PROJECT REPORT

Title: Aureobasidium rot in sweet cherry, 2003

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No information exists on the ability of *Aureobasidium pullulans* to infect fruit postharvest. This study was conducted to determine the frequency of occurrence of *A. pullulans* in cherry packing water systems and if fruit could be infected after exposure to *A. pullulans* in those systems. *Aureobasidium pullulans* colonizes stylar tissue of sweet cherries during the growing season, and may remain latent until fruit senesces at which time it may spread to healthy tissue causing a lesion to develop (Dugan and Roberts, 1994). In studies on the effects of MAP liners on extension of shelf life of cherries in 1999 and 2000, *A. pullulans* was the predominant cause of decay in commercially packed fruit (45% and 50.3% of fruit decayed in 1999 and 2000, respectively) (Kupferman and Sanderson, 2001).

The water systems at a packinghouse were assessed for microbial contamination in 2002. A relatively high density of *A. pullulans* was present and few other fungi were found (unpublished data). This packinghouse had a shipment of fruit rejected due to decay. Although the predominant disease in that rejected lot of fruit is not known, our previous experience suggests that *A. pullulans* was the most likely cause.

Significant findings

- *Aureobasidium pullulans* was found in cherry water systems in all 7 packinghouses surveyed
- In each water system, *A. pullulans* was recovered about 45% of the time.
- Highest population densities were found in dump tanks.
- Population densities diminished in succeeding water systems.
- *Aureobasidium pullulans* was not recovered in water systems with total chlorine >50 ppm.
- Both wounded and unwounded fruit were susceptible to infections caused by *A. pullulans* in water systems at densities >100 CFU/ml.
- Stemless cherries were less susceptible to infection than stemmed cherries.

Materials and methods

Packinghouse survey

Water samples were collected from each independent water system at seven Washington State cherry packinghouses. Chlorine in the water was neutralized with sodium thiosulfate. Water temperature, pH and total chlorine were measured in each water system at the time of sampling. Water samples were cultured on potato dextrose agar (PDA) containing antibiotics (chloramphenicol and gentamycin sulfate) and fungal colonies enumerated to determine the amount of *Aureobasidium* spp. and other pathogenic fungi present in each water system.

Fruit inoculations

Treatments were applied to commercially harvested Bing cherry fruit obtained from three North Central Washington packinghouses. Fifty fruit from each of the packers were used for replicates. Fruit were surface disinfested by dipping them in 100-ppm chlorine for 5 min. Cherries were either wounded (0.5mm diam x 2mm deep) a single time on the equatorial axis or unwounded. Stems were removed from an additional subset of fruit just before inoculation.

Three isolates of *A. pullulans* that had been previously recovered from cherry fruit were grown for 7 days on PDA at room temperature for inoculum. Fruit were inoculated by dipping them into aqueous suspensions of *A. pullulans* (0, 100, 1000, and 10,000 cfu/ml). Following inoculation the cherries were placed into aluminum pans lined with a damp paper towel, covered with plastic lids and stored for 2 weeks at 40°F before being assessed for lesion development. Isolations were made from lesions to assure that *A. pullulans* was the causal agent. The trial was repeated once. Results were similar for each experiment so data were combined for statistical analyses.

Results and discussion

Packinghouse survey

Aureobasidium pullulans was found in about equal occurrence (about 45%) in each of the waters systems sampled (Table 1). The only type of water system in which *A. pullulans* was not recovered was a single sample taken from a flume into which Captec had been added. However, dump tanks were the most heavily contaminated followed by hydrocoolers (Table 2). Each succeeding water system following the dump tank tended to be less contaminated. *Penicillium* spp., especially *P. expansum* was the only other fungus recovered frequently from water systems. *P. expansum* was the second most frequent cause of decay in previous experiments with MAP in 2000 (35% of decayed fruit).

Aureobasidium pullulans population densities varied widely among packinghouses and among sampling times within a packinghouse (Table 2). This variation was most closely correlated with the use of chlorine in the water systems, which ranged from no chlorine to 80-100 ppm in each system (Table 3). Temperature and pH of water systems also varied markedly among packinghouses. Highest populations of both *A. pullulans* and *Penicillum* spp. were recovered from water systems with no chlorine. Neither fungus was recovered from water that contained >60 ppm total chlorine (fig. 1).

Fruit inoculations

Fruit inoculated with *A. pullulans* in water developed Aureobasidium rot lesions. Fruit treatment had a differential effect on disease development. Disease incidence in fruit with stems increased with inoculum density >100 CFU/ml in both wounded and unwounded fruit (fig. 2). The incidence of decay in fruit inoculated with 0 and 100 CFU/ml was not significantly different. About 90% of lesions developed in the wounds of wounded fruit (Table 4). In unwounded fruit, most lesions developed at the stylar end, but about 29% of lesions developed on the sides.

In fruit with stems removed, most lesions developed at the point of stem attachment (Table 4). However, in stemless fruit, the incidence of decay was very low and there was no significant difference in the incidence of decay with inoculum dose (fig. 2). It is possible that removal of the stem caused a resistance response in the fruit, but the reason for this phenomenon is unclear.

Conclusions

This is the first report of sweet cherry spoilage caused by postharvest inoculation by *A. pullulans*. Inoculum loads >100CFU/ml in packinghouse water systems may cause significant decay losses. Chlorine at >60 ppm was effective at eliminating *A. pullulans* inoculum from water systems.

Literature cited

Dugan, F. M. and Roberts, R. G. 1994. Etiology of preharvest colonization on Bing cherry fruit by fungi. Phytopathology 84:1031-1036.

Kupferman, E, and Sanderson, P. G. 2001. Temperature management and modified atmosphere packing to preserve sweet cherry quality. Postharvest Information Network, July 2001. 9 pp.

Table 1. Frequency of occurrence of *Aureobasidium pullulans* and *Penicillium* spp. found in cherry packing water systems.

Water system	*Aureobasidium pullulans*	*Penicillium* spp.
Hydrocooler (n=12)	41.7 %	25.0 %
Dump tank (n=18)	44.4 %	38.9 %
Cluster cutter (n=15)	46.7 %	46.7 %
Flume (n=12)	41.7 %	8.3 %
Fungicide TRT (n=1)*	0.0 %	0.0 %

* Captec in flume water at full label rate

Table 2. Density of *Aureobasidium pullulans* and *Penicillium* spp. in Washington State cherry packinghouse water systems.

	Aureobasidium pullulans		*Penicillium* spp.	
Water system	cfu/ml	(± s.d.)	cfu/ml	(± s.d)
Hydrocooler (n=12)	222.2	655.9	5.0	12.0
Dump tank (n=18)	1284.8	3810.4	67.8	234.1
Cluster cutter (n=15)	106.4	202.4	7.3	9.6
Packing water (n=12)	11.1	15.3	0.6	1.9
Fungicide TRT (n=1)*	0.0	0.0	0.0	0.0

* Captec in flume water at full label rate

Table 3. Physical characteristics of Washington State cherry packinghouse water systems.

Water system	Total chlorine (ppm)	pH	temp °C
Hydrocooler (n=12)	33.3 (0 – 80)*	5.8 (2.8 – 7.1)	4.9 (0.2 – 16.0)
Dump tank (n=18)	38.4 (0 – 102)	7.1 (6.2 – 7.6)	8.9 (2.2 – 15.0)
Cluster cutter (n=15)	32.6 (0 – 100)	7.2 (6.8 – 7.5)	7.9 (1.8 – 16.8)
Packing water (n=12)	34.2 (0 – 90)	6.4 (4.0 – 7.5)	4.0 (0 – 17.9)
Fungicide TRT** (n=1)	0.0	7.2	3.0

* Mean and range

** Captec in flume water at full label rate

Table 4. Locations of *Aureobasidium* rot lesions on cherry fruit surfaces.

Fruit treatment	Cheek	Stem end	Stylar end	Wound
Stemless/unwounded	11.1 %	82.2 %	6.7 %	--
Stem/unwounded	28.6 %	8.4 %	63.0 %	--
Stem/wounded	5.4 %	1.5 %	3.5 %	89.6 %

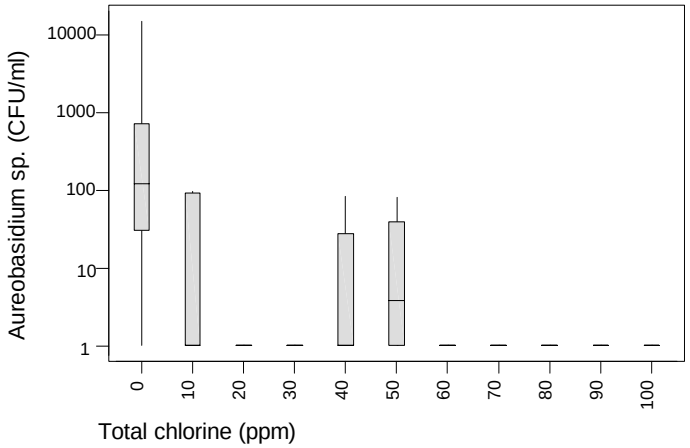

Figure 1. Effect of chlorination on inoculum loads of Aureobasidium pullulans and Penicillium spp. in cherry packinghouse water systems.

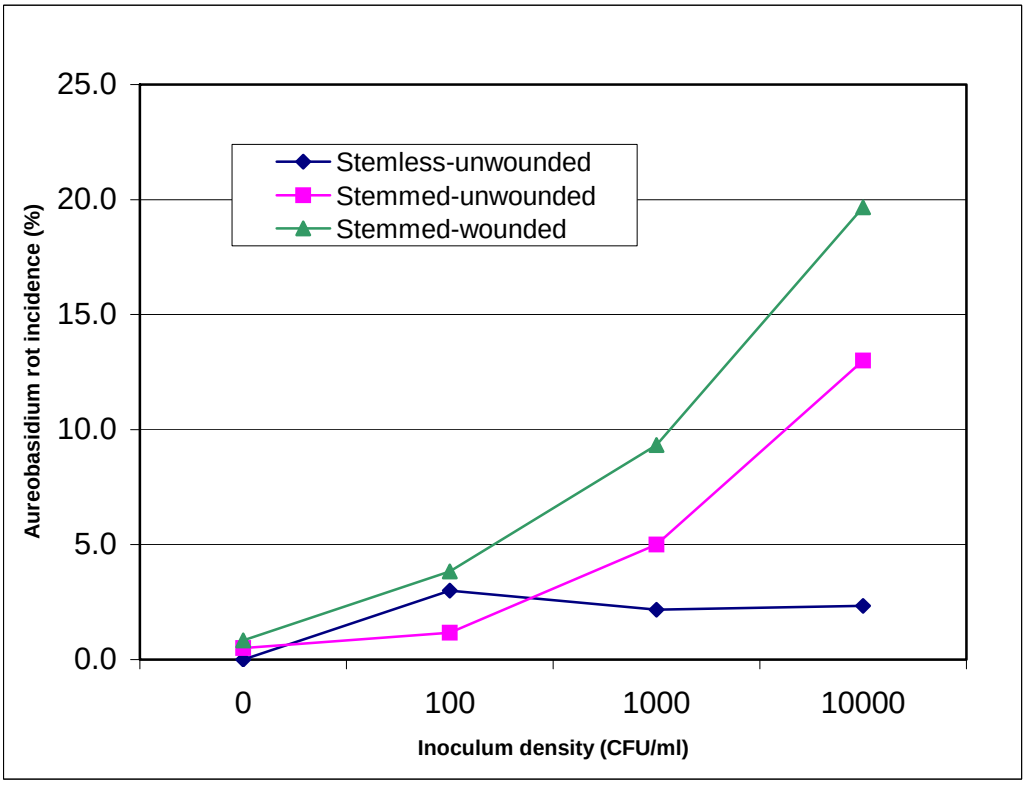

Figure 2. Incidence of decay in sweet cherry resulting from inoculation with increasing doses of *Aureobasidium pullulans*