CONTROLLING *ESCHERICHIA COLI* WITH IN-FIELD APPLICATION OF COPPER FUNGICIDES

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Introduction

The Food Safety Modernization Act (FSMA) seeks to protect humans from food-borne pathogens. The U.S. Food and Drug Administration (FDA) has proposed rules to implement the FSMA that include regulations on limits on the microbial content in agricultural irrigation water. Under the proposed rules, if water quality standards are exceeded, produce could still be considered safe for human consumption if appropriate remedial actions are taken. One such action could be the use of currently registered copper fungicides that also have bactericidal effects. Therefore, we conducted a field trial to compare generic *Escherichia coli (E. coli)* levels on onion bulbs from plots that had been treated with a copper fungicide just before. The objective of this study was to determine the effect of copper sulfate on *E. coli* levels in irrigation canal water and in the water at the ends of furrows of an onion field.

Materials and Methods

The experiment was conducted at the Malheur Experiment Station, in a field of 'Vaquero' onions that was dedicated for *E. coli* research. Double rows of onions were planted on 44-inch beds and each plot was 3 beds wide. There were four irrigation treatments with each treatment replicated five times. The irrigation treatments were: 1) drip irrigated with well water; 2) drip irrigated with ditch water; 3) furrow irrigated with ditch water; 4) furrow irrigated with ditch water with enhanced *E. coli* levels. Each of these plots was subdivided into two plots for the fungicide trial. Subplots were 33 ft long and 3 beds wide. One subplot of each main plot was left untreated and the other subplot was treated once on August 23, 2014 with Kocide® 2000 DF (53.8% copper hydroxide) at 1.5 lbs/acre. The application was made with a CO_2-powered backpack sprayer, with a 4-nozzle boom and 11004 nozzles, set at 30 psi and 35 gal/acre.

Onions were lifted on August 21, 2014 (2 days before fungicide application) and then sampled on four dates: August 22, the day before the fungicide application was made; 2) September 3; 3) September 10; and 4) September 16.

On each date, a row of 60 onions was sampled from each plot; no onions were discarded. Onions were topped and placed in wire baskets and then placed in plastic storage bags. To minimize potential cross contamination, workers wore gloves, and used sterilized knives and baskets in the

field. Knives and baskets were sterilized with bleach and workers exchanged gloves as they moved from plot to plot. Because we anticipated that irrigation treatments would have different amounts of *E. coli,* we sampled them in order of expected *E. coli* levels, going from low to high (i.e., drip-well water, drip-ditch water, furrow, and furrow with *E. coli* enhanced water). Within irrigation treatments, we expected treated onions to have lower *E. coli* levels than untreated onions. Therefore, we sampled the treated plots first.

Onions were transported to Western Laboratories, Inc. (Parma, ID) for *E. coli* analysis. In the laboratory, the roots, small remnants of soil, skins, and outer peel of the onions were removed from the bulbs and weighed. The skins, peels, roots, and soil were then thoroughly washed in 1 L of water. A 100 ml sample of the wash water was used to estimate a Most Probable Number (MPN) of generic *E. coli* present on the exteriors of the onions, using IDEXX Colilert® +Quanti-Tray/2000® (IDEXX Laboratories, Westbrook, ME) (Edberg and Edberg 1988, Edberg et al. 1990). The *E. coli* MPN per onion bulb exterior was then calculated based on the number of onions in each sample. The onion interiors were tested for the presence of *E. coli* and *Salmonella* for the September 16 sample date. Bulb interiors were tested as described in the paper "Survival of *Escherichia coli* on Onion During Field Curing and Packout" in this report.

Results and Discussion

No *E. coli* was detected in onions sampled before the copper fungicide treatment. This suggests that onions are not likely to become contaminated with *E. coli* under current production practices.

Only two samples of the subsequent samples collected after treatment were found to have *E. coli.* One sample was from an untreated plot and the other sample was from a copper fungicide-treated plot. The mean amount of generic *E. coli* per onion bulb in both of those cases was 0.8 MPN/100 ml of water.

The bulb interiors were negative for all microbial contaminants.

Given the absence of *E. coli* in the untreated bulbs, it is not possible to gauge the effectiveness of copper-based fungicides in controlling *E. coli* on onions. The lack of *E. coli* on untreated onions is still an important finding.

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