Alternative postharvest chemicals for use on avocado

K.R. Everett
The Plant and Food Research Institute for New Zealand Ltd
Private Bag 92169
Auckland 1142
New Zealand
Email: Kerry.Everett@plantandfood.co.nz

Introduction

There are two types of postharvest disease that affect avocado fruit, those that infect through the sides of the fruit (body rots) and those that infect through the stem-end wound (stem-end rots). Many different fungi can cause these diseases, but world-wide the most common body rot is anthracnose, caused by *Colletotrichum gloeosporioides*, and the most common stem-end rots are caused by *Phomopsis* spp. or other members of the Botryosphaeriaceae. These diseases are best controlled in the field, but postharvest application of fungicides can improve disease control.

Two postharvest chemicals have been used to control avocado rots for the last c. 40 years: prochloraz (Hartill et al., 1986, Muirhead et al., 1982, Rowell, 1983) and thiabendazole (Darvas & Kotze, 1981, Zauberman et al., 1975, Darvas, 1978). Prochloraz is applied by either dipping or spraying fruit in-line (Muirhead et al., 1982, Everett & Korsten, 1996), and thiabendazole is typically applied in wax used to coat the fruit (Darvas & Kotze, 1981, Darvas et al., 1990). There are maximum residue levels (MRLs) for avocados for both these fungicides, but not in all importing countries.

In New Zealand it has been shown that it is important to apply prochloraz as soon as possible after harvesting for best control (Table 1). The effect of delaying prochloraz application was also shown in a study simulating breaks in the cool storage chain in New Zealand, where there was a significant increase in stem-end rots if prochloraz was applied after fruit had been coolstored for 2 days, compared with application within 3 hours of harvest (Everett & Korsten, 1998).

Table 1: Effect of increasing time after harvesting before prochloraz application on isolation of fruit rot pathogens from avocado fruit.

	Hours from picking fruit to prochloraz treatment					
	not dipped	**0**	**2**	**4**	**12**	**24**
no. isolations/ 100 fruit	73	28	27	35	34	40

From Everett (2002).

Alternative chemicals

Synthetic fungicides

A number of other chemicals applied postharvest have been tested to control avocado rots, including azoxystrobin, benomyl, benzalkonium, bitertanol, carbendazim, flusilazol, guazatine, imazalil, kresoxim-methyl, phosphonate and propiconazole (Everett, 2002, Muirhead et al., 1982, Fischer et al., 2011). Some chemicals were as effective as prochloraz, such as boscalid/pyraclostrobin (Everett et al., 2007). These chemicals are not used because prochloraz is usually more effective, and also because of the cost of registration.

Sanitisers, elicitors, CO_2 and plant extracts

Some markets do not allow postharvest applications of prochloraz, and waxing is no longer used by all industries. This has stimulated a search for alternative control options, especially alternatives that are generally regarded as safe (GRAS) and do not require registration. A variety of products including elicitors such as methyl jasmonate, plant extracts such as neem oil, food grade antioxidants (butylated hydroxyl anisole and dibutylhydroxytoluene (BHT)), permanganate, quaternary ammonium compounds such as didecyldimethylammonium chloride, sanitisers such as chloride, chlorine dioxide, ethanol (10% and 76%), hypochlorite (100 ppm, 100 ppm pH 7, 150 ppm pH 8.5, 200 ppm pH 7), and CO_2 shock (30% for 24 hours, or for 3 days) were tested but did not control rots as effectively as prochloraz when applied postharvest (Everett, 2002, White et al., 1999).

Food-grade antioxidants (BHT and butylated hydroxyl anisole) were reported to reduce stem-end rots effectively for 'Ettinger' and 'Fuerte' fruit in Israel (Prusky, 1988), but had no effect on rots on 'Hass' avocado fruit in New Zealand (Everett, 2002). This highlights the need to test postharvest compounds reported to control rots on other cultivars of avocado in other countries before adopting new protocols.

Biological control agents

Several biocontrol agents have been developed and tested postharvest in South Africa, Australia and New Zealand, but these agents have not been fully commercialized, in part because of inconsistent efficacy (Coates et al., 1996, Everett et al., 2007, Havenga et al., 1999, van Eeden & Korsten, 2004). Other biocontrol agents have been reported for control of postharvest rots in avocados from Sri Lanka (Adikaram & Karunaratne, 1998, Madhupani & Adikaram, 2017), Colombia (Ramirez et al., 2015), Mexico and Chile (Campos-Martinez et al., 2016, Guardado-Valdivia et al., 2018), and China (Bi et al., 2019), and in Israel, non-pathogenic strains of *C. gloeosporioides* and *C. magna* have been used for biocontrol (Yakoby et al., 2001, Yakoby et al., 2002, Prusky et al., 1994). To date, none of these biocontrol products have been successfully commercialized. In general, biocontrol has not been able to replace synthetic fungicides (Carmona-Hernandez et al., 2019) because of inconsistent efficacy (Droby et al., 2009, Droby et al., 2016, Wisniewski et al., 2010).

Manipulation of ripening

There is a strong relationship between degree of ripeness and stem-end rot expression for avocado fruit rots in 'Hass' fruit in New Zealand (Figure 1). There is a linear relationship between fruit firmness and stem-end rot incidence (Everett & Pak, 2002), which means the more ripe (less firm) avocados are, the more rots there are. This has also been demonstrated for avocado rots in California (Smilanick et al., 2002, Arpaia et al., 2015).

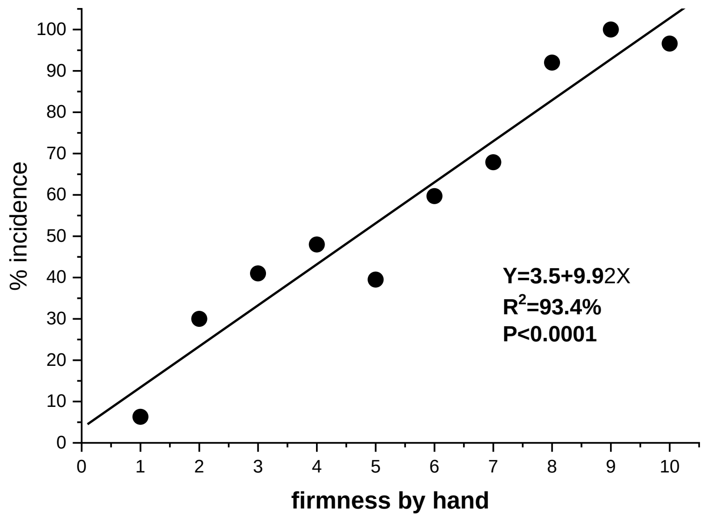

Figure 1. Incidence of stem-end rots in uninoculated 'Hass' avocado fruit compared with fruit firmness (ripeness).

Ripening does not proceed at the same rate for every fruit after harvest, but follows a normal distribution (Figure 2). If fruit are assessed at a particular time point after harvest (or removal from a coolstore), every individual fruit will be at a different stage of the ripening process. If a treatment retards ripening, the stem-end rot incidence would reduce by 10% per firmness point, based on the relationship between fruit rot and fruit firmness (Figure 1). Therefore it is important that all fruit are assessed at optimal ripeness, or a score of five for fruit firmness according to the International Avocado Manual (White et al., 2009), to ensure results are not confounded by test compounds that delay ripening.

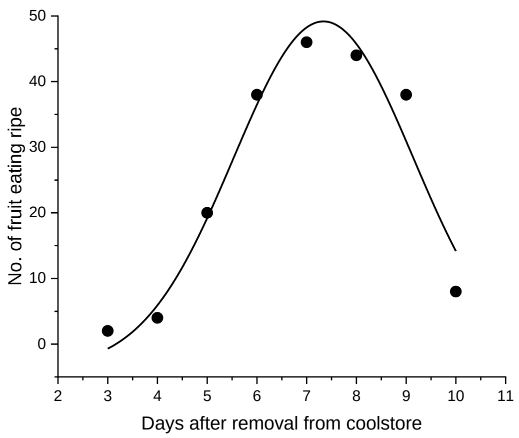

Figure 2. Typical 'Hass' avocado ripening curve after 28 days of coolstorage at 5.5°C in New Zealand.

It has been convincingly shown that increasing the rate of ripening by post-storage application of ethylene reduces the incidence of both stem-end rots and anthracnose (Sarananda et al., 2004, Fitzell & Muirhead, 1983). Presumably the time available for the pathogens to penetrate into the fruit to cause a visible rot is reduced when the fruit ripens more rapidly, therefore the incidence of rots also reduces (Arpaia et al., 2015, Smilanick et al., 2002). Ethylene pre-ripening is used commercially in California and other international markets (CBI, 2017, Murphy & Khilstadius, 1991). Arpaia et al. (2018) in California demonstrated that applying ethylene before cold storage resulted in more rots, probably because of acceleration of ripening during coolstorage resulting in increased susceptibility to decay.

Postharvest application of an elicitor (*Acremonium strictum* Elicitor Subtilisin; AsES) was reported to stimulate ripening, and reduced the growth of pathogens (Perato et al., 2018). The fruit were evaluated 12 days after treatment. Application of AsES accelerated ripening by increasing ethylene production, and also reduced the lesion size of natural infections observed on treated 'Torres' fruit. The mechanism was possibly similar to that of applying endogenous ethylene to accelerate ripening, which also reduces rots. However, testing AsES on naturally infected fruit that are evaluated at optimal eating ripeness is required to validate these results.

Essential oils

There are favourable reports in the literature regarding the *in vitro* efficacy of essential oils for controlling avocado rots (Combrinck et al., 2011). However, the assessments made on fruit were conducted only at a single time point, with improvement in both quality and firmness reported (Obianom & Sivakumar, 2018, Sarkhosh et al., 2017, Bill et al., 2016, Regnier et al., 2010, Sellamuthu et al., 2013). Essential oils retarded ripening, and any effects observed may have been due to the negative relationship between firmness and fruit rots (firmer fruit equals fewer rots) when fruit were evaluated at the same time point after treatment.

1-Methylcyclopropene (1-MCP)

Another chemical that has been shown to reduce rots in some countries, but to exacerbate rots in others, is 1-MCP (Daulagala & Daundasekera, 2015, Berry et al., 2015, Dogan et al., 2017, Adkins et al., 2005). Adkins et al. (2005) showed days to ripen were extended by increasing the concentration of 1-MCP, but correlated with this was an increase in stem-end rots. Stem-end rots have been shown to grow through the stem-end and into the fruit flesh during coolstorage (Everett & Pak, 2002) (Figure 3); therefore, delaying ripening allows more time for the stem-end rots to grow through the cut stem into the fruit. However, if orchards with a very low incidence of stem-end rots are selected for 1-MCP treatment, then better results would be expected.

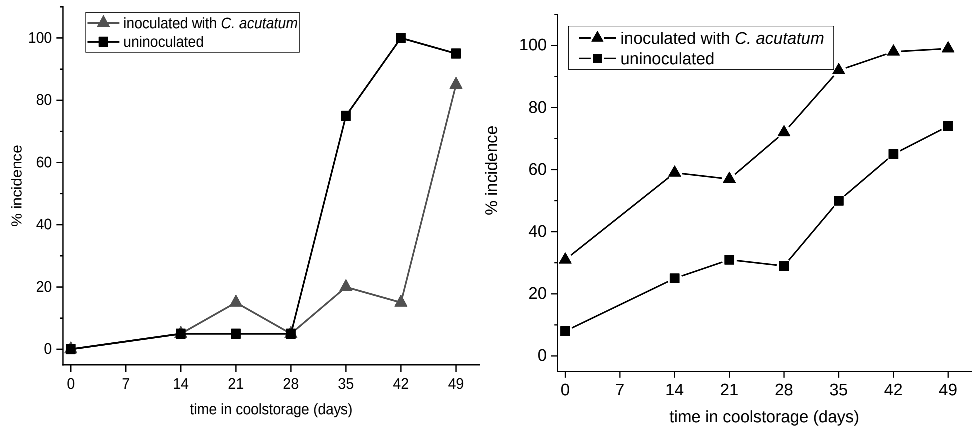

Figure 3. 'Hass' avocado fruit were placed in the coolstore at 5.5°C and 20-fruit samples removed after 0, 14, 21, 28, 35, 42 and 49 days. Fruit were cut immediately and assessed for stem-end rots a) or ripened at 20°C then assessed for stem-end rots b). Data from Everett and Pak (2002); graph b) was redrawn.

<u>Postharvest secateur sterilisation</u>

A very simple measure that has been shown to reduce stem-end rot infections at harvest is to sterilize secateurs between every cut when harvesting (Hartill and Everett 2002) (Figure 4).

Theoretically this could be achieved by wiping clippers on a cloth soaked with 70% ethanol or methanol, or other sanitizers, between every cut during harvest, but this has not been tested.

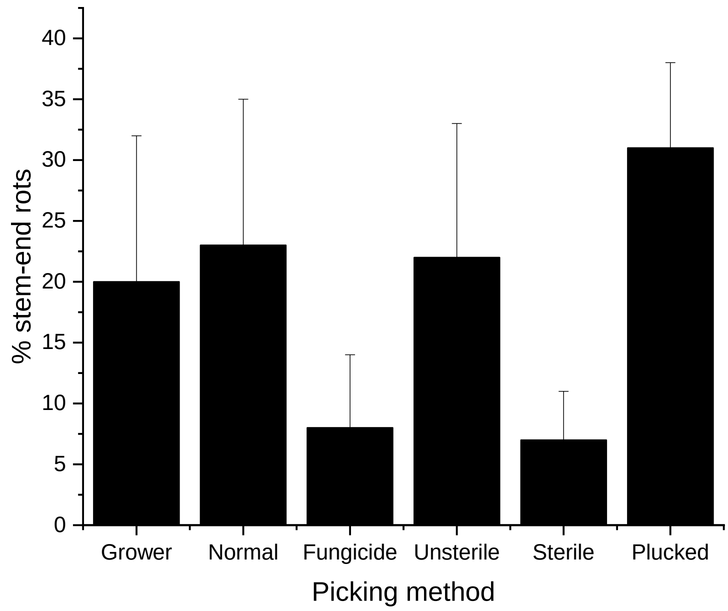

Figure 4. Comparison of different picking methods on incidence of avocado stem-end rots. Grower=using unsterilized clippers and picking into a bucket; Normal = using unsterilized clippers and a picking bag; Fungicide = dipping clippers in a fungicide (500 ppm benomyl + 500 ppm prochloraz) after sterilizing by flaming in 95% ethanol; Unsterile = using unsterile clippers but wrapping fruit in tissue then placing in a tray separated by cardboard partitions; Sterile = clippers sterilized by flaming in ethanol as before and fruit packed individually; Plucked= fruit removed from stem at harvest. Redrawn from Hartill and Everett (2002).

<u>Chitosan</u>

Other reports of promising compounds for rot control in the literature include chitosan (Chavez-Magdaleno et al., 2018, Xoca-Orozco et al., 2017). However, fruit firmness was also reported to be improved following chitosan application, and it was not clear if fruit were assessed at optimal eating ripeness (Chavez-Magdaleno et al., 2018, Correa-Pacheco et al., 2017, Marques et al., 2016, Xoca-Orozco et al., 2018). Because of a possible interaction between inhibition of ripening (firmness) and the development of rots, independent testing of chitosan on naturally infected fruit evaluated at optimal eating ripeness is recommended before commercial use.

<u>Carboxyl methylcellulose</u>

Carboxyl methylcellulose was shown to inhibit rots when incorporated with moringa extracts as a coating for avocado fruit, but also to increase shelf life (Tesfay et al., 2018). Once again, because of a possible interaction between ripening rate and rots, this should also be tested on naturally infected fruit assessed at eating ripeness.

Others

Application of a surface-inhabiting fungus, *Pestalotiopsis neglecta*, as a spore suspension was reported to delay anthracnose and stem-end rot expression (Adikaram & Karunaratne, 1998). Application of the anti-transpirant, Vapor Gard® (96% di-1-p-Menthene), was reported to reduce rots, but also delayed ripening, once more indicating a possible interaction between the effect on degree of ripeness and development of rots (Awad, 1992). In this report, gamma irradiation with 0.04 or 0.08 kGy increased rots (Awad, 1992). Another report of controlling anthracnose, but also delaying ripening, was for a beeswax coating (Bustan & Lahav, 2012).

Conclusions

There is good evidence that postharvest application of prochloraz or thiabendazole can reduce postharvest rots. No alternative chemicals for postharvest treatment evaluated so far show better control than prochloraz. A strobilurin fungicide, boscalid/pyraclostrobin, showed similar efficacy, and depending on cost of registration and availability of MRLs in importing countries, may be able to replace prochloraz in the future. Sterilizing secateurs between every cut during harvest, ensuring fruit are not subjected to temperatures above 25°C for 24 hours or longer during and after harvest, followed by postharvest ethylene ripening shows the most promise for maintaining good fruit quality. The efficacy of compounds such as essential oils and chitosan needs to be proven on naturally infected fruit assessed at the same degree of ripeness before commercial use. For reasons that are not clear, some treatments control rots only in the country in which they are tested, thus it is important to test compounds in the country and on the avocado cultivar on which they will be used, rather than rely on results produced on different cultivars in another country. 1-MCP has been shown to improve postharvest quality in the absence of fruit rots; therefore, this compound needs to be tested thoroughly on fruit from several different orchards in the country in which it will be used before commercial use is adopted.

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