

Control of Postharvest Decay by Fumigation with Acetic Acid or Plant Volatile Compounds

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ABSTRACT

Stored pome, stone fruit and berry crops are subject to postharvest decay if they are not protected against plant pathogens such as *Botrytis cinerea*, *Penicillium expansum*, *Monilinia* spp., or *Rhizopus stolonifer*. Decay in table grapes primarily caused by *B. cinerea* is prevented by frequent fumigations with sulfur dioxide over the storage period. Although there are many advantages to the use of fumigation, it is used infrequently for the control of postharvest decay. Studies on a wide range of materials that can be used as fumigants has identified several that appear to be good candidates for use on berries, pome fruit, and stone fruit to prevent postharvest decay. In this review the focus is on two classes of naturally occurring chemicals used as fumigants, acetic acid and plant volatile compounds. The first that is discussed is acetic acid usually applied as a vapor of glacial acetic acid or occasionally as vinegar. Details are presented on its use for both large and small volumes of produce as well as its use as a sanitizing agent for storage rooms and bins. Results from several published studies with a wide range of crops and under various conditions of temperature and humidity are summarized. These results provide a good picture of the efficacy of AA vapor and its potential to cause phytotoxicity on certain crops. Two compounds identified as plant volatiles, hexanal and 2-*trans*-hexenal, are discussed in detail. In this review the emphasis is placed on their ability to inhibit postharvest pathogens and their use in an overall postharvest strategy in combination with 1-methylcyclopropene (1-MCP).

Keywords: blue mold, brown rot, gray mold, hexanal, 2-*trans*-hexenal

Abbreviations: AA, acetic acid; AIT, ally isothiocyanates; CFU, colony-forming units; GC, gas chromatograph; LDPE, low-density polyethylene; MAP, modified atmosphere packaging; 1-MCP, 1-methylcyclopropene; ITC, isothiocyanates; RH, relative humidity

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INTRODUCTION

Many crops are stored for several months before they are sold and consumed. Low temperature storage of pome fruit, grapes, and carrots is the preferred method of preserving these crops although even under these conditions they are subject to decay by several plant pathogens (Eckert and Ogawa 1988). The storage life of many crops depends on treatment with an antifungal agent. Most postharvest fungicides are applied by either drenching, line sprays or by fumigation. Fumigants have many attributes that make them effective pesticides. Bond (1973) states that fumigants can diffuse through space and penetrate into protected places that are inaccessible to liquid or solid pesticides. Table grapes are fumigated with sulfur dioxide in storage, every 7 days to prevent the spread of *Botrytis cinerea* from infected

berries (Luvisi *et al.* 1992). Initial fumigation requires a higher rate of sulfur dioxide than subsequent treatments in order to control spores on the grape surface (Smilanick and Henson 1992). The use of sulfur dioxide fumigation on grapes and other crops such as litchi is not without problems, the foremost being it leaves undesirable residues (Sivakumar *et al.* 2007). It has no effect on established infections and can produce off-flavors and bleached skin spots on the berries (Narayanasamy 2006). For reasons such as these research has continued on the search for fumigants with antimicrobial properties. Acetaldehyde vapor at 0.5% (v/v) controlled blue mold of apples caused by *Penicillium expansum* when applied for 2 h (Stadelbacher and Prasad 1974). The fungicidal action was shown to be a function of concentration and exposure period. Mattheis and Roberts (1993) tested acetaldehyde, propanol, and butanal on

cherries and found that although these fumigants controlled *P. expansum* conidia from germinating they were phytotoxic to cherries. Another fumigant that looked promising for control of *B. cinerea* on table grapes was hydrogen peroxide (Forney *et al.* 1991). Grapes held in 0.27 mg L⁻¹ hydrogen peroxide for 24 h at 20°C required 10.5 min to kill 99% of the spores (Rij and Forney 1995). Another promising class of fumigants is gaseous allyl isothiocyanates (AIT) that inhibit bacteria and postharvest fungal pathogens at concentrations ranging from 100 to 1000 µg AIT L⁻¹ (Delaquis and Sholberg 1997). Mari *et al.* (2008) showed that 4-methylthiobutyl-ITC was the most effective isothiocyanate that was tested having the lowest ED₉₅ of 0.10 mg L⁻¹ for *Monilinia laxa* conidia and 0.52 mg L⁻¹ for mycelium. In reviewing non-conventional methods for control of postharvest pear diseases Mari *et al.* (2003) divide emerging technologies into the following three components; 1) application of natural antagonistic microorganisms, 2) application of natural antimicrobial substances such as the ITC compounds, and 3) application of sanitizing products. Sanitizing fumigants include products such as chlorine dioxide and ozone used in circulation water or as fumigants for use in storage rooms (Sholberg 2004; Linton *et al.* 2006). Although many of these products are promising and need further research this review will focus only on acetic acid (AA) including vinegar, and the plant volatiles, hexanal and 2-*trans*-hexenal used as fumigants for direct control of plant pathogens on produce and/or as sanitizers for the postharvest environment. Acetic acid vapor was previously reviewed by Sholberg *et al.* (1998) but plant volatile compounds do not appear to have been reviewed for control of postharvest decay. Since the first review on AA vapor many new research articles have been published on it and a better understanding of the conditions for its use have emerged.

FUMIGATION WITH ACETIC ACID

Characteristics of acetic acid

Acetic acid is considered to be a “generally recognized as safe” (GRAS) compound and is comparable to other GRAS compounds such as hydrogen peroxide, bicarbonate and carbonate salts, chlorine and sugar analogs because they leave low or non-detectable residues, degrade rapidly, and metabolize quickly in plant tissue (Barkai-Golan 2001). Acetic acid has been used for many years in the food industry to inhibit microbial growth and as an acidulant (Doores 1990, 1993). The mode of action of an acid is related to the undissociated portion of the molecule and is more important than any change in pH brought about by the addition of acid. Therefore AA is more potent as a fumigant because it exists as mixtures of undissociated monomers and dimers (Seaton 1993). Dissociated forms of weak acids are not absorbed by microorganisms to any great extent (Doores 1990). Research shows that short chain organic acids such as AA affect the cell membrane by interfering with the transport of metabolites and maintenance of membrane potential (Freese *et al.* 1973; Davidson and Juneja 1990). The inhibitory effect is due to the conduction of protons through membranes, effectively destroying the proton motive force which is needed for substrate transport (Freese and Levin 1978). Killing results from holes in the cell membrane. The concentration of AA vapor can be monitored in at least three different ways in storage rooms (Sholberg *et al.* 2003a). Commercial gas detector tubes (Matheson Safety Products, East Rutherford, NJ) are sensitive to the presence of very low quantities of AA vapor but can only be used once. These tubes are practical for determining if a storage room is safe to enter after fumigation but are not practical for monitoring AA during fumigation. Solid-state sensors (International Sensor Technology, Irvine, CA) were extensively tested and found to be accurate but prone to failure when needed most to measure AA concentration at a remote site (Sholberg *et al.* 2003a). Usually this required recalibrating the sensor. The advantages of these types of sensors are that they give a continuous

readout of AA concentration and can be interfaced with a computer for control of the fumigation process. The third method used for measuring AA vapor concentration is a gas chromatograph (GC) outfitted with a flame ionization detector and fused silica capillary column. Results with the GC are reliable and generally accurate.

Pome fruit fumigation

The first report on the use of AA vapor to control postharvest decay on pome fruit was published in 1995 so it is a relatively new technology (Sholberg and Gaunce 1995). Decay of ‘Golden Delicious’, ‘Red Delicious’, and ‘Spartan’ apples caused by *B. cinerea* or *P. expansum* could be prevented by fumigation of the apples with glacial AA vapor produced by wetting filter paper with the acid and allowing it to evaporate in an air tight container with some air circulation (Fig. 1). It was possible to fumigate the apples to kill plant pathogenic spores on the apples without damaging the fruit. In subsequent studies involving larger volumes of fruit this was not always the case and is a major factor limiting the use of AA commercially. Sholberg and Gaunce (1995) showed that relative humidity (RH) needed to be high for AA vapor to kill spores of *B. cinerea* or *P. expansum* on ‘Spartan’ apples. For example at a rate of 2.0 mg L⁻¹ AA vapor and 17% RH the area decayed by *B. cinerea* was 46.0 mm in diameter while at 98% RH the decayed area was zero. It was also shown that the higher the concentration of spores contaminating the apple, the higher the concentration of AA that was needed. Interestingly any of the three major short-chain organic acids, formic (mol wt 46), acetic (mol wt 60), or propionic (mol wt 74), could be used to reduce postharvest decay (Sholberg 1998b). Decay of pome fruit caused by *P. expansum* was reduced from 98% in the control to 16, 4 or 8% by AA, formic, and propionic acid vapor, respectively. Generally the same number of micromoles of each acid is effective so less formic acid is required than AA or propionic to control decay. However, formic acid is much more phytotoxic than either AA or propionic. Vinegar is a dilute form of AA and can be vaporized to reduce postharvest decay in harvested crops (Sholberg *et al.* 2000). Vapors of several common vinegars containing 4.2 to 6.0% AA prevented blue mold of apples contaminated with spores of *P. expansum*. The amount of vinegar needed to fumigate fruit is substantial when compared to the amount of glacial AA needed. For the use of vinegar to be commercially viable as a postharvest treatment, a more potent form of vinegar is needed of at least 10% or higher AA concentration. Studies on AA fumigation of large quantities of apples have been conducted to control postharvest decay and the results have been mixed (Sholberg 1998a; Sholberg *et al.* 2001). Generally the results were acceptable indicating that AA fumigation of apples in bins could be



Fig. 1 Acetic acid fumigation of apples to control blue mold. The apples were inoculated with *P. expansum* and the bottom two apples were fumigated with AA vapor.

accomplished to reduce blue mold decay without affecting fruit aroma. In these trials the fruit was fumigated at 10°C and fumigated from one to eight times to determine the margin of safety for the use of AA commercially. Lenticel damage increased tremendously after four to eight fumigations on 'McIntosh' apples although the treatment had no effect on the scald apple disorder. Unfortunately, it has become apparent that lenticel burning varied uncontrollably with concentration of AA, quantity of fruit, container type and apple cultivar so the treatment is not practical with large volumes of fruit in its present form. Thus the direction of AA fumigation in the pome fruit industry should be on its use as a sanitation method for storage rooms and bins. A similar conclusion was reached with the AA fumigation of large quantities of pears (Sholberg *et al.* 2004). For control of gray mold in d'Anjou pears a concentration of around 200 $\mu\text{L L}^{-1}$ of AA was needed to control decay at 2°C but lenticel blackening occurred on some pears at 300 $\mu\text{L L}^{-1}$ of AA leaving very little room for error. Acetic acid fumigation is much more manageable at warmer temperatures so under different conditions of temperature with accurate monitoring equipment it might be possible to fumigate large quantities of pome fruit safely and this can only be determined by further research.

Stone fruit fumigation

Roberts and Dunegan (1932) were the first researchers to document the use of AA vapor for control of peach brown rot. They found that glacial AA prevented spores of *Monilinia fructicola* from germinating but also blackened the fruit after a few minutes exposure to the acid. Sholberg and Gaunce (1996) revisited the use of AA vapor for control of brown rot and *Rhizopus* rot on peaches, nectarines, apricots and cherries. This time the results were more successful with examples of decay control on all four crops. Decay by *Monilinia fructicola* and *Rhizopus stolonifer* on 'Harbrite' peaches was prevented by as little as 1.4 or 2.7 mg L^{-1} AA, respectively and 2.0 mg L^{-1} AA controlled *M. fructicola* on 'Tilton' apricots (Fig. 2). However, AA concentrations of 2.7 mg L^{-1} and higher caused injury to both peaches and nectarines that appeared as brown streaks on peaches and reddish brown streaks on nectarines. Peaches treated before harvest with either captan or iprodione had less decay than fumigation with acetic acid alone. Probably because the fungicides applied before harvest prevented the development of latent or quiescent infections that were not affected by AA vapor (Jenkins and Reinganum 1965). Additional research on fumigants for the control of brown rot on apricots and plums was carried out by Liu *et al.* (2002). In these trials the authors compared AA to thymol for efficacy against *M. fructicola*. Both materials controlled brown rot of apricots and plums but thymol was more effective although thymol caused greater phytotoxicity as indicated by



Fig. 2 Acetic acid fumigation of apricots to control brown rot. The apricots were inoculated before treatment with conidia of *M. fructicola*, treated with acetic acid and incubated at 20°C for approximately 1 week. The apricots on the left were left untreated and those on the right were fumigated with AA vapor.

surface browning. The use of AA vapor to prevent decay of sweet cherries was examined by Sholberg (1998b) on eight sweet cherry cultivars including an unnamed white cherry cultivar. Likewise Chu *et al.* (1999) fumigated 'Hedelfingen' cherries with AA vapor inoculated with *B. cinerea* and placed the cherries in MAP bags made of 35- μm -thick, low density polyethylene after fumigation. The AA vapor controlled *M. fructicola*, *R. stolonifer*, and *P. expansum* in the Sholberg (1998b) study although propionic acid was less damaging to the fruit than AA or formic acid. In the Chu *et al.* (1999) study AA vapor reduced gray mold in cherries stored for 10 weeks in MAP although as noted previously for apricots and plums, thymol was more effective than AA vapor. However, thymol caused more stem browning and imparted a medicinal odor to the cherries.

Berry and miscellaneous crop fumigation

Storage life is an important consideration in the marketing of table grapes and strawberries. Moyls *et al.* (1996) conducted a study to determine if MAP using 38- μm -thick, low density polyethylene and fumigation with AA would increase the shelf-life of these crops. The combined process of AA fumigation followed by storage at 0°C for table grapes and 5°C for strawberries was very effective reducing decay to very low values in both crops (Fig. 3). Grapes were protected from spoilage for up to 2 months and strawberries for up to 2 weeks. Strawberries have also been fumigated with vinegar without the use of MAP to prevent infection by *B. cinerea* (Sholberg *et al.* 2000). White vinegar containing 5% (v/v) AA reduced decay by 50% when compared to the inoculated control. Acetic acid fumigation of table grapes in relatively large quantities showed that there was no significant difference between AA fumigation and sulfur dioxide fumigation for the control of *B. cinerea* (Sholberg *et al.* 1996). The use of AA fumigation has not been adopted commercially for use on table grapes even though it provides some useful benefits such as elimination of sulfur dioxide residues. Some other crops fumigated successfully with AA are citrus fruit to control *P. italicum*, and *P. digitatum* (Sholberg and Gaunce 1995; Sholberg 1998b); and kiwi and tomato fruit to control *B. cinerea* (Sholberg and Gaunce 1995). Acetic acid fumigation has also been used in food processing to reduce microflora on coleslaw and increase its shelf-life (Delaquis *et al.* 1997).

Sanitation with acetic acid vapor

Sanitation refers to the reduction of initial inoculum and hence, initial disease intensity (Madden *et al.* 2007). Reducing the inoculum will reduce the probability of new infec-



Fig. 3 Acetic acid fumigation of grapes to prevent bunch rot. The grapes were inoculated with conidia of *B. cinerea*, and the bottom grape bunch was fumigated with 8.0 mg L^{-1} AA vapor. Both untreated and treated grape bunches were placed in MAP bags at 0°C for 74 days when they were photographed.

Table 1 Some fumigants used to prevent decay in harvested fruit crops compiled from various sources (Barkai-Golan 2001; Mari *et al.* 2003; Linton *et al.* 2006).

| Fumigant | Application method | Dosage | Crop and pathogen |
|---------------------|------------------------------------------------------------------|---------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------|
| Acetaldehyde | Liquid acetaldehyde was injected into an air tight chamber | 0.5 to 2.0% (v/v) of air for 1 to 3 h | Pome fruit for <i>P. expansum</i> |
| Acetic acid | Glacial AA heated and the vapor blown throughout the enclosure | 2 to 4 mg L ⁻¹ for 2 h or more | Pome, stone fruit, berries, vegetables and seed for <i>B. cinerea</i> , <i>Penicillium</i> spp., <i>Monilinia</i> spp. and others |
| Ally-isothiocyanate | The pure product is allowed to evaporate in a closed container | 5 mg L ⁻¹ for 24 h | Pears and mung beans for <i>B. cinerea</i> and other pathogens |
| Chlorine | Dissolved in water | 100 ppm for 1 to 10 min | Pome and stone fruit for <i>B. cinerea</i> , <i>Penicillium</i> spp., <i>Monilinia</i> spp. and others |
| Chlorine dioxide | Generated at the site | 10 to 50 ppm for 1 min | Pome and stone fruit for <i>B. cinerea</i> , <i>Penicillium</i> spp., <i>Monilinia</i> spp. and others |
| Hexanal | Pure hexanal heated and the vapor blown throughout the enclosure | 100 to 450 ppm for 24 to 48 h | Pome and stone fruit for <i>B. cinerea</i> and <i>P. expansum</i> and others |
| 2-trans-Hexenal | The pure product is allowed to evaporate in a closed container | 12.5 µL L ⁻¹ for 24 to 48 h | Pome fruit, berries and grapes for <i>B. cinerea</i> and <i>P. expansum</i> |
| Nitrous oxide | Released as a gas | N ₂ O (80%) and O ₂ (20%) in atmosphere | Pome for <i>Penicillium</i> spp. |
| Ozone | Generated at the site | 1.0 ± 0.05 ppm for 2 weeks | Citrus for <i>Penicillium</i> spp. |
| Sulfur dioxide | Generated at the site | 100 ppm-hour for several minutes to 2 h | Table grapes for <i>B. cinerea</i> |
| Thymol | The pure product is allowed to evaporate in a closed container | 8 to 30 mg L ⁻¹ for 20 to 25 min | Stone fruit for <i>B. cinerea</i> and <i>M. fructicola</i> |

Table 2 Effect of AA fumigation on contamination by *Penicillium* spp. of various materials used in packinghouses.

| Material | <i>Penicillium</i> spp. CFU/cm ² | | Percent reduction in contamination |
|----------------------|---------------------------------------------|----------------------------------|------------------------------------|
| | Before AA fumigation ^z | After AA fumigation ^z | |
| Plywood | 145 ± 383 | 3.9 ± 7.2 | 97 |
| White fire retardant | 3817 ± 4889 | 0.5 ± 1.2 | 100 |
| Gray fire retardant | 12.8 ± 3.5 | 3.3 ± 5.8 | 74 |
| Galvanized steel | 1.0 ± 0.1 | 0.2 ± 0.1 | 80 |
| Overhead pipes | 509000 ± 74800 | 0.0 ± 0.0 | 100 |
| Floor | 237 ± 469 | 1.8 ± 3.5 | 99 |
| Air ^y | 13 ± 16 | 0.5 ± 0.3 | 96 |

^zFive swab samples were taken for each wall material before and after fumigation to determine the mean value of *Penicillium* spp. for each wall material. See Sholberg and Stokes (2006) for the method of determining the microbial count.

^yAir was sampled in 20 L quantities and count is based on the mean of five samples.



Fig. 4 Acetic acid fumigation of storage rooms to remove contamination by postharvest pathogens. Samples 10 cm² in area were swabbed from at least five different areas on storage room walls before and after fumigation with AA. The wash water from the swab samples was plated on acidified potato dextrose agar and the plates were incubated at 20°C for 1 to 2 weeks. The plates on the left were made from swab samples taken before fumigation and those on the right from swab samples taken after fumigation.

tions and growth of lesions. Furthermore, increasing time between infection and inoculum production can lower the disease rate. The vapor of AA is an excellent sanitizer because it is very effective in killing spores of postharvest pathogens such as *B. cinerea* and *P. expansum* and thereby reducing inoculum and slowing disease progress (Sholberg 2004). Several sanitizers have been tested on fruit crops with varying levels of success (Table 1).

A preliminary trial using AA vapor in a large empty storage room (1983 m³) containing six empty wooden apple

bins showed that AA vapor had potential for sanitizing large storage rooms and bins contaminated with postharvest pathogen spores (Sholberg 2004). Prior to treatment the levels of mold contamination were assessed and walls and bins were inoculated with fungal spores. Acetic acid vapor reduced contamination to very low levels on walls and bins (Fig. 4). Many different materials are used in construction of storage rooms such as plywood, galvanized steel, and fire retardant foam and these materials carry different populations of pathogens (Sholberg and Stokes 2006). Plywood and a white foam material generally are contaminated with high numbers of *Penicillium* spp. Fumigation of these material with AA vapor reduced populations on plywood from 145 to 4 colony-forming-units (CFU)/cm² and the white fire retardant from 3817 to 0.5 CFU/cm² (Table 2).

Contamination of picking bins by postharvest pathogens increases the probability of decay in storage because the water used to float fruit out of these bins becomes contaminated and spreads pathogenic spores to healthy fruit (Sanderson 2000). Several trials in packinghouses to determine if AA fumigation of bins reduces mold contamination have been conducted in packinghouse rooms (Sholberg 2004). Generally the results have been good with reduction in contamination levels of *Penicillium* spp. in bins (Table 3). Acetic acid fumigation reduced *Penicillium* spp. propagules on bins to zero in three out of five fumigations conducted in packinghouse rooms. There are many reasons that the treatment does not always work but can usually be traced to problems with maintaining the correct concentration of AA vapor over a period of at least 2 h or too low an ambient temperature or relative humidity.

In addition to these more conventional sanitation practices AA fumigation has been used to sanitize dormant trees and shoots (Sholberg *et al.* 2005a). Most interesting was the ability of AA vapor to destroy all sources of the powdery

Table 3 Effect of AA fumigation on contamination of commercial apple bins by *Penicillium* spp.

| Packinghouse room number | Number of bins per fumigation | <i>Penicillium</i> spp. CFU/cm ² | | Percent Reduction |
|--------------------------|-------------------------------|---------------------------------------------|---------------------|-------------------|
| | | Before AA fumigation | After AA fumigation | |
| 61 | 8 | 29.5 ± 36.7 | 2.3 ± 5.8 | 92 |
| 63 | 3 | 8.0 ± 10.3 | 0.0 ± 0.1 | 100 |
| 85 | 6 | 29.9 ± 44.7 | 8.5 ± 9.8 | 72 |
| 125 | 3 | 6.4 ± 6.8 | 0.0 ± 0.0 | 100 |
| 129 | 6 | 10.9 ± 16.2 | 0.0 ± 0.1 | 100 |

^aFive swab samples were taken from each bin before and after fumigation to determine the mean value of *Penicillium* spp. for each bin. See Sholberg and Stokes (2006) for the method of determining the microbial count.

mildew fungus in overwintering 'Jonagold' apple buds. When 96 'Jonagold' apple shoots known to be infected by *Podosphaera leucotricha* were fumigated with 12 mg L⁻¹ AA vapor for 2 h at 20°C the shoots were rendered free of powdery mildew infection although the nonfumigated shoots from the same trees were all infected.

FUMIGATION WITH PLANT VOLATILES

Origin of plant volatiles

Lipoxygenases commonly found in plant tissues catalyze peroxidation of polyunsaturated fatty acids (linoleic acid or linolenic acid) to various primary and secondary oxidation products (Hildebrand 1989). The two most important products that have been studied for antifungal activity from this group of compounds are hexanal and 2-*trans*-hexenal. Hexanal is derived from linoleic acid (C18:2) and 2-*trans*-hexenal is derived from linolenic acid (C18:3) via the lipoxygenase pathway. These compounds are thought to have an important role in the formation of flavors and aromas of many plant products (Hildebrand *et al.* 1988). In some foods the presence of lipoxygenase flavors and aromas are desirable but in the case of hexanal in soybeans it is considered an off flavor compound and a major goal of plant breeding is to limit its development. However these products have other uses in the plant and could be involved in plant growth and development, senescence or wound responses and pest resistance. The specific use of these plant volatiles for control of postharvest pathogens in several different crops is described below.

Berry and grape fumigation with plant volatiles

Vaughn *et al.* (1993) found that of 15 volatiles released from raspberries and strawberries during ripening, 1-hexanol, *E*-2-hexenal and 2-non-anone inhibited three different fungi including *B. cinerea* at 0.1 µL/mL. They concluded that such natural volatile compounds could function in control of postharvest diseases of berry crops. In succeeding studies hexanal, and related compounds, 1-hexanol, (*E*)-2-hexen-1-ol, (*Z*)-6-nonenal, and (*E*)-3-nonen-2-one exhibited potential as postharvest fumigants for the control of *B. cinerea* at very low levels of concentration (8-48 µL L⁻¹) (Archbold *et al.* 1997). Gardini *et al.* (1997) found that antifungal activity of hexanal depended on its vapor pressure based on studies they conducted on inhibition of *Aspergillus niger* in a model system. Their research also showed that warmer temperatures enhanced the antifungal activity of hexanal by increasing the vapor pressure. Studies by Fallik *et al.* (1998) showed that (*E*)-2-hexenal can stimulate or prevent mycelial growth of *B. cinerea* depending on concentration. For example when ripe strawberries were inoculated with 10⁶ CFU mL⁻¹ of *B. cinerea* and exposed to 10 or 100 µL (*E*)-2-hexenal at 2°C for 7 days before transferring to 22°C for 3 days, only the 100 µL treatment prevented growth of *B. cinerea*. Table grapes fumigated with 100 or 200 µL (*E*)-2-hexenal also had less mold growth (Archbold *et al.* 1999). Differences in metabolism of (*E*)-2-hexenal among strawberry, blackberry and grape samples and the resulting headspace concentrations may partly explain stimulation or inhibition of *B. cinerea* infecting these crops (Archbold *et al.* 2000). The authors speculated that it may be necessary to

use crop-specific volatiles or at least be aware that a volatile compound may not be useful for all crops.

Pome fruit fumigation with plant volatiles

Considerable research has been conducted on the use of hexanal vapor for control of postharvest decay and enhancement of fruit aroma in apples. Hexanal vapor inhibited hyphal growth of *P. expansum* and *B. cinerea* on potato dextrose agar, a medium used to grow postharvest fungi, and on apple slices after 48 h exposure at a rate of 100 ppm (Song *et al.* 1996). Hexanal applied at a concentration of 450 ppm or more killed both fungi after they were exposed to hexanal for 48 h because the fungi did not revive when moved to hexanal-free air. Hexanal was actively converted to aroma volatiles in 'Jonagold' and 'Golden Delicious' apple slices, with hexanal and hexylacetate production strongly enhanced after 20 to 30 h. The possibility of combining hexanal with MAP was also examined for apple slices. Permeability data for low-density polyethylene (LDPE) film indicated that hexanal would escape rapidly from such packaging. In order to overcome this problem more study on films or release of hexanal in MAP packages will be needed. Song *et al.* (1998) discovered that after 16 h apple slices treated with hexanal did not have any residue of hexanal. The absence of residue could be an important factor if attempts were made to register hexanal as a postharvest fungicide. The effect of hexanal on the bacterial and yeast populations of apple slices showed that hexanal totally inhibited mesophilic bacteria, prolonged the lag phase of psychrotrophic bacteria, and inhibited molds and yeasts (Lanciotti *et al.* 1999). Hexanal also prevented browning of the apple slices when used under MAP conditions. Blue mold caused by *P. expansum* is the most important postharvest disease of apples and an important disease of pears (Jones and Aldwinckle 1990). Neri *et al.* (2006a) found that of nine plant volatile compounds they tested for activity against *P. expansum* on 'Conference' pear, *trans*-2-hexenal was the best inhibitor of conidial germination while hexanal was somewhat less effective. When applied 24 or 48 h after inoculation *trans*-2-hexenal was effective but not after 2 h indicating that it probably only inhibited germinating or germinated spores (Neri *et al.* 2006b). An exposure length of 8 h was required to reduce fruit patulin content, a mycotoxin produced by *P. expansum*, 6 hours longer than what was needed to reduce decay. Further studies with *trans*-2-hexenal at a rate of 12.5 µL L⁻¹ on several apple and pear cultivars treated 24 h after inoculation with *P. expansum* showed that blue mold was controlled 50 to 98% depending on the cultivar, and patulin was reduced, while fruit appearance, color, firmness, soluble solids or titratable acidity were not affected (Neri *et al.* 2006c). Similar trials to determine antimicrobial activity and effect on pome fruit quality have been carried out with hexanal. Hexanal reduced spore viability of *P. expansum* in a concentration and time dependent manner *in vitro* on potato dextrose agar and *in vivo* on 'Golden Delicious' apples exposed to hexanal vapor for 48 h (Fan *et al.* 2006). It was suggested that investigations are needed to assess hexanal treatment under commercial apple storage conditions. A strategy was developed by Sholberg and Randall (2007) to use hexanal as part of a postharvest disease management strategy for control of blue and gray mold of pome fruit. The strategy depended on pretreatment



Fig. 5 Hexanal fumigation of peaches to prevent brown rot. The peaches were inoculated with conidia of *M. fructicola*, treated with hexanal and incubated at 20°C for approximately 1 week. The peaches on the left were not treated and those on the right were fumigated with hexanal vapor.

of fruit 2 weeks before harvest with a systemic fungicide known to be effective against *B. cinerea* and *P. expansum* such as cyprodinil or pyrimethanil (Sholberg *et al.* 2003b, 2005b). Immediately after harvest the fruit is fumigated with hexanal in bins (3.0 × 2.5 × 3.6 m) with 2 or 4 mg L⁻¹ for 24 or 18 h, respectively. The use of this strategy on pears produced the highest number of fruit free of mold contamination and on apples decay was reduced from 10 to 1% (Sholberg and Randall 2007). Since 2002 the apple industry in the United States has used 1-MCP to retain fruit firmness in stored fruit and to extend shelf life. It is possible that the combination of hexanal and 1-MCP could be effective in preventing postharvest decay of stored pome fruit. Spotts *et al.* (2007) evaluated the effects of prestorage treatment with 1-MCP, hexanal, and 1-MCP + hexanal on decay of d'Anjou pears in long-term storage. They found that hexanal reduced snow mold rot, an occasional disease on pears stored in the Pacific Northwest United States and Canada, but increased blue mold (Jones and Aldwinckle 1990). It is thought that 1-MCP reduces decay by inhibiting fruit ripeness and possibly by enhancing enzymes that play important roles in disease-defensive systems of plants (Liu *et al.* 2005). A combination of 1-MCP and hexanal at optimized rates could reduce decay, control superficial scald, and allow normal ripening of the fruit.

Stone fruit fumigation with plant volatiles

Although research reports on use of plant volatiles for control of postharvest stone fruit diseases are few in number they show that results similar to berries and pome fruit should be expected. Caccione *et al.* (1995) used hexanal to control brown rot caused by *Monilinia laxa* and *Rhizopus* rot caused by *Rhizopus stolonifer* on peaches, nectarines and plums. Hexanal at 2,500 ppm was as effective as benzaldehyde but produced phytotoxic symptoms when used at higher concentrations. Hexanal was used in Canada on peaches to control brown rot caused by *M. fructicola* and compared to AA vapor (Fig. 5) (Spiers 2001). Hexanal looked promising for decay control on peaches and was much safer to use than AA vapor that damaged the fruit surface but many questions would need to be answered before it could be recommended for commercial use.

CONCLUSIONS

Acetic acid vapor is an excellent product for destroying mold spores on packinghouse walls and fruit bins. It can also be used to prevent postharvest decay caused by important plant pathogens such as *B. cinerea* and *P. expansum*, but is prone to damaging pome and stone fruit. Light skinned apples and pears are especially susceptible to

damage from AA vapor and should only be fumigated in small quantities. On the other hand berry crops and grapes tolerate AA vapor well and it would be feasible to fumigate them in commercial quantities to prevent postharvest decay. Finally, vinegar usually contains 5% AA (v/v) and could be used to fumigate small quantities of fruit where the remaining excessive water vapor would not lead to problems with condensation of water vapor.

Plant volatiles such as hexanal and *trans*-2-hexenal are produced naturally in fruit through the lipoxygenase pathway and have been found to prevent postharvest decay in berries, pome fruit, and stone fruit. Their effectiveness depends on concentration and temperature because growth of *B. cinerea* can be stimulated or inhibited by low or high concentrations, respectively. Several studies with *trans*-2-hexenal on *P. expansum* showed that it could lower patulin levels in 'Conference' pears. Semi-commercial studies with hexanal applied after pretreatment of pome fruit with a preharvest fungicide was shown to be an effective strategy for controlling postharvest decay.

In conclusion plant volatiles are less phytotoxic than AA vapor but require significantly longer periods for fumigation and are not as active as AA vapor against many microorganisms such as *P. expansum*. When choosing AA vapor or a plant volatile fumigant to control decay these attributes should be taken into consideration.

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