

CPS 2012 RFP FINAL PROJECT REPORT

Project Title Assessment of sanitation techniques for tree fruit storage bins

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Objectives

Evaluate current sanitation practices for reducing overall microbial levels and food safety risks in tree fruit storage bins (wood and plastic).

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FINAL REPORT

Abstract

Tree fruit are the leading agricultural commodity in the Pacific Northwest. To protect the supply of tree fruit, it is important to assess and understand microbial contamination risks and efforts to control them in the production chain. Knowledge gaps exist regarding the ability of current bin sanitation practices to reduce foodborne pathogen risk. The objective was to evaluate bin sanitation effectiveness for wood and plastic bins. An industry survey in the Pacific Northwest indicated that wood bins are primarily used for apples and pears; plastic bins and totes, when used, are more common for cherries and soft fruits, like peaches and apricots. Laboratory studies identified that 3M Scotch-Brite[™] pads were the most consisent sampling technique compared to cotton swabs and metal files for biofilm removal on wood and plastic bin surfaces. Due to background flora present in samples from commercial tree fruit bins. CHROMagar™ ECC media was used to recover total coliforms and generic E. coli from samples. The following treatments were examined as cleaning and sanitation practices for wood and plastic bins: chlorine dump tank treatment, peroxyacetic acid treatment, pressure washing alone and water pre-rinse followed by cleaning and sanitation, heat treatment alone and chlorine dump tank treatment followed by heat treatment. The most effective strategies to reduce the number of sampling sites positive for total coliforms for wood and plastic bins were a water pre-rinse followed by cleaning and sanitation; heat treatment also warrants further investigation. Results indicated that soiled areas could overwhelm effective cleaning and sanitation strategies, and visual evaluation was unlikely to target all contaminated areas accurately.

Background

Tree fruit are the leading agricultural commodity in Pacific Northwest. Washington alone produces almost 60% of the apples and 40% of the pears grown in the United States (Smith, 2012). Although literature on minimizing pathogen contamination along the tree fruit supply chain has increased, most of the studies focused on microbial quality, harvesting methods or antimicrobial treatment of tree fruits (Annous, 2001; Beuchat, 1997; Du, 2003; Errampalli et al., 2005; Rodgers et al., 2004; Sapers, 1999; Sapers, 2002, Wang et al., 2007; Wisniewsky et al., 2000). Significant knowledge gaps exist regarding the role of harvesting and storage bins as a potential source of pathogen contamination and the ability of sanitation practices to reduce potential food safety risks. However, several studies indicate that bins serve as a potential reservoir for numerous plant pathogens that influence fruit quality (Cossentine et al., 2004; Higbee, et al., 2001; Randall, et al., 2011; Sanderson, 2000). Therefore, the potential exists for bins to harbor and transfer foodborne human pathogens such as Shiga-toxin producing *E. coli* (STEC), *Salmonella spp.* and *Listeria monocytogenes*.

Some industry studies on wood and plastic surfaces indicated that traditional sampling methods may not accurately recover and reflect microbial levels, which may have been related to the presence of biofilms (personal communication with industry representatives). Similarly, biofilms were apparently present in a study of cleaned and sanitized plastic cutting boards in a food service setting, which presented challenges in achieving accurate microbial enumeration; both surface scraping and firm swabbing were utilized during the study (Neth et al., 2008). Therefore, sampling methods for removal of biofilms needed to be investigated in this study to achieve accurate results.

Several factors may influence potential food safety risks associated with bin handling, including type of material (wood versus plastic), bin storage design (nested versus non-nested stacking), type of tree fruit being handled and fruit production practices (organic versus

conventional). Bruised apples have been shown to support growth of *E. coli* O157:H7 (Digman, 2000). However, the ability of different bin materials to influence fruit injury, and potentially food safety, is unclear. Few differences were observed in final fruit grade between the plastic and wood bins (Bollen et al., 2001). Wood bins are subject to weathering, resulting in rough surfaces that can harbor various pathogens, whereas plastic bins have been cited as more resistant to weathering with surfaces that are easier to clean and sanitize (Waelti, 1992). However, long-term integrity and cleanability of plastic bins has not been thoroughly studied. In a study of acetic acid fumigation of fruit storage bins to control diapausing codling moth (*Cydia pomonella* {L.}) larvae, increased pathogen mortality rate was observed in plastic bins as compared to wood bins (Randall et al., 2011). Current bin sanitation is primarily for control of plant pathogens; verification of control of potential foodborne human pathogens with current practices is warranted. Buyer requests for the industry to shift to plastic bin usage are increasing; nevertheless, investment in plastic bins represents a significant economic impact on the produce industry. There is a lack of scientific evidence to prove that plastic bins reduce food safety risks associated with human pathogen contamination.

Assessing pathogen risks associated with tree fruit bins and the effectiveness of bin sanitation is important to protect the supply of tree fruit and other produce. Development of appropriate methods to accurately assess microbial levels and bin sanitation effectiveness is critical. Assessing current bin sanitation methods and providing insight into sampling methods will deliver information for the produce industry to guide best practices to reduce microbial risks associated with bin handling and sanitation.

Research Methods and Results

<u>Methods</u>

Industry survey: An initial survey instrument was prepared, and individual interviews were conducted with three packinghouse food safety managers. Requested information included: prevalence of use for wood and plastic bins, number of years wood and plastic bins are typically used (anticipated useful life), types of bin storage design (nested versus non-nested stacking), bin storage conditions, handling during harvest and transport, use of bin liners, use of recycled bins and bin sanitation practices in the orchard and at the packinghouse.

Laboratory examination of sampling methods: Two experimental methods assessed three sampling techniques for wood and plastic bin surfaces and three enumeration methods for generic *E. coli* (ATCC 8739). *Experimental m*ethod 1 involved establishing biofilms of generic *E. coli* (ATCC 8739) on wood and plastic bin pieces as well as glass as a control treatment; wood and plastic bin pieces were submersed in dilute tryptic soy broth (TSB) and held at room temperature, with agitation, for 6 days, with nutrient addition every other day. Experimental method 2 was more likely to reflect potential bin contamination under commercial conditions and involves inoculation of 200 µl of generic *E. coli* (ATCC 8739) on wood and plastic bin pieces; the pieces are allowed to dry and incubated for up to 6 days at room temperature. In method 2, an examination of nutrient addition as well as 1, 4 and 6 days after inoculation. Sampling techniques for microbial recovery compared cotton swabs, metal files and 3M Scotch-BriteTM pads. Enumeration methods that were compared included violet red bile agar (VRBA), VRBA with tryptic soy agar (TSA) overlay (for injured cell recovery) and 3M E. coli/Coliform PetrifilmTM.

Preliminary industry experiments: Preliminary experiments were conducted in two facilities and plastic bins were provided by a third facility. The ability to examine bins that had been exposed to field conditions and contained fruit was examined. Wood bins were categorized by age and

condition into four categories (new 2013, 2009-2012, 2004-2008, 1995-2003). Plastic bins were not used at this facility, so another industry partner provided plastic bins for the experiment. The partner confirmed that all of the plastic bins were approximately 12-15 years old (1998-2001), so categories based on condition (undamaged and damaged) were assigned. Wood and plastic bins were sampled at the packinghouse bin storage area and randomly assigned to treatments: rapid return from the orchard and packed immediately, held in the orchard for several weeks and packed immediately, refrigerated storage prior to packing and control atmosphere prior to packing).

Approximately 80 bins were examined throughout the season; in some cases, a single bin was delivered to an orchard to be filled more than once. Bins were sampled for total coliform and generic *E. coli* levels prior to leaving bin storage and after treatment in the dump tank. Empty bins that had been stored in the bin storage lot without shipment to orchards and filling were examined immediately prior to and after dump tank treatment as a control. Based on observation of bin handling practices, it was determined that the entire bin could be considered a food contact surface since the bins are completely submersed in dump tank water. For examination of microbial levels prior to or after sanitation, bins were sampled at three one-inch² locations on surfaces that would be in direct contact with tree fruit as well as at three one-inch² locations on surfaces that would contact water in the dump tank. Total coliform and generic *E. coli* were quantified on VRBA with TSA overlays and 3M *E. coli*/Coliform PetrifilmTM.

For each examination of microbial levels prior to or after sanitation, bins were sampled at three one-inch² locations on surfaces that would be in direct contact with tree fruit as well as at three one-inch² locations on surfaces that would contact water in the dump tank. Total coliform and generic *E. coli* were quantified on VRBA with TSA overlays and 3M *E. coli*/Coliform Petrifilm[™] for Facility 1 and on CHROMagar[™] ECC and 3M *E. coli*/Coliform Petrifilm[™] for Facility 2. It was determined that sampling immediately after sanitation provided accurate microbial results when compared to sampled collected 24 hours after bin sanitation.

For each examination of total coliform and generic *E. coli* levels prior to and after cleaning and sanitation practices, bins were sampled at three one-inch² locations on surfaces that would be in direct contact with tree fruit as well as at three one-inch² locations on surfaces that would contact post-harvest agricultural water, such as water in dump tanks. Total coliforms and generic *E. coli* were quantified on VRBA with TSA overlays and 3M *E. coli*/Coliform PetrifilmTM for Facility 1 and on CHROMagarTM ECC and 3M *E. coli*/Coliform PetrifilmTM for Facility 2. It was determined that sampling immediately after sanitation provided more accurate microbial results when compared to sampling 24 hours after bin sanitation.

Methods for examination of industry bin sanitation practices:

For all cleaning and sanitation practices examined, bins were evaluated for condition and cleanliness. The same sampling sites could not be sampled more than once due to the aggressive nature of the sampling. The sampling team photographed and documented descriptions for each sampling site prior to and after treatment. The sampling team selected six representative sites prior to and after treatment based on visual evaluation in an effort to accurately evaluate the potential for a treatment to reduce microbial levels. Bins were sampled at three one-inch² locations on surfaces that would be in direct contact with tree fruit as well as at three one-inch² locations on surfaces that would contact post-harvest agricultural water, such as water in dump tanks. Total coliforms and generic *E. coli* were quantified on CHROMagar[™] ECC. Time of immersion in water systems is provided in minutes (min) and seconds (sec).

Examination of a chlorinated dump tank and bin washing system: Wood and plastic bins from the facility were categorized into age categories. For wood bins, four categories were identified (2000-2007, 1990-1999, 1980-1989, 1970-1979). For plastic bins, three categories were identified (2007-2009, 2004-2006, 2001-2003). Bins from each category were assigned to one of two treatments (dump tank and bin washer or bin washer only). Bins were sampled prior to sanitation, after dump tank treatment and after bin washer treatment as appropriate. Validation of a CHROMagar[™] ECC (see identified challenges below) was also performed. The facility measured chlorine activity using oxidation-reduction potential (ORP) measured in millivolts (mV) and time of submersion was recorded manually. Approximately 65 bins were examined.

Examination of a peroxyacetic acid dump tank system: For wood bins, four categories were identified (new 2014, 2000-2013, 1990-1999, 1980-1989, less than 1979). Plastic bins were not available at this facility and based on the results of one replication, further experiments were not performed. The facility targeted a concentration of 80ppm peroxyacetic acid in the dump tank system.

Examination of a chlorinated dump tank followed by a heat treatment system: For wood bins, four categories were identified (new 2014, 2000-2013, 1990-1999, 1980-1989, less than 1979). Plastic bins were not available at this facility so another industry partner provided plastic bins; however, the plastic bins could not be accommodated in the dump tank system, so only the heat treatment system was examined for plastic bins. For plastic bins, three categories were identified (2007-2009, 2004-2006, 2001-2003). Facility 3 utilized a chlorinated dump tank followed by a heat treatment. The heat treatment was a separate piece of equipment designed specifically for heat treatment of bins using a water immersion with chemical wetting agent targeting 168°F for a minimum of approximately 2 minutes. The facility measured chlorine activity using oxidation-reduction potential (ORP) measured in millivolts (mV), and time of submersion was recorded manually. Temperature in the heat treatment system was available from a thermometer (°F) and time in the system was recorded manually.

Examination of pressure washing with water alone, as well as pressure washing followed by cleaning and sanitizing. For wood bins, four categories were identified (2000-2007, 1990-1999, 1980-1989, 1970-1979). For plastic bins, three categories were identified (2007-2009, 2004-2006, 2001-2003). Bins from each category were assigned to treatments: pressure washing with water or pressure washing followed by cleaning and sanitizing. Different cleaning strategies were recommended by a partnering chemical supplier. All bins had water applied with a non-pressurized hose prior to cleaning and sanitizing. For wood bins, a powdered oxygen bleach was prepared with water in a 2:1 ratio as a cleaning solution. For plastic bins, an alkaline cleaner containing quaternary ammonium chlorides was prepared in a 2:1 ratio with water as a cleaning solution. During cleaning, areas that were heavily soiled were brushed. Following cleaning, the bins were rinsed with water and a sanitizer was applied. For both wood and plastic, a peroxyacetic acid (PAA) solution, diluted 1:100 was prepared to yield a target of 1200ppm, was prepared as recommended by the product label. Bins were rinsed with water approximately 15 minutes after application of the sanitizer. Microbial sampling was performed approximately one hour after the final water rinse for post-sanitation samples.

Results

Industry survey: Results from preliminary interviews indicated that a standardized, written survey was unlikely to result in collection of accurate information. Multiple individuals within a facility (food safety/quality, production, sanitation, orchard managers, among others) needed to

be consulted to gain accurate information. Therefore, an initial interview with food safety personnel was performed. The outcomes of this interview were documented and areas where additional information was needed were highlighted; the respondent was asked to consult with other company personnel and update the document. Individual interviews required more time for data collection, but it is expected that more accurate information was gained.

Results of the individual interviews reflect practices with bins associated with the Pacific Northwest tree fruit industry. Individual interviews were conducted with twelve food safety managers representing different packinghouses and anonymous poll was taken at an industry food safety meeting where more than twenty packinghouses were represented. Bins are typically owned by the packinghouse; therefore, the packinghouses are viewed as having the primary responsibility for cleaning and sanitation practices. Growers that find dirty or damaged bins are asked to return the bins to the packinghouse.

Results indicated that the majority of tree fruit packinghouses used wood bins for apples and pears. Bin liners were typically utilized for specific varieties that are more prone to bruising. Plastic bins and storage containers, when used, were more common for cherries and small plastic totes placed inside wood bins were more common for soft fruits. Bins were typically stored outside at packing facilities and were delivered to growers near harvest. The most common methods of bin cleaning and sanitation were immersion in a chlorinated dump tank system or hydrocooler, pressure washing or visual inspection with treatment of soiled bins.

Laboratory examination of sampling methods: Optimal sampling method varied with inoculation method, bin material and sampling time after inoculation (Figure 1). The optimal sampling method may depend on the cells' physiological state and stage of biofilm formation, which would not be known at the time of sampling in a commerical setting. It was determined that 3M Scotch-Brite[™] pads were the most consistent sampling method compared to cotton swabs or a metal file for biofilm recovery for both wood and plastic bin surfaces. Laboratory results, in the absence of background flora, indicated that VRBA with TSA overlay produced the most accurate results for microbial quantification (data not shown).

The experiments were designed to determine the optimal sampling technique and recovery media. However, it is interesting to note that generic *E. coli* levels tended to decline on wood pieces when the droplet method was used (with and without nutrient addition) in Figure 1A and 1C. For plastic pieces using the droplet method without nutrient addition, generic *E. coli* levels increased on day 1 and remained fairly stable through day 6 (Figure 1B); nutrient addition on day 4 promoted additional growth (Figure 1D). When the suspended method was used, both wood and plastic bin pieces supported biofilm development (Figure 1E and 1F).

Preliminary industry experiments: Results from the first facility indicated that microbial recovery was more accurate when bins were sampled immediately after exposure to chlorinated dump tank water than 24 hours after exposure to chlorinated dump tank water. This result may be influenced by a variety of factors, microbial die-off, absorption of microorganisms into the wood surface, among other factors. Bin tracking from the packinghouse to the orchard and return to the packinghouse was a successful strategy at this facility and allowed for the examination of bins that had been exposed to field conditions and contained fruit prior to exposure in the dump tank; empty, control bins that were stored at the packinghouse and sampled immediately before and after sanitation were also examined. Microbial levels were lower than expected; generic *E. coli* was recovered less frequently so total coliform levels were used for analysis of cleaning and sanitation effectiveness. Additionally, the background flora from industry samples proved challenging to differentiate from coliforms on the VRBA with TSA overlay media; therefore, CHROMagar[™] ECC was validated as an appropriate method in another series of experiments at a commercial facility.

Data indicated that wood bins had lower percentage of sampling sites positive for total coliforms prior to distribution to orchards (15% for wood versus 31% for plastic). After return from the orchard and treatment in a dump tank system, the number of sites positive for total coliforms were higher for both wood (39% increase) and plastic bins (13% increase); note the difference in magnitude is partially due to the plastic bins being more heavily soiled prior to distribution to the orchards. Challenges with dump tank management were observed in two facilities associated with this data which also likely contributed to the preliminary results. The project team identified additional project partners to further evaluate chlorinated dump tanks as a potential cleaning and sanitizing treatment. This study examined more than 80 bins over a five month period.

Examination of industry bin sanitation practices: Total coliforms were used to evaluate the ability of cleaning and sanitation practices to reduce microbial levels on bin surfaces; generic *E. coli* were recovered from bins infrequently and at levels that were too low to evaluate the effectiveness of cleaning and sanitation practices.

Examination of a chlorinated dump tank and bin washing system:

Dump Tank Treatment with Chlorine (average 774mV ORP):

- Wood Bins: 52% (11/21) had more sampling sites positive for total coliforms, 33% had no change and 14% had fewer sites positive for total coliforms.
- Plastic Bins: 56% (9/16) had no change in the number of sites positive for total coliforms, 25% had fewer sites positive and 19% had more sampling sites positive for total coliforms.

Results from this study indicated that immersion in a chlorinated dump tank system did not reduce the number of sites with total coliforms present for wood or plastic bins. Bins ranged from 1 min to 4 min 57 sec in the dump tank treatment. Although a bin washing system was evaluated at this facility, challenges with evaluating this system were encountered due to management practices; therefore, this data was not included in the results. This study examined 65 bins over a three month period and ended near the beginning of cherry harvest. Based on these results and results from the preliminary industry experiment above, it was determined that additional practices besides the most common industry practice should be examined. Due to timing and convenience for industry partners, additional experiments could not be performed until the beginning of apple season in August of 2014.

Examination of a peroxyacetic acid dump tank system:

Peroxyacetic Acid (PAA) Treatment (target 80 parts per million, ppm) in a Dump Tank:

- Wood bins: 72% had more sampling sites positive for total coliforms, 17% had no change and 11% had fewer sampling sites positive for total coliforms.
- Plastic bins were not available for testing.

The use of peroxyacetic acid in a dump tank system did not appear to improve the ability of a dump tank system to serve as a cleaning and sanitizing system for apple storage bins based on one replication; full replications of this experiment were not pursued due to project deadlines. Bins ranged from 1 min 37sec to 4 min 35 sec in the dump tank treatment.

Examination of a chlorinated dump tank followed by a heat treatment system:

Dump Tank Treatment with Chlorine* (average 829mV ORP):

• Wood Bins: 45% (12/29) had more sampling sites positive for total coliforms, 41% had fewer sampling sites and 14% had no change. Plastic bins could not be accommodated in the dump tank system.

Hot Water Treatment Following Chlorine Treatment:

• Wood bins after treatment in a chlorine dump tank system (noted above*) followed by hot water treatment: 68% had fewer sampling sites positive for total coliforms, 27% had no change and 5% had more sampling sites positive for total coliforms. Treatment in the dump tank system in this experiment ranged from 2 min 16 sec to 5 min 30 sec.

Hot Water Treatment Alone:

- Wood bins after hot water treatment alone: 48% (14/29) had no change in sampling sites positive for total coliforms, 41% had fewer sampling sites and 10% had more sampling sites positive for total coliforms.
- Plastic bins after hot water treatment alone: 57% had no change in sampling sites positive for total coliforms, 37% had fewer sampling sites positive for total coliforms and 6% had more sampling sites positive for total coliforms.

Hot water treatment warrants further examination as a sanitizing system for wood and plastic bins. Although the highest percentage of wood and plastic bins that received only the heat treatment demonstrated no change in sampling sites positive for total coliforms, the next most frequent response observed was fewer sites positive for total coliforms. In some cases, it appeared that soiled areas may have protected microorganisms during treatment (see photo set 1). Furthermore, for wood bins that were treated with chlorine prior to heat treatment, the majority (68%) had fewer sampling sites positive for total coliforms. Heat treatment ranged from 1 min 15 sec to 3 min 20 sec in the hot water treatment.

It should be noted that several experiments in this project, including this one, indicated that chlorinated dump tank systems did not appear to be effective as a cleaning and sanitation step. In this series of experiments, the chlorinated dump tank system appeared to enhance the ability of the heat treatment system to reduce the number of sampling sites positive for total coliforms on wood bins that received both treatments. There are several possibilities for this observation. Wetting the surface of the bins appears to enhance the recovery of microorganisms, so the exposure to the dump tank system prior to heat treatment may increase the ability of the heat treatment to reach microbial populations. No efforts were made to remove soil from the bins in this experiment; it is also possible that the dump tank treatment removed sufficient amounts of soil from some of the bin surfaces to enhance the effectiveness of the heat treatments that were not observed immediately after removal from the dump tank system. For this experiment, 86 bins were sampled over a three month period.

Examination of pressure washing with water alone, as well as pre-rinse followed by cleaning and sanitizing.

Pressure Washing with Water:

• Wood bins: 90% (9/10 bins) had more sampling sites positive for total coliforms after pressure washing with water.

• Plastic bins: 40% (4/10 bins) had no change, 30% had fewer sampling sites positive and 30% had more sampling sites positive for total coliforms after pressure washing with water.

Water pre-rinse followed by Cleaning and Sanitizing (average 1043ppm PAA):

- Wood bins: 65% (11/17) had fewer sampling sites positive for total coliforms after sanitation while 35% had more sampling sites positive for total coliforms.
- Plastic bins: 47% (8/17) had fewer sampling sites positive for total coliforms after sanitation while 29% had no change and 24% had more sampling sites positive for total coliforms.

A water pre-rinse (low pressure) followed by cleaning and sanitation steps were effective in reducing the number of sampling sites positive for total coliforms; this effect was more pronounced for wood bins compared to plastic. Pressure washing with water did not appear to be an effective strategy for wood bins; however, the results for plastic bins not as pronounced, given the limited data set (10 wood and 10 plastic bins examined). The project timeline limited the number of experiments that could be performed for this portion of the study. For this experiment, 54 bins were examined over a two month period, ending in December 2014.

Unexpected Outcomes: The terminology of cleaning and sanitation is not consistent or well understood among industry personnel; this lead to challenges in communication during the survey. Several facilities had challenges with dump tank management. These unexpected outcomes were managed through selection of partnering facilities.

Outcomes and Accomplishments

- This study was one of the first to examine cleaning and sanitation practices of tree fruit storage bins for produce safety; it is important to establish a baseline by evaluating current practices to determine appropriate action and recommendations. This baseline will assist in directing future research and validation of cleaning and sanitation practices.
- Laboratory studies identified the optimal sampling technique for removal of biofilms from wood and plastic storage bin surfaces.
- Preliminary industry studies identified the optimal media for recovery of total coliforms and generic *E. coli* from wood and plastic bins in the presence of background flora.
- Education on the difference between cleaning and sanitation is needed to ensure effective communication on food safety issues and implementation of appropriate practices. Increasing knowledge of chlorine chemistry and the importance of microbial control of cross-contamination using chlorine in dump tank systems is warranted. This not only includes facility personnel connected with food safety management, but also individuals connected with production, sanitation and upper management.
- Methods for wood and plastic bin cleaning and sanitation were identified.

Summary of Findings and Recommendations

Examination of industry bin sanitation practices:

• Data collection focused on establishing a baseline using current practices.

- Based on observation of bin handling practices, it was determined that the entire bin could be considered a food contact surface since the bins are completely submersed in dump tank water and could contribute to microbial levels and organic load in the dump tank system.
- Most current bin cleaning and sanitation practices observed did not involve efforts to remove heavily soiled areas prior to treatment, which can overwhelm effective cleaning and sanitation practices.
 - This observation makes drawing conclusions about the data collected in industry settings challenging, as some effective treatments may have appeared less effective on individual bins that heavily soiled (see Photo set 1).
- The data from this study indicated that a water pre-rinse followed by cleaning and sanitation would be an effective strategy for reduction of total coliforms on wood and plastic bins.
- Heat treatment appears to be a promising treatment for wood and plastic bins warrants further investigation.
- Development of a dry-dump mechanism that does not reduce fruit quality could also be an effective strategy to reduce organic load in dump tank systems and maintain dry surfaces for wood bins.
- Dump tank systems with chlorine or peroxyacetic acid for the majority of wood and plastic bins did not reduce the number of sites with total coliforms present.
- Pressure washing with water alone for the majority of wood and plastic bins did not reduce the number of sites with total coliforms present.
- Challenges with dump tank management, chlorine chemistry and water recycling issues were identified in this study. It is recommended that every facility review and evaluate practices related to dump tank management during packing as well as equipment cleaning and sanitation practices.
- Information gained through this project identified that tree fruit storage bins interface with the farming and fruit packing environment. It is reasonable that different standards and expectations for cleanliness and sanitation exist between these two environments. A challenge identified is that cleaning and sanitation currently rests with bin ownership at the packinghouse. However, the potential for collection of additional soil levels in the farming environment is anticipated. The presence of heavy soils on bins returning from the orchard could present challenges for sanitation in the packinghouse environment (storage and packing), challenges to dump tank management with chlorine sanitizers and challenges with protection of microorganisms during standard bin sanitation practices at the packinghouse. Adjustments to current cleaning and sanitation practices appear to be warranted. Greater communication and partnership between growers and packers to share responsibility for bin sanitation may also be warranted to address this complex agricultural issue.

Industry survey:

• The majority of the industry used wood bins for apples and pears; bin liners are primarily used for varieties that are more easily damaged. Plastic storage containers (bins or totes), when used, are more common for use with cherries and soft fruits, like apricots, peaches and plums.

• The most common methods of bin cleaning and sanitation were immersion in a chlorinated dump tank system or hydrocooler, pressure washing or visual inspection with treatment of soiled bins.

Laboratory and commercial examination of sampling methods:

- For the removal of biofilms from wood and plastic storage bin surfaces, 3M Scotch-Brite[™] pads were the most consistent sampling method. Prior to sampling, it is necessary to treat the 3M Scotch-Brite[™] pads with ethanol to remove antimicrobials that are present in the 3M Scotch-Brite[™] pads.
- Although laboratory studies found that Violet Red Bile Agar (VRBA) with an overlay was the most accurate method compared to VRBA and 3M E. coli/Coliform Petrifilm[™]; Preliminary studies in industry settings found an increased number of false positives with the Violet Red Bile Agar (VRBA) with an overlay. Therefore, CHROMagar[™] ECC was used in the remainder of the industry experiments.

APPENDICES

Publications and Presentations

Presentations:

Utah State Horticulture Association Annual Meeting. 2015. Spanish Fork, UT. Wood and plastic bin sanitation and other considerations for packinghouse food safety.

Washington State Horticultural Association Annual Meeting. 2014. Richland, WA. Validation of packingline food safety interventions and assessment of sanitation techniques for storage bins.

Great Lakes Fruit, Vegetable and Farm Market Expo. 2014. Grand Rapids, MI. How effective are different sanitation techniques for tree fruit storage bins?

Center for Produce Safety, Annual Symposium. 2014. Newport Beach, CA. Assessment of sanitation techniques for tree fruit storage bins.

Center for Produce Safety, Annual Symposium. 2014. Rochester, NY. Assessment of sanitation techniques for tree fruit storage bins. Poster presented and abstract published in the Center for Produce Safety Symposium Program. 25.

Pacific Northwest Horticultural Council. Food Safety Committee Meeting. 2014. Research Review and Update.

Publications for peer review and extension publications are being prepared based on project outcomes.

Budget Summary

Funding supported personnel, sampling supplies and processing as well as travel. Approximately \$101,575 was used to support technician and staff salary and benefits as well as wages and benefits for undergraduates. Project supplies amounted to \$54,808 and travel expenses were \$14,864. Total project funding was sufficient; however, personnel needed for this study was greater than anticipated.

Tables and Figures

Figure 1. Generic *E. coli* levels on wood and plastic bins pieces inoculated using conditions to reflect commercial conditions (droplet method) or for healthy biofilm conditions (suspended method) with microbial recovery using cotton swabs, metal files or 3M Scotch-Brite[™] pads on the day of inoculation or 1, 4 and 6 days later and enumerated on VRBA with TSA overlay.

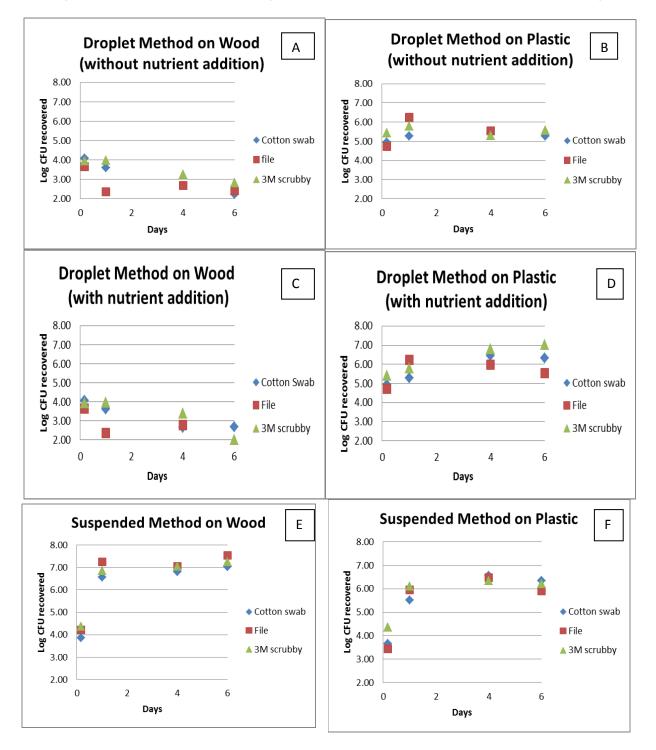


Photo Set 1. Wood bin, 2014. All six sampling sites less than 10 cfu/inch² prior to treatment; after heat treatment, 1 heavily soiled sampling site had 14,700 total coliforms cfu/inch². Plastic bin, 2008. 5/6 sampling sites had less than 10 cfu/inch² prior to heat treatment and one site had between approximately 40 total coliforms cfu /inch². After heat treatment 5/6 sampling sites had less than 10 total coliforms cfu/inch² and one soiled site 14,700 total coliforms cfu/inch². Both of these bins were classified as having more sampling sites positive for total coliforms after treatment.

2014 Pre- Heat 6/6 <10 CFU/in² coliforms 2014 Post-Heat Treatment 5 samples <10, 1 sample >10,000 CFU/in² coliforms



2008 Plastic Bin Pre- Heat 5 samples <10, 1 sample >10 CFU/in² Generic *E.coli*

2008 Plastic Post-Heat 5 samples <10, 1 sample >10,000 CFU/in² Generic *E.coli*



Suggestions to CPS

A section to mention acknowledgements would be appreciated.

Acknowledgements: Several industry partners donated staff time and facility resources to conduct research experiments; this involved several planning meetings, use of line time during normal production and troubleshooting efforts. Numerous industry partners contributed to this project with their time to respond to initial and follow-up interviews. Dr. Ines Hanrahan with the Washington Tree Fruit Commission provided insight and was involved in project planning and implementation for bin tracking systems. Dr. Hanrahan and several WTFRC interns were significant contributors and assisted with data collection throughout the study. The Northwest Horticultural Council also provided insight and recommended industry contacts throughout the study. Sincere thanks to all involved in data collection and analysis, including Tonia Green, Kim Thayer, Lauren Walter, Rachelle Unrau, Joel Small, Jessica Lie, Dr. Yen-te Liao, Eric Hughes Kyu Jeong, and numerous undergraduate students as well as Dr. Achyut Adhikari for collaboration on grant writing for this project. The opportunity to work with this team and the Pacific Northwest tree fruit industry was absolutely tremendous. A sincere thanks to representatives of Birko and Pace International for donations and recommendations. The support provided from the Center for Produce Safety and input from the technical committee is greatly appreciated.

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