



**CPS 2009 RFP  
FINAL PROJECT REPORT**

**Project Title**

Evaluation and optimization of postharvest intervention strategies for the reduction of bacterial contamination on tomatoes

**Project Period**

October 1, 2009 – October 31, 2011

**Principal Investigator**

Keith Schneider  
Department of Food Science and Human Nutrition  
University of Florida  
Gainesville, FL 32611  
Email: [keith29@ufl.edu](mailto:keith29@ufl.edu)  
Phone: 352-392-1991 ext. 309

**Co-Principal Investigators and Cooperators**

Yaguang Luo  
US Department of Agriculture, Agricultural Research Service  
10300 Baltimore Ave., Bldg. 002, Beltsville, MD 20705  
Email: [yaguang.luo@ars.usda.gov](mailto:yaguang.luo@ars.usda.gov), Phone: 301-504-5706

Steven A. Sargent  
Horticultural Sciences, University of Florida, Gainesville, FL 32611  
Email: [sasa@ufl.edu](mailto:sasa@ufl.edu), Phone: 352-392-1928

Renee Goodrich-Schneider  
Department of Food Science and Human Nutrition, University of Florida, Gainesville, FL 32611  
Email: [goodrich@ufl.edu](mailto:goodrich@ufl.edu), Phone: 352-392-1991

Billy L. Heller, Jr., Chief Executive Officer  
PTG Management Company Pacific Tomato Growers, Ltd., Palmetto, FL

Anthony DiMare  
DiMare Fresh, Tampa, 8150 Eagle Palm Drive, Riverview, FL

---

**Objectives**

- 1. Determine the effect of tomato dump tank water management standards on Salmonella infiltration.*
- 2. Determine the correlation among water quality measurement parameters, and assess functional limits where wash water replenishment or replacement is needed.*

3. *Determine whether a dry-dump system utilizing an overhead spray rinse with a brush washer, is equal to or better than a flume system in removing surface pathogens from the surface to tomatoes.*
4. *Develop specific operation limits and monitoring recommendations for implementation in commercial settings, and ensure this information is disseminated to growers, packers and other stakeholders.*

## **FINAL REPORT**

### **Abstract**

Tomatoes have been implicated in several salmonellosis outbreaks due to possible contamination through bacterial internalization during post-harvest handling. This project was conducted: (1) to determine the effect of tomato dump tank water management standards on *Salmonella* infiltration; (2) to investigate the correlation among water quality measurement parameters, and assess functional limits where wash water replenishment or replacement is needed; and (3) to determine whether a dry-dump system utilizing an overhead spray rinse with a brush washer, was equal to or better than a flume system in removing surface pathogens from the surface to tomatoes. In objective 1, mature green tomatoes at 90 °F (32.2 °C) were immersed in water containing approximately 10<sup>6</sup> CFU/ml *S. enterica*. Tomato varieties (Mountain Spring, Applause, BHN961, BHN 602, Sunbrite), post-stem removal time (0 hr-16 hr), temperature differentials ((-) 30 °F to (+) 10 °F), and immersion time (2min-15min) were evaluated for their effects on *S. enterica* internalization. For objective 2, the changes in water quality and sanitizer concentration were closely monitored during dump tank wash operations at three major tomato packinghouses in Florida. Additional laboratory tests were conducted to evaluate the dynamics of chlorine concentration and ORP reading as impacted by organic load simulated with tomato extract. For objective 3, the efficacy of sanitizers in an overhead spray and brush roller system was examined for reducing *Salmonella* on unwaxed, mature green tomatoes. All internalized *S. enterica* cells were found within the core tissue segments immediately underneath the stem-scars. Tomato variety, post-stem removal time by variety interaction and immersion time significantly affected the incidence of *S. enterica* internalization (P = 0.0001), while temperature differential in the test range (-10, 0, and 10 °F) had no significant effect. Increasing the time interval between stem removal and immersion greatly reduced pathogen internalization into tomato fruits except in the variety of Mountain Spring. Also, water quality declined continuously during packing house operations, as determined by significant increases in chemical oxygen demand (COD) and turbidity. Free chlorine and ORP varied widely among different packinghouses. Although the dump tank operations and chlorine dosing rates also varied widely among the packing houses, all of them maintained at least 25 ppm free chlorine in the dump tank throughout our visit. Although ORP values were positively correlated with free chlorine and negatively correlated with pH, this linear correlation was significant only within a small range of free chlorine concentrations, indicating that ORP has limited usage as an indicator for sanitizer effectiveness. The results from the final objective demonstrated the ability of sanitizers in the laboratory model overhead spray system to reduce *Salmonella* on tomato surfaces.

### **Background**

*Salmonella enterica* is a common etiologic agent of foodborne illness in the US. It is estimated that food borne *S. enterica* causes 1 million infections, almost 10,000 hospitalizations, and over 350 deaths annually in the US. Traditionally, most salmonellosis cases and outbreaks have been attributed to foods of animal origin such as meat and poultry products, eggs, and dairy products; recently, however, *S.*

*enterica* outbreaks are increasingly linked to consumption of fresh fruits and vegetables. Tomato fruit is a common vehicle of produce-associated salmonellosis in the US and has been linked with a number of outbreaks since 1998.

Investigations of outbreaks associated with tomatoes suggest that contaminated wash water in tomato packing facilities was one of the factors contributing to microbial contamination. Pathogen internalization can occur through the porous tissues of the stem scar. Once internalized, bacteria are sheltered by tomato tissues and become difficult to remove or inactivate. Thus, preventing pathogen internalization is a critical step in reducing food safety risks. Current post-harvest handling practices in typical tomato packing houses include immersion of tomatoes for up to 2 min in a heated, chlorinated water dump tank, with water temperature maintained at least 10 °F (5.6°C) higher than the incoming tomato pulp temperature. This practice, intended to reduce risk of pathogen internalization, was developed based on previous research on tomato soft rot development studies. Yet, the effect of these conditions on the prevention of *Salmonella* infiltration remains undetermined.

Previous studies also demonstrated that in addition to temperature differential, tomato variety and the time between harvest and dump tank water immersion may impact tomato stem-scar water uptake and soft rot development. Smith et al. reported that Florida 47 had significantly greater water uptake than Sebring when held under the same conditions, and greater water uptake occurred when the fruits of both varieties were dipped in water at 2 hr after harvest than at 8, 14, and 26 hr after harvest. However, no studies have evaluated these post-harvest handling conditions on *S. enterica* internalization. Lastly, the use of a traditional flume has been postulated as a potential source of cross contamination, thus a brush roller washing system equipped with an overhead spray bar was constructed to determine if this system could provide similar or better efficacy and a lab-scale model flume system. Therefore, the aims of this project were: (1) to examine the effects of four major factors (tomato variety, temperature differential between tomato pulp and bacterial suspension, post-stem removal time, and immersion time) on the internalization of *Salmonella enterica* serovar Thompson in tomato fruit (Objective 1); (2) to monitor the changes in water quality and sanitizer concentration during dump tank wash operations at three major tomato packing houses in Florida and evaluate the dynamics of chlorine concentration and ORP reading as impacted by organic load simulated with tomato extract (Objective 2); and (3) determine whether a dry-dump system utilizing an overhead spray rinse with a brush washer, is equal to or better than a flume system in removing surface pathogens from the surface to tomatoes (Objective 3).

## **Research Methods and Results**

### **Objective 1. Determine the effect of dump tank handling conditions on *Salmonella* infiltration**

#### ***Methodology:***

**Mapping the distribution of internalized *Salmonella enterica* cells:** Tomatoes were submerged in the bacterial suspension for 30 min at a -30 °F temperature differential between tomato pulp and bacteria suspension to ensure that *S. enterica* infiltrated into most of the tested tomatoes. After removal from the bacterial suspension, each tomato was thoroughly surface sanitized to ensure that all surface attached bacteria were killed to avoid cross-contamination. After drying on a sterile petri dish, the internal tissues of tomatoes from six different locations, as shown in Figure 1, were excised for microbial analysis, which was conducted using a modified most probable number (MPN) procedure, and confirmed.

**Determining the incidence and severity of *Salmonella* internalization as impacted by tomato variety, temperature differential, immersion time, and the post-stem removal time:** Freshly harvested mature green tomatoes were submerged in water containing suspensions of *Salmonella enterica* (Newport, Thompson) at the inoculation levels of  $10^6$  CFU/ml (2010) and  $10^5$  CFU/ml (2011). Immersion time ranged from 2 min to 15 min, temperature differentials between water and tomato pulp ranged from -30 to +10°F, and time delays between stem removal and submerging ranged from 0 to 16 hours. Upon removal from the bacterial suspension, tomatoes were thoroughly surface sterilized (Xia et al., 2011). The tomatoes were halved transversely and the central core tissues excised using a sterile cork borer. The segments underneath the stem-scar were stomached and enriched for *S. enterica*. *S. enterica* positive samples were further enumerated using an MPN procedure and confirmed serologically or via the GFP marker. Since internalized *S. enterica* cells were mainly located in the core tissues underneath the tomato stem-scar, this core tissue segment was used for all pathogen internalization studies.

**Key findings:**

The incidence and extent of *S. enterica* internalization varied significantly among different locations and/or tissue types (Figures 1). *S. enterica* was only detected in the core tissue segments (Figure 1, segments I, II, and III), especially the segment immediately beneath the stem-scar (Figure 2, segment I), and the internalization incidence and populations declined steadily in the core tissue samples with distance from the stem-scar. *S. enterica* was not detected in any of the other internal tissues, including locular cavity, tissues proximal to the blossom end and pericarp tissue samples. Also, the internalization depth of *Salmonella enterica* into tomato tissues was impacted significantly by immersion time (Figure 2). Since the majority of internalized *S. enterica* cells were located within the core tissue segment beneath the stem scar, all follow-up internalization studies used core tissue segment within 2-22 mm below the stem-scar.

Pathogen internalization was impacted by tomato varieties and time interval between stem removal and *Salmonella* suspension immersion. Under the current recommended condition of no more than 2 layers of tomatoes, and no more than 2 min of immersion time, tomato varieties and post-stem removal time significantly affected the incidence of *S. enterica* internalization, while temperature differential over the range of -10 to +10 °F had no significant effect (Figure 3). In general, Mountain Spring was less susceptible to *S. enterica* internalization than were Applause and BHN961. Increasing the time interval between stem removal and immersion greatly reduced pathogen internalization in BHN961 and Applause, while it had no effect in Mountain Spring tomatoes. The variety, interactions between variety and post-stem removal time, and interaction between temperature differential and post-stem removal time had significant effects on the populations of internalized *S. enterica*.

The incidence and extent of *S. enterica* internalization was significantly impacted by the range of temperature differential and immersion time and their interactions (Figure 4). Significantly more severe pathogen internalization was observed with a 15 min of immersion time than a 2 min immersion time. With 15 min immersion time, negative temperature differentials of -10 and -30 °F generated significantly higher *Salmonella* internalization than did 0 and + 10 °F differentials. However, with 2 min immersion time, there was no significant difference for pathogen internalization between temperature differentials from 10 to -30 °F for both varieties.

Two *Salmonella enterica* serovars, Thompson and Newport were used for 2010 and 2011 harvest seasons, respectively. The inoculum level used for 2010 was approximately  $10^6$  CFU/ml, which was

reduced to  $10^5$  CFU/ml in 2011. Also, due to the limitations in the availability of tomato varieties from commercial farms in Maryland, two sets of tomato varieties were used for 2010 and 2011. To validate the test results from different years, varieties, and *Salmonella enterica* serovars, additional experiments were conducted to evaluate the effect of *Salmonella enterica* serovars, and inoculation levels using high tunnel grown tomatoes (Variety: Applause) in 2011. Test results confirmed that, when immersed immediately after stem-removal, tomatoes have a very high incidence of *Salmonella* internalization, even without temperature differential. This occurs regardless of *Salmonella enterica* serovars or inoculation levels (Fig 5). However, inoculum level of both serovars had a substantial effect on the populations of *Salmonella* internalized with larger populations of *Salmonella* detected inside the tomatoes for inoculum level of  $10^6$  CFU/ml than for  $10^5$  CFU/ml.

## **Objective 2. Determine the correlation among water quality measurement parameters and their associations with dump tank sanitizer efficacy**

### ***Methodology:***

#### **Packinghouse survey:**

Surveys of three major commercial tomato packinghouse operations in Florida during 2010 and 2011, focused on dump tank water quality. Water samples were collected every 10-30 min for 4 to 8 hrs per day. Free and total chlorine, ORP, pH, turbidity, and total dissolved solids (TDS) were tested every 10-30 min, and chemical oxygen demand (COD) was determined every 2 hours. The volume of water in the dump tank and the weight of the tomatoes washed were recorded.

#### **Laboratory simulation studies:**

The impact of tomato dump tank operation conditions on water quality, correlation between free chlorine and ORP, and effect of chlorine level on pathogen inactivation and cross-contamination were investigated in a laboratory setting using tomato juice extract to manipulate the organic load.

**Correlation among ORP, free chlorine, and pH:** Chlorinated water solutions with different levels of free chlorine were prepared by adding varying amounts of Clorox to water. Solution pH was adjusted to 6.5 using citric acid and sodium hydroxide. Water quality was monitored in terms of pH, free chlorine, ORP and COD.

**Water quality and chlorine concentration changes as impacted by organic load:** Commercial wash operation was simulated with steady increase in organic load and periodical replenishment of free chlorine. Wash solution with 60 ppm free chlorine was prepared using Clorox. Tomato extract was added to the wash water in small increments. Wash solution changes in free chlorine, ORP, COD, and turbidity were monitored. When the free chlorine level dropped down to approximately 0.5 ppm, additional Clorox was added to the wash solution to bring the free chlorine level back to 60 ppm. This process including tomato extract addition and chlorine replenishment was repeated two more times. Wash solution pH was constantly maintained at 6.5 with citric acid and sodium hydroxide.

**Determination of chlorine concentration needed to prevent *Salmonella enterica* cross-contamination during washing:** Pre-marked non-inoculated and inoculated (with *S. enterica* serovar Thompson) tomatoes were washed together in solutions with free chlorine levels ranging from 0.2 to 50 ppm. After a 2 min exposure, the un-inoculated tomatoes were retrieved from the wash solution,

and the populations of *S. enterica* on stem scar and smooth surface were enumerated using a modified MPN procedure.

### **Key findings:**

#### **Packinghouse Survey:**

During the survey in Florida, we found that the productivity varied from 120,000 to 180,000 lb of tomatoes/hr among three packinghouses, and water temperature was consistently maintained at least 10 °F higher than tomato pulp. Tomatoes were dumped into the tank beginning about 0.5-5 hr after harvest, sometimes 24 hr. For these three packinghouses, the residence time of tomatoes in the dump tank was less than 2 min. The data from the Florida survey (Figure 7) indicates that water quality declined continuously during packing house operations, with a significant increase in TDS, COD and turbidity over time, which was in line with the observation of the accumulation of dirt and debris. The pH values in these packinghouses were different, ranging from 6.0 to 7.5, and mostly kept stable except periodically for the packinghouse II. The pH in the packinghouse II fell to 3.5. Free chlorine and ORP varied widely among different packinghouses, as influenced by the specific parameters of the packinghouse operations: rate of tomatoes washed and the dosage of chlorine in the dump tank etc. Despite wide variation in dump tank operations and chlorine dosing rates among the packinghouses, all packinghouses maintained at least 25 ppm free chlorine in the dump tanks throughout the period of our visit.

#### **Laboratory simulations:**

**Relationship between free chlorine, ORP, and pH:** The ORP values of chlorinated water decreased with the increase in pH and decrease in free chlorine concentration, but this linear relationship was maintained only when the free chlorine was below 10 ppm (Figure 7). In addition, large variation in ORP readings was observed even at the same pH and free chlorine level. In the presence of high organic load, the free chlorine concentration and ORP readings also gradually changed over time (data not shown). These findings suggest limitations in using ORP as an indicator for chlorine concentration, especially when the pH control system malfunctions and high organic load accumulates in wash water.

**Water quality and chlorine concentration changes as impacted by organic load:** Water quality declined rapidly over the continuous addition of tomato extract, as evidenced by the rapid increase in turbidity and COD (Figure 8). This was accompanied by a sharp drop in free chlorine concentration. Interestingly, the ORP readings were quite stable initially (Figure 8D), followed by a rapid decline when the free chlorine level fell below 10 ppm. A similar pattern was observed when the wash solution was replenished with Clorox. Furthermore, although we were able to restore the free chlorine concentration to around 60 ppm, accomplishing this task required increasing amounts of Clorox with increasing organic load.

**Determination of chlorine concentration needed to prevent *Salmonella enterica* cross-contamination during washing:** In the absence of chlorine in the wash solution, cross-contamination was noted on both tomato surface and stem-scar of the un-inoculated tomato washed with inoculated tomatoes (Figure 9). No *S. enterica* was detected on any of the tomato surface washed in solution containing free chlorine. However, *S. enterica* was detected in the stem-scar areas on tomatoes washed in solutions containing 0.2 to 5 ppm free chlorine. When the free chlorine concentration was increased to 10 ppm, no *S. enterica* cells were detected in any of the un-inoculated tomatoes. This underlines the

critical importance of maintaining sufficient sanitizer concentration to prevent pathogen cross-contamination.

**Objective 3. Determine whether a dry-dump system utilizing an overhead spray rinse with a brush washer, is equal to or better than a flume system in removing surface pathogens from the surface to tomatoes**

*Methodology:*

**Sodium hypochlorite and sanitizer efficacy studies:**

Fate of *Salmonella* on tomato surfaces was determined after overhead spray treatment with NaOCl (25, 50, and 100 mg/L) and a water control. In a sanitizer efficacy study, NaOCl (100 mg/L), ClO<sub>2</sub> (5 mg/L), PAA (80 mg/L), and a water control were tested. Five inoculated tomatoes were sprayed for 5, 15, 30, and 60 s per treatment. Tomatoes were placed in the valley between two rotating nylon brush rollers (46 cm long and 12 cm dia) rotating at 180 rpm. Three spray nozzles (Spraying Systems Co., Wheaton Ill., U.S.A.) 13 cm above rollers released a cone shaped spray at 12 psi and 21.4 ml/s flow rate.

**Flume and overhead spray comparison study**

Inoculated tomatoes were placed in a Precision circulating waterbath (Jouan, Inc., Winchester, Va., U.S.A.) measuring 38.7 cm by 30.5 cm and 19.0 cm deep containing either 10 L of NaOCl (100 mg/L) or 10 L water with 0.1% Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub> at 25 °C. The water bath simulated the flumes used in tomato packinghouses. Five tomatoes were tested per time of 5, 15, 30, and 60 s. Each treatment time was tested sequentially with no change of NaOCl or water between times. Flume water was tested for *Salmonella* at time 0 and after the 15 and 60 s treatment times.

*Key findings:*

**Efficacy of NaOCl concentrations in overhead spray system**

Significant reductions of *Salmonella* occurred after 15 s with NaOCl (Table 1). At 15 s, 100 mg/L NaOCl achieved a  $4.0 \pm 1.8$  log<sub>10</sub> CFU/ml reduction, which was significantly different from all other concentrations and water. A 3-log<sub>10</sub> unit reduction was also achieved by 50 mg/L NaOCl at 30 s and 25 mg/L NaOCl at 60 s. Even water reached a 3-log<sub>10</sub> CFU/ml reduction at 60 s. A 5 log<sub>10</sub> unit reduction was seen for 100 mg/L NaOCl at 30 s and 50 mg/L at 60 s.

**Efficacy of NaOCl, ClO<sub>2</sub>, and PAA in overhead spray system**

After only 5 s, PAA reached a 2.8 log<sub>10</sub> CFU/ml reduction of *Salmonella*. Conversely, NaOCl, ClO<sub>2</sub>, and water each had a 1.9 log<sub>10</sub> CFU/ml reduction. All sanitizers reached a 3-log<sub>10</sub> unit reduction at 15 s, including water (Table 2). PAA consistently achieved about a 1-log<sub>10</sub> unit higher reduction than the other sanitizers for 5, 15, and 30 s treatment. Increasing treatment time to 30 s did not significantly increase reduction by ClO<sub>2</sub> or NaOCl, but did for PAA. At 60 s, average log<sub>10</sub> reductions by sanitizers were all significantly higher than water and were not significantly different from each other.

## Flume and overhead spray comparison study

The flume water control did not produce significantly different reductions in *Salmonella* depending on spray time, with an average reduction of 1.0 log<sub>10</sub> CFU/ml. Flume data was compared to overhead spray NaOCl (100 mg/L) data from the sodium hypochlorite efficacy study. Overhead spray NaOCl treatments of at least 15 s significantly reduced more *Salmonella* from tomatoes compared to flume treatments. At 15 s, NaOCl in the flume and overhead spray had an average reduction of 1.3 and 4.0 log<sub>10</sub> CFU/ml, respectively (Table 3). At 30 s, reduction by NaOCl was enhanced to 5.6 log<sub>10</sub> CFU/ml in the overhead spray but only to 3.2 log<sub>10</sub> CFU/ml in the flume. Increasing spray time to 60 s did not result in a significantly higher reduction in either system.

Concentration of *Salmonella* was tested in the flume. At time 0, *Salmonella* was undetectable in flume water. *Salmonella* was recovered from flume water at an average of  $4.5 \pm 0.4$  log<sub>10</sub> CFU/ml after the 15 s treatment and  $5.1 \pm 0.2$  log<sub>10</sub> CFU/ml after the 60 s treatment, though these populations were not significantly different from each other. NaOCl (100 mg/L) effectively eliminated *Salmonella* in the flume as populations were undetectable throughout the study.

**Objective 4. Develop functional limits that operators can use to ensure that effective sanitizer concentrations and other control parameters are present in the dump tank or spray cleaning system. This will be part of our extension outreach program which will include conducting specialized training on proper washing protocols as support for GAPs/GMPs metrics development and subsequent implementation into food safety systems.**

The major findings of this project are summarized below. This information has been shared with the tomato growers and packers during the CPS symposium and several Florida grower meetings. During meetings with growers, both Drs. Luo and Schneider shared their findings on 1) internalization of *Salmonella*, 2) internalization as a function of variety, 3) dump tank conditions, 4) relationship between free chlorine, pH and ORP, 5) potential cross-contamination, and 6) effectiveness of the overhead spray system. In addition to the information sharing at workshop events both Drs. Luo and Schneider have prepared scientific publications sharing their results with the scientific community (see Appendix section – Publications and Presentations)

## Outcomes and Accomplishments

All goals set forth in the original proposal were met. The outcomes of each of the objectives (and sub-objectives) are:

- 1) Internalized *S. enterica* cells were found only within tomato core tissue segments immediately underneath the stem-scars. The internalization incidence and populations of *S. enterica* declined in the core tissue samples with distance from the stem-scar.
- 2) The incidence of *S. enterica* internalization was a function of tomato variety, post-stem removal time, and immersion time, as well as the interaction between tomato dump tank temperature differential and immersion time.
- 3) Three large packinghouses in Florida were surveyed in 2010 and 2011. It was observed that water quality declined continuously during packing house operations, with a significant increase in chemical oxygen demand (COD) and turbidity. Free chlorine and ORP vary largely among different packing houses, as influenced by the specifics of the packing house operations, including the rate of tomatoes washed, and the dosage of chlorine into the dump tank etc.



However, all of them maintained at least 25 ppm free chlorine in the dump tanks during our visits.

- 4) Laboratory simulation studies indicate that there is a relationship between free chlorine, pH and ORP in water containing a typical organic load. In general, ORP decreases with the increase in pH and decrease in free chlorine concentration. However, this correlation was only maintained within a small range of free chlorine concentration, suggesting a limitation in using ORP as a chlorine indicator.
- 5) Free chlorine is degraded rapidly by organic loads; although free chlorine concentrations can be brought back to the 60 ppm range, increasing amounts of sodium hypochlorite are needed with increasing organic load in the wash solution.
- 6) Pathogen cross-contamination occurred readily during tomato washing in the absence of free chlorine (or any other sanitizer). Cross-contamination was found on tomatoes washed in solution containing up to 5 ppm free chlorine, but not in the solutions containing 10 ppm free chlorine. This suggests that maintaining sufficient sanitizer concentration in the wash solution is critical to prevent pathogen cross-contamination.
- 7) The overhead spray system could achieve a 3- to 5- $\log_{10}$  unit reduction of *Salmonella* from tomato surfaces with specific sanitizers and spray times.
- 8) The overhead spray system could provide benefits over conventional flumes including higher pathogen reduction, and less sanitizer and water use, all of which help to decrease tomato packing costs and keep the tomato industry a viable part of Florida's economy.

**Tables and Figures (optional)**

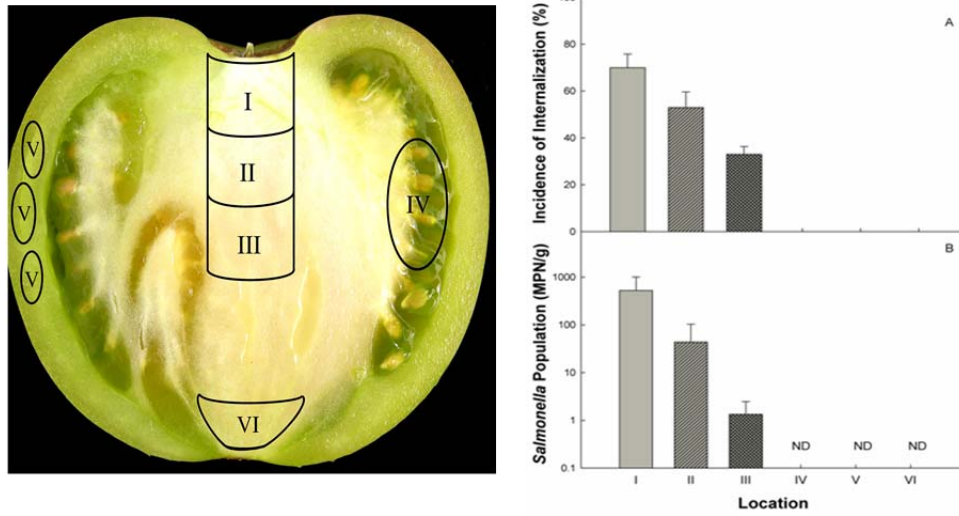


Figure 1. Frequency and extent of *Salmonella* Thompson internalization at various locations within tomato fruit. I - tomato core tissue segment within 2-12 mm below stem-scar; II - tomato core tissue segment within 12-22 mm below stem-scar; III - tomato core tissue segment within 22-32 mm below stem-scar; IV - tomato locular cavity; V - tomato pericarp tissues; VI - tomato internal tissues below the blossom end. ND - non-detectable at a detection limit of 0.38 MPN/g.

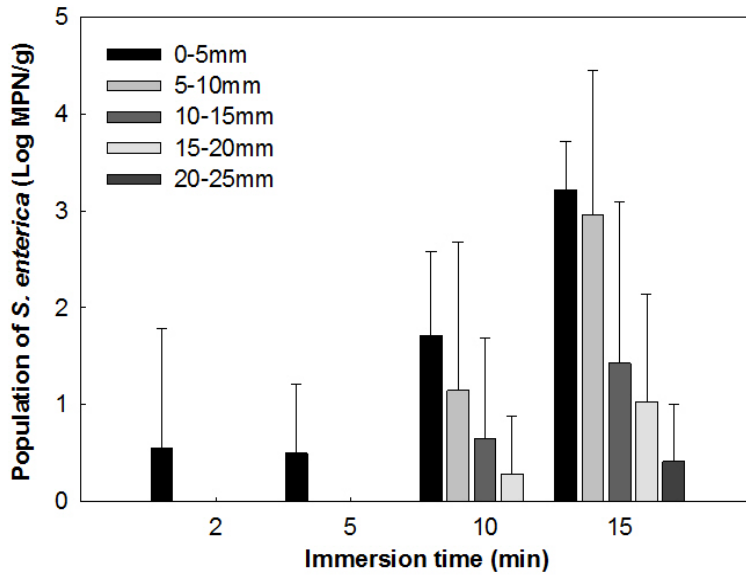


Figure 2 Distribution of internalized *S. enterica* serovar Thompson as affected by immersion time.

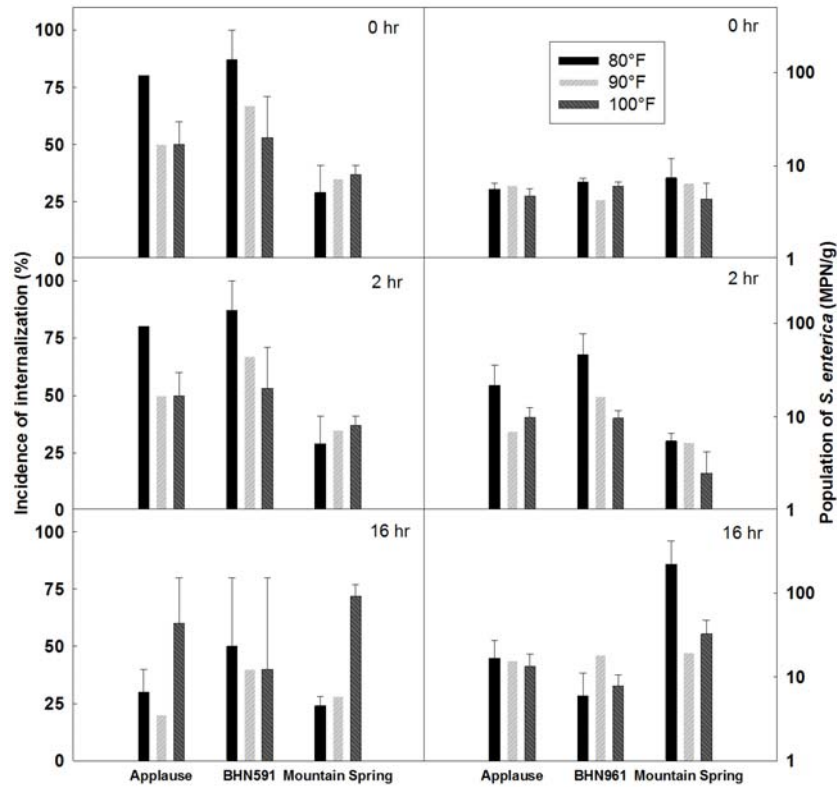


Figure 3 Incidence and extent of *Salmonella* Thompson internalized in tomatoes positive for pathogen internalization as affected by variety, temperature differential between tomato pulp and bacterial suspension, and post-stem removal time.

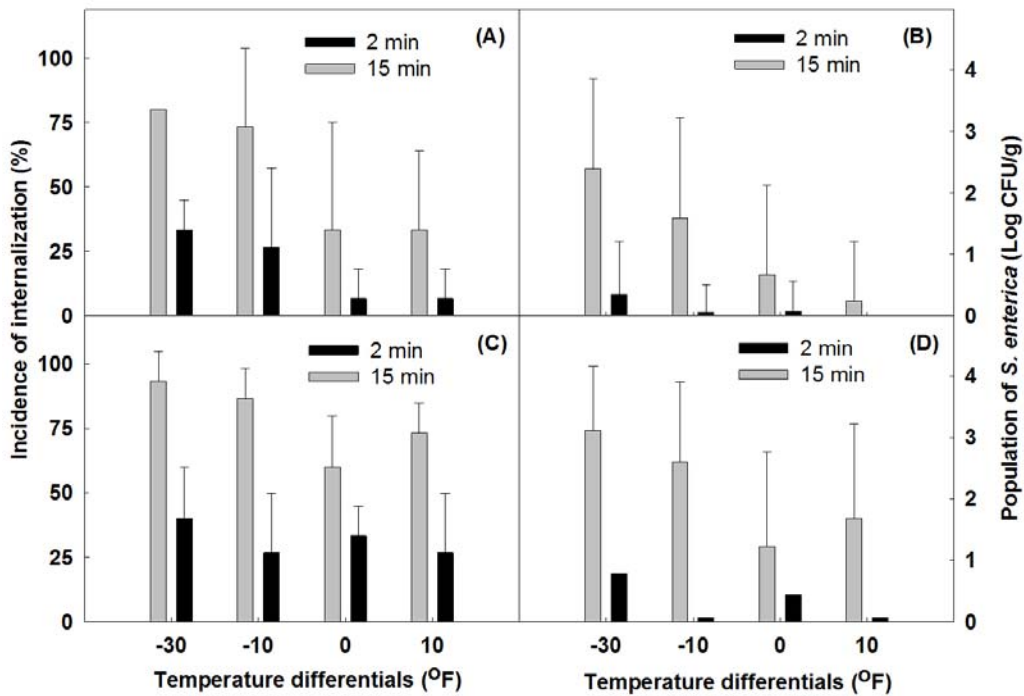


Figure 4 Incidence and population of internalization of *Salmonella* Newport in tomatoes as affected by temperature differential between tomato pulp and bacterial suspension, and immersion time. (A) and (C), BHN602; (B) and (D), Sunbrite.

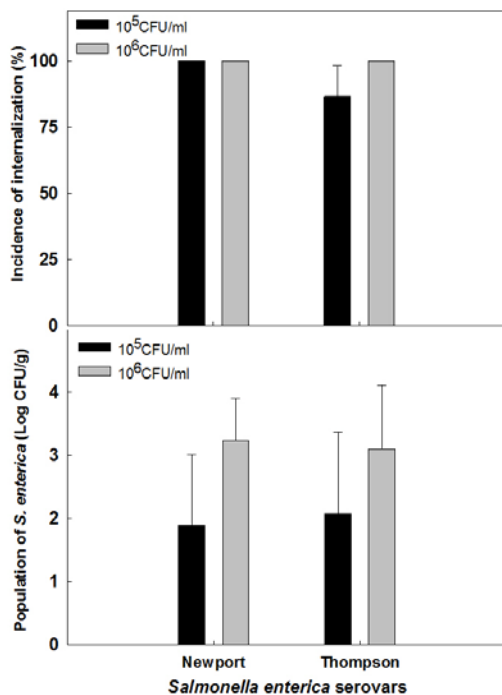


Figure 5 Effect of *Salmonella* strains and inoculation levels on the infiltration of *Salmonella* Newport into tomatoes.

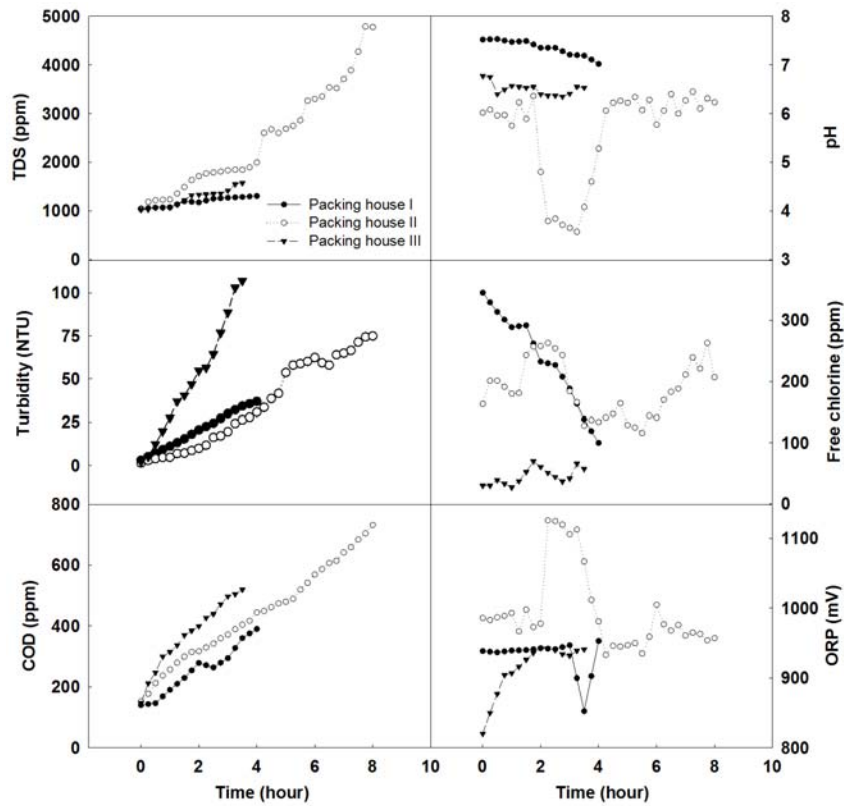


Figure 6. Tomato dump tank water quality of three packinghouses in Florida

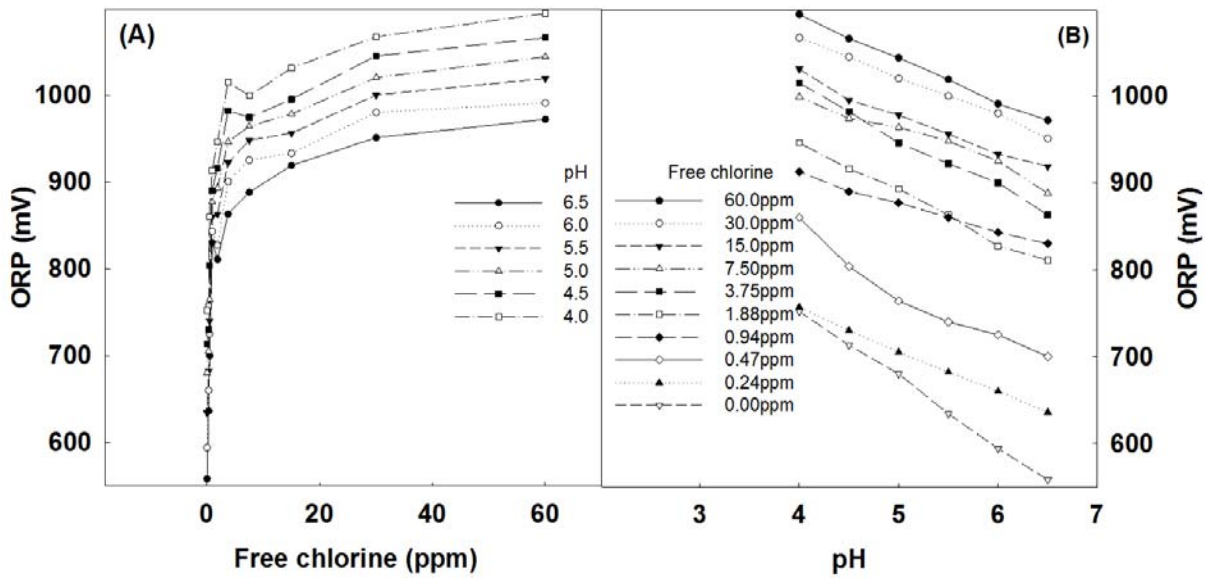


Figure 7. The relationship between ORP, free chlorine, and pH of chlorinated water enriched with tomato juice.

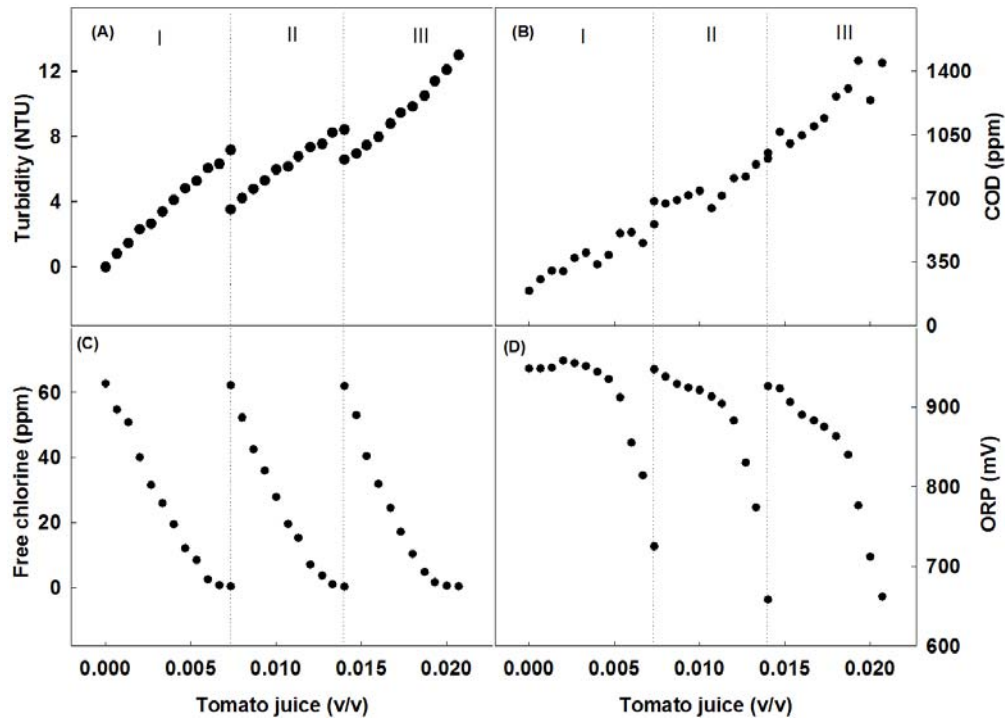


Figure 8. The changes of water quality during multiple additions of tomato extract and Clorox. (A) Turbidity; (B) COD ; (C) free chlorine; (D) ORP. I, the first stage; II, the second stage; III, the third stage.

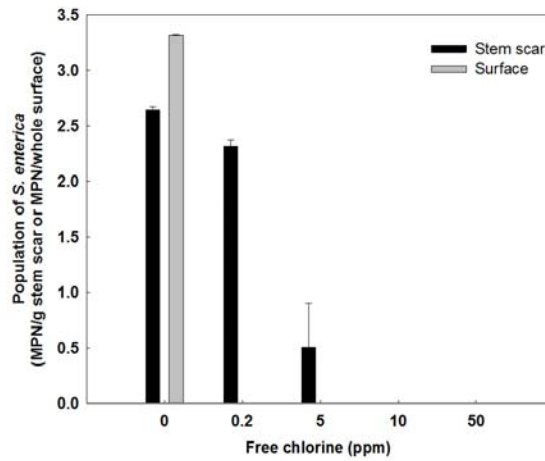


Figure 9. Cross-contamination of *Salmonella enterica* Thompson on tomatoes during washing.

Table 1 – Efficacy of 25, 50, and 100 mg/L sodium hypochlorite overhead spray treatment at 25°C

Time (s)	<i>Salmonella</i> log <sub>10</sub> reduction (log <sub>10</sub> CFU/ml) from tomatoes <sup>a</sup>			
	100 mg/L	50 mg/L	25 mg/L	Water control
5	1.7 ± 0.5 a, z	1.4 ± 0.8 a, z	1.0 ± 0.2 a, z	1.4 ± 0.4 a, y
15	4.0 ± 1.8 a, y	2.8 ± 1.1 b, y	2.0 ± 0.1 b, yz	2.3 ± 0.4 b, xy
30	5.6 ± 0.4 a, x	4.2 ± 0.8 b, x	2.5 ± 0.7 c, y	2.5 ± 0.2 c, x
60	5.5 ± 1.0 a, x	5.0 ± 1.2 ab, x	4.2 ± 1.1 b, x	3.0 ± 0.4 c, x

<sup>a</sup>Values are mean ± standard deviation of triplicate experiments of 5 tomatoes each (n=15). Means with same letter in the same row (abc) or in the same column (xyz) are not statistically different (p<0.05).

Table 2 – Efficacy of sanitizer overhead spray treatment at 25°C

Time (s)	<i>Salmonella</i> log <sub>10</sub> reduction (log <sub>10</sub> CFU/ml) from tomatoes <sup>a</sup>			
	NaOCl 100 mg/L	ClO <sub>2</sub> 5 mg/L	PAA 80 mg/L	Water control
5	1.9 ± 0.6 b, z	1.9 ± 0.7 b, z	2.8 ± 0.9 a, y	1.9 ± 0.7 b, y
15	3.5 ± 1.1 b, y	3.5 ± 0.7 b, y	4.7 ± 0.5 a, x	3.2 ± 0.8 b, x
30	4.1 ± 1.2 b, y	3.9 ± 0.3 b, y	5.5 ± 0.1 a, x	3.4 ± 1.1 b, x
60	5.5 ± 0.2 a, x	4.9 ± 0.3 a, x	5.5 ± 0.1 a, x	3.8 ± 0.7 b, x

<sup>a</sup>Values are mean ± standard deviation of triplicate experiments of 5 tomatoes each (n=15). Means with same letter in the same row (ab) or in the same column (xyz) are not statistically different (p<0.05).

Table 3 – Efficacy of simulated flume treatment at 25°C

Time (s)	<i>Salmonella</i> log <sub>10</sub> reduction (log <sub>10</sub> CFU/ml) from tomatoes <sup>a</sup>	
	100 mg/L NaOCl	Water control
5	0.8 ± 0.9 a, y	0.5 ± 0.3 a, x
15	1.3 ± 1.1 a, y	1.0 ± 0.8 a, x
30	3.2 ± 2.6 a, x	1.4 ± 0.8 b, x
60	3.3 ± 2.6 a, x	1.3 ± 1.1 b, x

<sup>a</sup>Values are mean ± standard deviation of triplicate experiments of 5 tomatoes each (n=15). Means with same letter in the same row (ab) or in the same column (xy) are not statistically different (p<0.05).

## **APPENDICES**

### **Publications and Presentations (required)**

Xia, X., Luo, Y., Yang, Y., Vinyard, B., Schneider, K., and Meng, J. 2011. Effects of Tomato Variety, Temperature Differential and Post-stem Removal Time on Internalization of *Salmonella enterica* Serovar Thompson in Tomatoes. Journal of Food Protection (in press).

Chang, A.S., and Schneider, K.R. 2011. Evaluation of Overhead Spray-Applied Sanitizers for the Reduction of *Salmonella* on Tomato Surfaces. Journal of Food Science (submitted)

### **Budget Summary (required)**