

Outcrossing Rate, Yield, and Selective Fruit Abscission in 'Ettinger' and 'Ardith' Avocado Plots

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ABSTRACT. The reciprocal effect of two avocado (*Persea americana* Mill.) cultivars—Ardith and Ettinger—on outcrossing rate and yield was studied in several orchards in Israel. Multilocus estimates of outcrossing rates were made using the isozyme loci *Mdh-1* (malate dehydrogenase) and *Aat-1* (aspartate aminotransferase) for 'Ettinger' progeny and *Lap-2* (leucine aminopeptidase), *Pgm-1* (phosphoglucosyltransferase) and *Tpi-1* (triosephosphate isomerase) for 'Ardith' progeny. When the two cultivars were in close proximity, estimated yields ranged from 10 to 20 t·ha⁻¹ and outcrossing rates ranged from 0.71 to 0.89 and from 0.87 to 0.90 for 'Ettinger' and 'Ardith', respectively. The effect of 'Ettinger' as a pollinizer was not restricted to adjacent 'Ardith' trees; it also reached more distant 'Ardith' trees. Thus, outcrossing rate in 'Ardith' was 0.82 at a distance of 30 m from 'Ettinger' in one orchard and 0.91 at a distance of 36 m in another orchard. These results confirm previous observations that 'Ettinger' is a highly potent pollinizer. Outcrossing rates in 'Ardith' and 'Ettinger' were found to increase from the young fruitlet stage to that of mature fruit. These findings provide evidence for selective abscission of selfed fruitlets. In addition, parentage analysis of abscised versus retained 'Ardith' fruit showed that 'Ardith' selfed fruit abscised at a much higher rate than outcrossed ones. The survival advantage of outcrossed fruit is probably related to the fact that selfed progeny have less-vigorous embryos than outcrossed progeny due to inbreeding depression.

The avocado (*Persea americana*) is considered to be an out-crossing species due to its protogynous dichogamy and the existence of two complementary flowering groups—A and B (Bergh, 1974; Davenport, 1986; Nirody, 1922; Stout, 1923, 1932). Planting complementary cultivars in orchards has therefore been recommended to achieve adequate pollination and fruit set (Bergh, 1977). However, although the unique flowering behavior of the avocado enhances the opportunity for cross-pollination, there is usually some overlap between female and male blooms within the same cultivar, which enables close-pollination as well (Davenport, 1986; Ish-Am and Eisikowitch, 1992). Self-pollination within flowers has also been reported (Davenport et al., 1994). Indeed, isolated single trees and solid blocks of a single cultivar are known to set good crops (Davenport, 1986;

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Hodgson, 1947).

In the present work, the reciprocal effect of two complementary avocado cultivars—Ettinger and Ardith—on outcrossing rate and yield was studied. 'Ettinger' (type A flowering group) is an old local selection that has become a prominent commercial cultivar in Israel (Ticho and Gefen, 1965). 'Ardith' (type B flowering group), a selection from the University of California Riverside's avocado breeding program, is now grown commercially in Israel (Blumenfeld and Elimeleh, 1986). Recently, it has been observed that close proximity between these two cultivars has a beneficial effect on the yield of both.

To evaluate the hypothesis that this effect is caused by cross-pollination between the two cultivars, we identified the pollen parent in fruit of 'Ardith' and 'Ettinger' using isozyme markers. In addition, we studied the effect of the pollen parent on outcrossing rate during fruit development.

Materials and Methods

PLANT MATERIAL. The studies were carried out in 'Ardith' and 'Ettinger' commercial plots located in various parts of Israel: Kfar Hanassi and Hagoshrim in the Upper Galilee; Nahshon, Na'an, and Tzora in the interior coastal plain, and Massuot Yitzhak, and Palmahim in the coastal plain. 'Ettinger' usually started to flower before 'Ardith', but their flowering periods overlapped for =2 to 3 weeks.

In Tzora, the 16-row 'Ettinger' plot was bordered on one side by a 10-row 'Ardith' block and on the other by a 5-row 'Ardith' block. Fruit were sampled from the first row, which was a mix of 'Ettinger' and 'Ardith', and from the eighth row, the maximum distance from the two bordering 'Ardith' blocks. In Massuot Yitzhak, the plot consisted of five rows of 'Ardith', in the middle row of which every third tree was 'Ettinger'. In Na'an, fruit were sampled from an 'Ettinger' row adjacent to an 'Ardith' block. In Nahshon, a plot consisting of a solid block of 'Ettinger' bordered on one side by a block of 'Ardith' was studied. In Hagoshrim, the 'Ardith' block consisted of four rows and was surrounded by a single row of 'Ettinger' trees. 'Ardith' fruit were sampled from trees adjacent to 'Ettinger'. In Kfar Hanasi, the 'Ardith' block consisted of 16 rows with a row of 'Ettinger' on two opposite sides. At each distance, fruitlets and fruit were randomly sampled.

The plot in Palmahim has a unique planting design. It had previously been a poor-yielding 'Ettinger' plot, which was topworked with 'Ardith' with only partial success. Consequently, about half of the trees were composed of 'Ettinger' and 'Ardith', about a quarter were pure 'Ettinger' and the other quarter pure 'Ardith'. Fruit were randomly sampled in four replicates from either mixed 'Ettinger' and 'Ardith' trees or trees of one cultivar adjacent to the other cultivar.

ISOZYME ANALYSIS. The embryos of the sampled fruitlets and fruit were used for parentage analysis (Degani et al., 1989). The isozyme systems were separated by standard horizontal starch gel electrophoresis (Brewer and Sing, 1970). Gels were sliced and stained for leucine aminopeptidase (LAP; EC 3.4.11.1) (Degani et al., 1986), malate dehydrogenase (MDH; EC 1.1.1.37) (Degani and Gazit, 1984), phosphoglucosmutase (PGM; EC 2.7.5.1) (Soltis et al, 1983), and triosephosphate isomerase (TPI; EC 5.3.1.1) (Goldring et al., 1987). Aspartate aminotransferase (AAT; EC 2.6.1.1) was resolved using Ashton buffer (Lundquist, 1979). Gels were run at 25 mA for the first 30 min, sample wicks were

removed, and electrophoresis was continued at a constant current of 50 mA for ≈ 4.5 h. Gels were stained for AAT activity with 50 mg aketoglutaric acid and 100 mg L-aspartic acid in 50 mL 0.1 M Tris- HCl-buffer pH 8.0; 50 mg fast blue BB salt and 5 mg pyridoxal 5-phosphate were added to the solution just before staining.

Five genes were used to estimate outcrossing: *Aat-1*, *Lap-2*, *Mdh-1*, *Pgm-1*, and *Tpi-1*. We found that the 'Ardith' genotypes for these isozyme loci are *FS*, *FF*, *FS*, *FF*, and *SS*, respectively, whereas 'Ettinger' genotypes *FF*, *FS*, *SS*, *FS*, and *FS*, respectively. Outcrossing rates in 'Ardith' fruit were determined using *Lap-2*, *Pgm-1*, and *Tpi-1*, for which 'Ardith' is homozygous and 'Ettinger' heterozygous. Outcrossing rates in 'Ettinger' fruit were determined using *Aat-1* and *Mdh-1*, for which 'Ettinger' is homozygous and 'Ardith' is heterozygous.

OUTCROSSING RATES. A multilocus estimate of outcrossing rate and its standard error were determined according to Shaw et al. (1981). In the present case, which is simpler than that dealt with by the latter authors, the maternal genotype and the potential parental genotype are known. All trees of each cultivar have the same genotype, since they were vegetatively propagated. In the isozyme loci used to detect outcrossing, the maternal parent is homozygous and the pollen parent is heterozygous; therefore, heterozygous progeny are definite outcrosses, whereas the pollen source of homozygous progeny is unidentifiable. However, assuming regular Mendelian genotype segregation, a percentage of homozygous progeny equal to that of the heterozygous progeny is expected to result from the outcrossing. The multilocus probability of not identifying an outcrossing event that actually occurred (α) can be estimated as the product of the proportions of homozygous progeny expected in outcrossing matings from all loci. Thus, instead of calculating allelic frequencies, simple genetic considerations yield α as $(1/2)^2$ and $(1/2)^3$ for two and three unlinked isozyme loci, respectively. The multilocus estimate of outcrossing is determined by $t = n/N(1 - \alpha)$, where n is the number of definitely outcrossed progeny (heterozygous individuals at one or more loci) in a sample of size N . Thus, for two isozyme loci, $t = n/N \times 4/3$ and for three isozyme loci $t = n/N \times 8/7$.

The difference between the outcrossing rates of two or more populations was analyzed by χ^2 test, using the actual numbers of fruit found to be heterozygous for at least one locus (n). This is a conservative method of analysis, as it can be shown that using the calculated numbers of outcrossed fruit, based on the estimates of t , leads to χ^2 results with greater significance.

Results

EFFECT OF DISTANCE FROM THE OUTCROSSING POLLEN PARENT ON OUTCROSSING RATE IN MATURE FRUIT AND YIELD. Outcrossing rates and yield in 'Ettinger' relative to its distance from 'Ardith' in five avocado plots in Israel are presented in Table 1. Close to 'Ardith', outcrossing rates were 0.71 to 0.89 and the estimated yield was 10 to 20 t-ha⁻¹. In the two orchards where sampling was also carried out at larger distances from 'Ardith', outcrossing rate and yield decreased markedly with increasing distance from 'Ardith'.

Table 2 presents outcrossing rates and yield in 'Ardith' relative to its distance from 'Ettinger'. In Nahshon we found a high rate of outcrossing (0.82) in 'Ardith' at a distance of 30m from 'Ettinger'. Moreover, in Kfar Hanassi, 91% of the 'Ardith' fruit were 'Ettinger' progeny at a distance of 36 m from the latter. (Table 3).

Table 1. Outcrossing rate in mature fruit and estimated yield of 'Ettinger' relative to its distance from 'Ardith'.

Orchard location	Date (September)	Distance (m)	Fruit assayed (no.)	Outcrossing rate ^z	Yield (t-ha ⁻¹)
Palmahim	1991	0-6	120	0.72 ± 0.09	20
	1992	0-6	200	0.71 ± 0.05	18
Na'an	1992	6	50	0.73 ± 0.09	10
Massuot Yitzhak	1992	6	50	0.89 ± 0.09	13
Nahshon	1992	12 ^y	35	0.27 ± 0.09	10
	1992	42	50	0	2
	1992	84	50	0.03 ± 0.03	2
Significance (χ^2)				**	
Tzora	1992	6	50	0.85 ± 0.09	13
	1992	48	50	0.16 ± 0.06	5
Significance (χ^2)				****	

^zMultilocus estimate of outcrossing rate and associated SE were determined according to Shaw et al. (1981).

^y'Ettinger' fruit were not sampled from the row adjacent to 'Ardith' because in this row the trees were very small or missing.

** , ****Significant at $P = 0.01$ and 0.0001 , respectively.

INCREASE IN OUTCROSSING RATE DURING FRUIT DEVELOPMENT. In Kfar Hanassi and Hagoshrim, 'Ardith' fruit were sampled on two dates—July 1994 (at a fruit mass of 1.7 to 3.5 g) and February 1995—at harvest time. In both plots, at a distance of 6 m from 'Ettinger', outcrossing rates in 'Ardith' increased significantly with fruit development, from 0.65 to 1.00 and from 0.70 to 0.91, respectively (Table 3). In Kfar Hanassi, at a distance of 36 m, outcrossing rate increased significantly, from 0.67 to 0.91 (Table 3).

Table 2. Outcrossing rate in mature fruit and estimated yield of 'Ardith' relative to its distance from 'Ettinger'.

Orchard location	Date (March)	Distance (m)	Fruit assayed (no.)	Outcrossing rate ^z	Yield (t-ha ⁻¹)
Palmahim	1992	0-6	174	0.87 ± 0.04	20
	1993	0-6	50	0.89 ± 0.07	15
Nahshon	1992	6	47	0.90 ± 0.07	15
	1992	30	50	0.82 ± 0.07	15
Significance (χ^2)				NS	

^zMultilocus estimate of outcrossing rate and associated standard error were determined according to Shaw et al. (1981).

^{NS}Nonsignificant at $P = 0.05$.

In another study carried out in Palmahim (1995), the outcrossing rates during fruit development were followed in 'Ettinger' and 'Ardith' (Table 3). The outcrossing rate of 'Ettinger' (average mass of fruitlet 1.5 g) was 0.52 in May. Two months later the rate had increased significantly to 0.77 and was 0.79 in mature fruit in September (Table 3). The outcrossing rate of 'Ardith' in May (average mass of fruitlets was 2.3 g) was 0.91, and 2 months later it had increased significantly to 1.00 (Table 3).

ABSCISSION OF 'ARDITH' FRUIT. Severe fruit drop frequently occurs in 'Ardith' in September to October. At this time of year, 'Ardith' fruit in the Palmahim plot reached an average mass of ≈140 g. To gain some insight into late fruit abscission, we collected abscised fruit from the ground under 'Ardith' trees as well as retained fruit from the same trees in 1992 and 1993. In both years we found that almost all surviving 'Ardith' fruit on the trees were hybrid 'Ettinger' progeny, whereas a high percentage of the abscised 'Ardith' fruit were selfs (Table 4).

Discussion

The high rate of outcrossing in mature fruit of adjacent 'Ettinger' and 'Ardith' trees (Tables 1-3) is consistent with the fact that these cultivars belong to complementary flowering groups with sufficient overlap between their flowering periods.

Table 3. Outcrossing rate in 'Ardith' and 'Ettinger' during fruit development.

Distance from pollenizer (m)	Date	Fruit assayed (no.)	Average fruit mass (g)	Outcrossing rate ²
Ardith-Kfar Hanassi				
6	July 1994	48	1.9 ± 0.1 ³	0.65 ± 0.08
	February 1995	50	238 ± 4	1.00 ± 0.05
Significance (χ^2)				
36	July 1994	40	2.4 ± 0.2	0.67 ± 0.1
	February 1995	49	213 ± 7	0.91 ± 0.07
Significance (χ^2)				
Ardith-Hagoshrim				
6	July 1994	39	3.5 ± 0.3	0.70 ± 0.09
	February 1995	54	236 ± 6	0.91 ± 0.06
Significance (χ^2)				
Ardith-Palmahim				
0-6	May 1995	200	2.3 ± 0.1	0.91(0.03)
	July 1995	176	46 ± 1.7	1.00(0.03)
Significance (χ^2)				
Ettinger-Palmahim				
0-6	May 1995	220	1.5 ± 0.1	0.52 ± 0.05
	July 1995	124	59 ± 3	0.77 ± 0.09
	September 1995	200	207 ± 5	0.79 ± 0.07
Significance (χ^2)				

²Multilocus estimate of outcrossing rate and associated standard error were determined according to Shaw et al. (1981).

³Means ± se.

***Significant at $P = 0.05$ or 0.001 , respectively.

The outcrossing rates in Palmahim were lower for 'Ettinger' than for 'Ardith' (Table 3). This might be due to the fact that 'Ettinger' started to flower before 'Ardith', and, consequently, when 'Ardith' started to flower, selfed 'Ettinger', fruitlets had already set. We assume that these early selfed fruitlets have some advantage over late outcrossed fruitlets, with $\approx 20\%$ of the 'Ettinger' selfed fruitlets surviving to maturity. On the other hand, 'Ardith' flowering overlapped that of 'Ettinger' from the start, and, therefore, selfed 'Ardith' fruitlets had no head start over outcrossed ones. Under these conditions, the strong survival advantage of 'Ettinger' progeny resulted in the failure and abscission of almost all selfed 'Ardith' fruit (Tables 2-3).

Table 4. Outcrossing rates in abscised vs. retained 'Ardith' fruit.

Fruit	Outcrossing rate ²			
	October 1992		September 1993	
	Fruit assayed (no.)	Outcrossing rate	Fruit assayed (no.)	Outcrossing rate
Abscised	50	0.57 ± 0.08	40	0.50 ± 0.1
Retained	30	0.91 ± 0.08	30	0.89 ± 0.1
Significance (χ^2)				
**				

²Multilocus estimate of outcrossing rate and associated standard error were determined according to Shaw et al. (1981).

**Significant at $P = 0.01$.

As pollinizers, 'Ardith' and 'Ettinger' differed in their long- distance effect on the rate of hybrid fruit production. Thus, in Nahshon, the outcrossing rate in 'Ettinger' was already low at a distance of 12 m (0.27) and was 0 at greater distances from 'Ardith' (Table 1). In Tzora, the effect of distance on outcrossing rate was remarkable: the outcrossing rate decreased from 0.85 to 0.16 as the distance from 'Ardith' increased from 6 to 48 m (Table 1). On the other hand, outcrossing rates of 'Ardith' by 'Ettinger' were high at close proximity and at the longer distances of 30 and 36 m (Tables 2-3). 'Ettinger' in Israel and 'Bacon' in California have been found to be highly effective pollen parents for 'Hass', even at large distances

(Degani et al, 1989, 1993; Goldring et al, 1987; Vrecenar- Gadus and Ellstrand, 1985). The high potency of 'Ettinger' as a pollen parent may be due to two factors: 'Ettinger' strongly attracts honeybees, which are the main insect pollinators of avocado in Israel (Ish-Am, 1994), and its pollen confers a high chance of survival on its progeny (Degani and Gazit, 1984).

In 'Hass', yield was correlated with the rate of outcrossing with 'Ettinger' (Degani et al., 1989) and 'Bacon' (Vrecenar-Gadus and Ellstrand, 1985). In the present study, we found a consistent decrease of hybrid percentage and yield in 'Ettinger' with increasing distance from 'Ardith'. This suggests that 'Ettinger' also requires cross-pollination to realize its full yield potential. This assumption is supported by the horticultural history of the Palmahim plot. It was originally an 'Ettinger' plot yielding consistently poor crops ($\approx 5 \text{ t-ha}^{-1}$). It was therefore top-worked with only partial success to 'Ardith'. When 'Ardith' started to bloom in 1990, the 'Ettinger' yield increased dramatically and has remained consistently high for the last 5 years. In a similar case (Robinson and Savage, 1926), a mature 'Fuerte' plot failed season after season to produce any fruit. When a young adjoining grove containing other cultivars first bloomed, the row of 'Fuerte' trees next to the new grove set a full crop of fruit. Cross-pollination of 'Fuerte' by cultivars of complementary flowering groups was suggested to be responsible for its high yield (Robinson and Savage, 1926). In Palmahim, we found high rates of outcrossing in 'Ettinger' and 'Ardith' along with high yields (Tables 1-2). These results support the conclusion that the presence of 'Ardith' and 'Ettinger' in close proximity to each other tends to increase their yield due to cross- pollination.

Besides cross-pollination, avocado yield is determined by several other important factors. A high rate of outcrossing does not necessarily ensure high yield, as shown in Table 1. Low yield, such as that found in Nahshon, could be the result of a very low rate of pollination. Inadequate pollination has been found to be a limiting factor in avocado fruit set wherever there are more attractive blooms of other species (i.e., *Citrus*) nearby, causing honeybees to desert the avocado bloom (Gazit, 1977; Ish-Am and Esikowitch, 1993; Vithanage, 1990).

A consistent and significant increase in the percentage of hybrid fruit from the young fruitlet stage to the mature fruit stage was found for 'Ettinger' and 'Ardith' (Table 3). In 'Ettinger' for example, 50% of the fruitlets and 21% of the mature fruit were selfed (Table 3). The failure of a sizable portion of the selfed 'Ettinger' and 'Ardith' progenies to develop into mature fruit in the presence of outcrossed fruit indicates the effect of selective abscission against selfed fruit. These results concur with those of a previous study, in which we demonstrated a greater survival rate of 'Ettinger' hybrids in 'Hass' (Degani et al., 1989).

In 'Ardith', selective abscission also occurred in 5- to 6-month- old fruit. Parentage analysis of abscised versus retained fruit in September and October showed that almost all surviving 'Ardith' fruit on the trees were hybrids of 'Ettinger', whereas a high percentage of the abscised 'Ardith' fruit were selfs (Table 4).

Greater retention of crossed vs. selfed fruit has been also reported for *Litchi chinesis* Sonn. (Degani et al., 1995), *Mangifera indica* L. (Dag et al., 1996), *Banksia spinulosa* Smith (Vaughton and Carthew, 1993), and other crops (Stephenson, 1981; Stephenson and Winsor, 1986). Limited resources are considered to be a major factor in fruit abortion (Lloyd, 1980; Stephenson, 1981; Lee and Bazzaz, 1982). It has been suggested that to match fruit production to available resources, maternal plants abort immature fruit on the

basis of differences in their embryo vigor (Vaughton and Carthew, 1993). Vigor is assumed to be an indicator of progeny fitness at later developmental stages and, by aborting less vigorous progeny, the average quality of the progeny is enhanced (Lloyd, 1980; Janzen, 1977). Selfed fruit with less-vigorous embryos due to inbreeding depression are likely to abscise at a higher rate than hybrid fruit with more vigorous embryos (Charlesworth and Charlesworth, 1987; Sedgley and Griffin, 1989; Vaughton and Carthew, 1993).

Literature Cited

- Bergh, B.O. 1974. The remarkable avocado flower. *Calif. Avoc. Soc. Yearbook*. 57:40-41.
- Bergh, B.O. 1977. Factors affecting avocado fruitfulness, p. 83-88. In: J.W. Sauls, R.L. Phillips, and L.K. Jackson (eds.). *Proc. First Intl. Trop. Fruit Course: The avocado*. Univ. of Florida, Gainesville.
- Blumenfeld, A. and M. Elimeleh. 1986. New avocado cultivar: 'Ardith' (in Hebrew with English summary). *Alon Hanotea* 40:741-743.
- Brewer, G.J. and C.F. Sing. 1970. *An introduction to isozyme techniques*. Academic Press, New York.
- Charlesworth, D. and B. Charlesworth. 1987. Inbreeding depression and its evolutionary sequences. *Annu. Rev. Ecol. System*. 18:237-268.
- Dag, A., S. Gazit, D. Eisenstein, R. El-Batsri, and C. Degani. 1996. Pollen parent effect on the selective abscission of 'Tommy Atkins' fruitlets. *Proc. 5th Intl. Mango Symp. Tel Aviv, Israel*, 1-6 Sept. 1996. (Abstr.)
- Davenport, T.L. 1986. Avocado flowering, p. 257-289. In: J. Janick (ed.). *Horticultural reviews*, vol. 8. Avi, Westport, Conn.
- Davenport, T.L., P. Parnitzki, S. Fricke, and M.S. Hughes. 1994. Evidence and significance of self-pollination of avocados in Florida. *J. Amer. Soc. Hort. Sci.* 119:1200-1207.
- Degani, C. and S. Gazit. 1984. Selfed and crossed proportions of avocado progenies produced by caged pairs of complementary cultivars. *HortScience* 19:258-260.
- Degani, C., A. Goldring, S. Gazit, and U. Lavi. 1986. Genetic selection during the abscission of avocado fruitlets. *HortScience* 21:1187-1188.
- Degani, C., A. Goldring, and S. Gazit. 1989. Pollen parent effect on outcrossing rate in 'Hass' and 'Fuerte' avocado plot during fruit development. *J. Amer. Soc. Hort. Sci.* 114:106-111.
- Degani, C., R. El-Batsri, and S. Gazit. 1993. Isozyme analysis of subtropical tree fruits. *Polish J. Chem.* 68:1031-1034.
- Degani, C., R.A. Stern, R. El-Batsri, and S. Gazit. 1995. Pollen parent effect on the selective abscission of 'Mauritius' and 'Floridian' lychee fruitlets. *J. Amer. Soc. Hort. Sci.* 120:523-526.
- Gazit, S. 1977. Pollination and fruit set in avocado, p. 88-92. In: J.W. Sauls, R.L. Phillips, and L.K. Jackson (eds.). *Proc. First Intl. Trop. Fruit Short Course: The avocado*. Univ. of Florida, Gainesville.
- Goldring, A., S. Gazit, and C. Degani. 1987. Isozyme analysis of mature avocado embryos

- to determine outcrossing rate in 'Hass' plot. J. Amer. Soc. Hort. Sci. 112:380-392.
- Hodgson, R.W. 1947. The California avocado industry. Calif. Agr. Ext. Serv. Circ. 43:5-93.
- Ish-Am, G. 1994. Interrelationship between avocado flowering and honey bees and its implication on the avocado fruitfulness in Israel (in Hebrew). PhD diss., Tel Aviv Univ., Tel Aviv, Israel.
- Ish-Am, G. and D. Eisikowitch. 1992. New insight into avocado flowering in relation to its pollination. Calif. Avoc. Soc. Yrbk. 75:125-137.
- Ish-Am, G. and D. Eisikowitch. 1993. The behavior of honey bees (*Apis mellifera*) visiting avocado (*Persea americana*) flowers and their contribution to its pollination. J. Apicult. Res. 32:175-186.
- Janzen, D.H. 1977. A note on optimal mate selection in plants. Amer. Nat. 111:365-371.
- Lee, T.D. and P.A. Bazzaz. 1982. Regulation of fruit and seed production in an annual legume, *Cassia fasciculata*. Ecology 63:1364-1373.
- Lloyd, D.G. 1980. Sexual strategies in plants. I. An hypothesis of serial adjustment of maternal investment during one reproductive session. New Phytologist 86:69-79.
- Lundquist, K. 1979. Allozyme frequency distribution in four Swedish populations of Norway spruce (*Picea abies* K.). I. Estimations of genetic variation within and among populations, genetic linkage and mating system parameters. Hereditas 90:127-143.
- Nirody, B.S. 1922. Investigations in avocado breeding. Annu. Rpt. Calif. Avoc. Assn. p. 65-78.
- Robinson, T.R. and E.M. Savage. 1926. Pollination of the avocado. USDA Circ. 387:1-16.
- Sedgley, M. and A.R. Griffin. 1989. Sexual reproduction of tree crops. Academic Press, London, p. 204-213.
- Shaw, D.V., A.L. Kahler, and R.W. Allard. 1981. A multilocus estimator of mating system parameters in plant populations. Proc. Nati. Acad. Sci. USA 78:1298-1302.
- Soltis, D.E., C.H. Hauffer, D.C. Darrow, and G.W. Gastony. 1983. Starch gel electrophoresis of ferns: A compilation of grinding buffers, gel and electrode buffers, and staining schedules. Amer. Fern J. 73:9-27.
- Stephenson, A.G. 1981. Flower and fruit abortion: Proximate causes and ultimate functions. Annu. Rev. Ecol. Syst. 12:253-279.
- Stephenson, A.G. and J.A. Winsor. 1986. *Lotus corniculatus* regulates offspring quality through selective fruit abortion. Evolution 40:453-458.
- Stout, A.B. 1923. A study in cross-pollination of avocados in southern California. Annu. Rpt. Calif. Avoc. Assn. 1922-1923, p. 29-45.
- Stout, A.B. 1932. Sex in avocados and pollination. Calif. Avoc. Assn. Yrbk. 1932, p. 172-173.
- Ticho, R.J. and B. Gefen. 1965. The avocado in Israel. Calif. Avoc. Assn. Yrbk. 49:55-60.
- Vaughton, G. and S.M. Carthew. 1993. Evidence for selective fruit abortion in *Banksia*

spinulosa (Proteaceae). Biol. J. Linnean Soc. 50:35-46.

Vithanage, V. 1990. The role of the European honeybee (*Apis mellifera* L.) in avocado pollination. J. Hort. Sci. 65:81-86.

Vrecenar-Gadus, M. and N.C. Ellstrand. 1985. The effect of planting design on outcrossing rate and yield in the 'Hass' avocado. Scientia Hort. 27:215-221.