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Differentiation Between Nucellar and Zygotic Citrus Seedlings by Leaf Shape[1]

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Summary. Leaf length divided by leaf width, (L/W), was studied as a criterion for differentiating between nucellar and zygotic citrus seedlings. Progeny of crosses of the variety 'King' as a seedparent (known to produce both nucellar and zygotic seedlings) fell into two classes: one with L/W similar to 'King' and the other midway between 'King' and the pollen parent. In several zygotic seedling populations, population means were intermediate between those of the parents. L/W can be an effective criterion for differentiating between nucellar and zygotic citrus seedlings where the parents differ appreciably.

Introduction

This study evaluated leaf length divided by width, (L/W), as a criterion for differentiating between nucellar and zygotic seedlings from polyembryonic citrus seed parents. Many citrus varieties produce seed containing both sexual and asexual embryos. Asexual embryos develop from nucelli, accompanying or replacing zygotic embryos. This results in two seedling populations — one zygotic, useful in breeding, and the other nucellar (maternal genotype) which is a useless burden in progeny tests. Nucellar seedlings are a major problem in citrus breeding (Cameron and Soost 1969).

If one parent in a desired cross produces only zygotic seedlings it can be used as the seed parent, avoiding the production of nucellar seedlings. Where both parents produce a proportion of nucellar seedlings, a criterion for distinguishing between nucellar and zygotic seedlings is required.

Leaf shape is a common criterion in distinguishing genotypes on a taxonomic scale, and because L/W is an objective measure of leaf shape, subject to evaluation for reliability, is was chosen for this study.

As nucellar seedlings have the maternal genotype, their leaf shape must vary around the maternal leaf shape mean. Zygotic leaf shape, assuming it is controlled mainly by genes having additive effects, will vary around the midpoint between the parents. Differentiation between nucellar and zygotic seedlings should be feasible where parental L/W's differ sufficiently, and where the influence of environmental heterogeneity can be adequately controlled. Support for this hypothesis would be provided by a bi-modal seedlings L/W distribution from a polyembryonic seed parent, one mode of the nucellar seedling population and the other of the zygotic seedling population.

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Materials and Methods

Leaves of varieties and seedlings from their crosses were measured (Table 1, 2).

Variety leaves were sampled from mature, grafted trees growing at Sarafand, using the ten outermost leaves of three branches of four trees per variety.

Table 1. *Leaf shape-length/width of citrus varieties at Sarafand*

Variety	Mean	Standard deviation	Polyembryonic
Avana	2.87	0.08	P
Clementine	2.57	0.13	—
Dancy	2.18	0.06	P
King	1.93	0.02	P
Orlando	1.95	0.02	P
Satsuma	2.36	0.10	P
Shamouti	1.98	0.08	P
Wilking	2.56	0.10	—

Table 2. *Leaf shape L/W of zygotic seedlings*

Parents		Mean of zygotes	Mean of parents
Seed	Pollen		
Clementine	Dancy	2.05	2.38
Clementine	Shamouti	2.15	2.27
Clementine	Wilking	2.54	2.56
Temple	Wilking	2.26	2.33

Seedling leaves were taken from one-year-old greenhouse seedlings. For each cross, ten leaves from each of ten seedlings were samples, but in crosses of 'King' as a seedparent with 'Avana' and 'Clementine', 100 seedlings were sampled to provide an adequate description of seedling population distribution. 'King' is known to produce both nucellar and zygotic seedlings (Cameron and Soost 1969). Crosses of seed parents which produce only zygotic seedlings were included in the study to test the assumption that the zygotic mean L/W is approximately midway between that of the parents.

Each leaf blade was measured along its midrib, and at right angles to its midrib at the widest point of the leaf blade, to the nearest mm. Curved leaves were measured along the arc, but grossly distorted leaves were not sampled.

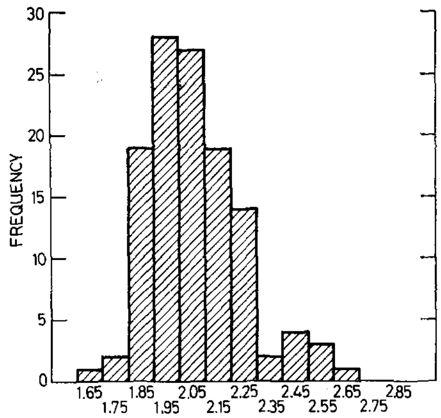
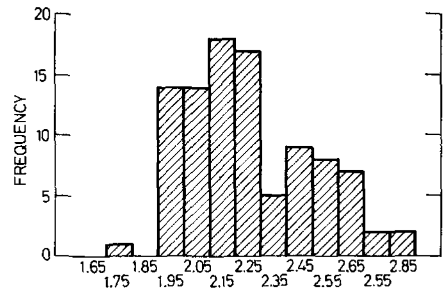


Fig. 1. Distribution of leaf length/width of seedlings from the variety 'King' crossed with 'Avana'

Fig. 2. Distribution of leaf length/width of seedlings from the variety 'King' crossed with 'Clementine'



Results and Discussion

'King' seedlings' L/W from crosses with 'Avana' and 'Clementine' fell into bi-modal distributions. One mode of 'King' × 'Avana' seedlings was in the 1.90 to 1.99 class. Since 'King's L/W was 1.93, the population represented by this mode may very likely be the nucellar population. The other mode, 2.40 to 2.49, is comparable to the mean of both parents, 2.40, and hence is probably the mode of the zygotic distribution. In the 'King' × 'Clementine' cross the mode most similar to that of 'King' was in the range 2.10 to 2.19, only slightly different from 'King', and the other mode, 2.40 to 2.49, was just a little higher than the mean of both parents, 2.25. The observations on the two classes, one apparently nucellar and the other apparently zygotic, support the hypothesis that L/W is an effective criterion for differentiating between nucellar and zygotic seedlings.

The apparently clear division between the zygotic and nucellar seedlings was enhanced by the relatively large difference in L/W of the parents in each cross, ca. 0.6. 'King' has a relatively wide leaf, while both 'Avana' and 'Clementine' have narrow leaves. Differentiation in crosses of parents a little more similar in L/W is probably possible if some error in identification is acceptable: error in rejectiing a zygote as a nucellar, or error in accepting a nucellar as a zygote. This type of error should be acceptable considering the ease of measurements vs. the expense of carrying a large number of nucellar seedlings in progeny tests. Where the number of seeds per pollination and the proportion of zygotic seedlings is low, it would usually be preferable to accept the error of including even the doubtful zygotes, while where the number of seeds per pollination and proportion of zygotic seedlings is high it would be preferable to reject the doubtful zygotes. Seed yield per cross and proportion of zygotes are also considerations in judging whether a specific cross is feasible in conjunction with seedling differentiation.

In crosses with seed parents whose seedlings are all zygotes, i. e. 'Clementine' and 'Wilking', the seedling means were similar to the mean of both parents (Table 2), supporting the assumption that the inheritance of L/W is largely controlled by genes whose effects are additive. This also supports the proposition from the 'King' crosses, that the seedling classes between the values of 'King' and the pollen parent are zygotic classes.

Environmental heterogeneity within a controlled experiment appears to have only a moderate influence on leaf shape compared with genetic effects. This was observed in the similarity of the proposed nucellar modes of the two 'King' crosses, and in the little variation of L/W among individual trees within a variety. To see if there was a large location influence, two varieties were observed at other locations. 'Satsuma' had L/W of 2.36 at Sarafand and 2.10 at Ein Shemer, while 'Dancy' was 2.18 at Sarafand and 1.90 at Bet Dagan. While these observed location differences were modest compared with varietal differences, varietal data should be obtained from trees growing in a common environment and under similar management in judging the L/W diversity of parents in a cross.

Conclusion

Nucellar and zygotic seedlings from crosses of parents differing sufficiently in L/W can be differentiated. Nucellar seedlings have a L/W similar to that of the seed parent while the mean of zygotic seedlings is approximately intermediate between the parents. The division between these two populations is facilitated by plotting frequency distribution.

Literature

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