IFAS Citrus Initiative Annual Research and Extension Progress Report 2006-07 Mechanical Harvesting and Abscission

Project title: Effect of Mechanical Harvesting Systems on Fruit Microflora

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Objectives:

- (1) Evaluate the microbiology of the surface and juice of citrus fruit collected by the OXBO pick up systems
- (2) Evaluate the amount of sand present on the surface of citrus fruit collected by the OXBO pick up system.

Progress on First Objective:

For Florida to effectively compete in the world citrus industry, significant reductions in harvesting costs will be necessary. Mechanical harvesting (MH) can be thought of a two-step process: (i) removal of fruit from the tree and (ii) collection of fruit (immediately, by a catch-frame device or during retrieval of fruit from the ground). This study evaluates the microbiological aspects of mechanically-handled fruit with respect to fruit surface microflora and corresponding fruit juice microflora.

Four replicates of mechanical harvesting were performed through May 31, 2007 and three treatments groups were evaluated from each replicate: (i) hand-harvested fruit (control), (ii) mechanically-harvested fruit (picked up directly from ground) (MH fruit), and (iii) mechanically-harvested fruit in combination with the OXBO pick-up machine (MH/PU fruit). Within each group of fruit (control, MH, and MH/PU) 75 oranges were collected, and 25 non-defective fruit were randomly selected from each group for analysis.

Thirty milliliters of buffer were added and the rub/shake/rub technique was sued to remove microorganisms from the fruit surface. Microbial analysis included total aerobic plate count (APC) on plate count agar (PCA), and acidophilic organisms count (AOC) on orange serum agar (OSA). Results are reported in Table 1 as colony forming units (CFU) per ml of wash buffer.

As an indicator of potential contamination, generic *Escherichia coli* and *Salmonella* testing was also done on pooled samples of 5 oranges (five pooled samples per test group). Pooled samples, rather than individual oranges were used due to the expense and time required to run these tests. The VIP *Salmonella* test kit (BioControl) and E*Colite TM (Charm Sciences) were used to test for *Salmonella* and *E. coli* respectively. Results are reported as presence or absence of these organisms in Table 2.

Following testing the fruit surface, all samples were stored at 4°C for 18 ± 2 h. Oranges were then placed in 85°C water for 2 min to sterilize the surface of the fruit and juiced by hand through cheese cloth into a sterile container. Parallel microbial testing was done for juice samples and is reported as CFU/ml juice in Table 3 and as presence or absence of these organisms in Table 4.

In general control fruit had fewer microbes on the surface of the fruit when compared to MH and MH/PU fruit, however no real trends can be attributed to harvest method for total fruit

surface microbial loads for all runs (Table 1). Significantly lower APC counts for control fruits are often expected as these fruits were not in contact with the soil surface, the source of many microorganisms on agricultural products. However, this was not true for all trials in 2006/2007, and is consistent with results reported in 2005/2006. This result suggests that dropping fruit to the ground and/pr picking it up mechanically does not necessarily result in higher microbial loads. Moisture, and other environmental variables, such as soil type and cover crop, may ultimately be very important in total number microorganisms that adhere to the dropped fruit. Trial 2 was conducted during a rain event, yet more variability between treatments existed in trials three and four, thus other factors, such as mechanical action of the machines, equipment sanitation, grove location and tree/fruit treatments during production may also lead to variability.

Results of the AOC analysis follow the same general trend as those for APC. In general, there are no significant differences among the three treatment groups. Many factors contribute to the surface microflora of a raw agricultural product. These include production practices, natural ecology of the fruit/microorganism system, equipment sanitation, geography and climate, and hygiene of harvest and packinghouse personnel. All of these factors may have impacted the results obtained from this study.

E. coli was detected in four of the pooled samples in MH and in three of the pooled samples for MH/PU fruit, and *Salmonella* was detected in one of the pooled MH/PU fruit, however there is no correlation between the presence of *E .coli* and *Salmonella*. The *Salmonella* strain isolated has been sent out for serotyping. Any fruit in contact with soil has the potential to become severely contaminated, although that was clearly not the case for many of the fruit that were picked up from the ground, either by hand or mechanically. However, this one result emphasizes the need for proper grove floor observation and maintenance, as well as equipment cleaning and care. Soil, other organic materials present on the orchard floor, and machines are potential sources of contamination of fruit surfaces.

In all cases, juice samples contained significantly less microflora than the corresponding fruit, often times being at or below the limit of detection (Table 3). For both APC and AOC in fruit juice, no real trends can be attributed to the harvest method. The interior of sound fruits harbor few microbes as is apparent by these results. The indicator organism *E. coli* and *Salmonella* were not detected in any of the juice samples, despite the presence of these organisms on the fruit surface in some samples.

No indication that fruit which come in contact with the ground are consistently and significantly higher in microbial surface contamination is indicated by the results of the four trials run during 2006/2007 study. This result is consistent with results reported in 2005/2006. Previously no *Salmonella* was detected on fruit surfaces. The overall level of *Salmonella* contamination on any raw agricultural product is quite low (1-3%; U. S. FDA), and the result of 1 out of 60 positive samples is not entirely unexpected. However, given the intimate contact of fruit from two of the treatment groups with the soil surface, this result is an encouraging one in the development of harvesting/collection systems that rely on some sort of pick up system. It should also be noted that the one *Salmonella* isolated occurred during a rain event, which have been observed to increase the likelihood of *Salmonella* isolation in other crops (Uesugi et al., *In Press*). The result for *E. coli* was also 7 out of 60 positive is significantly higher than the 0 out of 60 samples reported in previous years, and indicates that there contamination loads on any of the sample groups of this indicator organism remains relatively low. While generic *E. coli* are not considered foodborne pathogens, their presence can be indicative of fecal contamination

from warm-blooded animals. However, no correlation between *E. coli* and *Salmonella* presence in this small sample group was noted.

There is practical importance to the surface microflora of oranges delivered to the processor. Contamination of raw materials is listed as the second most serious food safety problem in the food processing industry, after deficiencies in employee training. However, incoming fruit to citrus processing plants is typically washed and sanitized, and the vast majority (>98%) of Florida-processed orange juice is pasteurized or similarly treated to inactivate spoilage enzymes and to microbiologically stabilize the product. Wider adoption of mechanical harvest/pick up systems will be somewhat determined by the quality of fruit delivered to the processor. This quality includes potential microbiological contamination as well as the typical measures of machine yield and efficiency, and economics. For these reasons, it is important to continue to collect fruit and juice microbiological quality information for any harvest/collection system that promises commercial viability, and due to the role environmental factors may play on this quality, to include more sampling sited in widely different groves and weather conditions.

This work has been presented at the Florida State Horticultural Society's annual meeting June 4th, 2007 in Palm Beach Gardens Fl, and will be published in the Proceedings of that meeting.

Progress on Second Objective:

Due to processor concerns with regards to the amount of sand present in mechanically harvested samples, experiments were undertaken to quantify the influence of mechanically harvesting on sand levels. Within the three test groups, previously described, collected from the field, ten fruit samples were selected and washed with tap water. The wash water was allowed to settle, and was filtered. Results of sand collection are shown in Table 5, and a visual representation of sand levels for trial three is shown in Figure 1. Although, results presented in Table 5 do not appear convincing that significantly increased the amount of sand on the MH/PU fruit), Figure 1 clearly demonstrated the increase in sand present following mechanical harvesting. Data from these four trials indicate that fruit surface sand levels are affected by harvesting method. However, this is the first year sand levels were measured, and other studies to compare this work to have not been identified. For these reasons, it is important to continue to collect fruit surface sand levels in the future in more sampling locations.

This work has been presented at the Florida State Horticultural Society's annual meeting June 4th, 2007 in Palm Beach Gardens Fl, and will be published in the Proceedings of that meeting.

Table 1. Fru	uit surface microflora in	n log colony formi	ng units (CFU) per	ml of orange wash	for
four trials of	f mechanical harvestin	g (MH) with and	without pick up (P	U) machines (n = $(n = 1)$	25
oranges).					
Trial	Control	MII	N/II/E	TT	

Trial	Control	MH	MH/PU
	Log CFU/ml on PCA		
1	3.6 ± 0.4	3.6 ± 0.6	3.5 ± 0.4
2	3.8 ± 0.4	4.3 ± 0.3	4.5 ± 0.4
3	3.0 ± 0.8	4.3 ± 0.4	4.4 ± 0.6
4	3.6 ± 0.4	4.7 ± 0.3	5.3 ± 0.5
	Log CFU/ml on OSA		
1	3.9 ± 0.6	3.8 ± 0.4	3.8 ± 0.6
2	4.1 ± 0.5	4.3 ± 0.4	4.4 ± 0.5
3	3.7 ± 0.4	4.5 ± 0.5	4.6 ± 0.4
4	3.8 ± 0.3	4.8 ± 0.5	4.1 ± 0.5

Table 2. Summary of fruit surface indicator (*E. coli*) and pathogenic organisms (*Salmonella*) for four trials of mechanical harvesting (MH) with and without pick up (PU) machines (n = 5 enrichments).

Trial	Control	MH	MH/PU
	E. coli enrichments		
1	0	3	3
2	0	0	0
3	0	1	0
4	0	0	0
	Salmonella enrichments		
1	0	0	0
2	0	1	0
3	0	0	0
4	0	0	0

Table 3. Juice microflora in log colony forming units (CFU) per ml for four trials of mechanical harvesting (MH) with and without pick up (PU) machines (n = 25 juice samples).

Trial	Control	MH	MH/PU
	CFU/ml on APC		
1	1.4 ± 2.2	0.3 ± 0.3	0.8 ± 2.7
2	> 0.02	0.02 ± 0.1	0.06 ± 0.2
3	> 0.02	1.1 ± 5.5	0.2 ± 0.6
4	0.3 ± 0.8	0.3 ± 0.6	2.0 ± 3.2
	CFU/ml on OSA		
1	0.3 ± 0.6	0.3 ± 0.5	0.6 ± 1.5
2	> 0.02	> 0.02	0.04 ± 0.2
3	> 0.02	0.04 ± 0.2	0.04 ± 0.3
4	0.1 ± 0.4	0.1 ± 0.3	0.1 ± 4.5

Trial	Control	MH	MH/PU
	E. coli enrichments		
1	0	0	0
2	0	0	0
3	0	0	0
4	0	0	0
	Salmonella enrichments		
1	0	0	0
2	0	0	0
3	0	0	0
4	0	0	0

Table 4. Summary of juice indicator (*E. coli*) and pathogenic organisms (*Salmonella*) for four trials of mechanical harvesting (MH) with and without pick up (PU) machines (n = 5 enrichments).

Table 5. Sand collected from fruit surface for four trials of mechanical harvesting (MH) with and without pick up (PU) machines (n = 10 oranges).

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Trial	Control	MH	MH/PU	
	Milligrams/cm ²			
1	0.02	0	0.058	
2	0.038	0.107	4.8	
3	0.032	0.11	0.36	
4	0.00	0.108	0.056	

