

AVOCADO POLLINATORS IN SOUTHEAST SPAIN

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In its native place, avocado original pollination vectors are stingless bees. In most areas of recent cultivation, avocado pollination is mainly performed by regular bees. However, some authors suggest that wind may play a role in avocado pollination in areas where female and male flower phases partially overlap. This argument is maintained in the stigma receptivity prolongation which offers chances to autogamy (self-pollination) and geitonogamy (pollination of a flower by another flower of the same flowering plant), driven by wind. In this work we have explored the importance of large and small insects and wind as avocado pollinators in South East Spain.

To do so, we compared pollen adhesion and the resulting fruit set in control flowers freely exposed with those produced in flowers bagged either with micro perforated plastic (allowing wind and small insects to enter) or with tissue paper (excluding all kind of insects). The results show that in our cultivation area only the activity of honey and bumble bees allows fruit set. The activity of thrips caused certain levels of pollen deposition on the stigmas and an initial swelling of avocado flower ovary, however, fruitlets soon abscised. The flowers enclosed in paper bags did not have pollen grains on the stigmas nor set any fruit. An introduction of commercial hives is therefore strongly recommended to achieve avocado pollination in South East Spain.

Key words: pollination, bees, bumble bees, trips, wind, fruit set.

VECTORES DE POLINIZACIÓN DEL AGUACATE EN EL SURESTE ESPAÑOL

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En su lugar de origen el aguacate es polinizado por pequeñas abejas sin aguijón, mientras que en las nuevas áreas de cultivo, la polinización del aguacate se realiza fundamentalmente por abejas. Algunos autores, sin embargo, sugieren que el viento puede contribuir a la polinización del aguacate en aquellas zonas donde las fases femenina y masculina de la flor se solapan. Este argumento se sostiene en la prolongación de la receptividad estigmática que ofrece oportunidades tanto a la autogamia (autopolinización dentro de una misma flor) como a la geitonogamia (autopolinización ente flores del mismo árbol). En este estudio se analiza la importancia de la polinización llevada a cabo por insectos de diferente tamaño y por el viento, comparando la adhesión de polen y el cuajado de frutos en flores expuestas a la

polinización libre, en ramos embolsados con bolsas microperforadas que permiten el acceso de pequeños insectos y en ramos herméticamente cerrados donde tan sólo el viento podría provocar alguna transferencia de polen. Los resultados indican que en el SE español tan sólo la actividad de abejas y abejorros permite el cuajado de frutos. Aunque la actividad de trips causó cierto nivel de polinización, los frutitos inicialmente cuajados cayeron pronto. Por el contrario, las flores embolsadas con papel de seda no adhirieron polen alguno y no cuajaron fruto. Por todo lo anterior se recomienda la introducción de colmenas para la polinización del aguacate en el SE español.

Palabras clave: polinización, abejas, abejorros, trips, anemofilia, cuajado de frutos.

INTRODUCTION

Avocado (*Persea americana* Mill.) is a fruit trees with a peculiar flowering process. This species has hermaphrodite flowers, but exhibits a sophisticated mechanism of dichogamy. Avocado flowers open twice, first time as female and second time as male. Between both phases, flowers close. This flower cycle, accomplished regularly in avocado native areas, promotes cross-pollination between cultivars that present complementary flower cycles (type A and B cultivars). In South East Spain, weather conditions during bloom often allow brief overlaps between female and male phases in a reduced proportion of flowers (Martínez et al, 1999, Cabezas, 2003). This situation may explain why farmers continue using solid orchards of 'Hass' with no pollinizers.

Avocado flower rewards attract different species of pollinators. In its native area, avocado is mainly pollinated by stingless bees (Gazit y Degani, 2002). In Spain, the proximity of many avocado orchards to undisturbed natural areas and greenhouses facilitates frequent visits of honeybees (*Apis mellifera*) and thrips (*Frankliniella occidentales*) (Cabezas, 2003). Some species of bumblebees (*Bombus terrestris* and *Bombus occidentalis*) have been also tested for avocado pollination because of their greater robustness (Neil and Pidduck, 2003), larger

working distance (Ish-Am et al, 1998) and positive effects on fruit size (Cuevas and Cabezas, 2005). A controversy about wind role in avocado pollination persists. Davenport (1998) indicates that in mild climate areas, such Florida, where an overlap between female and male floral phases occurs a chance for wind pollination exists. The argument is that stigma receptivity prolongation offers chances to autogamy (self-pollination) and geitonogamy (close-pollination), driven by wind. Davenport (1998) suggests that in these areas wind is the main pollinator while insects play a secondary role in avocado pollination.

This experiment was carried out with the aim of determining the importance of wind and insect visitors of avocado flowers in the processes of pollination and fruit set in cultivar 'Hass' cultivated in South East Spain.

MATERIAL Y METHODS

The experiments were carried out in a multivarietal plot located in the Experimental Station of Cajamar Foundation in El Ejido (Almería, Spain). Three pollination treatments were implemented: open pollinated flowers (unrestricted access), bagged flowers using micro-perforated plastic bags (where wind and small insect have access), and bagged flowers using tissue paper (where only wind may facilitate the transport of pollen although limited between enclosed flowers). Micro-perforated bags present 28 pores per cm² with a pore diameter of 400 µm, allowing windborne pollen to pass. Small sized insects as thrips also entered micro-perforated bags. Tissue paper bags were hermetic. Bagging was done before first flower opening (phenostage E) and kept until tepal withering (phenostage G) (Cabezas et al, 2003a).

Experimental design was randomized blocks were four trees acted as blocks and replications. Trees were selected at random among those with high level of flowering. No hives were introduced in the plot, although nearby greenhouses provided a number of bees and bumblebees (used as pollinators in vegetable crops) and thrips (plague of protected crops).

On these trees pollen adhesion and fruit set was determined. Pollen adhesion was estimated by the number of pollen grains per flower and the percentage of pollinated flowers in a random sample of 48 flowers per treatment collected at first (F1c) and second flower closure (F2c) (Cabezas et al, 2003a). Fruit set as the number of fruitlets per shoot in a simple of 8 shoot per tree was determined 35 (initial fruit set) and 70 days after bloom (final fruit set). Analyses of variance and mean separation using Duncan test was carried out.

RESULTS AND DISCUSSION

Partial or complete isolation of flowers from insects significantly diminished pollen adhesion (Table 1). Open pollinated flowers averaged 3 pollen grains per flower. These pollen grains germinated in a high proportion (Figure 1). Open pollinated flowers were visited by bees (*Apis mellifera*) (Figure 2a) and in a lesser extend by ants (*Formica* sp), houseflies (*Musca domestica*) and bumblebees (*Bombus terrestris*) (Figure 2b). Bagging with micro-perforated plastic limited the number of pollen grains on flower stigma (0.6 pollen grains per flower). In micro-perforated

bags was very common, however, to see active colonies of thrips (*Frankiniella occidentalis*) (Figure 3a). The complete isolation of flowers using tissue paper bags precluded insect pollination and no pollen grains were found in the stigmas of these flowers (Table 1). The hermeticity seems confirmed by the total absence of any kind of insect within the bags; only wind movement might have contributed to pollen transfer among flowers in these conditions.

The number of pollen grains adhered to the stigma was higher in the second than in the first flower closure. That was true in open pollinated flowers as well as in partial isolated flowers. This fact implies some level of pollination during male phase. The increase in pollen adhesion during male phase was proportionally greater in partial isolated flowers often visited by thrips. Cool weather conditions during blooming allow in our conditions a short overlap of female and male phases, specially at the beginning of the season when temperatures are low (Cabezas et al, 2003b). This overlap allows certain levels of self-pollination. Several authors have observed self-pollination during the male phase (Davenport, 1989; Sedgley, 1977), although this last author rejects the chance of fertilization in this scenario.

Flower isolation did not cause a significant effect on initial fruit set ($p=0.13$). Open pollination provided a higher number of fruitlets per shoot than bagged shoots did, although surprisingly shoots with restricted pollination set initially a high number of fruitlets (Table 1). All trees behave at this respect similarly, and no block effect was detected ($p=0.13$). Differences among treatments in final fruit set were, however, high and significant ($p<0.01$). The values indicate that only open pollination is able to guarantee adequate levels of final fruit set. In bagged shoots no one fruitlet reached the stage of final set (70 days after bloom). No effects of the block (tree) was detected on final fruit set ($p=0.45$). The contrasting response to pollination treatments when estimated at the stage of initial versus final fruit set suggest that early measurements of fruit set is not adequate for estimating yield in our weather conditions.

Therefore, only open pollinated flowers showed a proper level of pollen adhesion and of final fruit set. All shoots in which bee pollination took place set fruit; results that did not occur in bagged shoots. Several authors find avocado pollen grains too heavy and sticky for wind pollination, indicating that only medium-sized insects may successfully carry out pollen transport (Robbertse et al, 1998; Visscher and Sherman, 1998). The combined activity of bees and bumblebees seems to provide higher levels of pollen adhesion and germination (Ish-Am and Eisikowitch, 1998; Cuevas and Cabezas, 2005). In micro-perforated bags where colonies of thrips were seen on the flowers (Figure 3a) pollen adhesion occurred. However, the number of pollen grains was low therefore questioning the efficiency of thrips as avocado pollinators. Moreover, most of the fruitlets initially set in partial isolated flowers abscised before 70 days after bloom, and those that remained attached were commonly small and misshapen, being considered not commercial at harvest (Figura 3b). Hoddle (2001) thinks that thrips visit avocado flowers just for eating pollen, having no pollinator function. Furthermore, Lovatt (1990) has observed that the few pollen grains delivered by thrips result

damaged. This pollen may enhance the initial development of the ovary in avocado, but it is not capable of achieving fertilization. It is important underline that temperature and humidity within the bags used in this experiment do not significantly differ of the external ones (Del Río and Caballero, 1999).

Finally, our results also inform that the total isolation of the flowers from insects (tissue paper bags) completely impeded pollen transfer to the stigma of the flowers, and although an initial enlargement of the ovaries took place all fruitlets soon abscised. Sedgley (1980) also has observed an initial development of unfertilized fruitlets of cultivar 'Hass'. Flowers with non-functional ovules may also produce parthenocarpic fruits under some conditions due to special hormonal stimuli (Tomer et al, 1980). Certainly the situation within tissue paper bags do not reproduce closely wind pollination, since pollen movement is greatly limited to flowers enclosed in the same bag. However, the nil fruit set in micro-perforated bags (where wind may play a role) suggests a negligible importance of wind pollination in our growing condition, in spite that higher pollen adhesion after the second flower closure suggests some extension of stigma receptivity during the male phase (Table 1).

CONCLUSIONS

Avocado depends on insect pollination under our growing conditions. Bees (*Apis mellifera* L.) and bumblebees are the main pollinators of avocado in South East Spain. The activity of these insects allowed high levels of pollen adhesion and fruit set. Short distance pollen transport is achieved by *Frankliniella occidentalis*, although its efficiency resulted low and did not allow fruit set to take place. Enclosed flowers where no insect had access did not present any pollen grain adhered to the stigmas, suggesting a negligible importance of wind pollination for avocado orchards in Spain.

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Table 1. Pollen adhesion and fruit set in open pollinated flowers and partial and completely isolated flowers bagged with micro-perforated and tissue papers bags, respectively.

Treatment	Pollinated		Pollen grains per flower	Initial fruit set (fruits/shoot)	Final fruit set (fruits/shoot)
	flowers (%)				
	♀ F _{3f}	♂ F _{4m}			
Open pollinated flowers	42	54	3.0	47.2	1.5
Partially isolated flowers	20	32	0.6	25.1	0.0
Completely isolated flowers	0	0	0.0	33.0	0.0

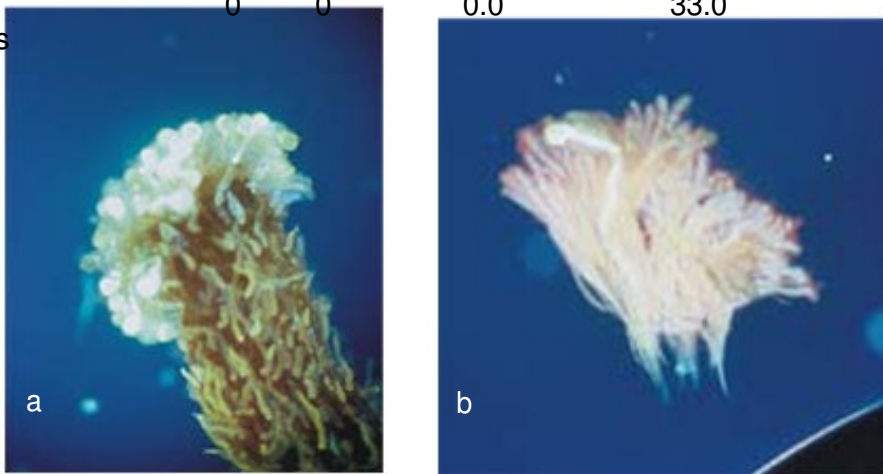


Figure 1. (a) Pollen germination in open pollinated flower (b) Pollen tube growth.



Figure 2. Bees (a) and bumblebees (b) visiting open pollinated flowers. Observe pollen load in corbiculae.



Figure 3. Partial isolated flowers. (a) Active colonies of thrips (*Frankiniella occidentalis*). (b) Small misshapen fruits resulting from thrip activity.