Research Title: The role of soil fertility on the severity of Fusarium wilt of watermelon (*Fusarium oxysporum* f. sp. *niveum*)

Project details: Fusarium wilt of watermelon, caused by the fungal pathogen *Fusarium oxysporum* f. sp. *niveum* (FON), has been a serious threat for the watermelon production in the United States. The pathogen causes pre- or post-emergence damping-off of young seedlings and wilting of mature plants in the field. A unilateral stem necrosis is a characteristic symptom of Fusarium wilt, which can be easily, visualized when runners or stems are sectioned. Under favorable conditions, yield losses more than 80% can be experienced. Several management strategies like seed treatment, crop rotation, weed control, and chemical management options are available but in most cases, they are inadequate to manage FON epidemic. Apart from these strategies, a different approach to the integrated disease management of FON is required. Being a soil borne fungus, soil fertility and soil mineral nutrition may play an important role in FON infection and severity of symptoms. However, very little information is available on this aspect. Hence, the overall goal of the proposed research is to determine the role of soil fertility on the severity of Fusarium wilt of watermelon.

Preliminary data on other pathosystem: In two years of field studies, we have developed several significant models based on cation concentrations of pepper foliage and soil and Bacterial Leaf Spot (BLS) severity. These models are comprised of Cu, Fe, Mn or Zn as major contributors alone or in different ratios. These cations also act as cofactors for superoxide dismutase (SOD) enzymes that detoxify reactive oxygen species produced in plants upon pathogen attack. As a result, hydrogen peroxide is formed, which acts as precursor for salicylic acid (SA) formation. SA has been proposed as the signal molecule to initiate the systemic acquired resistance (SAR) pathway. We have found a strong evidence of SOD enzymes involvement in these models as seen by the effects of increased levels of Cu, Fe or Zn on the relative gene expression for the three major classes of SODs (Cu-Zn SOD, MnSOD and FeSOD) in pepper tissues. We also observed that increased levels of SA, MnSOD activity in plants showing less BLS severity than in plants with severe BLS symptoms, thereby providing evidence of a SAR response. Furthermore, the BLS-cation models also provided good fit for relative expression of the SOD genes; Cu-ZnSOD (P= 0.05; adj.R^2=0.92) and MnSOD (P= 0.009; adj.R^2=0.98). Relative expression of the NPR1 gene (non-expresser pathogenesis-related protein, another component of SAR pathway) fit a model consisting of Cu-ZnSOD, FeSOD, and MnSOD expression levels (P< 0.0001; adj. R^2=0.93) indicating a relationship between SOD genes and NPR1 gene.

This phenomenon was also observed in *Nicotiana tabacum* when challenged with *Tomato spotted wilt virus* (TSWV). Tobacco plants with low TSW severity ratings had higher expression levels of MnSOD and NPR1 genes than compared to plants with high disease severity (data not shown). Moreover, highly significant and predictive TSW models were developed using Cu, Fe, Mn or Zn cations alone or in combination with one another. One of the models was validated, when we predicted TSW severity in a field study (2014). Based on a survey of soil cation concentrations at the Bowen Farm, UGA, Tifton, we predicted TSW severity (%) by plugging concentrations of key cations in predictive TSW model. Based on model predictions, two sites (approximately 100 m apart) were selected as high- and low-TSW risk areas prior to tobacco planting. When plants were rated for TSW prior to harvesting, plants at the low-risk sites had 4.5% TSW severity compared to 33.1% disease severity at the high-risk site. These data validated our model in terms of explaining the patterns of disease variation in the field. Such disease predictions should also be evaluated for BLS severity in pepper using models developed from soil mineral analysis. Preliminary data indicate that such predictions are possible.

The objectives are:
1) Determine if soil mineral nutrition/fertility is related to Fusarium wilt of watermelon severity under field conditions.
2) Validate field observations in a controlled environment in pot cultures with key cation ratios in watermelon that induces systemic acquired resistance.
3) Quantify salicylic acid (SA), an early product in the SAR pathway, from watermelon plants treated with key cation ratios.

Objective 1. Watermelon transplants (cv. Sugarbaby) will be planted in the field (Black Shank Farm, Coastal Plain Research Station, Tifton, GA) and at planting, seedlings will be inoculated with conidial suspension of 10^5 conidia per ml of FON race 1. After 2 weeks of inoculation, plants will be scored based on different severity level (low, moderate and severe). Composite soil samples from each severity level will be sampled and elemental ratios will be determined. Elemental ratios will be correlated with levels of disease severity.

Objective 2 and 3. Key cation ratios identified in the objective 1 will be used for the pot culture experiment under greenhouse conditions. Watermelon seedlings (cv. Sugarbaby) will be established in pots with pasteurized sand. After two days of planting, key cation ratios will be applied in the form of foliar or soil chelates. A conidial suspension of 1ml of FON containing 10^5 conidia/ml of FON race 1 will be drenched near base of the seedling. Seedlings treated with sterile water will serve as negative control. After three weeks of inoculation, seedlings will be scored for FON severity. Concurrently, leaf samples from each treatment will be taken and send for mineral analysis. Leaf samples will also be analyzed for the SA concentrations using spectroscopy technique. Later, cation ratios will be correlated with levels of disease severity and SA concentrations. Several regression models will be made to deduce relationship between cation ratio in watermelon seedlings and FON severity.

Timeline of the project: The timeline of the project is one year **(Jan, 1, 2016 to Dec, 31, 2016)**

Dates of periodic reports to be submitted: 06/1/2016 and 12/31/2016

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NWA priority area: Fusarium wilt of watermelon.

Budget:

1. Funds are requested for laboratories supplies necessary plating and PCR confirmation of FON, and Gas chromatography for Salicylic acid, field plot material and supplies include: fertilizer, onion transplants, fungicides and herbicides. Amount: $6,500

2. Funds for buying transplants, greenhouse operation and soil/plant tissue chemical composition analysis (@$24/sample) = $3000.

3. Salary for an undergraduate worker for 6 months with $9/hr for 20hr a week. Amount: $4730

Total funds: $14,230

Addendum

Fusarium wilt of watermelon caused by a soilborne fungus (*Fusarium oxysporum* f. sp. *niveum*) (FON) has been an emerging threat to watermelon production in the United States. In Georgia (2015), outbreaks of Fusarium wilt have been reported from 12 counties resulting in huge economic losses. Various management options including crop rotation, seed treatment, chemical management, use of cultivar resistance, cover crop have been routinely employed by the growers; however, Fusarium wilt continue to occur every year. We are proposing a different approach to the integrated disease management of FON. Being a soil borne fungus, soil fertility and soil mineral nutrition may play an important role in FON infection and severity of symptoms. However, very little information is available on this aspect. Hence, the overall goal of the proposed research is to determine the role of soil fertility on the severity of Fusarium wilt of watermelon. In this proposal, we aim to investigate relationship of soil fertility to Fusarium wilt severity and host resistance. We have various level of success in understanding this concept on different diseases including bacterial spot of pepper and sour skin of onion. We will use our experience and expertise in understanding this concept in FON. Hopefully, various factors of soil fertility may influence host resistance in watermelon against FON and in future may translate to a management strategy.

Budget justification:

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