

## **POLLEN/STIGMA INTERCOMPATIBILITY BETWEEN DIFFERENT AVOCADO CULTIVARS**

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### **ABSTRACT**

*Pollination in avocados have been studied by various authors in different countries, but very little information is available on the situation in South Africa, especially about pollen/stigma interactions of different cultivars. In reciprocal, in vitro hand-pollinations between three different cultivars done at Westfalia Estate, we could confirm previous statements that avocados are natural out breeders, but poor self-pollinators. 'Fuerte E' proved to be the best pollen donor for 'Ettinger', while 'Fuerte A' was the best pollen donor for 'Hass'. 'Ettinger' was the best "all rounder" pollen parent and also gave the best results with 'Fuerte A' and 'Fuerte E'. Results obtained from open-pollinated flowers collected over a period of one month, indicate that fruit set is restricted to a few "favourable" days per month.*

### **OPSOMMING**

*Bestuiwing by avokado's is deur verskeie outeurs in verskillende lande ondersoek, maar daar bestaan nog baie min inligting oor die situasie in SuidAfrika, veral ten opsigte van die stuifmeel/stempel interaksies van verskillende kultivars. A/a resiproke, in vitro handbestuiwings wat tussen drie verskillende kultivars op Westfalia Landgoed uitgevoer is kon ons vorige uitsprake bevestig dat avokado's natuurlike uitteilers, maar swak selfbestuiwers is. 'Fuerte E' was die beste stuifmeelouer vir 'Ettinger' terwyl 'Fuerte A' die beste stuifmeelouer vir 'Hass' was. 'Ettinger' was die beste "algemene" stuifmeelouer en het ook die beste resáltate met 'Fuerte A' en '-E' gelever. Resultate met oopbestuifde blomme wat oor 'n tydperk van een maand versamel is, dui daarop dat vrugset slegs tot enkele "gunstige" dae per maand beperk is.*

### **INTRODUCTION**

In his paper on "Avocado flower pollination", Peterson (1955) referred to Nirody (1922) and Stout (1932) as authors who started the study on avocado flower behaviour and pollination. Since then, a number of papers related to flowering and fruit set were published by different authors in the "California Avocado Society Yearbook". More recently, Gazit and co-workers have published extensively on avocado pollination and fruit set in Israel, (Gazit 1977; Degani & Gazit 1984; Gazit & Gafni 1986; Degani, Goldring, Gazit & Lavi 1986; Goldring, Gazit & Degani 1987; Degani, Goldring & Gazit

1989) while Sedgley studied avocado pollination in Australia and Davenport is continuing the pollination work in California. In spite of all these publications, pollination and fruit set in avocado is still unsatisfactory. Very little is also known about pollination under South African conditions. The possible reason for the avocado to be so arduous, is the fact that the flowers are dichogamous and the existence of A-type and B-type cultivars (Gazit 1977). To illustrate the amplitude of the problem, Degani et al. (1986) state that "Atypical mature avocado tree has about one million flowers during each flowering season: yet, it usually yields only a few hundred mature fruits". In this paper we report on the results of preliminary experiments and observations that were done during the 1993 flowering season at Westfalia Estates.

## **MATERIAL AND METHODS**

### ***In vitro* pollination**

The following cultivars were used in this experiment: 'Ettinger', 'Hass' and 'Fuerte'. At Westfalia single tree records of production are being kept,

and over a period of several years, constantly high producing trees, 'A'trees', and constantly low producing trees, 'E'-trees, have been identified. For the purpose of this paper we are referring to these trees as "cultivars", 'Fuerte A' and 'Fuerte E'. For each cultivar, 16 x 20 female phase flowers were collected just at the very beginning of a thesis. For each combination, four replications of 20 flowers for each combination were put in agar containing petri dishes, with their pedicells stuck into the agar. Flowers in the late female phase were collected in the same way and kept in a growth cabinet at 27°C until they reopened in the male phase. The latter flowers were used as pollen donors during hand-pollinations. The female phase flowers of each cultivar were reciprocally hand-pollinated with pollen of each of the other cultivars. Collection of the flowers had to be carefully planned for having fresh pollen available for the different combinations. After being pollinated, the flower-containing petri dishes were incubated at 27°C for a period of 1416 hours. The flowers of each replication were separately fixed in a Carnoy solution for 12 hours before being transferred to 70% ethanol. The pistil of each flower was carefully removed and prepared for observation with an epifluorescence microscope. The number of pollen grains on the stigma, number of pollen tubes in the style, number of pollen tubes penetrating the ovary and number of pollen tubes entering the ovule were recorded for each pistil.

### **Open pollination**

Four replications of 20 flowers in the male phase were collected from each of the cultivars 'Ettinger', 'Hass', 'Fuerte A' and 'Fuerte E'. Flowers were fixed in Carnoy and prepared for viewing with epifluorescence optics. Pollen tube performance was scored as described above.

### **Effect of environment on pollination**

For a period of one month, flowers in the male phase were collected on a daily basis from the cultivars 'Hass' and 'Fuerte', prepared for epifluorescence microscopy and the pollen tube growth recorded. During this period, the maximum and minimum daily temperatures, as well as the number of bees per tree noticed while collecting flowers were also recorded.

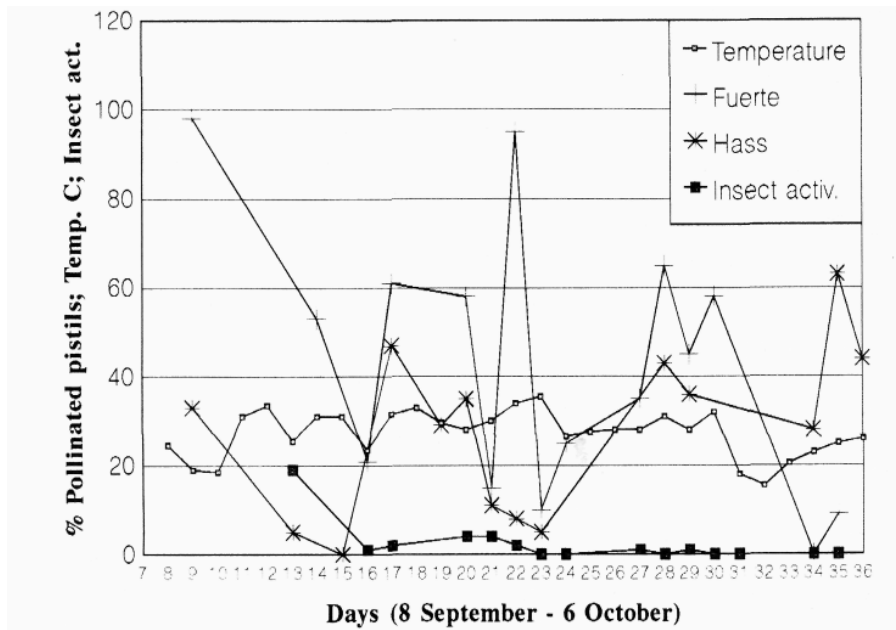


FIG 1:  
Daily % open pollinated Hass and Fuerte flowers related to temperature and insect activity

## RESULTS

The results of the first two experiments are given in Tables 1 and 2 and those for the third experiment in Figure 1. Table 1 depicts the percentages of pistils in which pollen tubes have progressed down to the ovary and ovule. A statistical analysis was done on the results and significant differences are indicated in the footnotes. Table 2 shows the percentage of successfully open-pollinated flowers on a specific day (8 September 1994), in terms of percentage of pistils with pollen tubes reaching different points.

TABLE 1. Percentage pistils with pollen tubes reaching a) the ovary and b) the ovule, 24h after reciprocal *in vitro* hand-pollinations between different avocado cultivars

PISTIL PARENT	POLLEN PARENT				
	Pollen t. response	Ettinger	Hass	Fuerte A	Fuerte E
Ettinger	In ovary	59.6	71.4	61.5	85.2*
	In ovule	17.7	17.7	10.8	51.9*
Hass	In ovary	68.1	50.0	92.1*	69.7
	In ovule	27.5	11.5	31.6	25.8
Fuerte A	In ovary	73.5	45.3	67.7	57.3
	In ovule	14.7	1.6-*	1.6-*	4.9-*
Fuerte E	In ovary	71.8	44.3	41.4-*	46.7
	In ovule	19.7	11.4	5.7-*	15.0

\* (Ettinger x Fuerte E) Ovary Odds ratio = 3.9 (P< 0.05)      -\* (Fuerte A x Hass)      Ovule Odds ratio = 13.6 (P< 0.02)

\* (Ettinger x Fuerte E) Ovule " " = 5.0 (P< 0.005)      -\* (Fuerte A x Fuerte A) Ovule " " = 13.2 (P< 0.02)

\* (Hass x Fuerte A) Ovary " " = 7.9 (P< 0.005)      -\* (Fuerte A x Fuerte E) Ovule " " = 4.2 (P< 0.025)

\* (Fuerte E x Fuerte A Ovary " " = 2.1 (P< 0.05)      -\* (Fuerte E x Fuerte A) Ovule " " = 3.6 (P< 0.05)

n Figure 1 pollination success is related to daily maximum temperatures and bee activity during the relevant period. Bee activity is expressed as the total number (x 2) of bees spotted on the side of the tree from which, and at the time when, flowers were collected.

TABLE 2. Pollination success in open pollinated flowers collected at two different stages, expressed as % pistils with a) pollen on stigma, b) pollen tubes in style and c) pollen tubes in ovary

PISTIL PARENT	POLLEN PARENT & COLLECTING STAGE							
	Ettinger		Hass		Fuerte A		Fuerte E	
	Male	Post-m	Male	Post-m	Male	Post-m	Male	Post-m
Pollen on St.	56.8	-	27.5	35.3	91.4	-	95.5	95.8
P.t in style	8.1	-	17.3	15.7	60.3	-	45.5	62.5
P.t. in ovary	0	-	9.8	0	41.4	-	18.2	45.8

P.T = pollen tube

St. = stigma

Post-m = Flowers collected after the male stage

## DISCUSSION

### *In vitro*-pollination

For each treatment the statistical programme compared the proportions of stigmas that contained pollen tubes in the ovary or style. The Ettinger x Ettinger combination was taken as the point of reference. As a pollen parent for 'Ettinger', 'Fuerte E' was significantly better than any of the other cultivars. The probability of obtaining successful pollination with this combination, was four times greater than the combination 'Ettinger' x 'Ettinger' (odds ratio = 3.9). The best pollen parent for 'Hass', judged on the basis of percentage pollen tubes reaching the ovary, was 'Fuerte A' and in this case the probability for successful pollination was eight times greater than the 'Ettinger' x 'Ettinger' combination (odds ratio = 7.9). Judged on the basis of pollen tubes reaching the ovule, however, this combination was not significantly better than the 'Hass' x 'Ettinger' combination. Although not statistically better than the 'Ettinger' x 'Ettinger' combination, 'Ettinger' proved to be the best pollen donor for 'Hass', 'Fuerte A' and 'Fuerte E', both in terms of pollen tubes reaching the ovary and pollen tubes reaching the ovule. The reciprocal combinations 'Ettinger' x 'Fuerte A' and 'E' as well as 'Fuerte A' and 'E' x 'Ettinger', all produced high figures in spite of the fact that both are B-type cultivars. This observation supports the results of Degani *et al.* (1989) who reported an out crossing rate of 40% where 'Ettinger' served as a pollen donor for 'Fuerte' and Gazit & Gafni (1986), who found that 'Ettinger' pollen gave good results with 'Fuerte' and 'Hass'. This clearly shows that cross-pollination between cultivars of the same flowering group is possible and can even be preferable.

Except for the combination 'Fuerte A' x 'Hass', all the combinations that produced figures significantly lower than the 'Ettinger' x 'Ettinger' combination, were 'Fuerte' x 'Fuerte'. Low figures were also obtained with the selfed 'Ettinger' and 'Hass'. This again supports

the statement by Degani *et al.* (1989) and references therein, that the avocado is considered as an out crossing plant.

### **Open pollination**

The results in Table 2 show that the success rate of open-pollination in 'Fuerte A as well as in 'Fuerte E' was much higher than in 'Ettinger' or 'Hass'. Looking at the percentage pollen tubes that had reached the ovule, the success rate in 'Fuerte' is even more dramatic if it is considered that the flowers were collected in the male phase, which means that in 'Hass', the time allowed for the pollen tubes to reach the ovule was at least 36 hours, while in 'Fuerte' and 'Ettinger', only about 20 hours were available (Peterson, 1956). 'Hass' flowers, which open in the female stage in the morning, are normally exposed for pollination from 09:00 to about 15:00, while in 'Fuerte' and 'Ettinger', the female stage stretches from about 15:00 to 18:00 and if conditions are unfavorable, they do not open at all. It is for this reason that B type cultivars are not suitable for the coastal regions of California (Peterson 1956; Lesley & Bringhurst 1951). When conditions are, however conducive for pollination, 'Fuerte' seems to have no problem. In 'Hass', however, pollination is a real problem which needs immediate attention.

### **Effect of the environment**

The success of open-pollination related to daily variations in environmental conditions for the cultivars 'Fuerte' and 'Hass', is shown in Figure 1. There is a vague relationship between the peaks of open pollination success by the two cultivars, but the most striking effect is the fact that only certain days are favourable for effective pollination. These days seem to coincide with average day temperatures of above about 28°C. Gazit (1977) stated that, "Normal flower behaviour only occurs during warm weather with minimal night temperatures above 12°C and maximum day temperatures above 22°C. As in the previous experiment, the open pollination success in 'Fuerte' was again considerably higher than in 'Hass'. These observations were very preliminary and will have to be repeated in different localities.

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