



Vineyard Development: Disease Testing Guidelines

By Judit Monis, Ph.D.

As the spring season progresses, vineyard managers and growers are planning new vineyard development functions. The most common activities include planting nursery produced bench grafts, rootstock rootings (for later field budding), and top working. This article describes a simple process to allow the best return on investment of disease testing.

There are important steps for ensuring a healthy vineyard with consistent grape quality and yield potential. An integral part of this process involves the health status assessment of field selections, nursery rootstock, and scion material prior to planting. Our laboratory specializes in testing grapevines for the presence of disease causing agents (i.e., pathogens). We have developed reliable procedures for the detection of the most important pathogens that cause disease in grapevines. It is critical that samples are collected at the right time of the year and at the appropriate vine location for accurate disease assessment. The results from disease testing will provide the information needed for sound vineyard development decision making.

Top working or grafting is an activity that allows the grower to change the scion variety of an established vine by budding or grafting. This activity allows sampling the most mature sections (cordon and trunk) of the vine since the top of the vine will be replaced. The portions above and below the graft union constitute the best type of sample for testing for the presence of fungal pathogens (vine decline and Esca) and viruses included in HealthCheck™ Panel A (leafroll

and rugose wood diseases). However, if the goal is to determine the presence of soil-borne pathogens, root samples will be required.

The ideal sample number to test prior to making top grafting decisions will depend on the diversity of vines present in the vineyard. If the vineyard is planted with vines from the same origin (i.e., same rootstock/scion combination from a common source) testing at least ten vines is recommended. If there are many rootstock/scion combinations, the vines are from unknown origin, there is suspicion of infection, or the vineyard is adjacent to an infected vineyard, a more exhaustive sampling regime will be required. Furthermore, the number of vines to sample will increase if mealybugs, nematodes, or other disease transmitting vectors are present in the target vineyard or neighboring vineyards. If no apparent symptoms are present, random and representative sampling is recommended. If disease symptoms are present in the vineyard, testing samples from both symptomatic and asymptomatic vines will aid the diagnosis.

The time to submit samples of bench graft and rooting material for testing should coincide with the time the grower decides which rootstock-scion combination will be planted. In most cases, nurseries are able to trace their mother vines and organize cuttings in specific bins. A representative sample should be collected from each bin of rootstock and scion material that will be used for grafting.

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President's Corner



Darrell Maddox, President

More Services

Not long after our acquisition by the Eurofins Scientific Group, I used this column to relay my enthusiasm about what I thought this meant for our clients. On the horizon, I see many of these benefits being implemented. Our integration into the Eurofins laboratory information management system (LIMS) is only a few months away and not only do we expect improved laboratory flow with this system, but we see it helping you with on-line ordering and real-time processing information. We also expect that we can handle your samples for GM testing very soon, providing you with another "one stop shop" service. Our DNA laboratory continues to work with their European counterparts to expand our marker library. We have also been discussing testing seed for pesticide residues and foodborne human pathogens. (Though ESTA still agrees with the position of the American Seed Trade Association that testing planting seed for foodborne pathogens is not scientifically valid, we recognize that some of you are still being asked for proof of testing for these pathogens.)

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Vineyard Continued

The correct sample number will depend on mother block history and budget. For non-destructive testing, samples will need to be submitted at different times of the year to cover the whole spectrum of grapevine disease causing agents. For example, the Pierce's Disease bacterium is best detected later in the summer season (petioles from mature leaves), while the viruses associated with decline are best detected in the spring season (young foliar shoots). Please see Figure 1 for more details.

Because of the potential of virus spread and fungal infections in vineyards it is important to test samples after field finishing or cold storage (especially for the detection of fungal pathogens). The testing is needed even if the vines are from a reputable certification program. Visual inspections are important but most likely cannot determine if a pathogen is present.

This spring season is the most appropriate for submission of samples for HealthCheck™ Panel B testing. HealthCheck™ Panel B includes the decline and degeneration disease causing viruses: Arabis mosaic virus (ArMV), Grapevine fanleaf virus GFLV, Tobacco ringspot virus (TRSV), and Tomato ringspot virus (ToRSV). These viruses are transmitted by nematodes. Work in our lab has shown that although dormant wood from heavily infected vines may be used, young tips and leaves collected in the spring better enables the detection of the viruses included in HealthCheck™ Panel B.

Please check our website for updates and call us for your specific testing needs.

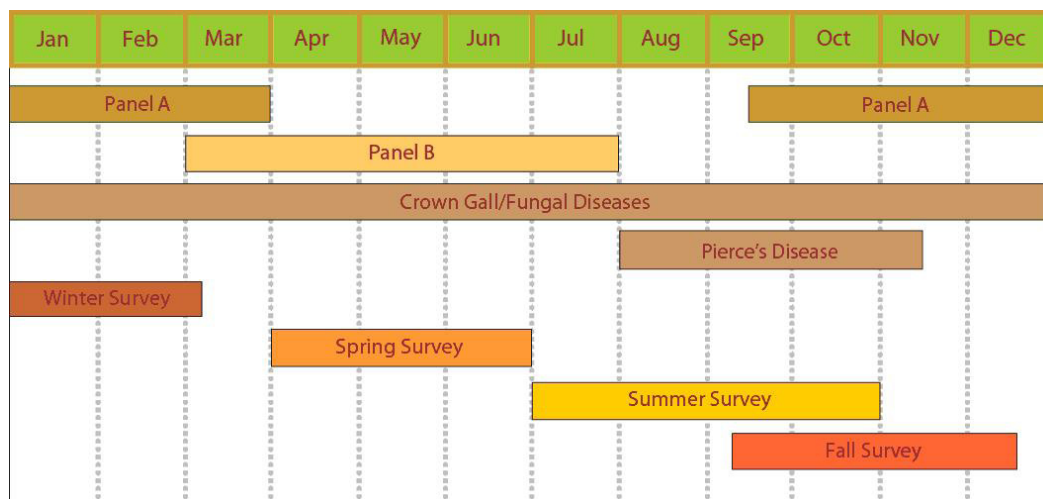


Figure 1: Schedule for Grapevine Testing and Vineyard Services

Changes in the Future of Seed Testing

By Linda Barbosa

For many years there have been two seed testing organizations in the United States, the Association of Official Seed Analysts (AOSA) and the Society of Commercial Seed Technologists (SCST). The AOSA is composed of seed analysts from government and university labs and the SCST is composed of analysts from private seed companies and private testing labs. Up until recently the AOSA was the only one authorized to accept new seed testing methods. There is now a proposal for consolidating the AOSA and the SCST into one seed testing organization.

The following is a short version of the proposal prepared by the Consolidation Task Force

More Services Continued

Access to the Eurofins technologies and other Eurofins services will continue to complement our list of seed quality, genetic and plant health services, but at the same time, we have worked especially hard to ensure that we have not lost touch with our primary mission to help your success by providing reliable,

Lead by Loren Wiesner, Retired Research Leader, ARS-USDA

Vision: To serve agriculture in the area of seed testing

Mission: To serve the seed industry by maintaining and encouraging the highest professional standards and proficiency among its members and promoting uniform seed testing standards through accreditation, research and method development and maintaining and updating the Rules for Seed Testing.

The Association will achieve this mission by:

- Educating and accrediting seed technologists
- Develop standardized seed

testing methods and maintaining them in the Rules for Testing Seed

- Producing and distributing educational materials for seed technologists
- Provide a forum for the exchange of ideas between seed technologists, researchers and the greater seed industry

Benefits to AOSA and SCST Members:

- Provides one organization that represents all seed technologists and seed researchers
- Indicates solidarity to the

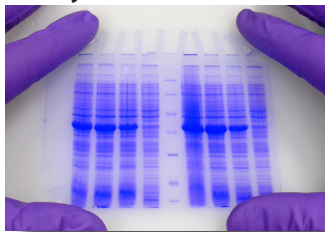
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Come See Us at Upcoming Events

- ESTA Seed Health Workshop: June 1 in Longmont, CO (Eurofins STA Technical Experts)
- AOSA / SCST Meeting: June 2-5, 2009 in Fort Collins, CO (Linda Barbosa)
- ASTA Annual Meeting: June 20-25 in Scottsdale, AZ (John Mizicko)
- American Phytopathological Society (APS) Annual Meeting: August 1-5 in Portland, OR (Judith Monis)
- International Council for the Study of Grapevine Viruses and Virus-Like Disease (ICGV) Meeting: August 31 - September 4 in Dijon, France (Judith Monis)

Hybrid Purity Lab: Genetic Services

By Russell Maxwell



At Eurofins STA Laboratories (ESTA), we routinely use protein analysis for hybrid purity, inbred uniformity and/or varietal identification testing. This is accomplished using a method of gel electrophoresis known as isoelectric focusing (IEF). Now, everyone knows that IEF is a technique for separating proteins by their electric charge differences. This results in multiple clean, clear, crisp bands that can differentiate between individual varieties, inbred parents and used to determine percent hybridity.

As I said, everyone knows this. But have you ever thought about what happens to your seed samples when they arrive at ESTA? From a customer's point of view, getting results is pretty straight forward, as it should be. You send in your samples for a hybrid purity test and in a couple of weeks, sooner if a 'rush order' is requested, you get your results. Simple. But have you ever wondered what happens with your samples in our lab?

When a sample arrives, it first goes to our receiving/data entry/administrative department, where the sample information is entered into our sample tracking database. The information entered includes; specific client data, species, variety, lot number, test requested and completion date. An order form is printed for review by

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Electronic Newsletter Coming Soon!

Eurofins STA is going green! In an effort to proactively save natural resources, we will begin sending our newsletter electronically. Starting with our next issue, the information you value will be conveniently emailed to your inbox and viewable on our website. Sign up to begin receiving our electronic newsletter today!

If you already receive our newsletter via email, there is no need to sign up again. If you do not, no problem. Just register at <http://www.eurofinsus.com/stagreen.html>.



The Move is Complete

By John Mizicko

It gives me great pleasure to announce that Eurofins STA Laboratories has finally completed the renovation of our greenhouses at our new facility at 7240 Holsclaw Road in Gilroy, CA. Although we had physically moved our staff and laboratory work to the new facility almost a year ago, we had to continue testing our BFB samples at the old facility. This was due to the fact that the greenhouses at Holsclaw Road were not set up to do BFB testing and we had to undergo extensive renovation of them to bring them up to our stringent requirements. Originally, we had planned to do this renovation in two major stages over a period of two years due to the large capital expenditure needed for the renovation. It was fortunate that the purchase of STA Laboratories by Eurofins

occurred shortly after our move from the Monterey Frontage Road facility. Recognizing the importance of the greenhouses to our BFB testing and of that testing to the success of STA Laboratories, Eurofins provided the assets needed to complete the renovation in one stage.

Work began in earnest in November and with a few unexpected delays was completed in mid March. We now have three state of the art greenhouses of approximately 3000 sq ft each. This expands our BFB dedicated greenhouse space from 7200 sq ft at the old facility to 9000 sq ft here at the Holsclaw site. The three old greenhouses were completely gutted, the ground was regraded, concrete floors were poured, heat retention cloth was installed, high

intensity lighting was installed, the misting systems were put in, new rolling benches were built

and installed, and a weather station-linked computer control system was added to monitor and control all aspects of the environment within each house. These houses are now tailor-made for our BFB testing program.

Since our new facilities have dedicated greenhouse and growth chamber space for other pathology testing, we no longer have to give up space in the BFB greenhouses for these other tests. At the old site, we had to dedicate 2-4 benches in each of the two houses for other pathology programs.

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Move Continued

This reduced the number of BFB samples we could plant in each house by 2-4 10K samples. With our new, renovated greenhouses, all the bench space is dedicated solely to BFB samples. The “pathology” greenhouse at our new site is divided into five separate sections with individually controlled temperature and lighting. We will be taking one of the sections in this “pathology” greenhouse and converting it to BFB testing. It currently only lacks a misting system and that should be in place by mid June. The addition of this section will increase our dedicated BFB space by another 1500 sq ft.

As you can see, Eurofins STA Laboratories continues to invest in the physical resources of our BFB testing program. We firmly believe that the greenhouse grow out method is still the most reliable for detecting seed borne BFB in cucurbit seed samples. It does not mean that we have given up on other methods such as IMS/PCR, but the greenhouse method is still the bench mark in the industry and the one we will always compare new techniques to.

Hybrid Purity Continued

personnel in the Hybrid Purity (HP) Laboratory, and if everything is in order the form is signed to acknowledge acceptance. A copy is then sent back to the client, and the sample is moved into the incoming samples area in the HP laboratory along with the Order Form and a generated Lab Sheet that is specific to the individual sample. Using a work-in-progress (WIP) list the technicians in the HP lab prioritize the samples for protein extraction. The variety is checked against our genetics database that contains the protocols (gels, stains, etc.) necessary for the sample(s) being tested.

The sample seeds are then loaded into a 96-well plate where the seeds are crushed and an extraction buffer is added to each well. This is another advantage of using IEF: we do not have to wait for germination to extract protein from the root or leaf. Depending on the species and staining method employed, the sample may require as long as 12 hours in the extraction buffer. Once the samples have completed the necessary extraction time the samples are loaded onto a 96-lane gel and run through the IEF process, which takes approximately 1½ hours. At the end of the IEF process, the gel is removed from the electrophoretic gel box and put into a glass or plastic casserole dish and placed on a circulating bath. At this time the separated proteins in the gel are stained which can generally be poured directly on the gel. If this is the case, it takes between 5- 30 minutes to visualize the bands. Some stains do require more time.

Once the gels are stained they are analyzed by the HP Associate Technician. The score and any other pertinent information is detailed on the lab sheet, dated and initialed by the analyzing technician. The information from the lab sheet is then entered into the sample tracking database which can then be used to generate a ‘Certificate of Analysis’. This certificate is then sent to the administrative department where it is either faxed or emailed back to the customer.

The HP lab and ESTA pride ourselves on customer service and customer satisfaction by producing accurate and reproducible data on a timely basis. If you have not tried our service, you should. And if you have any questions, please contact us. We will be more than happy to answer any questions you may have.

While it gave me great pleasure to announce the opening of our new greenhouses, it saddens me to announce that Anita Castro Sparks, who so adeptly led the BFB program here at our California site, has moved back to the University of Georgia. Anita has taken a position as Lab Manager for Dr. Ron Walcott – she is back where Eurofins STA had obtained her from. So although she no longer works for Eurofins STA, she is still closely linked to the seed industry and the BFB issue. We will continue to draw on both her and Ron’s experience and expertise with this significant pathogen. Eurofins STA Laboratories is now in the process of locating a replacement for Anita to head up the BFB program here in California. In the interim, the program will be run by Aline Reno, who was Anita’s technician and had been trained by Anita and was (and is) an integral part of the BFB program. If you get the chance, stop by and see the new greenhouses and meet Aline.

Changes in the Future Continued

- seed industry, North America and our international colleagues
- Reduce duplication in association administration, leadership committee work and is a more efficient use of financial resources
- Enhances the ability to determine seed research needs and conduct research to improve seed testing evaluations through improved communications
- Better able to develop standardized seed testing methods

Benefits to the Seed Industry

- Streamline communications when the industry needs to communicate concerning their seed testing needs
- Provide the seed industry with standardized uniform seed evaluation methods
- Continued high quality research that meets their seed evaluation needs
- Allow for one accreditation system for all of the seed technologists
- A standardized and uniform set of Rules for use in seed quality assessment, seed labeling, seed law enforcement and certification of seed will be developed by one organization



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