FREQUENTLY ASKED QUESTIONS
Annual Ryegrass (*Lolium multiflorum*) Testing

Production of certified annual ryegrass (ARG) is growing. ARG is harvested early in July and has to be tested as fast as possible. In years when harvest is delayed, timeliness is even more critical. This generates many questions about the speed of testing so that seed can be tested, labeled, and shipped just in time.

EARLY TAGGING PROGRAM

In order to respond to customers that cannot wait for the germination/fluorescence (Germ-Fl) test, the OSU Seed Certification Service has developed an *Early Tagging Program for annual ryegrass destined for OECD markets*. This program includes additional field inspection and allows tagging based on the ploidy level of leaf samples from the production field and purity and TZ tests on an official seed sample. Germ-Fl and ploidy tests on official certification samples are still required to verify the correctness of the early tagging decision. Those interested in this program should contact OSU Seed Certification at 541-737-4513.

FREQUENT QUESTIONS ON TESTING

Any test that requires germinated seedlings such as the fluorescence test (Fig.1) or ploidy by cytometry test (Fig. 2) requires time. The lab has developed many accelerated procedures or options to assist its customers with this process as described in the following questions.

1. How can I find out when my germination/fluorescence test will be finished?

Ideally, samples are planted within 1-2 days from arriving to the lab. The date of planting and number of days pre-chilled (if applicable) is posted on the eCertification website. Depending on the speed of germination of each sample, the test can be completed in 14-21 days from planting. Customers can check the status of their samples on the web at any time.

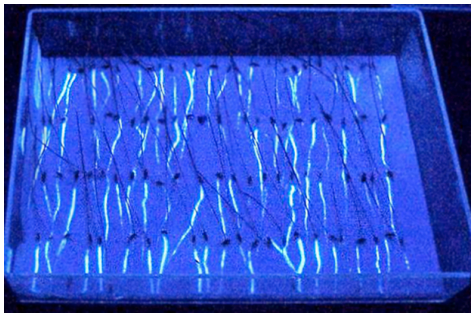

Fig 1. A fluorescence test to distinguish annual and perennial ryegrass.

2. Can you germinate my sample without chilling?

The AOSA Rules indicate to pre-chill dormant seed. Chilling of freshly harvested seed breaks dormancy, and helps the sample to achieve its maximum potential germination. Germination of non-chilled freshly harvested seed results in slow, irregular germination which sometimes is insufficient to meet certification standards. Approximately two months after harvest, dormancy of ARG disappears naturally; thus, the OSU Seed Lab stops pre-chilling on Labor Day.

3. Do any germination-fluorescence test of ARG end before 14 days?

Yes, a testing rule was approved in 2011 allowing the Germ-Fl test of ARG to be finished before 14 days if the sample has reached maximum germination. In practice, the lab ends the test if the sample has reached maximum germination and when the fluorescence is higher than the VFL or satisfactory for certification. Based on the new procedure, about 85% of Germ-Fl tests are ended on first count (7 days after pre-chilling or 14 days from planting). Samples that germinate slowly and/or have low fluorescence are continued for an additional 7 days.

4. Does the purity report change when the fluorescence test is reported?

The initial purity report is a mechanical purity only. Once the Germ-Fl test is completed, the final purity report is issued reflect the percentages of annual and perennial ryegrass detected by fluorescence.

5. Can ARG samples for OECD be certified with TZ?

Yes, but only when the seed lot has followed the *OECD Early Tagging Program*. The usual Germ-Fl test is still required, but tagging does not wait for the germination and fluorescence test results.

6. Is a ploidy test required on all ARG?

A ploidy test is required when certifying tetraploid varieties in the Oregon or OECD certification programs. Some commercial seed growers ask for a ploidy test to determine potential diploid contamination in their fields or finished products. Breeders use this test to assure they are selecting pure tetraploid lines or original seeds.

Fig. 2. Flow cytometry is used at OSU seed lab to determine the ploidy level in ryegrass.

7. Can Seed Certification accept ploidy results on leaf sample from the field?

Yes, but this procedure is used only for the *OECD Early Tagging Program* to have ploidy results available before harvest. This procedure requires obtaining a random sample of green leaves from production fields by Certification inspectors. However, the ploidy test done on leaf sample from the field does not negate the requirement for a follow-up ploidy test being done on an official seed sample after cleaning the crop for verification of the early field test.

8. Should I test for ploidy a lot that comes labeled as tetraploid from overseas?

Yes, if the seed from a foreign source is going to be planted and increased under the Oregon Certification program, it is best to confirm the ploidy level of the pre-control sample before planting the field. In the past, some ryegrass samples were imported as tetraploid and were found to be diploid. A ploidy test is now required on all OECD ARG pre-control samples, which can save time, money, and headaches.

9. What happens if someone forgets to order a ploidy test?

It saves time to order the ploidy test along with purity and Germ-Fl tests at the same time. If you forget, this will result in further delays. If you are listed on the sample as the grower, cleaner, or CC, you can see if a ploidy test was ordered on your sample using the eCertification website or by calling the Seed Lab.

We hope that this information is helpful. If you have any other questions regarding testing ARG, please contact the OSU Seed Lab at seedlab@oregonstate.edu or call 541-737-4464. For certification related questions, please contact OSU Seed Certification at 541-737-4513.