot unless excluded species are ruled out
• Intercept samples (18% were of prohibited noxious weed seeds):
  • Seeds found by dog-team intercepts of parcels (13)
  • From vehicles at CA Border Inspection stations (25)
  • County high-risk interceptions (36)
Dust-like seeds

• Very small (0.5 mm or less), found in a variety of flowering plant families, including Orobanchaceae and other families formerly grouped into the snapdragon family

• Some non-native taxa of Orobanchaceae (e.g., *Orobanche*) are Federal noxious weeds parasitic on important crop plants such as tomatoes and hemp

• Very expensive and difficult to control due to long-term seed bank

• Morphological identification of dust-like seeds is challenging
Seed of Calceolaria (slipper flower) found in a Brassica vegetable sample

- Seed 0.3 mm
- submitted for identification by a major seed company lab
- DNA sequence - 99.7% identity match to *C. corymbosa*, a native South American wildflower.
- Plant is cultivated globally as an ornamental flower
- Seed may have lodged in dividing equipment

(Photo: D. Meyer, CDFA Seed Lab)
Garlic mustard, noxious weed new to California

- Noxious weed in the northern U.S.
- Discovered in California in the last two years
  - identified by the CDFA Seed Lab
- Under eradication by the U.S. Forest Service
- Develops a lasting seed bank
- Inhibits growth of crop plants
- Should be kept out of farmland and agricultural seed supply
Thanks!

- Bob Price, Senior Seed Analyst, CDFA Seed Lab

*Mantisalca salmantica*, daggerflower, an “A” rated pest sold in craft stores. Photo: D. Meyer, CDFA Seed Lab
Overview

✓ Seed Lab (SL) Functions

➤ SL Label Compliance Testing Activities

➤ Seed Testing: Regulatory Perspective

➤ Training and Methods Development: Essential Activities

➤ Benefits to the Seed Industry
Label Compliance Testing-1

Sample number and distribution

- Total (July 2021-April 2022): 528
  - (88% of 600 target)
Label Compliance Testing-2

Sample number and distribution

Sample number

FY 21-22

Q1
Q2
Q3
Q4
Sample number and distribution

Label Compliance Testing-3

![Bar chart showing sample number distribution for FY 21-22 and FY 20-21 for quarters Q1, Q2, Q3, and Q4.](image)
Label Compliance Testing

- Labelling standards
- Purity
- Germination
Label Compliance Testing

Percentage of samples with at least one type of violation

5-year average (18.2%)
Label Compliance Testing-6

Percentage of samples with at least one type of violation

2018
Label Compliance Testing - 7

Percentage of samples with at least one type of violation
Label Compliance Testing

Percentage of samples with at least one type of violation

![Bar chart showing percentage of samples with violations from 2018 to 2020. The chart shows a decrease in the percentage of violations over the years.]
Label Compliance Testing

Percentage of samples with at least one type of violation

![Bar chart showing percentage of samples with at least one type of violation for years 2018 to 2021. The chart indicates a trend of increasing violation percentages over the years.]
Label Compliance Testing-10

Percentage of samples with at least one type of violation
Label Compliance Testing-11

Most common violations:

- PVP notification;
- Seed kind;
- Non-registered labeler
Label Compliance Testing-12

Purity/Germination Violations:

- Pure seed percentage (5%, n=26)
- Inert matter percentage (3%, n=14)
- Other crop seed percentage (1%, n=4)
- Germination percentage (1%; n=4)
Other activities

- **Service testing (for fee)**
  - 281 samples
    - 171 rice for certification
    - 26 seed IDs
    - 19 custom tests
- **Feed mill inspections (48)**
- **Detections/identifications**
- **Training/methods development**
- **Research/investigations**
- **Other services**
What is Seed Testing?
Regulatory Perspective

Rules: application of standard procedures to evaluate physical, physiological and other characters of seed lots.
Seed Testing

Rules (standard procedures):

- For all aspects of seed testing
  - Association of Official Seed Analysts (AOSA)
  - Federal Seed Act (FSA)
  - International Seed Testing Association (ISTA)
  - Other country-specific rules (e.g., Canadian M&P)
SOPs vs. Rules

SOPs:

- Documentation, safety measures, equipment inspections, training requirements, etc.
- Testing procedures (Rules) must be the same across labs
Table 2A. Weights for working samples

<table>
<thead>
<tr>
<th>Kind of seed</th>
<th>Unit</th>
<th>Minimum weight for one sample</th>
<th>Minimum weight for two samples</th>
<th>Maximum weight of one sample</th>
<th>Maximum weight of two samples</th>
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Table 6A. Methods of testing for laboratory germination

<table>
<thead>
<tr>
<th>Kind of seed</th>
<th>Substrate*</th>
<th>Temperature (°C)</th>
<th>Percent germination at 7 days</th>
<th>Specific requirements and notes</th>
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*Substrate: sand, soil, vermiculite, perlite, or other suitable material.

**Temperature (°C): 20-30 for most seeds; 15-25 for hard seeds.

***Percent germination: 7 days after sowing, except for hard seeds which require 10-15 days.

****Specific requirements: Good moisture supply, no light, and a humid atmosphere.

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<th>First count (days)</th>
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<th>Specific requirements and notes</th>
<th>Dormant seed[^1]</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Lupinus albus</em></td>
<td>B, T</td>
<td>20</td>
<td>3</td>
<td>10</td>
<td>Hard seeds: see sec. 6.2d and 6.9m(6)</td>
<td></td>
</tr>
<tr>
<td>Kind of Seed</td>
<td>Substrata&lt;sup&gt;a&lt;/sup&gt;</td>
<td>Temperature (°C)</td>
<td>First count (days)</td>
<td>Final count (days)</td>
<td>Specific requirements and notes</td>
<td>Dormant seed&lt;sup&gt;f&lt;/sup&gt;</td>
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<tr>
<td><em>Tetragonia tetragonoides</em></td>
<td>T</td>
<td>15; 20</td>
<td>5</td>
<td>21</td>
<td>Soak fruits overnight (16 hr), air dry 7 hr; plant in very wet towels. Do not rewater unless later counts exhibit drying out</td>
<td>On 21&lt;sup&gt;st&lt;/sup&gt; day scrape fruits and test for 7 additional days</td>
</tr>
</tbody>
</table>
Seed Testing-1

- AOSA, ISTA, FSA; 1960-1975
Rules used by most of the world
What explains this huge increase???

AOSA, ISTA, FSA, 1960-1975

AOSA+, FSA, Canadian M&P, ISTA+

Present Rules
Large increase in types of tests
  - Moisture content, cultivar purity, TZ, vigor, etc.
Approximately 700 species
More than one method for the same test (some species)
  - Artificial substrate used to be cotton sheets/blue blotter
  - Now 10 substrates plus soil
Regulatory labs must be familiar with all the Rules for all the listed species
Example: fava beans sample, with different germination test results (range of 17%) by three labs, using different Rules (ISTA, AOSA, Canadian M&P), substrates and temperatures.
Job Description:
- Analyze corn and soybean seed samples for the following tests:
  - Accelerated aging
  - Warm and cold germinations
  - Purity analysis
  - Inert/foreign material
  - Noxious weed identification
- Compile and analyze data from seed testing and provide feedback to seed production team.
- Work within the seed production team to audit quality concerns, identify quality issues and create processes for quality improvement
- Audit current processes and make changes as needed on an annual basis. Create new processes, including the development of new methodologies for seed testing, as required to achieve the objectives of the lab.
JOB REQUIREMENTS

1. Education and Training:
   ○ Bachelor Degree
   ○ Registered Seed Technician is preferred or ability to obtain RST accreditation in less than two years.

2. Technical Requirements:
   ○ Excellent verbal and written communication skills.
   ○ Possesses knowledge of various seed testing methods for corn, soybeans and wheat is preferred.
   ○ Possesses a positive attitude.
   ○ Ability to establish priorities, work independently, work as a team member, and proceed with objectives without supervision.
   ○ Possess a high-level attention to detail, organized, and be able to consistently deliver timely and accurate results.
   ○ Proficiency with computers, Microsoft Office and spreadsheets.
What are the added costs of maintaining the ability to test and apply all the Rules?

- Significant but not substantial
  - Thousands of dollars annually for the different Rules
  - Inefficient use of resources (space/equipment)
What are the added costs of maintaining the ability to test and apply all the Rules?

- Significant but not substantial
  - Thousands of dollars annually for the different Rules
  - Inefficient use of resources (space/equipment)
  - A wide range of planting materials
  - Other reference materials
As a regulatory lab we often test and re-test under different conditions.

- We have to get it right!
- Whatever we report is considered the true evaluation of the seed lot.
CA Food and Agricultural Code. 52363(b):
“A written report which is issued by the State Seed Laboratory that shows the analysis of any such sample is prima facie evidence of the true analysis of the entire lot from which the sample was taken.”
As a regulatory lab we often test and re-test under different conditions.

- We have to get it right!
- Whatever we report is considered the true evaluation of the seed lot.
- Not every sample gets re-tested

- Added costs of testing, re-testing and investigations: Time!
Do All Regulatory Labs Retest?

- Some, but not all regulatory labs!
- Our policy (germination): a label is in compliance until proven otherwise

At least two tests under same or different conditions, and no single test produces results indicating label is in compliance
Different Conditions

- Replant using other options in Rules (substrates, temp., etc.)
- Replant after removing seed treatment
- Replant under grow lights/growth chamber
- Add germination promoting chemicals
- Disinfect seeds
- Test viability by TZ
Retesting/Multiple Testing Frequency

- About 8% of all regulatory samples for germination
- Service: about 5% of all samples for germination
- Investigative: every sample
- Time of actual testing can be more than 10x ‘normal’ sample testing time (5x on average)
Treated Squash Sample Retest-
Germination Label Violation

- Planted according to Rules
- Planted according to Rules (repeat test)
- Planted in sand-extended test
- Planted in peat pellets-extended test
Investigative Sample - Fava Beans

- Planted according to ISTA Rules
- Planted according to Canadian M&P Rules
- Planted according to FSA/AOSA
- Planted in peat pellets
- Seeds planted with and without disinfection with bleach
- Germination and abnormal types determined for each planting method, separated into root and shoot abnormalities
Testing a ‘New’ Species

Part of SL SOP (based on previous experience)

- Plant according to most common method in Rules
- Plant using alternative methods
- Plant in soil/peat pellets
- Based on species, determine need for dormancy breaking treatments
Fees for Investigative Samples?
- In-house training of Environmental Scientists and Associate Seed Botanists
- Training of analysts from other USA labs (mostly for certification)
- Training workshops
  - Purity
  - Vigor testing
  - Germ. diagnostics
  - Germ. Methods
  - Moisture testing
  - Seedling Evaluation
  - Referee testing
  - Statistics in seed testing
Training-2

- Seeds of most species do not behave ‘normally’ when germinated on artificial media
- Many things can go wrong during a germination test, unrelated to seed quality
- Variation in germination testing remains high for many reasons
Safflower
Purity violations are always more common than germination violations

- Tolerances are wider for germination
- Tolerance: maximum expected difference due to chance

“A larger and more arbitrary tolerance must be allowed in the results of germination tests [compared to purity results].” USDA, 1928.

Label: 85%; Test: 78% = results are in tolerance
Validating suitable methods for germination testing and seedling evaluation is as critical as good training.

The impact of proposed new rules on the CA seed industry is always a consideration.
Method Validation-2

- Modifications of existing methods
- Additions of test options
- Selecting best methods for ‘new’ species
- Introduction of new methods (research)
Benefits to the Seed Industry

- Reliable evaluations through rigorous protocols
- Training
- Research
- Information
- Validation (referee) studies and Rules proposals
  - Involvement in different AOSA and ISTA activities to promote uniformity within and across organizations.
Examples-1

- Reduced period of tall fescue germination test (21 to 14 days)
- Reduced germination period of radish germination test (7 to 6 days)
- Identification of species with hard seeds
Examples-2

- Improve ROA (report of analysis) for transparency and accuracy (ongoing); ASTA request
- Review accuracy of florescence levels in determining annual/perennial ryegrass (initiated)
- Improve accuracy of seed counts for flower/wild species
- Development of testing and evaluation protocols and reference website for wild/native species (ISTA Technical Working Group)
The American Seed Trade Association (ASTA) and the Association of American Seed Control Officials (AASCO) resolved to investigate the uniformity of seed lab analyses.

Blinded samples of known composition (9 species) were sent to several labs (regulatory and commercial).

I was asked to provide statistical analysis, interpretation and recommendations.
In general, purity analyses results were consistent among labs.

Germination results indicated significant variation among labs for many species. Differences in results among labs were as high as 36%.

TZ test results also varied. Differences among labs for the same tested species were as high as 60%.
Significant deficiencies in the ability of labs to detect and classify noxious weeds, weeds and other crop seeds.

Significant deficiencies in the ability of labs to correctly identify foreign seeds.

A large number of determinations of foreign seed numbers were significantly lower than ‘spiked’ results. In many cases, contaminants were entirely undetected.

ROA compliance was inconsistent.
Recommendation

- Concerned organizations (AOSA and SCST) should resolve to improve uniformity in all areas of seed testing, as well as the content of official seed test reports.
- ASTA asked AOSA to work on improving test uniformity!
AOSA and SCST members formed several working groups (WG)

The CA Seed Lab is represented in several WG and leading the effort in the two most critical areas: purity and germination testing

Germination WG: Seedling Evaluation Reference and Training Database
Seedling Evaluation Database

- Funded by the USDA; a 5-year project.
- Much more than a database; a systemic and quantitative way of understanding and improving seedling evaluation
Seedling Evaluation Database - Objectives

Objectives:

- Understand why analysts’ results vary
- Resolve many of the gray areas related to seedling evaluation
- Provide uniform training to all analysts (ISTA as well)
- Reduce errors and variation in germination results that can negatively impact on producers/sellers
- Increase repeatability of evaluations for regulatory purposes
2 □ Poor picture quality; reliable evaluation not possible (advance to next picture)

□ Normal; all essential structures present; defects not so severe (advance to next picture)

□ Abnormal; select all abnormal structures and select abnormality reason(s) for each. Optional: provide additional notes in the comments box below.

- Shoot Abnormal reason 1 Abnormal reason 2
- Root Abnormal reason 1
- Seedling Abnormal reason 1 Abnormal reason 2
- Borderline/Doubtful; seedling falls along the normal/abnormal boundary. Select all borderline structures. Optional: explain your answer in the comments box below.

□ Shoot □ Root

□ Shoot □ Root

Comments (optional)
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Comments (optional)
Which is more uniform, rep 1 or rep 2?
Field bean
Thank you!