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Tree vigour influences disease susceptibility of 'Hass' avocado fruits

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Abstract. Differences in tree vigour as a result of feeder root destruction by *Phytophthora cinnamomi* were found to significantly ($P < 0.05$) influence disease susceptibility, fruit ripening rates and fruit size of 'Hass' avocado. Fruits from root rot-affected, non-vigorous trees had less anthracnose and took longer to ripen. However, they were probably unmarketable due to their small fruit size compared with fruits from healthy vigorous trees. Fruits from non-vigorous trees also had less pepper spot than vigorous trees, but only on the inside of the tree canopy. The increased disease resistance of fruits from non-vigorous trees was related to a 40% increase in the concentration of Ca in the fruit flesh.

Additional keywords: Colletotrichum gloeosporioides, Phytophthora, pepper spot, nutrition.

Introduction

Anthracnose, caused by the fungus *Colletotrichum gloeosporioides*, is the most serious postharvest disease of avocado (*Persea americana*). Stem-end rot, another important postharvest disease of avocado, can be caused by a number of different fungi including various *Dothiorella* spp., *Lasiodiplodia theobromae* and *Phomopsis perseae*. *C. gloeosporioides* can also cause rots at the stem-end. In this paper, the effect of tree vigour on fruit susceptibility to postharvest diseases and pepper spot in 'Hass' avocado was investigated. Pepper spot is a newly described preharvest disease that is also caused by *C. gloeosporioides* (Willingham *et al*. 2000).

Differences in avocado tree vigour most commonly arise from differences in Phytophthora root rot infection levels caused by the fungus *Phytophthora cinnamomi*. Other studies on avocado in South Africa have shown that tree vigour can have a significant impact on fruit mineral accumulation (Cutting and Bower 1990; Witney *et al*. 1990). Fruits from less vigorous trees were shown to have a higher concentration of Ca, which is important for sound fruit quality (Chaplin and Scott 1980; Wills and Tirmazi 1982) and disease resistance (Conway *et al*. 1992). In this paper, we investigated the effects of tree vigour on mineral accumulation and disease susceptibility.

Methods

This study was conducted over two cropping seasons in a commercial orchard of avocado, cv. Hass, at Mt Tamborine in south-east Queensland. Eight or nine apparently healthy (vigorous) and eight or ten unhealthy (non-vigorous) trees in the same orchard block were selected for this study during the 1996/97 and 1997/98 seasons, respectively. The trees classified as non-vigorous had an average rating of 7 and the trees classified as vigorous had an average rating of 1 on the 0 (vigorous and healthy) to 10 (dead) scale used in Phytophthora research (Darvas *et al*. 1984).

Assessments of tree flowering, vegetative flushing, crop load and canopy leaf cover were made in November 1997, after fruit set and the spring vegetative flush had occurred. A rating scale of 1–5 was used for all criteria, where 1 = none, 2 = light, 3 = moderate, 4 = heavy and 5 = very heavy.

Prior to harvest in August 1998 (season 2), the incidence of pepper spot (percent fruit affected) was assessed in a sample of 12 fruits per tree. The aspect (north, south, east or west) and the position (inside or outside) of the fruit within the tree canopy were recorded.

In October 1997 (season 1), 20 mature fruits were harvested from each single tree replicate of each treatment (i.e. vigorous and non-vigorous). The sample size was increased to 40 fruits in August 1998 (season 2) so that the effect of fruit aspect on disease could be determined. Twenty fruits from the north side and 20 fruits from the south side of the tree were harvested in this second season. Following harvest, fruits were ripened at 22°C and 65% RH, and assessed for postharvest diseases. Fruits were assessed daily for firmness and judged ripe when soft enough to yield to gentle hand pressure. The number of days each fruit took to ripen was recorded as a measure of fruit shelf life. Ripe fruits were sliced longitudinally into quarters, peeled and examined for anthracnose and stem-end lesions. Anthracnose and stem-end lesion severity were estimated as the percentage of fruit surface area affected by either disease. The incidence of anthracnose and stem-end lesions was calculated as the percentage of fruits affected by either disease. The percent acceptable fruit was calculated as the percentage of fruits with 5% or less anthracnose severity and no stem-end lesions. The cause of lesions at the stem-end was determined by isolation onto potato-dextrose agar amended with 0.5%

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Table 1. Effect of 'Hass' avocado tree vigour on tree flowering, vegetative flushing, crop load and canopy leaf cover (1997/98 season)

Treatment	Flowering (1–5)[A]	Flushing (1–5)	Crop load (1–5)	Leaf cover (1–5)
Vigorous	4.1[B] a	3.3 a	2.1 a	4.2 a
Non-vigorous	3.7 a	2.2 b	1.8 a	2.5 b
SE	0.25	0.27	0.37	0.15

[A]A rating scale of 1–5 was used where 1 = none, 2 = light, 3 = moderate, 4 = heavy and 5 = very heavy.
[B]Within columns mean values followed by the same letter are not significantly different at $P = 0.05$.

streptomycin (SPDA). Cultures were incubated at 25°C for ~2 weeks and identified.

To measure mineral concentrations, six fruits from each of eight replicate trees were collected from the outer canopy around the tree at a uniform height (~1.5 m). Sampling was from the vigorous and non-vigorous treatment trees at harvest in August 1998. Fruit flesh was oven dried at 60°C to a constant weight, ground to a fine powder using an electric coffee grinder (Philips, Groningen, The Netherlands) and digested in 5:1 (v/v) nitric-perchloric acid (Baker and Smith 1974). Total mineral nutrient concentrations were then measured by inductively coupled plasma atomic emission spectrophotometry (ICPAES).

Statistical analyses were conducted using Genstat 5 release 4.1 data analysis software (Lawes Agricultural Trust, Rothamsted Experimental Station) for a completely randomised design analysis of variance (season 1) or a split-plot design with tree replicates as whole plots split for fruit aspect (season 2). For the pepper spot data, a split-split-plot design analysis with tree replicates as whole plots split for fruit aspect and fruit position was conducted. Arcsine angular or square root transformations were made on percentage or rating data, respectively. However, examination of residual plots indicated transformation did not show improved distribution of residuals. Hence, untransformed data are presented. Pair-wise testing between means was done using the least significant difference (lsd) procedure at $P = 0.05$.

Results

Non-vigorous trees assessed in late spring 1997 produced significantly ($P < 0.05$) fewer vegetative flushing terminals and had a significantly ($P < 0.05$) less dense canopy (i.e. lower leaf cover rating) than vigorous trees (Table 1). There were, however, no significant ($P < 0.05$) differences in flowering intensity or crop load between the two treatments (Table 1).

In 1997, fruits from non-vigorous trees took significantly ($P < 0.05$) longer to ripen, had a significantly ($P < 0.05$) lower incidence of anthracnose and a higher percentage of acceptable fruit than fruits from vigorous trees (Table 2). In 1998, the same trend was apparent but the differences were not significant ($P < 0.05$). There was, however, a significant ($P < 0.05$) interaction between treatments and fruit aspect in 1998. Larger differences in anthracnose incidence and fruit acceptability (Table 3) were evident between treatments on the north side than the south side of the tree. Fruits from non-vigorous trees had 25% lower incidence of anthracnose and 20% more acceptable fruit (Table 3) than those from vigorous trees on the north side. However, similar disease incidence for anthracnose and acceptability were recorded on the south side. Although fruits from non-vigorous trees had less anthracnose and took longer to ripen, they probably could not be considered marketable due to their small fruit size (Table 2).

Table 3. Effect of tree vigour on the incidence of anthracnose and acceptable fruit in 'Hass' avocado fruits harvested from the north side and south side of the tree (1997/98 season)

Treatment	Aspect North	Aspect South
Anthracnose incidence (%)[A]		
Vigorous	42.5[B] a	40.9 a
Non-vigorous	16.6 b	35.5 a
Acceptable fruit (%)[C]		
Vigorous	67.5 b	77.6 ab
Non-vigorous	87.8 a	77.2 ab

[A]Aspect standard error for comparing within a treatment is 3.99, for all other comparisons the standard error is 7.54.
[B]Mean values followed by the same letter are not significantly different at $P = 0.05$.
[C]Aspect standard error for comparing within a treatment is 4.75, for all other comparisons the standard error is 5.94.

Table 2. The effect of tree vigour on fruit diameter, shelf life, severity and incidence of anthracnose and stem-end lesions of 'Hass' avocado fruits ripened at 22°C and 65% RH

Treatment	Fruit diameter (mm)	Shelf life (days)	Anthracnose Severity (%)	Anthracnose Incidence (%)	Stem-end lesions Severity (%)	Stem-end lesions Incidence (%)	Acceptable fruit[A] (%)
Harvested October 1997							
Vigorous	–	12.1[B] (± 0.47)b	33.3 (± 5.0)a	95.2 (± 1.8)a	10.5 (± 1.5)a	72.2 (± 5.5)a	7.6 (± 4.0)b
Non-vigorous	–	13.7 (± 0.50)a	30.5 (± 4.8)a	88.5 (± 1.7)b	13.6 (± 1.5)a	61.5 (± 5.2)a	21.5 (± 4.0)a
Harvested August 1998							
Vigorous	71 (± 0.97)a	11.3 (± 0.32)a	5.2 (± 1.4)a	41.7 (± 7.0)a	1.5 (± 0.56)a	7.9 (± 2.3)a	72.6 (± 4.9)a
Non-vigorous	62 (± 0.97)b	11.9 (± 0.32)a	3.6 (± 1.4)a	26.0 (± 7.0)a	1.7 (± 0.56)a	7.9 (± 2.3)a	82.5 (± 4.9)a

[A]Acceptable fruit was calculated as the percentage of fruits with 5% or less anthracnose severity and no stem-end lesions.
[B]Mean values (± s.e.) within columns followed by the same letter are not significantly different at $P = 0.05$.

Table 4. The effect of tree vigour on the incidence (%) of *Dothiorella* spp. (stem-end rot) and *Colletotrichum gloeosporioides* (anthracnose) fungal pathogens isolated from stem-end lesions of 'Hass' avocado fruits ripened at 22°C and 65% RH

Treatment	*Dothiorella* spp.	*Colletotrichum gloeosporioides*
	Harvested October 1997	
Vigorous	42.4[A] (± 5.0)b	21.3 (± 5.1)a
Non-vigorous	59.3 (± 4.7)a	8.2 (± 4.8)a
	Harvested August 1998	
Vigorous	60.9 (± 17.2)a	16.7 (± 7.0)a
Non-vigorous	60.0 (± 17.2)a	3.1 (± 7.0)a

[A]Mean values (± s.e.) within columns followed by the same letter are not significantly different at $P = 0.05$.

Table 5. Effect of tree vigour on the incidence of pepper spot on 'Hass' avocado fruits positioned on the inside and outside of the tree canopy (1997/98 season)

Fruit position	Treatment		Row mean
	Vigorous	Non-vigorous	
Inside	96.6[A] (± 3.2)a	83.4 (± 3.4)b	90.4 (± 2.5)B
Outside	98.9 (± 3.2)a	100.0 (± 3.4)a	99.4 (± 2.5)A

[A]Mean values (± s.e.) followed by the same case letter are not significantly different at $P = 0.05$

No significant ($P < 0.05$) differences were found between the treatments for the severity or incidence of stem-end lesions (Table 2). However, when the causal organisms of these stem-end lesions were identified and the data segregated accordingly, a significant ($P < 0.05$) difference was found between treatments for pathogen predominance. In 1997, fruits from non-vigorous trees had a significantly ($P < 0.05$) higher incidence of *Dothiorella* spp. at the stem-end than fruits from vigorous trees (Table 4). A general trend was evident in both seasons with stem-end rot pathogens detected more frequently than the anthracnose pathogen at the stem-end of fruits from non-vigorous trees compared to those from vigorous trees (Table 4).

On average, the incidence of pepper spot was higher on fruits positioned on the exterior of the tree canopy (outside fruits) than fruits positioned on the interior of the tree canopy (inside fruits) (Table 5). Treatment differences in pepper spot were apparent only between inside fruits, with lower incidences of pepper spot on non-vigorous inside fruits than on vigorous inside fruits (Table 5). Positional effects were also only apparent in the non-vigorous treatment, with lower incidences of pepper spot on non-vigorous inside fruits than on non-vigorous outside fruits (Table 5).

Fruits harvested from vigorous trees had a significantly ($P < 0.05$) lower (~40%) concentration of Ca and higher concentrations of Mg and K compared with fruits harvested from non-vigorous trees (Table 6). These differences resulted in a less favourable balance of nutrients (i.e. lower Ca + Mg/K ratio and higher K/Ca ratio, Table 6). No significant ($P < 0.05$) differences in percentage flesh dry matter were evident between the two treatments (Table 6).

Discussion

Tree vigour was found to significantly impact on disease susceptibility. The increased disease resistance to anthracnose and longer shelf life of fruits from the non-vigorous trees may have been due to their higher concentration of Ca and more favourable balances of mineral nutrients, in particular the higher ratio of Ca + Mg/K.

Fruit mineral nutrition, especially Ca, is very important for sound fruit quality (Chaplin and Scott 1980; Wills and Tirmazi 1982). High fruit Ca concentration has been correlated with low levels of postharvest disease in apple (Conway and Sams 1983, 1984, 1987; Sams *et al*. 1993; Conway *et al*. 1999), peach (Conway *et al*. 1987) and avocado (Hofman *et al*. 1999). The ability of Ca to retard ripening and senescence and reduce the postharvest development of diseases is thought to be due to strengthening of cell walls and maintenance of membrane selective permeability and integrity (DeMarty *et al*. 1984). Calcium strengthens cell walls by cross-linking the pectic polymers in the cell wall and middle lamella (Carpita and Gibeaut 1993), thereby making them less accessible to attack by fungal pectolytic enzymes (Bateman and Lumsden 1965; Stockwell and Hanchey 1982; McGuire and Kelman 1986; Conway *et al*. 1992). Restricting tissue maceration and the subsequent release of free Ca^{2+} ions from the cell wall may also reduce decay by preventing host signalling and the activation of enzymes involved in pathogenesis (Pagel and Heitefuss 1990).

Table 6. The effect of tree vigour on fruit flesh mineral concentrations (g/kg FW) and percentage dry matter (DM) of 'Hass' avocado fruits harvested in August 1998

Treatment	Ca	Mg	K	Ca + Mg/K	K/Ca	% DM
Vigorous	76.7[A] b	269.3 a	4150 a	0.09 b	65.5 a	27.2 a
Non-vigorous	127.6 a	253.1 b	3672 b	0.11 a	30.1 b	28.3 a
SE	4.8	5.2	120	0.004	3.5	0.6

[A]Mean values within columns followed by the same letter are not significantly different at $P = 0.05$.

Fruits from the more vigorous trees possibly had a lower concentration of Ca due to the presence of more vegetative shoots which can out-compete fruits for Ca (Kirkby and Pilbeam 1984; Ho *et al*. 1993). Similar reductions in fruit Ca accumulation from more vigorous avocado trees were observed by South African researchers (Cutting and Bower 1990; Witney *et al*. 1990). However, Cutting and Bower (1990) were unable to relate the reduced fruit Ca accumulation in more vigorous trees to any significant changes in fruit quality, such as an increase in physiological storage disorders. The observed effect of tree vigour on fruit size in this study may have also been contributing to the higher concentration of Ca in smaller fruits from non-vigorous trees. An inverse curvilinear relationship between fruit Ca concentration and mean fruit mass has been shown to exist in apple (Perring 1979). In mango, an increase in tree vigour, associated with an increase in the leaf to fruit ratio, has been shown to cause a significant reduction in fruit Ca accumulation and an increase in postharvest disease susceptibility (Simmons *et al*. 1998).

Fruits from non-vigorous trees were also found to be less susceptible to pepper spot, but only on the inside of the tree canopy. This observation was most likely because the incidence of pepper spot was, on average, lower on the inside of the tree. Inside fruits were shaded and less sunburnt, which can exacerbate pepper spot (Willingham *et al*. 2000). Thus, larger treatment differences were evident. Pepper spot may be more prevalent on fruits positioned on the sunny exposed side of the tree as the fruits natural defences may be compromised in stressed tissue (Joyce *et al*. 1998).

The increase in the presence of stem-end rot fungi in non-vigorous fruits may have been due to the significant reduction in vegetative flushing. Any factor that reduces the vegetative growth of a tree, such as water stress, has been hypothesised to favour the colonisation of the fruit pedicels and stem-end tissue by endophytic stem-end rot fungi (Pusey 1989). Although, endophytic colonisation of avocado fruit stem-end tissue has not been proven, it is believed that an infection process occurs in avocado similar to that shown in mango (Johnson *et al*. 1992).

This study has demonstrated that factors that influence mineral (particularly Ca) accumulation into the fruit such as differences in tree vigour due to root rot, can have a significant impact on anthracnose and pepper spot susceptibility. Canopy density was also shown to be an important factor for pepper spot development as less pepper spot was observed on fruits positioned on the inside of the tree canopy of non-vigorous trees compared to vigorous trees. These fruits would be less sun-stressed but also in conditions that were perhaps less conducive (i.e. reduced humidity due to less dense canopy) for spore germination and infection.

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