IFAS Citrus Initiative
Annual Research and Extension Progress Report 2012-13

Project Title: Fate of Indicator Organisms on Citrus in the Field.
Investigator: M.D. Danyluk
Objective(s) Pursued: Evaluate the fate of coliforms and *E. coli* sprayed onto Hamlin and Valencia trees in the field until *E. coli* populations can no longer be detected.
Progress on Objectives:
Detailed Accomplishments in 2012-13:

To evaluate the fate of coliforms and *E. coli* that may be sprayed onto oranges close to harvest if low quality water is used, low quality water (ca. 2.5 and 4 log CFU/ml *E. coli*; low count (l) and high count (h) water) was applied to three citrus trees the highest application rate that may be applied. Experiments were conducted at the Citrus Research and Education Center on Hamlin fruit in October, November, December, January, and February, and to Valencia fruit in March, April, and June (8 months). Oranges (3 x 10 fruit samples) from canopies of each of three trees (n=90) were removed before and immediately following spraying, at 2 and 6 h following application, and approximately every other day until *E. coli* was no longer detectable by enrichment in two subsequent samples or no fruit remained on the trees. Coliform/*E. coli* were enumerated initially by plating onto *E. coli*/coliform chromogenic agar, and then enriched for using the Coliert Quanti-tray/2000 MPN standard testing kit to allow for a limit of detection of 1 MPN/10 oranges. Temperature, rainfall, relative humidity and other environmental factors were obtained for trial periods from FAWN.

Results for the 2012-2013 trials are included below, including initial inoculum loads (Table 1), and survival of *E. coli* and coliforms on citrus trees, by month (Figures 1-7). A summary of E. coli survival on citrus, including March, April, May, and June 2012 are also included in Figure 8. In four months, March, June, and October 2012, rainfall close to inoculum application appears to have increased survival, or allowed for growth of organisms. Rainfall, and changes of relative humidity, especially within 48 hours of spraying are the major environmental factors contributing to the survival of organisms on fruit. In January, 2013 a slower than expected decline of E. coli could not be attributed to any environmental factors.

Table 1. Microbial counts in low quality microbial waters generated for use in field trials.

	High Count Water (log CFU/ml)		Low Count Water (log CFU/ml)	
Month	TPC	*E. coli*	TPC	*E. coli*
October	5.6	4.3	3.9	0.9
November	6.6	4.0	5.7	2.6
December	5.3	3.9	4.9	2.3
January	6.0	5.1	4.8	2.5
February	5.5	4.5	6.0	2.6
March	5.2	4.4	3.7	2.0
April	5.2	4.0	3.6	2.3

* At no point were *E. coli* or coliform populations detected in CREC well water used as the mixing water to establish the low microbial quality water.

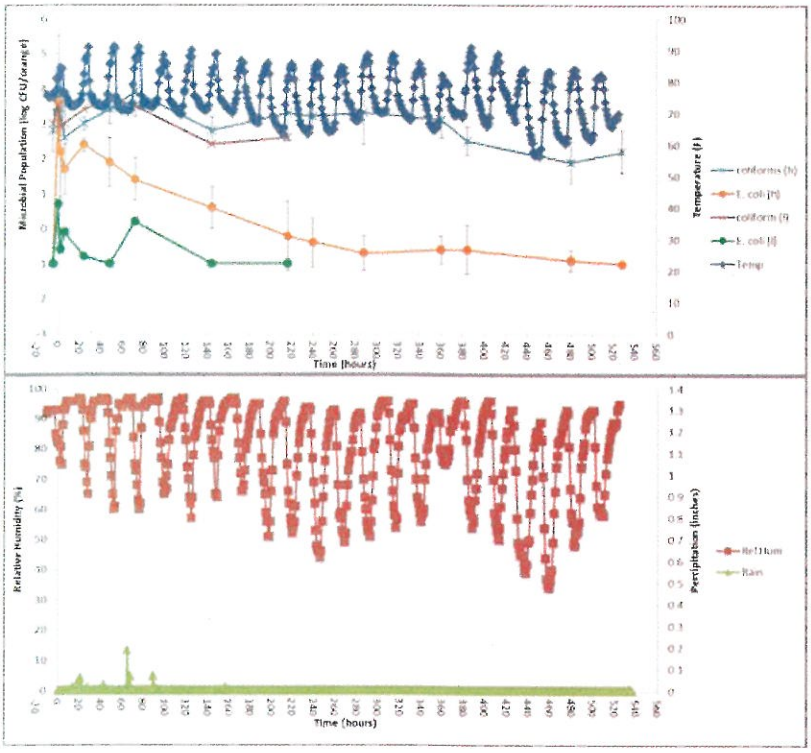

Figure 1. Fate of coliforms/E. coli on citrus in October, 2012

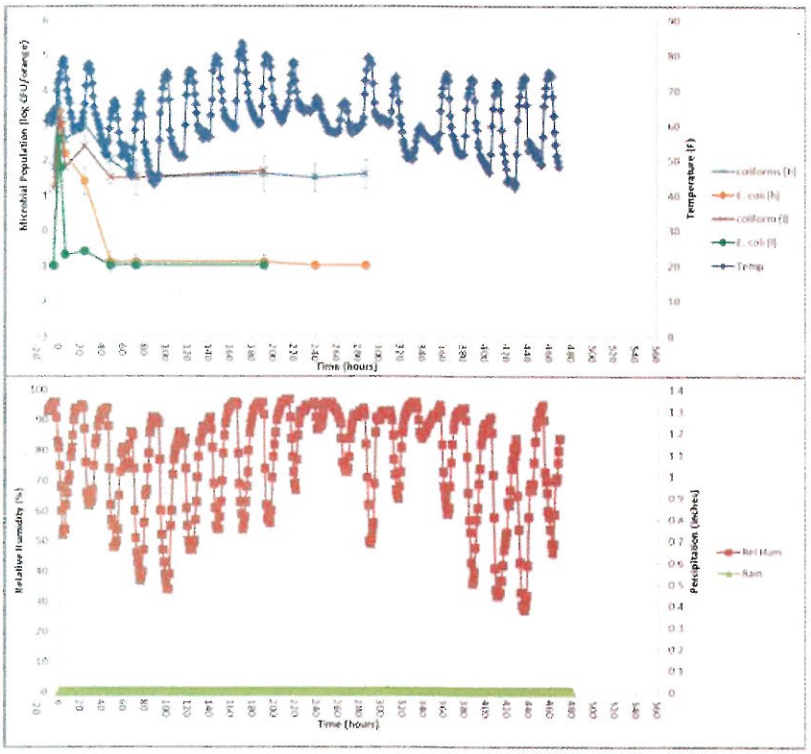

Figure 2. Fate of coliforms/E. coli on citrus in November, 2012

Areas where progress exceeded expectations:

The microbial modeling research group, led by Don Schaffner at Rutgers University has been engaged to help with microbial modeling of field survival to aid in the long term analysis of the data and the development of a model to help establish appropriate pre-harvest intervals on products.

Areas where progress didn't meet expectations:

Due to other commitments in May (cantaloupe packinghouse investigations by FDA), no trial was run in May, 2013. The June, 2013 spray trial is ongoing; the results of this trial are not reported here.

Impact of accomplishments towards overall goals of funding:

The overall goal of this funded project is to generate data on the survival of generic *E. coli* on citrus trees in the field over the course of the typical citrus harvest season. The data generated over the course of the

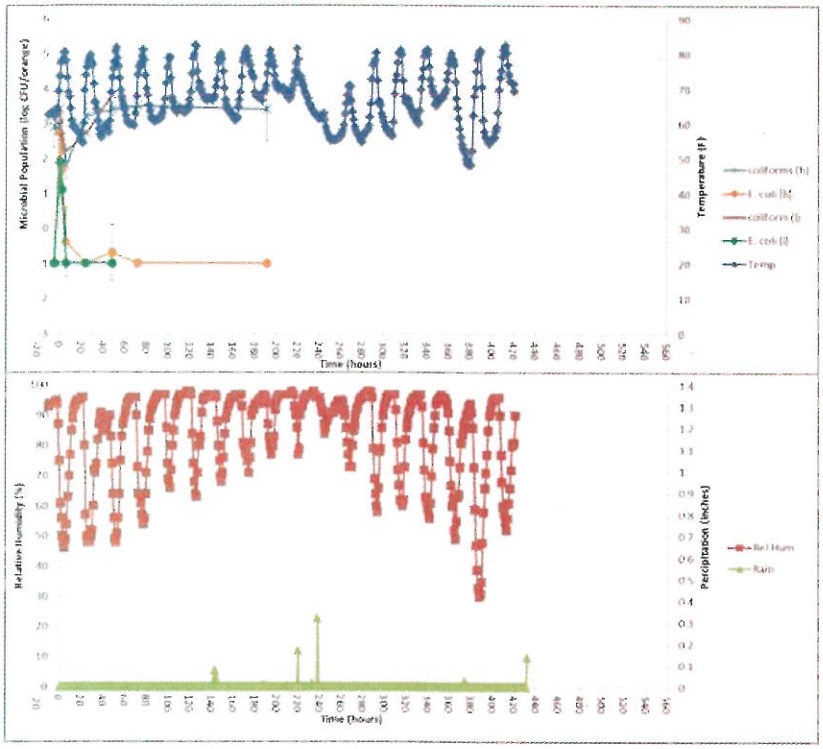

Figure 3. Fate of coliforms/E. coli on citrus in December, 2012

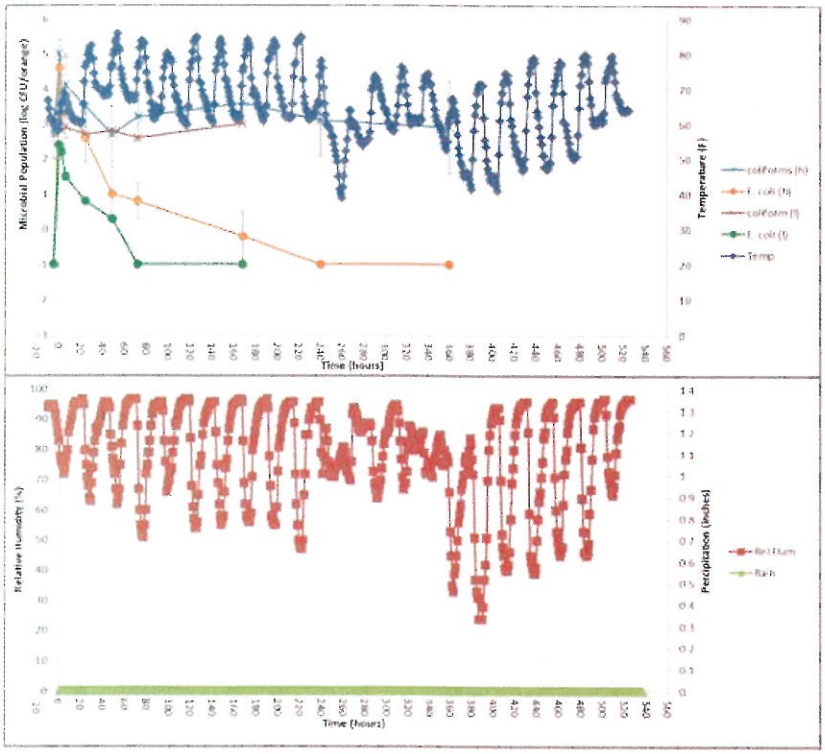

Figure 4. Fate of coliforms/E. coli on citrus in January, 2013

2012-2013 harvest season will be combined with data generated during the 2011-2012 harvest season to begin to set the framework for establishing a pre-harvest interval for the use of crop contact water that may not meet microbial water quality standards proposed in the Produce Safety Rule under the Food Safety Modernization Act.

Presentations associated with 2012-13 efforts:

Danyluk, M.D. and T.M. Spann. 2012. Fate of Indicator Organisms on Orange Trees in the Grove, Packinghouse Day and Indian River Postharvest Workshop, Lake Alfred and Fort Pierce, FL, 2012

Danyluk, M.D. 2012. Update on FSMA, Packing Line Studies and Field Indicator Survival Data. Indian River Citrus League, Vero Beach, FL, 2012

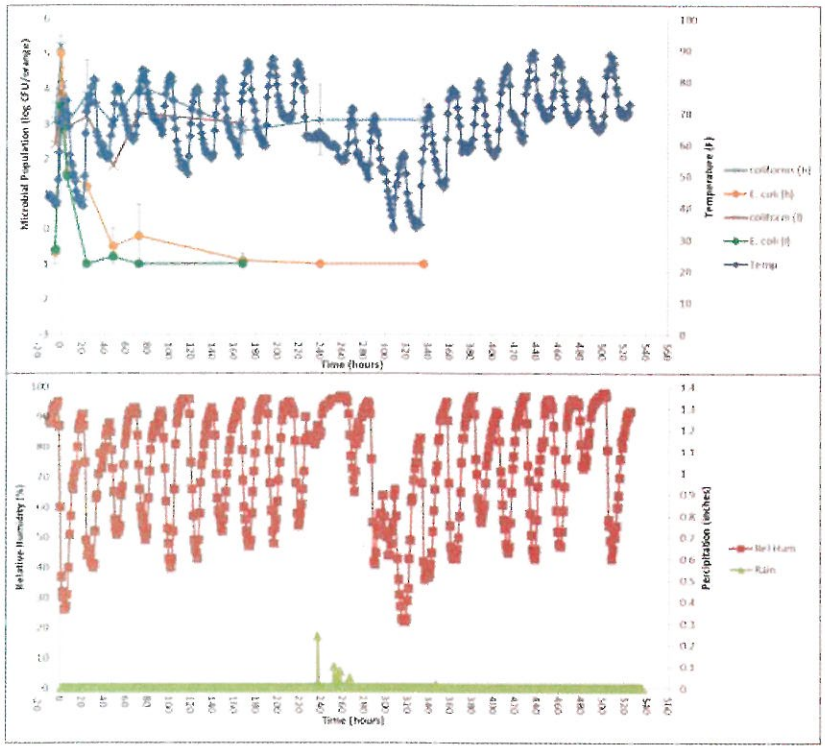

Figure 5. Fate of coliforms/E. coli on citrus in February, 2013

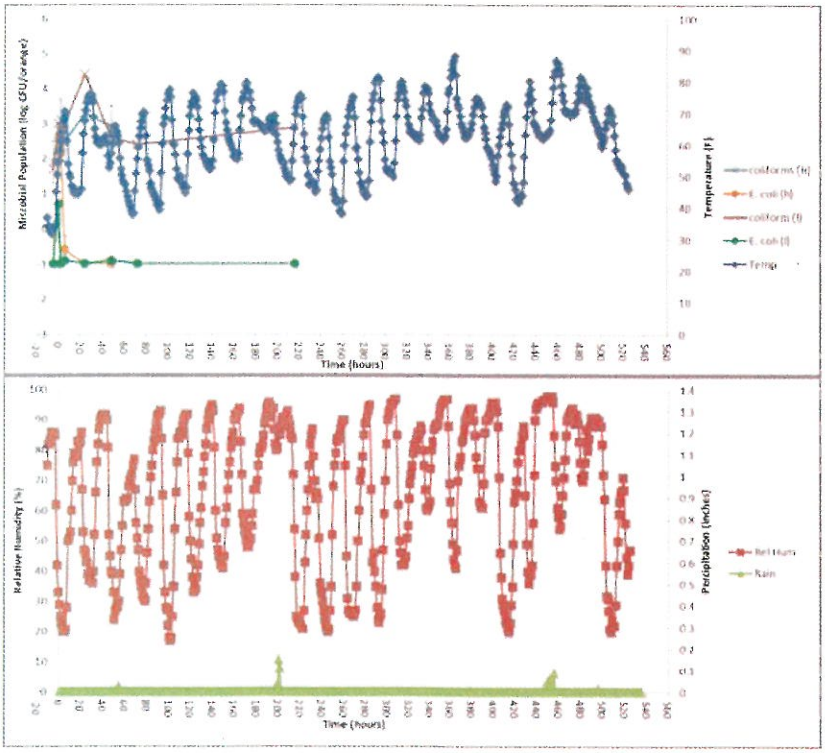

Figure 6. Fate of coliforms/E. coli on citrus in March, 2013

Mootain, G.K., L.M. Friedrich, T.M. Spann, D.W. Schaffner and M.D. Danyluk. 2013 Fate of indicator organisms on oranges following the application of contaminated foliar sprays. Florida State Horticultural Society, Sarasota, FL, 2013.

Mootian, G.K., L.M. Friedrich, T.M. Spann, D.W. Schaffner, and M.D. Danyluk Fate of Indicator microorganisms on oranges following application of low microbial quality water in foliar sprays. To be presented at the 2013 International Association for Food Protection Annual Meeting in Charlotte, NC.

Refereed and non – refereed publications from 2012-13 efforts: None in 2012.

Next steps:

An additional year of funding is being requested to replicate field trials during the 2013-2014 harvest season. A recently published framework document

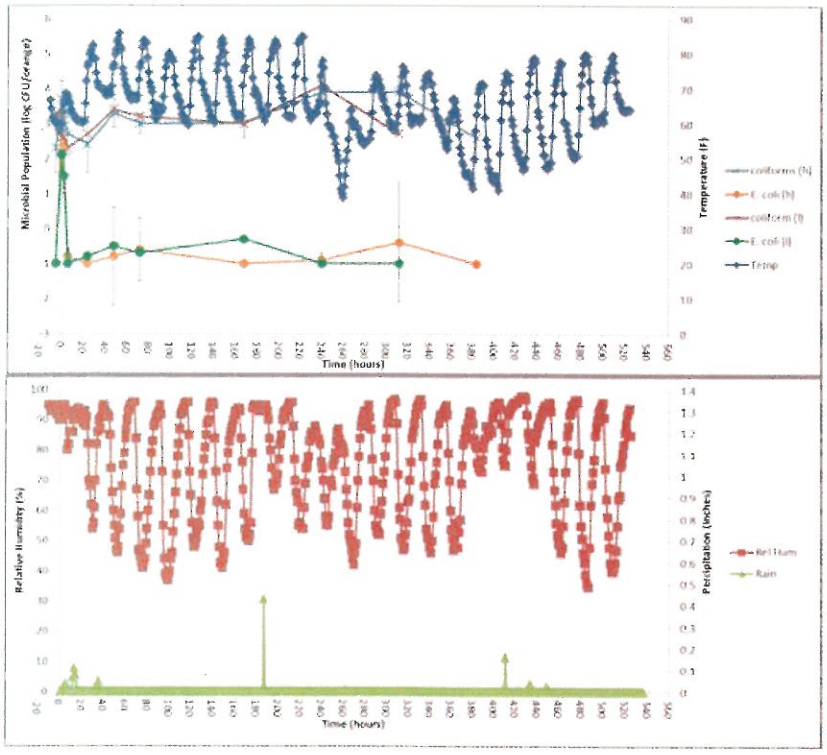

Figure 7. Fate of coliforms/E. coli on citrus in April, 2013

to evaluate research related to low microbial quality agricultural waters recommends multiple years of field trials to establish recommendations for pre-harvest intervals.

Additional funding in 2013-2014 is requested to hold a 1 day workshop for Florida growers, packers and Extension Agents, discussing available technologies to improve the microbial quality of agricultural waters.

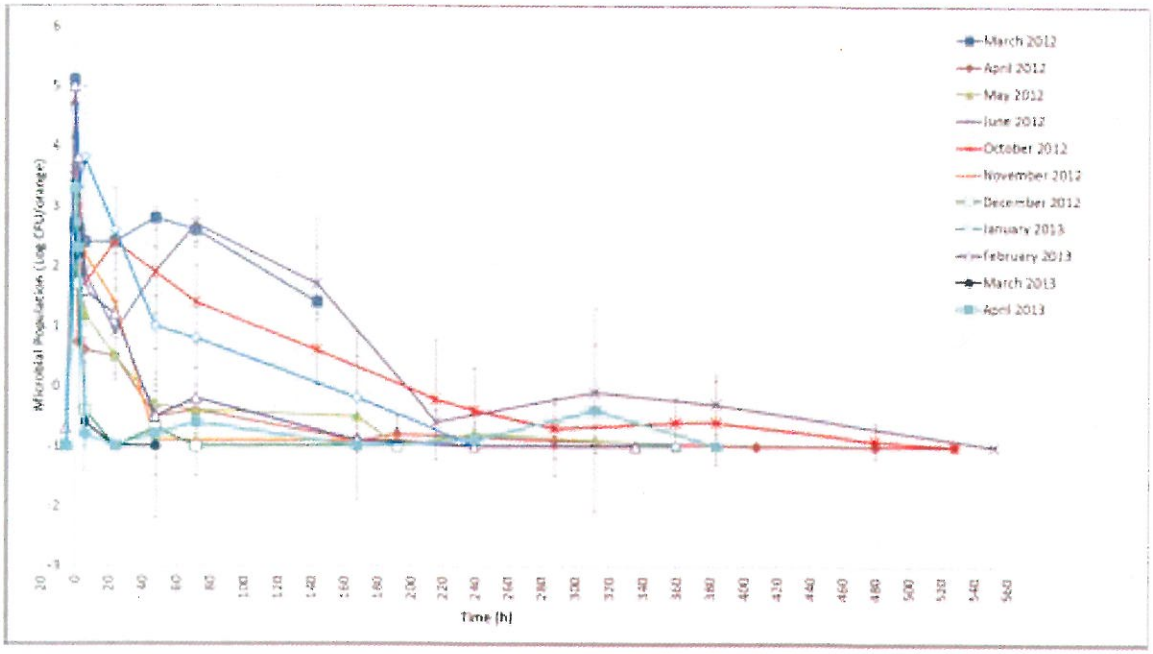

Figure 7. Summary of E. coli decline on citrus trees in March – June, and October – December, 2012 and January – April, 2013.