top variety. Although the present experiment is
limited to the use of Rough lemon and sour
orange rootstocks and interstocks, the results
indicate that the rootstock was the controlling
factor influencing tree growth, yields, and fruit
quality and that the interstock did not alter the
rootstock influence in any of these respects. It
should be obvious, however, that a change of
top variety would be disastrous if the new top
introduced a virus into a combination in which
the interstock or rootstock were intolerant of
the introduced virus.

ACKNOWLEDGMENT

The author is grateful to E. James Koch of
the Biometrical Services, ARS, Plant Industry
Station, Beltsville, Maryland, for the statistical
analyses herein.

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EFFECTS OF POLLEN SOURCE ON FRUIT
CHARACTERISTICS AND SET OF FOUR CITRUS HYBRIDS

C. J. HEARN, P. C. REECE AND R. FENTON
Crops Research Division,
Agricultural Research Service,
U. S. Department of Agriculture,
Orlando

ABSTRACT

The citrus hybrids, 'Robinson', 'Lee', 'Page',
and 'Nova' were released during the last 10
years by the USDA. The 'Orlando' and 'Clem-
entine', parents of these hybrids, are commonly
unfruitful when planted in solid blocks. This
suggests that their hybrids might perform like-
wise. Each hybrid was found to be self-incom-
patible when self-pollinated. Only Page set a
significant amount of fruit without cross-pollina-
tion. Robinson produced only a limited amount
of viable pollen but the other hybrids produced
good quantities of viable pollen.

When cross-pollinated the effects on fruit
set, seed content and fruit diameter were sig-
ificant. Lee, Orlando, and 'Temple' generally
were the most effective pollinizers of the varie-
ties tried. The Lee pollen had a metaxenia effect
on Page fruit.

INTRODUCTION

Self-incompatibility has been reported as
characteristic of 'Clementine' (Citrus reticulata
Blanco) tangerine (4); and 'Orlando' (C. para-
disi Macf. X C. reticulata) (2); and 'Minneola' (C. para-
disi X C. reticulata) (5) tangelos.

Robinson and Nova varieties are hybrids of
Clementine X Orlando (7) (10) and Page is a
hybrid of Minneola X Clementine (9). When
these varieties are released, growers were ad-
vised that they might require cross-pollination
in order to set good crops of fruit. That Rob-
inson requires cross-pollination was shown by
previously reported experiments (8).

Experiments were designed to determine
whether Lee, Nova and Page require cross-
pollination, and also whether the pollen of dif-
ferent varieties might affect their fruit char-
acteristics. Although the crop and fruit size
may be increased, cross-pollination usually in-
creases seediness. A linear relationship be-
tween fruit size and the number of seeds has
been reported in 'Valencia' [C. sinensis (L.)
Osbeck] (1) and Orlando (8).
Material and Methods

Bearing trees of Lee, Nova, and Page were individually caged with honey bees to determine whether these hybrids are fruitful when self-pollinated. In another experiment the flowers of Lee, Robinson, Nova and Page were emasculated and hand pollinated with pollen of other citrus varieties. To substantiate unexpected results, Page pollinations were repeated. The fruits were harvested when mature, then the diameters were measured and the seeds per fruit counted; finally, the percentage of fruit set was calculated. The correlation between fruit diameter and number of seeds was analyzed statistically by comparing two sample means with unpaired observations and by using the “t” test (12).

Pollen germination in 20 percent sucrose and 1 percent Bacto-agar at room temperature was studied. The pollen was obtained by dehiscing the anthers under an incandescent lamp. Germination was observed over a 36-hour period using the hanging drop method.

Table 1. The average percent germination of pollen of six citrus varieties in 1968

<table>
<thead>
<tr>
<th>Pollen Variety</th>
<th>Average percent germination</th>
</tr>
</thead>
<tbody>
<tr>
<td>Page</td>
<td>45.0 a/</td>
</tr>
<tr>
<td>Temple</td>
<td>40.3 a</td>
</tr>
<tr>
<td>Orlando</td>
<td>39.1 a</td>
</tr>
<tr>
<td>Lee</td>
<td>38.1 ab</td>
</tr>
<tr>
<td>Nova</td>
<td>30.2 b</td>
</tr>
<tr>
<td>Robinson</td>
<td>6.7 c</td>
</tr>
</tbody>
</table>

1/ Any two means not followed by the same letter are significantly different at the .01 level.
2/ Germination percentages are the average of 1800 pollen grains germinated in 36 hours.

Result and Discussion

Page, Temple, Orlando and Lee pollen germination percentages were of about the same order (45.0, 43.3, 39.1 and 38.1 respectively) whereas Nova pollen gave poorer germination (30.2%). In these experiments Robinson produced a limited quantity of inferior pollen (6.7% germination). On the basis of these results, Robinson would be an unsatisfactory pollinizer (Table 1).

ROBINSON

On Robinson tangerine, pollen of Orlando and Lee tangelos induced the largest size fruit, whereas Temple pollen induced the largest percent of fruit set (Table 2). The large set of fruit produced by Temple pollen resulted in smaller fruit. Robinson fruits resulting from Lee pollen contained fewer seeds than those pollinated from Orlando and Temple. Fewer fruit were set, and fewer seeds resulted, from Page pollen. This suggests that Page would be inferior to Temple as a pollinizer. Nova pollen, likewise, gave poor results. The poorer results with Page and Nova pollens may be due to cross-incompatibility in some degree. The presence of genetically controlled cross-incompatibility has been reported (11). In the field, Orlando and Temple have been satisfactory pollinizers of Robinson.

NOVA

In these experiments Nova set few fruits without cross-pollination (Table 2), a factor which suggests that Nova is self-incompatible.

Table 2. Effect of pollen from different varieties on fruit size, seed number and fruit set of Robinson and Nova

<table>
<thead>
<tr>
<th>Pollen Variety</th>
<th>ROBINSON 1967</th>
<th>NOVA 1966</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Avg. Fruit Dia. cm</td>
<td>Avg. Seeds Per Fruit</td>
</tr>
<tr>
<td>Lee</td>
<td>6.35 a/</td>
<td>20.2 c</td>
</tr>
<tr>
<td>Orlando</td>
<td>6.35 a</td>
<td>23.2 b</td>
</tr>
<tr>
<td>Temple</td>
<td>6.20 b</td>
<td>25.2 a</td>
</tr>
<tr>
<td>Page</td>
<td>6.00 c</td>
<td>9.7 d</td>
</tr>
<tr>
<td>Nova</td>
<td>5.90 c</td>
<td>4.8 e</td>
</tr>
</tbody>
</table>

1/ Any two means not followed by the same letter are significantly different at the .01 level, except those with letter followed by an asterisk are significantly different at the .05 level.
2/ Few fruits set in the absence of cross-pollination.
Nova set the largest fruits when Orlando pollen was used. However, the number of seeds per fruit and the percent fruit set was the same when Orlando, Lee or Temple pollen was used. Nova tangelo set fewer fruits from Page pollen and these fruits contained fewer seeds than those set from other pollens.

**LEE**

Lee tangelo set few fruits without cross-pollination, and those that set were seedless (Table 3). Lee pollen is viable and effective on other varieties, suggesting that Lee is self-incompatible. Orlando and Page pollens were equally effective in setting fruit of Lee. Although fruit size from Nova pollen was good, the lower percentage fruit set (as compared to pollen from Page and Orlando) indicates that these varieties would be more effective pollinizers.

**PAGE**

Page tangelo trees caged with bees in 1966 set a good crop of parthenocarpic fruit, but were very small (Table 4).

Page set more and larger fruit when pollinated with Lee pollen than with Temple or Orlando pollen. The average number of seeds per fruit induced by Lee, Orlando and Temple pollen differed slightly. Regression analyses (not illustrated) showed that a positive linear relationship existed between the number of seeds and the fruit size when a single pollen source was used. These results agree with work previously reported (1) (8). A possible cross-incompatibility between Page and Robinson, and between Page and Nova is shown by the fact that few or no seeds resulted when Page flowers were pollinated by those two varieties (Table 4).

Fruit size was approximately equal when Page was pollinated with Orlando or Lee pollen (Table 4). However, Orlando pollen induced larger fruits than Temple, although fruits resulting from Temple pollen contained more seeds. In 1967, the performance of Lee and Nova pollens may have been hampered by poor condition of the pollen. In both years, Nova was a poor pollinizer of Page. Page fruits set from Lee pollen in 1967 contained the smallest number of seeds, but the average fruit size was

---

**Table 3. Comparison of fruit size and number of seeds when Lee tangelo was self-pollinated and when cross-pollinated with Nova, Orlando and Page pollen**

<table>
<thead>
<tr>
<th>Pollen Variety</th>
<th>Avg. Fruit Dia. cm</th>
<th>Avg. Seeds Per Fruit</th>
<th>No. of Flowers</th>
<th>No. of Fruit Set</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nova</td>
<td>6.93 a*</td>
<td>25.1 b</td>
<td>1005</td>
<td>32</td>
</tr>
<tr>
<td>Page</td>
<td>6.60 ab</td>
<td>26.6 ab</td>
<td>403</td>
<td>55</td>
</tr>
<tr>
<td>Orlando</td>
<td>6.55 b</td>
<td>29.8 a</td>
<td>572</td>
<td>67</td>
</tr>
<tr>
<td>Lee*</td>
<td>6.38 b</td>
<td>0</td>
<td>-</td>
<td>25</td>
</tr>
</tbody>
</table>

1/ Any two means not followed by the same letter are significantly different at the .01 level, except those with letter followed by an asterisk which are different at the .05 level.

2/ A Lee tree was caged with honey bees to obtain self-pollination. Few fruits set in the absence of cross-pollination.

**Table 4. Effect of pollen from different varieties on fruit size, seed number and fruit set of Page**

<table>
<thead>
<tr>
<th>Pollen Variety</th>
<th>Avg. Fruit Dia. cm</th>
<th>Avg. Seeds Per Fruit</th>
<th>Percent Fruit Set</th>
<th>Avg. Fruit Dia. cm</th>
<th>Avg. Seeds Per Fruit</th>
<th>Percent Fruit Set</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lee</td>
<td>6.60 ab</td>
<td>23.3 b</td>
<td>40.6</td>
<td>6.50 ab</td>
<td>9.7 d</td>
<td>13.62</td>
</tr>
<tr>
<td>Temple</td>
<td>6.17 b</td>
<td>24.8 a</td>
<td>19.0</td>
<td>6.45 b</td>
<td>24.0 a</td>
<td>17.1</td>
</tr>
<tr>
<td>Orlando</td>
<td>6.17 b</td>
<td>19.8 c</td>
<td>26.5</td>
<td>6.65 a*</td>
<td>22.2 b</td>
<td>23.6</td>
</tr>
<tr>
<td>Robinson</td>
<td>5.90 c</td>
<td>0</td>
<td>11.1</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Nova</td>
<td>5.57 d</td>
<td>1.0 d</td>
<td>25.6</td>
<td>6.20</td>
<td>2.2</td>
<td>1.02</td>
</tr>
<tr>
<td>Page</td>
<td>5.62 d</td>
<td>0</td>
<td>- 3/</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

1/ Any two means not followed by the same letter are significantly different at the .01 level, except those with letter followed by an asterisk which are significantly different at the .05 level.

2/ It is suspected that Nova and Lee pollens had become partially inviable before pollination.

3/ A good crop of fruit was set parthenocarpically.
among the largest set by the three effective pollen varieties.

Page fruits set by Lee pollen and containing the same number of seeds were larger than fruits set by Orlando and Temple pollens in both 1966 and 1967 (Table 5). This suggests a metaxenia effect. Metaxenia in Citrus was first reported in Greece (6) in 1957. These experiments showed that the pollen varieties influenced the shape, color, reaping and quality of the fruit.

The comparative effectiveness of pollinizer varieties when used in combination with Robinson, Lee, Page and Nova is shown in Table 6. Temple, Orlando and Lee as pollinizers were effective in all cross-pollinations tested. Page was less effective than the above mentioned pollinizer varieties, whereas Nova and Robinson generally were ineffective as pollinizers.

Since cross-pollination of citrus is dependent upon insects it is essential that honey bees be present to assure satisfactory fruit set.

Results of cross-pollinations are based on hand pollination. In a large commercial grove the results might differ somewhat.

### SUMMARY

Pollen germination studies revealed that Robinson produces a limited quantity of pollen which is of poor quality and, therefore, is a poor pollen source. Hand pollination tests further showed Robinson to produce poor pollen.

Results of controlled pollinations of Robinson, Lee, Nova and Page showed that each is self-incompatible; Page, however, did set considerable parthenocarpic fruit.

Lee and Orlando appeared to be the most effective pollinizers for Robinson, but Temple

<table>
<thead>
<tr>
<th>Pollen Variety</th>
<th>Pollinated</th>
<th>Lee</th>
<th>Orlando</th>
<th>Page</th>
<th>Nova</th>
<th>Robinson</th>
</tr>
</thead>
<tbody>
<tr>
<td>Robinson</td>
<td>++/</td>
<td>++</td>
<td>++</td>
<td>42/-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Lee</td>
<td>++/</td>
<td>++</td>
<td>-3/</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Page</td>
<td>++/</td>
<td>++</td>
<td>+45/</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Nova</td>
<td>++/</td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
</tbody>
</table>

1 = good combination
2 = fair combination
3 = poor combination
4 = untried combination but probably good
5 = Lee pollen appears to lead to larger Page fruits than other pollen varieties

was nearly as good. The most effective pollinizers for Lee were Orlando and Page.

On Nova, Orlando pollen gave the best results, followed closely by that from Temple, Lee, and Page.

The largest Page fruit that we produced from hand pollinations in 1966 resulted from Lee pollen, and the next largest resulted from Orlando and Temple. In 1967, Page fruit pollinated by Lee or by Orlando were the same size. Page fruits set from Temple pollen were smaller than those from Orlando pollen. Regardless of pollen source, there was a positive linear relationship between fruit size and number of seeds per fruit. On the basis of fruit size and seed number, Lee would be considered the best pollinizer for Page.

Page fruits resulting from Lee pollen were larger than those with the same number of seeds resulting from other pollen varieties. This size increase was apparently due to a metaxenia effect of the Lee pollen. Nova and Robinson were poor pollinizers of Page, perhaps because of cross-incompatibility.

### LITERATURE CITED


### Table 5. Comparative average diameter of Page fruit set from hand pollination with Lee, Orlando and Temple pollens grouped into classes according to number of seeds per fruit in 1966 and 1967

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>(cm)</td>
<td>(cm)</td>
<td>(cm)</td>
<td>(cm)</td>
<td>(cm)</td>
<td>(cm)</td>
</tr>
<tr>
<td>5-8</td>
<td>5.04/-</td>
<td>6.45</td>
<td>-</td>
<td>6.45</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>9-12</td>
<td>6.35</td>
<td>5.63</td>
<td>-</td>
<td>6.27</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>13-16</td>
<td>6.25</td>
<td>5.92</td>
<td>-</td>
<td>6.35</td>
<td>5.95</td>
<td>5.84</td>
<td>-</td>
</tr>
<tr>
<td>17-20</td>
<td>6.35</td>
<td>-</td>
<td>6.15</td>
<td>6.48</td>
<td>5.95</td>
<td>6.12</td>
<td>-</td>
</tr>
<tr>
<td>21-24</td>
<td>6.63</td>
<td>6.86</td>
<td>6.28</td>
<td>6.63</td>
<td>6.03</td>
<td>6.32</td>
<td>-</td>
</tr>
<tr>
<td>25-28</td>
<td>6.78</td>
<td>6.35</td>
<td>6.83</td>
<td>6.30</td>
<td>6.98</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>29-32</td>
<td>6.83</td>
<td>-</td>
<td>6.65</td>
<td>7.21</td>
<td>6.38</td>
<td>6.99</td>
<td>-</td>
</tr>
<tr>
<td>Avg. No. of seeds</td>
<td>23.3</td>
<td>9.7</td>
<td>19.8</td>
<td>22.2</td>
<td>24.8</td>
<td>24.0</td>
<td>-</td>
</tr>
<tr>
<td>Avg. fruit dia. cm</td>
<td>6.60</td>
<td>6.30</td>
<td>6.17</td>
<td>6.65</td>
<td>6.17</td>
<td>6.43</td>
<td>-</td>
</tr>
</tbody>
</table>

1/ Few or no fruits occurred in seed number groups left blank.
INFLUENCE OF 2,4-D ON GLUCOSE METABOLISM OF CITRUS LEAVES

F. J. Leal and R. H. Biggs

Abstract

Since 'Pineapple' and 'Valencia' oranges have a different organ-abscission response to 2,4-dichlorophenoxyacetic acid (2,4-D), investigations were initiated to determine the underlying cause for this difference. A comparison of the influence of 2,4-D on respiration and glucose metabolism of the two citrus cultivars was made by measuring O₂ uptake, and the ratio of ¹⁴CO₂ evolving from tissues metabolizing glucose-1-¹⁴C (g-1-¹⁴C) and glucose-6-¹⁴C (g-6-¹⁴C).

The O₂ uptake of 'Pineapple' leaves was increased nearly two-fold by 2,4-D treatment, whereas, with identical treatments, the respiration of 'Valencia' leaves was not significantly affected. From the ratio of g-6-¹⁴C to g-1-¹⁴C metabolized by 'Pineapple' and 'Valencia' tissues, there was evidence that both the regular glycolytic (EMP) and the pentose phosphate pathways (PPP) were operative in healthy, fully mature leaves. 'Valencia' leaves had the most pronounced PPP as evidenced by a ratio of 0.1 as compared to 0.5 for 'Pineapple' leaves during the first 60 minutes after detachment. The ratio increased with time after detachment of the leaves of 'Valencia' but remained about 0.5 for the duration of measurements for 'Pineapple' leaves. There was evidence that 2,4-D slightly increased the ratio with 'Pineapple' immediately after addition of the chemical.

Introduction

Abscission of citrus organs is a serious horticultural problem from several standpoints. If it occurs at the wrong time, overthinning of young fruits and pre-harvest drop occurs; on the other hand, if abscission is prevented, there are problems of too many fruits per tree and of harvesting the fruits.

Research on agents that influence pre-harvest drop of citrus has demonstrated that auxin would delay abscission of fruits on certain varieties (5,6,7,9,12,16,19,20) and not on others (17, 18). Moreover, the response of a particular cultivar varied with the geographical location (17,18) and from year to year (18). Recently, it has been shown with tests of 2,4-D at low concentration on explants of several cultivars that they all responded to 2,4-D, even those previously found to respond very little to field application of 2,4-D (21). The difference in the response to 2,4-D could be due to a difference in uptake, translocation, metabolism of 2,4-D per se, and/or tissue responses to 2,4-D.

As a part of a project to determine the patterns of uptake, translocation, and metabolism of 2,4-D in cultivars of citrus susceptible and non-susceptible to inhibition of the abscission processes, the present report will demonstrate that 2,4-D does alter the metabolism of a susceptible cultivar to a much greater extent than a non-susceptible cultivar under certain conditions. The indicators of metabolism measured were O₂ uptake, CO₂ evolving from tissues metabolizing G-1-¹⁴C and G-6-¹⁴C. Previously, it has been shown that 2,4-D will stimulate respiration (8) (11) (13) and that 2,4-D caused an increase in the amount of glucose catabolized via PPP (8) (11).

Materials and Methods

Plant material was 'Pineapple' and 'Valencia' orange leaves from 8-year-old trees growing in experimental plantings at the University of Florida. Care was exercised in obtaining healthy, fully mature leaves of approximately the same...