Relationship of Fruit Ripeness to Infestation in 'Sharwil' Avocados by the Mediterranean Fruit Fly and the Oriental Fruit Fly (Diptera: Tephritidae)

DAVID H. OI AND RONALD F. L. MAU

Department of Entomology,
University of Hawaii,
Honolulu, Hawaii 96822

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ABSTRACT Harvested and unharvested 'Sharwil' avocados, *Persea americana* Mill., were individually exposed to gravid females of Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), or Oriental fruit fly, *Dacus dorsalis* Hendel. Infestations of 0–30% were obtained from avocados exposed at 0–2 postharvest; infestations of 66.7–100% at 3–7 d postharvest. Percent infestations of 15.8 and 4.8% were obtained from unharvested avocados exposed to *C. capitata* and *D. dorsalis*, respectively. Mean puparial recoveries ranged from 0 to 4.8 puparia per exposed fruit from the unharvested avocados and avocados exposed at 0–2 d postharvest, and recoveries ranged from 7.7 to 135.5 from avocados exposed at 3–7 postharvest. The hard avocado skin seemed to provide a physical barrier which resulted in lower infestations of both fruit fly species in unharvested avocados, and in avocados that were within 3 d postharvest.

KEY WORDS Insecta, fruit flies, *Persea americana*, maturity

THE PRESENCE of fruit flies in Hawaii prevents the shipment of avocados, *Persea americana* Mill., from Hawaii to the continental United States without methyl bromide fumigation (Anonymous 1976). However, treatment with methyl bromide at required dosages causes discoloration, pitting, and decreased shelf life in many avocado varieties, resulting in unmarketable fruit (Akamine 1963, Ito & Hamilton 1980).

Although certain thick-skinned avocado varieties are thought to be resistant to fruit fly infestations, studies have shown that many of these varieties were occasionally infested (Back & Pemberton 1918; Willard & Mason 1929). Armstrong et al. (1983) reported that unharvested, mature green avocados of var. Sharwil were resistant to the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), the Oriental fruit fly, *Dacus dorsalis* Hendel, and the melon fly, *D. cucurbitae* Coquillett. They suggested that the skin and callus formation in damaged skin provided an effective barrier to fruit fly infestations. From their study, it seemed that the hardness (or impenetrability) of the avocado skin prevented fruit fly infestations. While unharvested 'Sharwil' avocados may be resistant to fruit fly infestations, it is possible that exposed, harvested fruits may still be potential hosts. Oi (1983) found that measurements of skin penetrability could not be used to define a stage in harvested 'Sharwil' avocados that was not susceptible to infestations of *C. capitata* or *D. dorsalis*. The objective of the present study was to examine maturity (or ripeness) as an alternate means of defining a stage in the 'Sharwil' avocado that is not susceptible to infestations of *C. capitata* or *D. dorsalis*.

Materials and Methods

Both species of fruit flies used in the studies originated from laboratory strains maintained at the USDA–ARS Tropical Fruit and Vegetable Research Laboratory in Honolulu, Hawaii. These strains had been in culture for several decades. Flies were reared in the laboratory based on procedures developed by Mitchell et al. (1965) and Tanaka et al. (1969). Laboratory conditions during rearing, fruit infestation, and observation were 25 ± 1°C and 70 ± 6% RH.

Postharvest Exposure to Fruit Flies. 'Sharwil' avocados were obtained from three locations—Ataraxia Farms, Honaunau, Hawaii; the Hawaii Agricultural Experiment Station (HAES) at Waiakea, Hawaii; and the HAES at Poamoho, Oahu. Fruits were harvested in the mature green stage and were shipped to the laboratory in Honolulu. Determination of the mature green stage was based on the lack of skin sheen and the traditional harvest season of the trees. The lack of shrivelling and the normal softening of the harvested fruit were indicative that the fruit had attained the harvestable, mature green stage (Yee 1978).

Avocados of varying degrees of ripeness were individually exposed to 50 gravid female and 25 male flies of either fruit fly species for 25 ± 0.6 h in waxed, cardboard rearing cages (3.8 liter). Ripeness was estimated on the basis of age (days) postharvest, which was similar to the method of Willard

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Fig. 1. Longitudinal section of the stem–fruit junction; *D. dorsalis* eggs were laid through the junction and into the avocado flesh.

& Mason (1929). Flies were 10–21 d old at the initiation of the exposure period. After exposure, the fruit were held for 5 d at room temperature, cut in half longitudinally, and examined for evidence of larval infestation. Fruit were then held over vermiculite for 9 d for puparial recovery. As a check for natural fruit fly infestations, fruit which were not exposed to flies in the laboratory exposures (63 fruit) were handled in a similar manner.

The percentage of infested fruit and the mean number of puparia recovered per fruit were determined based on the number of fruit exposed per stage of ripeness. The percentage of infested fruit indicated the extent, or amount, of infestation that occurred, and the mean number of puparia provided a conservative estimate (because of possible larval mortality) of the severity of the infestation.

Observations of exposed avocados revealed that many eggs were laid in the junction of the stem and the fruit (Fig. 1). As a consequence of abscission between the stem and the fruit during ripening, a slight space would develop at this junction, thus providing access to the avocado flesh. To evaluate the relative importance of this oviposition site on infestations, the stem–fruit junction was sealed with melted paraffin. Avocados with the paraffin applications were denoted as "treated" and those without paraffin as "untreated."

Totals of 65 and 74 untreated avocados were exposed to *C. capitata* and *D. dorsalis* respectively. For the treated fruit, totals of 38 and 48 fruit were exposed to *C. capitata* and *D. dorsalis* respectively. The numbers of fruit exposed at each stage of ripeness are shown in Tables 1 and 2.

Preharvest Exposure to Fruit Flies. This study was conducted at the HAES at Poamoho, Oahu, in 1982, using 'Sharwil' avocado trees that were bearing fruit in the harvestable, mature green stage. Individual, unscarred fruit and an adjacent branch with leaves were enclosed in screen cages (24 cm by 17 cm diameter). Each fruit was exposed to 35 gravid female and 10 male flies of either fruit fly species. Food and water were also added to each cage. Controls consisted of individually caged fruit, which, in addition to being exposed to the same number of flies, were also cut with a knife to expose the flesh to oviposition. After a 3-d exposure period, the fruits were harvested and taken to the laboratory for determination of percent infestations and mean puparial recoveries.

Table 1. Percent infestations and mean puparial recoveries per stage of ripeness (days postharvest) from 'Sharwil' avocados exposed to *C. capitata*

Ripeness (days postharvest)	n^a	% infested	Mean no. puparia recovered[b]
Untreated[c]			
7	9	66.7	7.7c
5	15	100.0	82.2ab
4	6	100.0	89.0a
3	10	100.0	30.0b
2	10	30.0	3.8cd
1	10	10.0	2.2cd
0	5	0.0	0.0d
Treated[d]			
9	5	100.0	79.4a
6	5	60.0	10.0b
5	8	63.0	25.5b
3	10	40.0	9.7b
1	10	30.0	1.7b

[a] Number of avocados exposed.
[b] Data used for analysis were transformed ($\log_{10}(y + 1)$); means shown are not transformed. Means followed by the same letter are not significantly different ($P = 0.05$; Duncan's multiple range test [SAS Institute 1982]).
[c] Stem–fruit junction not sealed with paraffin.
[d] Stem–fruit junction sealed with paraffin.

Table 2. Percent infestations and mean puparial recoveries per stage of ripeness (days postharvest) from 'Sharwil' avocados exposed to *D. dorsalis*

Ripeness (days postharvest)	n^a	% infested	Mean no. puparia recovered[b]
Untreated[c]			
11	5	0.0	0.0b
7	15	86.7	135.5a
6	5	100.0	121.0a
5	14	92.9	89.0a
3	10	100.0	125.0a
2	10	30.0	4.8b
1	10	20.0	0.6b
0	5	20.0	2.2b
Treated[d]			
10	7	86.0	24.7a
7	5	40.0	98.0a
6	5	20.0	21.0a
5	9	56.0	16.6a
3	12	67.0	57.8a
1	10	40.0	5.3a

[a] Number of avocados exposed.
[b] Data used for analysis were transformed ($\log_{10}(y + 1)$); means shown are not transformed. Means followed by the same letter are not significantly different ($P = 0.05$; Duncan's multiple range test [SAS Institute 1982]).
[c] Stem–fruit junction not sealed with paraffin.
[d] Stem–fruit junction sealed with paraffin.

Totals of nine cut and 19 intact, attached avocados were exposed to *C. capitata*, while 21 intact and two cut, attached avocados were exposed to *D. dorsalis*. The low number of cut fruits exposed to *D. dorsalis* was a result of an inordinate amount of fruit abscission, possibly caused by excessive handling during incision.

Analysis. Analyses of the postharvest exposure data were performed separately among treated and untreated avocados for each fly species. Analyses of variance and Duncan's multiple range tests (SAS Institute 1982, 119–138) were used to evaluate the effect of ripeness on mean puparial recoveries. Because the variances of puparial counts among the stages of ripeness were heterogenous, counts were logarithmically transformed ($\log_{10}(y + 1)$). For the preharvest exposures to *C. capitata*, the chi-square test for independence was used to compare the percentages of cut and intact, attached avocados that were infested or uninfested. Because of the small sample size of cut avocados, Fisher's exact test (Steel & Torrie 1980, 504–506) was used to compare percentages for the *D. dorsalis* exposures. A 5% significance level was used for all analyses.

Results and Discussion

No puparia were recovered from any of the avocados held to check for natural infestations.

Postharvest Exposures. Exposures of the untreated avocados to *C. capitata* and *D. dorsalis* generally resulted in increased percentages of infestation and puparial counts as fruits ripened (Tables 1 and 2). More than 66% of the untreated fruit from each ripeness category were infested when exposed to flies of either species from 3 to 7 d postharvest. Mean puparial recoveries were significantly higher from fruits exposed to *C. capitata* at 3–5 d postharvest than fruit exposed at 0–2 and 7 d postharvest. Mean *D. dorsalis* puparial recoveries were significantly higher from fruit exposed at 3–7 d postharvest than at 0–2 and 11 d postharvest. The large increases in percent infestation and mean puparial recoveries beginning at 3 d postharvest were probably associated with the exposure of flesh at the stem–fruit junction which occurred at this time.

The lower infestation percentages and puparial recoveries (at 7 and 11 d postharvest) from fruits exposed to *C. capitata* and *D. dorsalis*, respectively, may be attributed to the decomposition of overripe avocados. Some of the avocados at these stages of maturity were beginning to display evidence of fungal decay and were probably unsuitable substrates for larval development. Puparial recoveries were consistently lower from the stages with fungus-infected avocados when compared with the other stages. The unsuitability of fungal-decayed overripe fruit for larval development of *C. capitata* has been reported by Back & Pemberton (1915) and for *D. dorsalis* by Bess & Haramoto (1961).

Infestations of both species were generally lower in the more recently harvested avocados (0–2 d postharvest). This concurs with reports of hard, mature green avocados being poor hosts of *C. capitata* (Back & Pemberton 1918, Willard & Mason 1929) and *D. dorsalis* (Manoto & Mitchell 1976). Resistance to *C. capitata* infestations in avocados of the Guatemalan race (the 'Sharwil' is a Mexican × Guatemalan race hybrid) was thought to be due to their tough, thick skin (Pope 1924) or to their oily flesh (Willard 1927).

Infestations of treated avocados exposed to *C. capitata* generally increased with ripeness, ranging from 30% for fruit exposed at 1 d postharvest to 100% for fruit at 9 d postharvest (Table 1). Infestations of treated avocados exposed to *D. dorsalis* did not follow an increasing trend with ripeness and ranged from 20% at 6 d postharvest to 86% at 10 d postharvest (Table 2). The mean number of puparia recovered from treated avocados exposed to *C. capitata* at 9 d postharvest was significantly higher than the recoveries from the other stages of ripeness (Table 1). Puparial recoveries from treated fruits exposed to *D. dorsalis* were not significantly different (Table 2).

Percent infestations and puparial recoveries were generally lower in the treated than in the untreated avocados of the same ripeness exposed to *C. capitata* and *D. dorsalis*. However, percent infestations were higher in the treated than in the untreated avocados exposed at 1 d postharvest. Treated fruits exposed at 3 d postharvest did not exhibit the dramatic increase in percent infestations and mean puparial recoveries found in the untreated fruits. Thus, the separation at the stem–fruit junction was important because it provided a suitable oviposition site for both fruit fly species. Fruits at all stages of ripeness among the treated avocados were infested by both fruit fly species. It was evident that the avocado skin was not a completely effective barrier to oviposition and larval development within the fruit. Therefore, harvested fruit must be protected from exposures to fruit flies, and postharvest operations such as packing and shipping should be accomplished within 3 d postharvest to minimize the risk of oviposition at the stem–fruit junction.

Preharvest Exposures. All of the cut avocados had evidence of infestation; puparia were recovered from all fruits except for one fruit exposed to *C. capitata* (88.9% infested). In contrast, only 15.8 and 4.8% of the attached, intact fruits were infested by *C. capitata* and *D. dorsalis*, respectively. These results were significant for the *C. capitata* exposures ($\chi^2 = 13.68$; df = 1; $P < 0.001$) and for the *D. dorsalis* exposures (probability of obtaining the observed or more extreme infestations was 0.012 by Fisher's exact test). Mean (±SEM) puparial recoveries from the cut and intact fruits, respectively, were 69.8 (±24.8) and 1.0 (±0.8) for *C. capitata* and 218.5 (±33.5) and 0.2 (±0.2) for *D. dorsalis*. Puparial recoveries from attached avo-

cados that were cut or intact showed that unharvested, mature green avocados were capable of supporting infestations of either species of fruit flies. The low infestation rates in the attached, intact fruits, and the correspondingly few puparia recovered per infested fruit ($\bar{x} = 6.3$ for *C. capitata*, 4.0 for *D. dorsalis*) indicated that the avocado skin was an important barrier to infestation.

Willard & Mason (1929), exposing various Guatemalan avocado varieties to *C. capitata*, obtained a 1.3% infestation rate in unharvested, caged fruits. Armstrong et al. (1983) did not find any infestations among unharvested, caged 'Sharwil' avocados exposed to either *C. capitata* or *D. dorsalis*. The contrasting results between this study and that of Armstrong et al. (1983) can probably be attributed to differences in fly numbers, exposure duration, and postexposure handling of the fruit. Armstrong et al. (1983) exposed unharvested avocados to five females per caged fruit for 24 h, whereas exposures of 35 females for 3 d, which were similar to the methods of Willard & Mason (1929), were used in this study. The higher fly densities and longer exposure period would favor successive ovipositions within the same oviposition puncture, which could possibly overcome callus formation around oviposition punctures. Successive ovipositions by several *D. dorsalis* females in the same oviposition puncture has been reported by Steiner (in Christenson & Foote 1960). Callus formation has been cited as a factor of fruit fly resistance in unharvested avocados by Smith (1973), Sarooshi et al. (1979), and Armstrong et al. (1983). Moreover, the higher puparial recoveries may be attributed to differences in postexposure procedures. Fruits were cut 5 d after fly exposure to facilitate larval survival and pupation (Manoto & Mitchell 1976), whereas Armstrong et al. (1983) left the fruits intact for 3 wk.

The apparent ineffectiveness of the skin barrier to fruit fly infestations in the harvested avocados exposed before the separation at the stem–fruit junction (0–2 d postharvest) may be associated with the onset of the ripening (or softening) processes which occur after fruit abscission. This phenomenon of postharvest ripening in avocados has been documented by Biale et al. (1954) and Burg & Burg (1962). Another possibility is the lack of sufficient callus formation around oviposition punctures in harvested avocados. Kay & Schroeder (1963) have demonstrated in vitro tissue regeneration of pericarp sections excised from harvested, immature, and mature green 'Hass' avocados. However, the effectiveness of this regeneration against fruit fly infestations would be dependent on the time period required for sufficient growth and hardening. Schroeder (1955) reported that in vitro cellular proliferation of pericarp tissue from mature avocados was observed "within three or four weeks."

One of the objectives of the present study was to determine if a particular stage of mature green 'Sharwil' avocados was not susceptible to fruit fly infestations. Identification of such a stage could provide a basis for relaxation of quarantine restrictions on the shipment of 'Sharwil' avocados from Hawaii to the mainland United States. The postharvest exposure studies suggested that less mature fruits were not as susceptible to infestations compared with more mature or ripe fruit. The field exposures tested this trend further by exposing unharvested fruit. While all stages of ripeness tested were infested by either *C. capitata* or *D. dorsalis*, exposures were made in enclosed cages under high fly densities, which may represent a worse-case scenario. The low number of infestations found in the unharvested, intact avocados under such extreme conditions suggested that possible resistance exists. However, this resistance seemed to be dependent on the integrity of the avocado skin.

Determination of sound, unblemished fruit by visual inspection would be difficult in the 'Sharwil' variety because of its rough skin texture. While the probability of fruit fly infestations of 'Sharwil' avocados may be remote under natural field conditions, the nature of resistance in this variety does not ensure that infestations could not occur. Harvest operations could render the fruit susceptible if the skin was damaged as a result of handling. In conclusion, it is apparent that 'Sharwil' avocados can serve as a potential host of *C. capitata* and *D. dorsalis* under reasonable normal harvesting and handling conditions.

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