A STUDY OF SELF-POLLINATION AND CROSS-POLLINATION IN AVOCADO (*PERSEA AMERICANA* MILL.) CV. HASS OF DIFFERENT VARIETIES.

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Abstract

The following study was carried out at "La Palma" Experimental Center of the Faculty of Agronomy of the Catholic University of Valparaiso, for the evaluation of the influence of different avocado varieties as pollinators of Hass. A Hass test field (planted 10x10m.) was used, containing sections with different pollinators planted in 1975. Pollinators are distributed in quincunce in the Hass planting, belonging to the Bacon, Edranol, Hass, Rincon and Zutano varieties. The control was a section with only Hass trees planted without pollinators. The aim was to determine for each combination and the control the percentages of self-pollination and cross-pollination in the descendants of the Hass fruits by analyzing the origin of parental pollen in fruit embryos using isoenzymes as genetic markers; thereby enabling a determination of which varieties behave better as pollinators of Hass under local conditions in the Quillota area. Electrophoresis test in starch gels at 11% concentrations were carried out using 14 isoenzymatic systems of GOT-1, GOT-2, PGI-2, PGM-1, PGM-2, LAP-1, LAP-2, EST-FL, MDIL, TPI-2, SKDH-1, SKDH-2, 6-PGD-2, 6-PGD-3 and 6-PGD-4. The analyzed fruit was collected from October 1994 to January 1995. The results showed Hass to be weak self-pollinator, being easily displaced by pollen from other parental plant when found associated to other varieties. The pollinators which provided the highest cross-pollination results in their combinations were Zutano, Edranol and Bacon cultivars, presenting no significant differences between them. Rincon variety proved a weak pollinator for Hass.

1.- Introduction

Avocados have many production problems, generally related to pollination, biannual production and fruit set problems, for that reason alternatives need to be found which would allow to improve or alleviate these issues, thereby attaining larger quantities of good-quality fruit at harvest.

Pollination problems are due to the avocado's particular flowering pattern, and despite complete flowers being present the phenomenon of dicogamia protoginea occurs where the sex verticils ripen out of time, thereby lessening the chances for self-pollination. Added to this is the fact of having two flowering patterns, A and B, which open at different times, and which possibly need to be complementary for adequate pollination.

For this reason, for a long time the need to in between trees which are complementary in their flower-type has been proposed, thereby achieving the maximum pollination and resulting a high level fruit production. However, it such behavior is not always observed because is strongly
influenced by climatic conditions. It is known that low temperatures cause delay in blossoming (Sedgley and Grant, 1983), or even the A and B patterns are not followed (Calvert, 1993).

It is necessary to evaluate what influence different pollinator trees have on the Hass tree and analyze whether there is a better pollinating variety for the cv. Hass, than the Hass itself. For this purpose use was made of one of the orchards belonging to the Faculty of Agronomy of the Catholic University of Valparaiso, where there are different fields of Hass in combination with other varieties.

The results of fruit production by Hass tress in these different combinations, in studies carried out several seasons ago, did not show great differences between them. This indicates that either there is no reason to use pollinators in the case of Hass, thereby allowing to plant dense orchards of this variety alone, and producing significant crops, as affirmed by Gazit (1977) and Hodgson (1947), or, that due to the proximity of other fields, there was probably significant cross-pollination.

The aim of this study was to determine the origin of the parental pollen fertilizing the Hass flowers, in each of the different combinations, through characterization of the isoenzymes present in the fruit embryo.

2. Material and methods

The study was carried out in one of the orchards at the "La Palma" Experimental Station at the Catholic University of Valparaiso, in Quillota Province, Chile (32° 50’ latitude south and 71° 13’ longitude west).

This orchard was planted in 1975, with a separation of 5x5 m except for the control Hass which was left finally at a separation of 10x10m. Later the diagonals were thinned, so at present the Hass trees are at a 10x10 m separation, with the pollinator at the center (quincuncial). The fields, planted at random with three repetitions, have the following combinations:

<table>
<thead>
<tr>
<th>Hass with Bacon</th>
<th>Hass with Hass</th>
<th>Hass with Zutano</th>
<th>Hass with Rincon</th>
<th>Hass with Edranol</th>
<th>Control Hass</th>
</tr>
</thead>
<tbody>
<tr>
<td>x x x x x</td>
<td>o o o o</td>
<td>x x x x</td>
<td>o o o o</td>
<td>x x x x</td>
<td>x x x x</td>
</tr>
</tbody>
</table>

x: Hass  
o: Pollinator

Each field at present has 32 trees (except the control field which has only 20), of which 20 correspond to Hass and 12 to the pollinating tree. The sampling material was obtained by selecting three of the six central Hass trees in each field to avoid, as far as possible, the influence of the neighboring fields.

Electrophoresis in starch gel was carried out between the months of October and November 1994, taking fruit of approximately 12 to 13 months of age. 15 fruits were collected per tree, choosing 5 large fruits (between 205 and 245g.), 5 medium fruits (173 to 204g.) and 5 small fruits (150 to 172g.): 45 fruits per field, 13 5 per treatment and 8 10 in total.

Nine enzymes with 15 isoenzymatic systems were analyzed in the samples, in order to be able to differentiate fruit-parentage as completely as possible. The enzymes which were analyzed
were: GOT-1, GOT-2; PGI-2; PGM-1, PGM-2; LAP-1, LAP-2; EST-FL; MDH; TPI-2; SKDH-1, SKDH-2; 6-PGO-2, 6-PGO-3 and 6-PGD-4.

3. Results

To determine the parent supplying the pollen of the progeny, it is the best to use isoenzymes whose phenotype is homozigote and different between parents, due to the fact that in this case all progeny can be differentiated. However this situation did not occur and isoenzymes had to be chosen which had heterozigote phenotypes, with which only one quarter or half of the descendants could be differentiated, but nevertheless it was possible to mark out possible parents in certain particular cases.

A small number of fruits could not be analyzed, some of large sizes, but mainly small ones whose embryos, without showing apparent symptoms of being dead or of being altered in any way, displayed a minimum of intensity in the electrophoresis dyes, or a total absence of enzymatic activity.

The results, showed a relatively high number (38%) of cases where it was not possible to identify a single pollen donor, due to the fact that its gene types did not provide enough information to permit discrimination from the other possible parents. Ruling out these doubtful cases could lead to a distortion of the real values of cross-pollination although in the full study each of the situations was analyzed. On proving that there are no substantial changes between the two analyses, we have preferred to give the results considering that when there are two possible parents the probability of occurrence was distributed equally between them, 50-50, and in the case of three possible parents the probability was distributed 1/3 to each one.

Analyzing the percentages of cross-pollination between treatments, it can be seen that the percentage of cross-pollination is extremely high in the different fields, with significant differences between them (Figure 1). These results show that in the fields there was at least 64.1% cross-pollination, corresponding to the treatment Hass/Hass, i.e., only 35.9% of the fruit produced by Hass trees were pollinated with pollen from this same variety.

Fields with pollinators in the quincuncial, such as Rincon, Bacon, Edranol an Zutano, display significant differences from the Hass/Hass treatment, and indicate between 84.5% and 97.4% cross-pollination. The control Hass is at an intermediate point, displaying significant differences only from the Hass/Zutano treatment.
Avocado orchards in Chile, including the orchard under study, are managed with bees, with between 3 and 6 hives placed in each hectare. It is, therefore, not surprising to find these results, bearing in mind the large radius of action these pollinating insects exert in free-pollination orchards, and taking into account the scant separation between the different fields.

Analysis of cross-pollination between each treatment (Table 1), shows that there is an interaction between different pollen-donating parents. In the treatments Hass/Control, Hass/Hass, and Hass/Rincon, the Zutano, Edranol and Bacon parent-trees participated strongly. On the other hand when these parent trees are present in the fields they themselves dominate strongly.

Table 1 - Interaction between different pollen-donating parents

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Hass</th>
<th>Pollen Parents</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Zutano</td>
<td>Rincon</td>
</tr>
<tr>
<td>H C</td>
<td>21.6</td>
<td>16.8</td>
</tr>
<tr>
<td>H/R</td>
<td>35.5</td>
<td>11.6</td>
</tr>
<tr>
<td>H/R</td>
<td>15.3</td>
<td>30.8</td>
</tr>
<tr>
<td>H/E</td>
<td>7.7</td>
<td>2.2</td>
</tr>
<tr>
<td>H/Z</td>
<td>2.6</td>
<td>74.1</td>
</tr>
<tr>
<td>H/B</td>
<td>16.3</td>
<td>12.1</td>
</tr>
</tbody>
</table>

The weakness of Hass as a self-pollinator has already been reported by Guil and Gazit (1992). There may be several explanations: first, Hass may have some problems with its pollen which makes self-pollination difficult; second, climatic conditions may not favor a good overlap of blossom states, thereby favoring cross-pollination; third, the genetic material which the Hass plant passes on to its descendants is in a disadvantage compared with other pollen donors, as far as it allows a high survival rate of small fruits.

The percentage of cross-pollination for each treatment, and by sizes, shows that in the total number of fruits, the Zutano, Edranol and Bacon varieties are statistically the same, but they differ from Hass, which in its turn differs from Rincon (Table 2).

Table 2 - Sizes distribution of Varieties

<table>
<thead>
<tr>
<th>Size Variety</th>
<th>Large</th>
<th>Medium</th>
<th>Small</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hass</td>
<td>36 b</td>
<td>44 bc</td>
<td>52 b</td>
<td>132 b</td>
</tr>
<tr>
<td>Zutano</td>
<td>78 c</td>
<td>58 c</td>
<td>55 b</td>
<td>191 c</td>
</tr>
<tr>
<td>Edranol</td>
<td>68 c</td>
<td>70 d</td>
<td>81 c</td>
<td>219 c</td>
</tr>
<tr>
<td>Rincon</td>
<td>11 a</td>
<td>13 a</td>
<td>5 a</td>
<td>29 a</td>
</tr>
<tr>
<td>Bacon</td>
<td>69 c</td>
<td>73 d</td>
<td>77 c</td>
<td>229 c</td>
</tr>
</tbody>
</table>

Total        | 262 | 259 | 270 | 791 |

Test: $\chi^2$ square, 5% 

It can be seen that Rincon has a small share as much in the different treatments as in sizes. It should not be forgotten that this is the only cultivar which has a type-A flower, like the Hass variety which is the one to be pollinated. However, if Hass did self-pollinate perhaps this variety's problem lies in the quality of pollen, as suggested by Degani and Gazit (1984).
All the other varieties have type-B flowers, which means that their blossom states are complementary with Hass, and perhaps with better pollen "quality", possibly implying a greater fruit set and a higher survival rate of fruits up to maturity. For that reason it can be deduced that the genotype of the embryo plays a very important role in differential-selection for fruit survival.

References