Project Title: Integrated management of bacterial spot and bacterial speck of tomato

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In the southeastern U.S., fresh-market tomato production accounts for over 50% of harvested acres and 63% of yield nationally. In 2004, 126,400 acres of tomatoes were harvested in the U.S. for the fresh market industry, representing 1.34 billion dollars (National Agricultural Statistics Service). However, tomato yield and quality are threatened by bacterial diseases for which effective control measures are limited. Bacterial spot of tomato, incited by *Xanthomonas campestris* pv. *vesicatoria*, and bacterial speck of tomato, incited by *Pseudomonas syringae* pv. *tomato*, have been identified as major production problems in the southeastern United States. Bacterial spot is a major problem in Florida where high temperatures and moisture conditions prevail during the growing season. In the past few years, this pathogen has become a significant problem in North Carolina, where it has been identified throughout all tomato production areas of the state. Bacterial speck may also cause serious losses in KY, NC, SC and TN. In the spring 2005, bacterial speck caused significant problems on tomatoes in Florida. Although bacterial spot is typically a major problem in Florida, the pathogen has not been known to cause significant damage to tomatoes in North Carolina until recently. Growers apply copper plus mancozeb at least twice weekly in an attempt to control this disease. However, the disease is not effectively controlled when environmental conditions are optimal. Furthermore, control is hampered by the presence of copper-tolerant strains and the endemic nature of the pathogen. Antibiotics such as streptomycin have been ineffective for many years as a result of the development of resistant strains. Bacterial speck has emerged as an economically significant disease in the mountains of North Carolina. The disease is now one of the most persistent bacterial disease problems found in the tomato growing regions of North America. Replicated trials in 2000 demonstrated a fruit speck incidence of up to 46% with a 36% associated decrease in marketable yield.

There is an identified priority to develop commercially acceptable IPM programs to limit extensive losses growers currently encounter. Our team is well positioned to address this priority and the industry is currently at a critical juncture - EPA is currently evaluating the widespread use of copper in agricultural systems through their Registration Eligibility Decision (RED) process and has expressed concern about the "amount applied and frequency of application". Several alternative disease management strategies have shown efficacy against certain bacterial pathogens. Among these, bacteriophages and the plant activator, acibenzolar-S-methyl (Actigard; Syngenta Crop Protection, Greensboro, NC) are most promising. In field studies where Actigard, a compound which induces natural protection of plants against pathogens in tissues remote from the initial treatment and which is termed systemic acquired resistance (SAR), was applied, bacterial spot disease severity was significantly reduced in fresh-market and processing tomatoes without significant yield responses. In another study conducted in Florida and supported by S-RIPM funding, Actigard effectively reduced disease without significantly improving yield. In greenhouse studies applications of Actigard stimulated defense responses as manifested by development of necrotic lesions on leaves which in further tests were confirmed to result from a hypersensitive response. Given that Actigard applications activate strong defense responses, further research is needed to reduce negative effects on yield responses.

Bacteriophages (bacterial viruses) have significant potential as biocontrol agents for controlling bacterial diseases pathogens. An approach that uses a mixture of bacteriophages was developed to overcome problems experienced in the past. We tested mixtures of bacteriophages for control of the bacterial spot disease on tomato on overhead irrigated tomato transplants and observed that irrigation water containing bacteriophages specific to the bacterial spot pathogen reduced disease incidence compared to copper. Furthermore, in field studies, bacterial spot

severity was significantly reduced in phage-treated plots compared to copper-mancozeb or the control plots, while yield was also significantly improved. As a result of previous funding from S-RIPM, formulated bacteriophages were identified which significantly enhanced longevity of phages under greenhouse and field conditions resulting in greater efficacy against the bacterial spot pathogen. In field tests, all formulations except the non-formulated phage significantly reduced disease severity attributed to bacterial spot compared to the control and standard copper-mancozeb treatment. We have been leaders in developing this technology and success in tomato systems will provide a basis for success in other pathosystems.

Although streptomycin resistance has been documented in *Xcv* populations, there are other experimental antibiotic bactericides that have shown activity against this pathogen. Kasugamycin, currently manufactured by Arvesta Corporation, is used in South America for managing bacterial pathogens on fruit trees. Efficacy studies with Kasugamycin in Georgia this past year on bacterial spot of pepper, also caused by *Xcv*, showed that this antibiotic was effective at controlling this pathogen in greenhouse studies (Dr. David Langston, University of Georgia, Tifton). Although this compound is still in the experimental stage, the Arvesta Corporation is working on the development of Kasugamycin as a systemic bactericide due to promising data collected from various field and toxicological studies.

Host resistance to pathogens is a critical component of integrated disease management systems. Varieties and genotypes with resistance to bacterial spot have been developed in the University of Florida breeding program, and are in various stages of development in North Carolina. Some tomato varieties carry the Pto gene for resistance to race 0 of the bacterial speck pathogen. Several different commercial varieties showed variable levels of tolerance to bacterial spot during the 2005 growing season in North Carolina. Further complicating the use of plant-derived resistance to control bacterial spot is the fact that *Xcv* composes a very heterogeneous group of strains. Actigard was shown to effectively delay race changes and may increase the durability of genotype-specific resistance. Therefore, deployment of varieties with moderate levels of resistance to bacterial diseases coupled with the use of SAR inducers may increase durability of resistance. SAR inducers in tomato show promise for future disease management.

Recently, as part of S-RIPM funding, we have initiated an intensive study focusing on an integrated approach for controlling bacterial spot (Jones, Momol and Olson). Several greenhouse experiments were conducted and included the use of various combinations of bacterial biocontrol agents (antagonistic and PGPR strains), bacteriophages specific to the target bacterium (race T3 strain of *Xcv*), and compounds that induce systemic acquired resistance (SAR) in the plant. Antagonistic and PGPR strains did not reduce disease severity attributed to the bacterial spot.

In this proposed project we plan to expand the use of these newly available tools to include multiple races of the bacterial spot pathogen and the bacterial speck pathogen. The main purpose of this project is to evaluate new and/or alternative bacterial disease management approaches and to integrate the most promising ones into comprehensive, environmentally and economically viable strategies to manage these two major tomato bacterial diseases. Results derived from the proposed studies will be used to help design a multi-faceted IPM system for controlling these important bacterial diseases of tomato in the southeastern United States.

Manner in which this project addresses Southern Region Priorities

Due to the destructive nature of these diseases, growers sometimes panic at the prospect of serious loss and apply copper bactericides judiciously for foliar bacterial diseases. Some of the proposed tactics in this project will help to reduce such wasteful use of pesticides. As field trial results become available, treatments that provide better bacterial disease suppression will give

growers more options to increase yield and improve the economics. Integrating biological and cultural control approaches within conventional production systems can help minimize pesticide resistance in pathogen populations and decrease adverse environmental and human health risks. This mission oriented project supports the long-range goal of improving the sustainability of U.S. agriculture by reducing high risk chemical use through environmentally reduced-risk tactics. Results of this research may also provide innovative, basic information for future studies on using bacteriophages and Kasugamycin in other bacterial pathosystems.

The PIs will directly extend results of the research to other extension professionals, and to growers in all major tomato productions areas of U.S., and will write publications on results of the research for refereed and trade journals so that the research will benefit research and extension personnel on bacterial speck and bacterial spot disease management of tomatoes nationwide. The results will be extended to national and international audiences at the American Phytopathological Society meetings, vegetable grower workshops and annual meetings, other related happenings (county extension programs), and tomato field days. Potential users of this research are farmers, consultants, IPM providers and agricultural extension personnel in the southeastern US. This work should produce economic, practical and effective management options for controlling bacterial speck and bacterial spot on fresh-market tomatoes.

b. Objectives

1. To Optimize Integrated Management of Bacterial Spot and Bacterial Speck with the SAR inducer, Actigard, which has Limited Effects on Plant Yield by:
    a. Identifing resistant lines to determine if they respond to PGPRs and if lower rates of Actigard can be applied to enhance disease control without affecting yield in the field.
    b. Comparing the effects of modified application strategies of the SAR inducer (Actigard) in combination with bacteriophage applications.
2. To Evaluate Kasugamycin (Arvesta Corporation) for Management of Bacterial Spot and Bacterial Speck by:
    a. Screening isolates of *Xcv* and *Pst* for sensitivity to Kasugamycin *in vitro*.
    b. Evaluating efficacy of Kasugamycin applications for management of bacterial spot and bacterial speck in greenhouse and field trials.
3. To Combine the Best Management Strategies in Objectives 1 and 2 for Bacterial Spot and Bacterial Speck in Field Experiments.

Description of ways the project addresses Southern Region Priorities (Part I.B.2)

Multi-state: The proposal addresses multi-state needs in terms of significantly improved disease management of major disease problems present throughout the Southern region including AL, FL, GA, KY, NC, SC and TN.

Biologically-based: This proposal is based on an integrated approach for implementing biological control using bacteriophages and host resistance to reduce disease losses. Furthermore, we are using this approach in an attempt to reduce the amount of copper and EBDC compounds applied.

Stakeholder-identified IPM priorities: We have included several letters from stakeholders in North Carolina and Florida indicating the need for significant improvements in disease control of bacterial diseases associated with tomato. Among these were letters from the Florida Tomato Committee, Glades Crop Care which works closely on IPM related aspects of disease and insect control with growers in Florida and Georgia, The GADSDEN TOMATO GROWERS ASSOCIATION, INC, in Gadsden County Florida, United Agri Products in North Carolina, and The North Carolina Tomato Growers Association.