# INFECTION CRITERIA FOR PATHOGENS CAUSING BODY ROTS IN AVOCADOS

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## ABSTRACT

Post-harvest body rots are initiated when the fruit are infected in the orchard. Temperature has a very strong influence on the infection process, and if will not occur if temperatures are too low. Experiments were carried out to determine whether or not avocado pathogens infect latently to cause body rots during winter in New Zealand. The aim of these experiments was to investigate if fungicides need to be applied in winter by determining the time at various temperatures in which spores of Colletotrichum acutatum and Colletotrichum gloeosporioides will form appressoria, of Colletotrichum acutatum, Colletotrichum gloeosporioides, and spores Botryosphaeria parva and Phomopsis sp. germinate. A gradient temperature plate was used to determine the time at each of 14 temperatures ranging from 10-30° C for either spore germination or appressorial formation to occur. Spore germination and appressorial formation by C. acutatum and C. gloeosporioides and spore germination by Phomopsis were inhibited by temperatures commonly occurring in avocado growing districts in New Zealand in winter (<14° C for 24 hours), but spore germination by *B. parva* was not inhibited. Based on these results, application of fungicides during winter should result in improved control of avocado fruit rots, since B. parva is the most common pathogen.

**Keywords:** temperature, spore germination, appressorial formation, *Colletotrichum*, *Botryosphaeria*, *Phomopsis*.

### INTRODUCTION

In New Zealand, body rots are caused by five pathogens, *Colletotrichum acutatum*, *C. gloeosporioides*, *Botryosphaeria parva*, *B. dothidea* and *Phomopsis* sp. The most commonly isolated pathogen from body rots in New Zealand is *B. parva* (Hartill, 1991; Everett and Pak 2001). In Israel and Australia *C. gloeosporioides* is the major pathogen causing body rots, and infects throughout the year (Peterson, 1978; Coates *et al.*, 1993; Prusky *et al.*, 1991). In both these countries, spores of *C. gloeosporioides* germinate to cause latent infections by appressoria (hard protective structures glued on the side of the avocado fruit). These appressoria remain latent until fruit ripen after harvest, when they resume growth and fungal hyphae (threads) penetrate into the fruit flesh to cause rots.

Studies in New Zealand have failed to confirm that latent infections by *C. gloeosporioides* or *C. acutatum* occur throughout the year (Everett and Hallett, 1997; Hartill 1988). Further, some results suggest that spraying fungicides in winter may not be important for rot control (Everett, 2000). Appressoria of New Zealand isolates of *Colletotrichum acutatum* and *C. gloeosporioides* do not form below 12° C (Everett,

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1998). It is possible that infection by some pathogens does not occur in winter in New Zealand avocado orchards because the temperature is too low for appressorial formation and/or spore germination.

Temperature studies of avocado pathogens in the literature are on isolates of *Colletotrichum gloeosporioides* from environments with a higher ambient temperature than is common in avocado growing areas of New Zealand. Dodd et al. (1991) comment that the isolates of *Colletotrichum gloeosporioides* from the Philippines require higher temperatures than those of Queensland, Australia (Fitzell *et al.*, 1984) to form appressoria. Thus the outcomes of overseas literature does not provide the information necessary to answer the question proposed in this study.

The aim of these experiments was to investigate if fungicides need to be applied in winter by determining the time at temperatures ranging from 10-30° C in which spores of *Colletotrichum acutatum* and *Colletotrichum gloeosporioides* will form appressoria, and spores of *Colletotrichum acutatum*, *Colletotrichum gloeosporioides*, *Botryosphaeria parva* and *Phomopsis* sp. germinate.

## MATERIAL AND METHODS

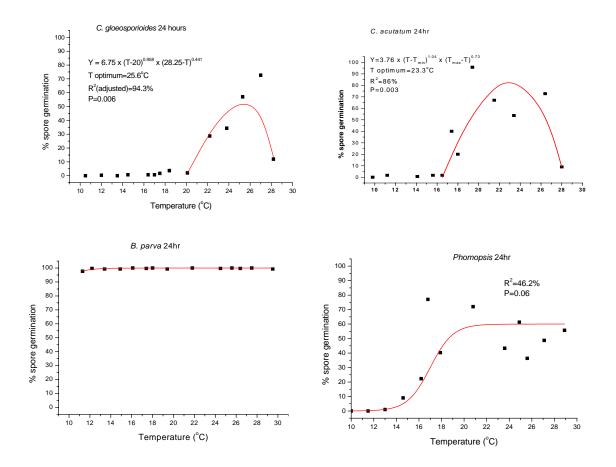
A 10  $\mu$ l aliquot of 10<sup>5</sup> spores/ml suspension of each of *Colletotrichum acutatum*, *Colletotrichum gloeosporioides*, *Phomopsis* sp. and *Botryosphaeria parva* were placed on water agar on a glass slide, then covered with a cover slip. Glass slides with spores were placed on a gradient plate on which the temperature ranges from 10-30° C, using the method described in Everett (1998), but using 3 replicate slides per temperature/fungus combination in a vertical line at right angles to the temperature gradient. Spore germination and appressorial formation were counted after 2, 4, 6, 8, 10, 12, 24, 48 and 96 hours, and 7 days after commencement of the experiment, for a total of 10 assessments, or until more than 95% of spores had germinated. There were 3 replicate slides per temperature, with a total of 42 slides per fungus. The same slides were assessed at each time.

Meteorological summaries of regional monthly average temperatures were obtained from the New Zealand Meteorological Service (NZMS, 1980).

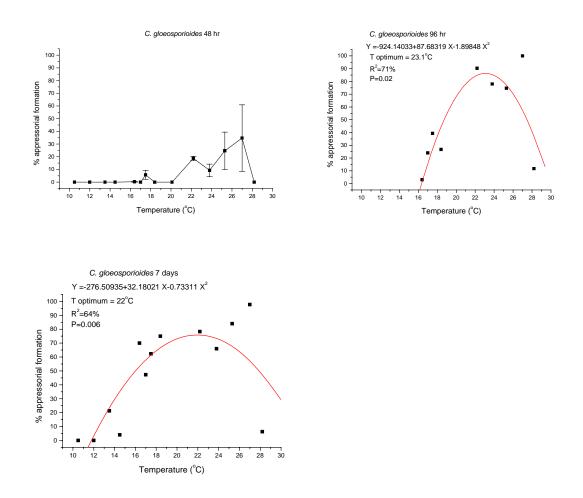
### RESULTS

Spore germination after 24 hours did not occur below 20° C for *C. gloeosporioides*, below 16.5° C for *C. acutatum*, or below 14° C for *Phomopsis* sp. Low temperatures did not inhibit spore germination by *B. parva* (Figure 1). Optimal temperatures for spore germination were 25.6° C for *C. gloeosporioides* and 23.3° C for *C. acutatum*.

Appressorial formation for *C. acutatum* and *C. gloeosporioides* was also inhibited by low temperatures. Temperatures above  $16^{\circ}$  C for longer than 48 hours continuously are required for appressorial formation by *C. gloeosporioides* (Figure 2), and above  $12^{\circ}$  C for longer than 96 hours for appressorial formation by *C. acutatum* (Figure 3). Optimal temperatures for appressorial formation for *C. gloeosporioides* is the range  $22-23^{\circ}$  C



**Figure 1.** Spore germination of *C. gloeosporioides*, *C. acutatum*, *B. parva* and *Phomopsis* sp. after 24 hours. A model was fitted when statistically significant to determine optimal temperature for germination.



**Figure 2.** Appressorial formation by *Colletotrichum gloeosporioides* in response to temperature after A) 48 hours, B) 96 hours and C) 7 days.

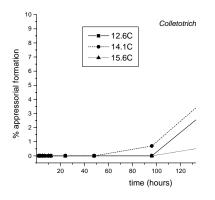
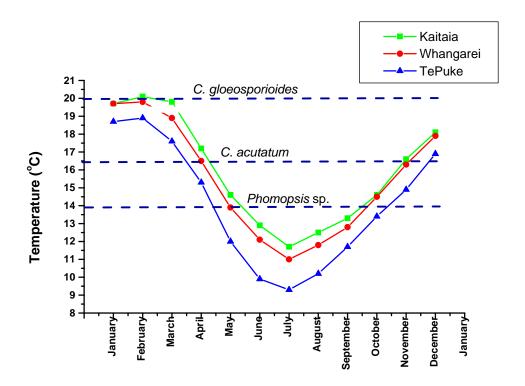


Figure 3. Appressorial formation by *C. acutatum* over time at 3 temperatures.

#### DISCUSSION

For three of the avocado pathogens (*Colletotrichum gloeosporioides, C. acutatum* and *Phomopsis* sp.) average temperatures over winter are too low for germination to occur during winter in the avocado growing districts in New Zealand (Figure 4). For these fungi infection is unlikely to occur over winter. However, for *Botryosphaeria parva*, only 24 hours above 11° C is required for spores to germinate. There is a relatively high probability that these temperature durations will occur in winter in all avocado growing districts. Overall most rots are caused in New Zealand avocados by *Botryosphaeria parva* (Everett, 2000), with a relatively higher proportion in the southern growing regions of New Zealand.



**Figure 4.** Mean daily temperatures in three districts in New Zealand. Horizontal lines represent the minimum temperatures required for germination by spores of the fungi tested after 24 hours.

It has been noted in the literature for isolates of *Colletotrichum gloeosporioides* from mango and *Stylosanthes* that there is some variation in response to temperature (Ela and Quimio, 1973; Estrada *et al.*, 2000; Lenné and Brown, 1991; Munaut and Maraite, 1998) between isolates. Differences in temperature response between isolates of *Colletotrichum gloeosporioides* from avocado, apple and *Stylosanthes* have also been noted by workers in this laboratory (Everett *et al.*, 2000). Because of this variation between isolates, further examination of the temperature response of a large number of isolates would provide more robust data. However, the dominance of *Botryosphaeria parva* as a pathogen of avocados in the cool climate of New Zealand in contrast to its relative infrequency in rots in avocados grown in the warm

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climates of Queensland Australia, South Africa and Israel, suggest that the results of this study are likely to be representative of the overall trends in New Zealand.

On the basis of this data, a fungicide programme based on summer spray applications should control all fungi except *B. parva*, explaining the predominance of *B. parva* in avocado fruit rots in New Zealand. Application of fungicides during winter is likely to further reduce rots of avocados in New Zealand by improving control of *B. parva*.

### CONCLUSION

Infection of avocado fruit by the two *Colletotrichum* spp. or *Phomopsis* sp. is unlikely to occur in winter due to the low temperatures over this period. However, infection by *B. parva* is unaffected by cool temperatures common in avocado growing districts in New Zealand during winter. It is not known to what extent infections over winter contribute to final rot levels, but winter fungicide applications should decrease the level of rots in ripe fruit.

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