Fungicides, Biocides, and Sanitizers for Managing Postharvest Pathogens in Apples

David A. Rosenberger
Deptartment of Plant Pathology
NY State Agric. Exp. Station, Cornell University

This work was supported in part by the New York Apple Research and Development Program.

Maintaining fruit quality and ensuring food safety are two critical objectives for operators of apple storages and packinghouses. Over the past ten years, the apple industry has acquired new options and tools for storing apples. However, selecting the best options and integrating the various components into a safe and cost-effective management scheme has added complexity to storage and packing operations. In addition, there is increasing scrutiny of postharvest practices for all fresh produce due to the repeated outbreaks of illness from food-borne pathogens in recent years.

> **"Over the past ten years, the apple industry has acquired new options and tools for storing apples. Fungicides can reduce postharvest losses while biocides are essential for controlling microbial populations in water flumes. Selecting the best options for preventing decays, preserving fruit quality, and ensuring food safety requires knowledge of the available options coupled with careful assessment of how selected options will mesh with market place requirements."**

To further complicate matters, many large apple buyers (retailers) are imposing their own audits and restrictions on how apples are grown, stored, and packed. Although the audits/restrictions from retailers are nominally related to concerns about food safety and sustainable production systems, the advertising components of retailer-driven audits (i.e., my store is "greener" than your store) often appear to outweigh valid concerns about food safety and sustainability. Nevertheless, these constraints must be factored into postharvest management strategies despite the fact that they add needless expense and complexity to apple storage and packing/sales operations.

The remainder of this article presents information on various options that can be used for managing postharvest decays of apples while also minimizing risks that fresh apples might carry human pathogens.

Fungicides, Cleaners, Biocides, and Sanitizers

Fungicides are used specifically to control fungal pathogens. In stored apples, the two major fungal pathogens are *Penicillium expansum*, the cause of blue mold, and *Botrytis cinerea,* the cause of gray mold. Apart from controlling fungi that, like *P. expansum,* can produce mycotoxins, fungicides have no value for controlling the microbial organisms involved in food safety issues.

Biocides and sanitizers have a broader range of biological activity because, when applied under appropriate conditions, they can kill virtually all fungal, bacterial, and protozoan organisms. Most sanitizers could also be called biocides, but the term "sanitizer" is commonly applied to products used to kill microorganisms on hard surfaces whereas "biocide" is used for products incorporated into aqueous solutions such as apple flotation tanks and water flumes in packinghouses. Neither sanitizers nor biocides will kill microbes protected within decaying organic matter (e.g., a rotten apple) or within organic films that can persist on hard surfaces. Thus, sanitizers are effective only when applied to surfaces that have already been cleaned using a detergent or other cleaner to remove debris and films that can protect undesirable microbes.

The sanitizers and biocides available for apple packinghouses are oxidizing agents that kill microbes by disrupting cell membranes. Bacteria and other microbial organisms that can cause food borne illnesses are easier to kill than are spores of *P. expansum*, so a biocide that controls bacterial pathogens will not necessarily eliminate fungal spores.

Fungicide Options for Managing Postharvest Decays

Blue mold was controlled from the early 1970's through the mid-1990s by drenching fruit after harvest with benzimidazole fungicides such as Benlate, Topsin M, or Mertect 340F. (Only the latter is still registered for postharvest applications today.) Even though *P. expansum* with resistance to benzimidazoles could be detected in many packinghouses by the late 1970s, these fungicides continued to control decays because the benzimidazole-resistant strains of *P. expansum* were controlled by diphenylamine (DPA), an antioxidant that was always included in postharvest drenches as a control for the physiological disorder known as storage scald. By the mid 1990s, populations of *P. expansum* in many large packinghouses had become resistant to the benzimidazole/DPA combination, and huge quantities of inoculum began to cycle from one year to the next on contaminated apple bins.

New fungicides registered in recent years have provided effective options for controlling *P. expansum* (Table 1). The four fungicides currently labeled for postharvest applications on apples are Captan, thiabendazole (Mertect 340F), fludioxonil (Scholar) and pyrimethanil (Penbotec). In addition, application of Pristine fungicide shortly before harvest can suppress postharvest decay pathogens if the residues from the last field spray are not eliminated by rainfall prior to harvest. All of these fungicides have different modes of action, a fact that is useful for fungicide-resistance management.

Fungi will never develop resistance to Captan because Captan attacks multiple biochemical sites to prevent spore germination. However, resistance to both Scholar and Penbotec could develop relatively quickly (perhaps after four or five years of continuous use). To preserve activity of these new fungicides, packinghouse operators should adopt one of the following resistance management strategies:

Table 1. Fungicides useful for managing postharvest pathogens in apples.

Trade name	chemical name	FRAC* group and comments on application method and/or usefulness
Captan	**captan**	Group M4*. Although relatively ineffective for protecting fruit if viable Penicillium spores enter fruit wounds, Captan can suppress decay by reducing spore viability and thus inoculum load in recirculating drenches. Captan should not be used alone, but it can be useful for reducing selection pressure for resistance to other fungicides when tank-mixed with other fungicides in postharvest drenching systems. Labeled rates per 100 gal: 2.5 lb for Captan-50, 1.6 lb for Captan-80, or 1.25 qt for Captec 4L.
Mertect 340F	**thiabendazole**	Group 1*. Used in postharvest drenching systems, it provides excellent control of Penicillium and Botrytis so long as benzimidazole-resistant strains of these pathogens are not present. Some benzimidazole resistant strains are still controlled when Mertect is applied with diphenylamine, but Penicillium populations in many storages are now completely resistant to the Mertect/DPA combination. Labeled rate: 1 pt/100 gal.
Penbotec	**pyrimethanil**	Group 9*. Used in postharvest drenching systems, it provides excellent control of Penicillium and Botrytis. To prevent fungicide resistance, do not use this product more than two years in succession and consider using it in combination with Captan. Labeled rate: 1 pt/100 gal.
Scholar	**fludioxonil**	Group 12*. Used in postharvest drenching systems, it provides excellent control of Penicillium and Botrytis and may suppress latent infections of some other summer fruit rot fungi (e.g., Botryosphaeria sp.) To prevent fungicide resistance, do not use this product more than two years in succession and consider using it in combination with Captan. Labeled rate per 100 gal (as per FIFRA Section 2(ee) label issued 1 Sept. 2006): 6 oz for Scholar 50WP; 10 fl oz for Scholar SC (new formulation with label pending).
Pristine	**pyraclostrobin plus boscalid**	Groups 7 & 11*. No registration for postharvest applications. Preharvest sprays applied to control black rot, white rot, bitter rot and flyspeck may also suppress blue mold. Preharvest sprays are especially beneficial for pears that will be stored for more than a few weeks. However, rains that occur between the last field application and harvest may reduce effectiveness against postharvest pathogens. Labeled rate: 1 lb/A.

* FRAC = Fungicide Resistance Action Committee; FRAC has classified fungicides into more than 60 groups based on their biochemical mode of action. Risks of developing fungicide resistant pathogens can be reduced by alternating among fungicide groups or by applying tank mixtures involving fungicides from different groups.

1. **Stop using postharvest fungicides.** This might be feasible if DPA can be applied to filled storage rooms via fogging so that fruit need not be exposed to recycling drench solutions after harvest. Blue mold incidence usually drops to insignificant levels if fruit are not exposed to recycling drenches after harvest. In some years, however, fruit not treated with a postharvest fungicide may come out of long-term storage with more than five percent of fruit infected with gray mold. Gray mold decays usually develop from quiescent infections that were present on fruit at harvest. The number of fruit carrying quiescent infections varies from year to year and is probably affected by fungicides applied between petal fall and harvest. Unfortunately, we have no way of predicting the incidence of gray mold that will develop on non-treated fruit. The Mertect/DPA combination has continued to control gray mold even in storages where it no longer controls blue mold, so we really don't have any good indicators of how much gray mold might develop in the absence of postharvest fungicide treatments. However, gray mold incidence will presumably be higher in years when extensive rainfall occurred at or shortly after petal fall (thereby allowing *B. cinerea* to colonize dying petals and then move into sepals) or in years with extensive wet periods during the several weeks prior to harvest.
2. **Vary the fungicide that is used from one year to the next.** Where Mertect or the Mertect/Captan combination is no longer effective, Penbotec should be used one year followed by Scholar the following year. Where the Mertect/Captan combination is still effective, a three-year rotation involving Penbotec, Scholar, and Mertect or Mertect/Captan could be employed. Because most of the inoculum for *P. expansum* cycles from one year to the next on field bins, the objective of rotating fungicides is to ensure that bins are not exposed to the same fungicides more than two or three times in direct succession. Mertect, Penbotec, or Scholar in drench water will not kill all of the spores on dirty bins because these fungicides seem to be more effective for preventing infection in fruit wounds than for killing inactive spores on bin surfaces. However, varying the fungicide that is used from year to year will decrease the likelihood that the population of *P. expansum* surviving from year to year on bin surfaces will become resistant to the postharvest fungicides.
3. **Use Captan in combination with Mertect, Penbotec, or Scholar** in postharvest applications. The rationale for this approach is explained in the next section.
4. **Combine the previous two options** so that Mertect, Penbotec, and Scholar are used in a two or three-year rotation and each of them is always used in combination with Captan. This is the optimal resistance management strategy, but it may not prove feasible if some markets will not accept fruit treated with Captan or one of the other fungicides. It may also be more costly than other options.

Recent Research on Captan as a Postharvest Fungicide

In the numerous postharvest tests that I conducted over the past 25 years, Captan rarely provided more than 50% control of blue mold. I therefore concluded that Captan was not very useful as a postharvest fungicide. In most of these tests, wounded apple fruit were exposed to spores of *P. expansum* and were then dipped into solutions of Captan. Recently, however, I realized that while Captan may be relatively inefficient for protecting apple wounds from infection, it might benefit postharvest disease control by killing the accumulated spores in the recirculating drench solutions, thereby reducing the inoculum load in the recycling drench water.

To test this hypothesis, we conducted a trial in fall of 2008 where we prepared Captan fungicide solutions, added a known quantity of *P. expansum* spores to those solutions, and then dipped freshly wounded Empire apples into the solutions immediately or after 6, 12, 24, 48, or 72 hours. The solutions were agitated before each set of fruit was treated and again at various intervals between the latter three treatments. Other treatments included for comparison were a water control containing the same inoculum and a second Captan/inoculum solution amended with soil and organic debris to simulate the conditions that occur as recycling drench solutions accumulate dirt and debris from bins being drenched. We suspected that the nutrients from this debris might be critical for stimulating early stages of spore germination for *P. expansum* and that Captan might kill spores more effectively after germination was initiated than when spores remained in a dormant state as they do in nutrient-free water.

Results from this trial showed that disease control with Captan used at the maximum labeled rate increased from less than 45% for fruit treated immediately after solutions were prepared to more than 75% for solutions that were held for 72 hours before fruit were treated (Figure 1). We also assessed the viability of spores in the Captan/inoculum solutions by dilution-plating some of the solutions each time that fruit were treated. Results from the dilution plating showed that spore viability decreased with increasing exposure time. These results support the hypothesis that Captan gradually reduces spore viability in aqueous solutions, thereby reducing inoculum loads and producing a concomitant reduction in fruit infections. Although we have not yet tested Mertect, Penbotec, and Scholar for their sporicidal capabilities, it is generally accepted that these fungicides are fungistatic rather than fungicidal. That means that spores exposed to these fungicides will not be killed and can germinate normally if the fungicide residues are removed or diluted. Mertect, Penbotec, and Scholar presumably work by preventing invasion of fruit tissue rather than by killing spores directly whereas Captan can kill spores directly.

Captan used alone will not provide adequate protection of wounded fruit going through a drencher system. However, other fungicides used in combination with Captan will perform better when fruit are exposed to the reduced inoculum levels that persist in Captan solutions, and selection for resistance to the other fungicides will also be significantly reduced. The value of Captan in postharvest drench solutions might be minimal if drenchers were emptied, cleaned, and refilled with fresh solutions on a daily basis. However, where drench solutions are used for extended periods of time, including Captan in the drench solutions could significantly enhance control of blue mold, especially if bins are still carrying large quantities of blue mold inoculum.

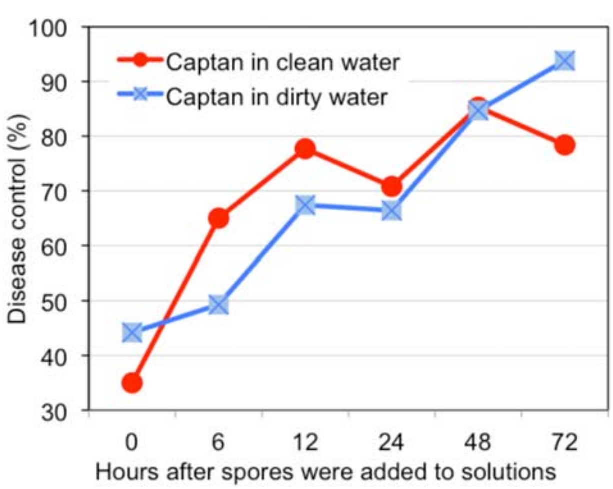

Figure 1. Effectiveness of Captan for controlling *Penicillium expansum* as affected by the delay between preparation of the treatment/inoculum solution and the time that freshly wounded Empire fruit were treated. Percent disease control was calculated by comparing disease incidence of these treatments with that of control fruit exposed to a clean-water spore suspension at 0-time (immediately after solutions were prepared).

Using Biocides and Sanitizers in Storages and Packinghouses

Despite availability of effective new fungicides, packinghouse operators should not ignore the importance of using sanitizers and biocides. A sanitizer should be used for annual cleaning/sanitizing of storage rooms and for periodic cleaning of hard surfaces in apple packinghouses. Biocides should be included in all packing line water dumps and water flumes to prevent accumulation of both *P. expansum* spores and potential human pathogens in these recycling flume waters. Note, however, that biocides should never be added to postharvest drench tanks that contain DPA because DPA is an anti-oxidant and it therefore is not compatible with biocides, most of which are oxidizers.

Numerous kinds of biocides and sanitizers are available for applications in apple packinghouses and storage rooms. However, the most effective and easiest-to-use sanitizer for hard surfaces will involve some kind of quaternary ammonium (quat) compound, and the most economical and easiest-to-use biocide is probably sodium hypochlorite (the active ingredient in chlorinated water). Other biocides that can be used in water flumes include peroxides (e.g., Stor-Ox), ozone, and chlorine dioxide. The latter two are gasses that must be injected into the water systems, and that may require more expensive equipment and safety monitoring that is necessary when using sodium hypochlorite.

Effectiveness of sanitizers and biocides is impacted by the following factors:

1. Product concentration.
2. Temperatures of the solution or surface to be treated.
3. Exposure time.
4. Introduction of organic matter.

The product label usually limits product concentration that can be used in packinghouses. Sanitizers and biocides are more active at higher temperatures than at lower temperatures, but it may be difficult to adjust temperatures for many packinghouse applications. For example, the temperature of packinghouse flume water is largely controlled by the temperature of apples coming out of storage. Heating that water to improve biocide activity may not be economically feasible.

Exposure time is adjustable in some situations and not in others. For example, exposure time of ephemeral sanitizers such as aqueous peroxide solutions can be extended by using a fog generator to continuously supply fresh product over a multi-hour period in closed, empty storage rooms or by applying chlorine dioxide to hard surfaces in foams that serve to prolong the contact time.

The introduction of organic matter is important because biocides react quickly with organic matter, thereby reducing the active concentration of biocide in solution. In apple packing operations, one way to compensate for loss of the biocide due to introduction of organic matter into water flumes is through the use of automated metering pumps that replenish the biocide as soon as oxidizing levels drop below a preset point.

When sodium hypochlorite is added to water flumes, the optimal concentration depends on a variety of factors. Although concentrations of up to 200 ppm of free chlorine are allowed on some product labels, concentrations above 100 ppm increase chances of injuring fruit. The standard recommendation has been to maintain the concentration of free chlorine between 50 and 100 ppm in water flumes where chlorine is added manually so as to ensure that an effective concentration will be maintained even if there is a sudden influx of organic debris that neutralizes some of the hypochlorite. Where automated systems are used to meter in chlorine and to maintain the appropriate pH in water flumes, free chlorine concentrations as low as 15 to 25 ppm are sufficient to kill microbes in the water solutions. When chlorinated water is used in large presort operations, using low concentrations of hypochlorite will minimize salt accumulations in water flumes. If high levels of sodium hypochlorite are maintained via metering pumps, salts can sometimes accumulate to phytotoxic levels in the water flumes on presort lines where water is not changed regularly.

For sanitizing hard surfaces, quaternary ammonium products (quats) are the preferred sanitizers because quats leave a biocidal film on treated surfaces whereas sanitizers such as sodium hypochlorite (chlorinated water) have no activity after they dry. The residual biocidal film that is left after quaternary ammonia sanitizers have been applied prolong the contact time, thereby increasing the control of microorganisms. With quaternary ammonium sanitizers, the labels may allow a higher concentration if hard surfaces receive a clean water rinse following application of the sanitizer. Only lower concentrations are allowed for surfaces that will not be rinsed. For most applications in the apple industry, the lower concentration without a water rinse will be both adequate and easier to use. Follow label directions carefully. Hard water can reduce the activity of quats and a water conditioner may be needed if the formulated product does not have a water conditioner incorporated into the formulation.

Exposure time can be a limiting factor for effectiveness of both quaternary ammonium sanitizers and hypochlorite solutions, especially in situations where solution temperatures drop below 70° F. For example, we conducted two bin sanitation trials with the quaternary ammonium sanitizer Deccosan 315 and found that spraying or drenching wooden and plastic bins with this sanitizer reduced the number of *P. expansum* spores on the bins by roughly 99.9 %. However, in a third trial with the same product, we achieved only a spore load reduction of only 70 to 80 % despite using similar methods. In retrospect, we suspect that the reduced effectiveness in the third trial resulted from preparing the quat solution with well-water (presumably about 55° F.) and then immediately treating bins under conditions where the bins dried rather quickly whereas the first two trials were conducted with quat solutions that had reached ambient summer temperatures before bins were treated.

The same temperature and exposure-time limitations allow bins coming out of chlorinated water dumps on packing lines to retain large numbers of viable *P. expansum* spores. The temperature of flume water on packing lines is usually between 43° and 50°F. because the water is constantly cooled by the introduction of the cold apples coming out of storage. At these temperatures, and assuming that the chlorinated water in the dump tank is adjusted to 100 ppm of free chorine, an exposure time of at least 15 minutes might be required for an effective kill of *P. expansum* spores on bin surfaces. Activity of the chlorinated water on wooden bin surfaces may be further reduced by interaction of the hypochlorite with wood fibers or with other adhering organic matter.

So if using chlorinated water in flumes and bin dumps does not fully sanitize bins, why is it recommended? Chlorinating water flumes is essential for preventing cross-contamination of large volumes of fruit by microbes that are introduced with the fruit from each bin that is emptied. Despite the fact that the solution temperature and exposure time may limit effectiveness of the sanitizer on bins and apples moving through the dump-tanks, microbes that are released into the water will be exposed for longer periods in the recycling water and will ultimately be killed by the hypochlorite. More importantly, bacteria are far more sensitive to hypochlorite than are spores of *P. expansum*, so bacteria introduced into the water flumes will be killed rapidly despite the cold solution temperatures. Using chlorinating flume water on packing lines should be a standard practice for food-safety reasons.

Unfortunately, all of the biocides that can be used in apple dump tanks and water flumes are oxidizers that will cause corrosion (i.e., rust) in any steel surfaces that are contacted by the biocide. Thus, the need for biocides in apple dump tanks due to increasing scrutiny of food safety issues will dictate that older steel tanks will eventually need to be replaced with stainless steel. Nails in wooden bins will also require more frequent replacement when wooden bins are repeated exposed to biocides, or growers will need to convert to plastic bins. Thus, using a biocide such as sodium hypochlorite (bleach) in water flumes will necessitate expensive changes in some older packing operations. Nevertheless, maintaining effective biocide levels in water flumes should be considered an essential practice for food safety reasons.

Acknowledgments

This work was supported in part by the Apple Research and Development Program. Additional funding came from USDA via support for Multistate Research Projects NE-103 and NE-1018 and from agrichemical companies that produce the postharvest fungicides labeled for apples.

David Rosenberger is a research and extension professor of Plant Pathology and superintendent of New York State Agricultural Experiment Station's Hudson Valley Lab. He leads Cornell's program in postharvest diseases of tree fruit crops.