Operating a State-wide Strawberry Disease Diagnostic Services Center

Principle Investigator
Steven T. Koike
Lab Director
TriCal Diagnostics
8100 Arroyo Circle
Gilroy, CA 95020
(831) 637-0195
SKoike@trical.com

Cooperating Investigators
Mark Bolda
UC Cooperative Extension, Santa Cruz County

Oleg Daugovish
UC Cooperative Extension, Ventura County

Tom Gordon
Department of Plant Pathology, UC Davis

Frank Martin
USDA-ARS, Salinas

Summary
This strawberry diagnostic program is a service-oriented extension activity. With the support of the California Strawberry Commission and the strawberry industry, we operate a diagnostic lab in Salinas that focuses on diagnosing problems for strawberry growers throughout California. This diagnostic service is intended to confirm the presence of common, well known strawberry pathogens as well as to be in the position to detect new diseases and problems if these might arise. We continue to offer culture-based soil tests for Verticillium and Macrophomina inoculum levels. Diagnostic services are available to all segments of the strawberry industry, ranging from large operations to limited resource growers, and includes transplant growers, production growers, pest control advisors, and other field professionals. Lab findings are communicated to growers and also to other strawberry researchers, increasing the opportunity for collaborative research. Our location on the central coast also facilitates timely analyses for those who submit samples from this region.
In 2016 through 2017, our lab continued to be called upon to confirm cases of the three major soilborne pathogens: *Macrophomina, Fusarium, and Verticillium*. While all three pathogens remain significant concerns for growers in all the coastal strawberry production regions from Ventura in the south through Santa Cruz in the north, there appeared to be significant increases in the number of *Fusarium* cases throughout the state. In 2016-2017, our lab confirmed numerous cases of anthracnose caused by *Colletotrichum acutatum*; the development of anthracnose was clearly linked to rainfall that occurred this season.

**INTRODUCTION**

The general goals of the project are the following:

1. **General strawberry diagnostics.** Appropriate strawberry pathogen assays were standardized and implemented at the UC Cooperative Extension lab in Salinas, California. Standard protocols were used for testing plants for *Macrophomina, Fusarium, Phytophthora, Verticillium, Rhizoctonia, Colletotrichum, Cylindrocarpon*, and *Xanthomonas*. Leaf blotch disease caused by Zythia is identified by examining leaf lesions for the characteristic fungal structures. Soil samples were tested for *Verticillium* and *Macrophomina* propagules using a dry sieve method and NP-10 semi-selective medium. Our diagnostic services also identified non-pathogen problems such as plant establishment problems due to stress and production issues, and fruit bronzing due to environmental factors.

2. **Statewide diagnostic services.** To increase services to growers in various strawberry production areas in the state, we enlisted the involvement of Cooperative Extension personnel in various counties and researchers at UC Davis and the USDA-ARS in Salinas, CA. Services were provided to strawberry growers, transplant producers, and pest control advisors from throughout the state.

3. **Coordinate with other researchers working with strawberries.** To integrate this extension effort with the overall research program for strawberries, we communicated with various UC, USDA, and Cal Poly SLO researchers, so as to inform them about pertinent lab findings. We are actively collaborating with Tom Gordon (UC Davis) and Frank Martin (USDA) on the *Macrophomina* and *Fusarium* soilborne disease concerns. We consulted with Bill Wintermantel (USDA) regarding possible virus disease cases.

4. **Focus on current soilborne issues.** Because of the importance of *Macrophomina* and *Fusarium* soilborne problems, we particularly focused on strawberry dieback cases and advised growers on this issue. This aspect of the diagnostic program, along with confirmation of Verticillium wilt, comprised the majority of the samples received in 2016-2017.
**METHODS**

Submitted plant samples are always tested for general pathogens using non-selective, non-specialized media (acidified corn meal agar and other agars for fungi; sucrose peptone agar for foliar bacteria). As deemed appropriate, other selective, specialized media are added to the isolation protocols (PARP for Phytophthora and Pythium, NP-10 for Verticillium and Macrophomina, FS for Fusarium, modified Mather’s for Colletotrichum). We particularly focused on the developing Macrophomina and Fusarium problems on strawberry. Collapsed strawberry plants were always examined specifically for both of these soilborne pathogens as well as for Verticillium. For soil, the dry sieve soil assay and NP-10 medium are used for both Verticillium and Macrophomina.

Recently, molecular detection methods have been developed that do not require DNA extraction, and the amplification of target DNA can be done at a single temperature (hence the term isothermal amplification) using a low-cost piece of equipment. One example of this method is recombinase polymerase amplification (RPA); using this technology the Martin lab has designed diagnostic assays that will detect Phytophthora, Macrophomina, and Verticillium of strawberry. This method can help the strawberry industry because soilborne disease diagnostics can be accomplished in a matter of hours instead of days. This RPA method is now being used on a regular basis in the UCCE lab. Subsequent research from the USDA Martin lab should later add RPA assays for the Fusarium wilt pathogen of strawberry.

For further information on this new RPA tool in the UCCE lab, see the following blog article by Koike and Bolda: Rapid diagnosis of soilborne diseases of strawberry and other crops: [http://ucanr.edu/blogs/blogcore/postdetail.cfm?postnum=24140](http://ucanr.edu/blogs/blogcore/postdetail.cfm?postnum=24140).

**RESULTS AND DISCUSSION**

Because the dieback symptoms caused by Verticillium, Macrophomina, Fusarium, and Phytophthora are very similar, laboratory testing will be required to identify which pathogen is involved. Using both culture and RPA methods, we regularly tested for and detected the soilborne pathogens Macrophomina, Phytophthora, and Verticillium. Culture tests can consistently recover the strawberry pathogen Fusarium oxysporum f. sp. fragariae; we are awaiting the development of an RPA assay for this Fusarium pathogen. The success of the new DNA-based RPA technique has made this method a standard part of our diagnostic services for soilborne pathogens. Our lab also handled and identified the standard foliar/fruit diseases such as angular leaf spot, leaf blotch, powdery mildew, gray mold, Rhizopus/Mucor fruit rot, and other miscellaneous problems.

**EXTENSION**

Diagnostic findings were communicated with growers and PCAs who submitted the samples. Disease developments were highlighted in our UC Cooperative Extension newsletter (Crop Notes) and also the extension blog sponsored by Farm Advisor Mark Bolda ([http://ucanr.org/blogs/strawberries_caneberries/](http://ucanr.org/blogs/strawberries_caneberries/)). Important findings from our diagnostic lab were also communicated to Spanish-only speakers by translating and posting the articles on the Spanish language blog ([http://ucanr.org/blogs/fresamora/](http://ucanr.org/blogs/fresamora/)).
ACKNOWLEDGMENTS

The California Strawberry Commission and the strawberry industry in California have been consistently supportive of this program. We thank the California Strawberry Commission for making this program possible. We thank the growers and pest control advisors who have participated in this service by submitting samples. We acknowledge the following persons for their assistance: Mark Bolda, Oleg Daugovish, Tom Gordon, Sofia Hernandez, Dan Legard, Frank Martin, Stacy Mauzey, Tim Miles, Cayla Tsuchida, Bill Wintermantel. We thank Eric Lauritzen (Monterey County Agricultural Commissioner) for his assistance with the greenhouse and laboratory facilities at UC Cooperative Extension—Monterey County.

REFERENCES


