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**Agricultural Experiment**  
**Station**

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AUBURN

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**A Field Method for Distinguishing**  
**Certain Orange Stock**

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BY  
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\*In cooperation with U. S. Department of Agriculture.

# A FIELD METHOD FOR DISTINGUISHING CERTAIN ORANGE STOCK. \*

BY

FREDERICK A. WOLF, *Plant Pathologist.*

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In the warmer parts of the states bordering on the Gulf of Mexico the planting of citrus orchards for the growing of citrus fruits on a commercial scale is becoming more and more extensive. This fact is strikingly manifest in the two lowermost counties of Alabama, Baldwin and Mobile. There is growing recognition that the climatic and edaphic conditions within this portion of the state are very favorable for orange culture. Along with this may be taken into account the fact that the past winter, because of its unusual rigor, has entailed severe losses to the orchardists and nurserymen of southern Texas. Some of them have, therefore, moved their nursery sites to these counties. Then, too, profitable crops of oranges have been grown in this section of our state. All of these reasons are contributory in accounting for the yearly increase in the planting of orange groves in Alabama.

Experimentation has shown that the Satsuma orange succeeds best for this region but only if it has been budded or grafted on Trifoliolate stock. There is reason to believe, however, that varieties other than the Satsuma as well as other citrus fruits can be grown successfully in southern Alabama. It is useless to make plantations of Satsuma trees budded on Sour orange stock for it will only result in the production of an inferior tree with a very inferior fruit. Attention is called to this limitation of the Satsuma to Trifoliolate stock in a recent circular <sup>(1)</sup> from the United States Department of Agriculture, Bureau of Plant Industry. It is also pointed out in this circular that, in 1887, Prof. Otto Penzig, <sup>(2)</sup>

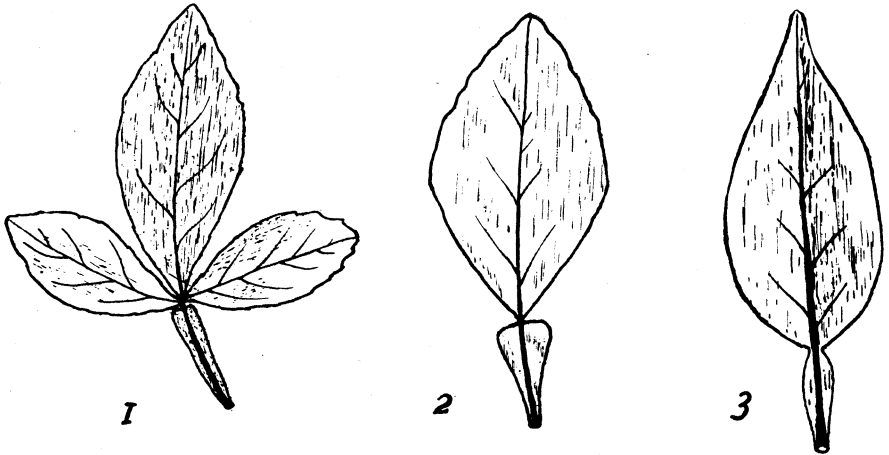
\*The subject matter of this circular was used as a minor thesis in the Department of Phanerogamic Botany, Cornell University, special thanks being due Dr. H. B. Brown, now Professor of Botany in the Mississippi Agricultural and Mechanical College.

1. Swingle, W. T. The limitation of the Satsuma orange to Trifoliolate orange stock. Circular of the Bureau of Plant Industry 46: 1-10, pl. 1, figs. 6 1909.

2. Penzig, O. Studi botanici sugli agrumi e sulle piante affine (con un atlanti in folio di 58 tavole). Annali di Agricoltura. 590 pp. 1887.

Director of the Botanical garden at Genoa, Italy, discovered a very evident difference between the pith of the stem of the Trifoliolate orange and the Sour orange. This structural difference is easily seen with the aid of the low power of a microscope. Sections of the pith of the stem of the Trifoliolate orange cut longitudinally show more or less imperfect transverse partitions made up of thick walled cells, the cells being irregular in size and shape (1. c. p. 187, pl. 13, fig. 6.) A similar section of the Sour orange shows none of these partitions, with a tendency to all the cells being uniform in size and arranged in rows lengthwise of the stem.

Upon the advent of this circular certain nurserymen appealed to the United States Department of Agriculture for help in the



Leaves of three species of oranges. Fig. 1, Trifoliolate; fig. 2, Sour; fig. 3, Yuzu.

determination of their stock. Much of this stock is grown in their own nurseries, part of it comes from other states, and some is imported from Japan. Some so-called Trifoliolate stock was sent to Mr. Swingle who reported that part of the material was not Trifoliolate nor did it seem to agree in structure with the Sour orange. It was evident that certain nurserymen were marketing a mixed lot of Trifoliolate orange stock. In view of this fact further inquiry was made by the writer and it was found that there is grown in Japan an orange upon which the Satsuma is budded which is known under the commercial name of Yuzu. The Yuzu orange is armed with spines much as the Trifoliolate but the leaves are entirely different so that they can easily be distinguished under

field conditions. When, however, all the leaves have fallen, as is the case when the trees are graded in the packing shed, determination is rendered much more difficult. An examination of the pith of the Yuzu shows a structure somewhat similar to the Sour orange except that the cells are not so regular in their arrangement, and they also vary in size. This makes the Yuzu easily separable from the Trifoliate orange. While at present it cannot be recommended that Satsuma oranges be budded on Yuzu yet the matter has been made the subject of experimentation in southern Texas with very favorable results. The Alvin Japanese Nursery Co., with a branch nursery at Grand Bay, Ala., finds \* that Satsuma on Yuzu will require a longer time to bring the trees into bearing but that that the fruits will be larger and the trees probably longer lived than on Trifoliate stock. Since, however, a very considerable expense is involved in the starting of an orange grove the planter does not wish to experiment. He insists on oranges budded on Trifoliate stock. These are known to grow well and be profitable. It might be noted that as a result of subsequent observation the percentage of Yuzu stock among the Trifoliate was found to be very small and that great care is now being exercised to prevent its importation and sale as Trifoliate stock.

This work was undertaken with the aid of Frederick W. Mally, formerly connected with the State Department of Agriculture of Texas, because of the various appeals, in the spring of 1910, for help in determining the kind of nursery stock. It was very evident from the first that it would be a physical impossibility to examine several million trees by making sections of the pith of the stem, and also that it would be impracticable financially. Then, too, such an examination makes it necessary that a cut be made which extends half way through the stem. Stock examined in this way is not readily saleable and is in a rather impaired condition even though the wound may soon heal over.

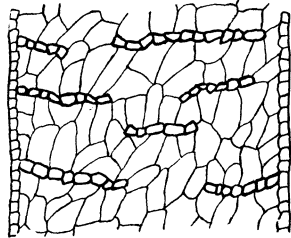


Fig. 4.—Pith of the Trifoliate orange in longitudinal sections showing the thick walled cells that make up the imperfect cross partitions.

\*This statement is used through the courtesy of the Alvin Japanese Nursery Company.

Since one does not have the leaves by which to distinguish species in the packing shed he must look to other parts of the plants for characteristic differences. It was found that the roots seem to possess characters which may be used in distinguishing the species. These are both morphological and histological. A typical stock of Yuzu has only two large roots with a few moderately thick smaller branches, plate 1, fig. 2, while a typical Trifoliolate stock has several large roots with numerous fibrous branches fig. 1. After examining thousands of trees in the packing shed we are of the opinion that, if the two species are grown under similar conditions, this character may fairly safely be used in distinguishing them. Specimens will be met with, of course, which cannot be determined on the basis of this character. They must be made the subject of a more detailed study.

These two species can be distinguished also, when they occur in mixed lots, by a difference in odor. When the cortex of the root is bruised a strong, penetrating odor, quite disagreeable to many will be found to characterize the Yuzu, while the Trifoliolate will have a faint, much milder odor. The attention of a considerable number of persons has been called to this fact and they all agree that they are markedly different.

These two simple methods can be supplemented by another which is practically as easy of application. This method is based on anatomical differences of the species. Since such differences are fairly constant within a given species when the plants are grown under the same conditions of soil and moisture their use can be relied upon as a scientifically accurate means for identification. These differences show well in thin cross-sections of the roots when examined under the low power of a microscope. By this means the differences are made so prominent that, when once they are fixed in mind, a worker, using a hand lens, may be certain of his determination of species.

Since a microscope is not always available, and some little skill is required in cutting suitably thin sections, some have been cut and photographs made of these sections as they would appear

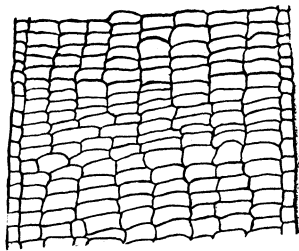


Fig. 5.—Longitudinal section of the pith of the Saur orange. Note the uniformity in the size of the cells, their serial arrangement, and the absence of thick walled cells.

when seen through a microscope. For this purpose one-year old roots about 4mm. in diameter (two-thirds the diameter of an ordinary lead pencil) were embedded in celloidin, sectioned and stained with safranin and Delafield's haematoxylin. Safranin stains red the lignified parts (wood fibers, vessels, and groups of bast fibers) and the suberized or corky parts of the outer bark. The medullary rays, cambial region, and the remainder of the bark are stained blue by Delafield's haematoxylin. Microphotographs were then made of sectors of such sections. (Plate II). In each case the tissues extending from the pith area or central area (no pith cells are present) to the outer bark are shown. In Fig. 3, which shows a portion of the root of Trifoliate orange, there is a rather large pith area made up of large cells. The xylem or wood contains numerous vessels, some of which are very large in diameter. These occur either singly or in groups of two to six.

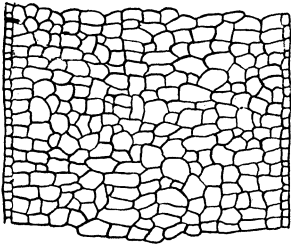


Fig. 6.—Pith of the Yuzu orange in longitudinal sections. Compare it with the sour orange, noting the lack of uniformity in size and in arrangement of the cells.

The cambium forms a sharp line of demarcation between the wood and the bark. Within the bark are three or four rows of groups of bast fibers, forming broken concentric rings. These groups are separated by relatively small distances, in the two inner rows, and are much more scattered in the outer rows. In all of the microphotographs the groups of bast fibers show as dark areas within the bark. Fig. 4 represents a similar sector of the root of the Yuzu orange. The pith area is essentially of the same size as in the Trifoliate orange, but the cells are larger in diameter. The vessels in the wood are not nearly so large and prominent, neither is there so evident a line of separation between the wood and the bark. Only a few scattered groups of bast fibers are present. This species is easily distinguished from the Trifoliate orange on account of the smaller diameter of the vessels in the wood. Fig. 5 is a similar sector of the root of the Sour orange. The pith area is so very small that the medullary rays almost converge at the center. The vessels are comparable in size with those of the Yuzu. The wood and the bark are quite sharply delimited. The groups of bast fibers are close together in the inner row, and there are only a few scattered groups farther out. This is a condition intermediate between the Trifoliate and the Yuzu.

The position of the elements that are used in this anatomical diagnosis as seen in an entire cross section, the relative sizes of the pith areas and vessels, and the number and the distribution of the groups of bast fibers, are represented in a diagrammatic way in Plate III. Fig 6 is a diagram of Trifoliolate orange, Fig. 7 of Yuzu and Fig. 8 of Sour. In each sketch (a) represents the outer bark or corky tissue, (b) one of the groups of bast fibers, (c) a vessel of the wood, (d) the cambial or growing zone, (e) a medullary ray, and (f) the pith area.

When one has familiarized himself with the points of difference in structure of the roots of these three species of oranges they can be distinguished with certainty with the aid of a hand lens. Technically trained experts are not necessary for carrying out the work. The accuracy and efficiency of this method of separation based entirely on anatomical differences has been demonstrated in our field work. Two men working together, one provided with a sharp budding knife with which to make a smooth, clean cut and the other with a hand lens, can easily determine from five to six thousand trees per day. Since it is necessary to grade all the trees in the packing shed, the determination may be made at the same time and need involve but little additional expenditure of time. As a result of the field tests, one need feel no hesitancy in stating that this method may be quickly learned and accurately employed by the nurserymen and growers.

As is well known the difference in species of plants do not always lend themselves as well to verbal description as to observation, both to the layman and to the scientist. When once the type has been fixed in mind, however, it has relatively clear-cut, well-defined characters. These characters may appear more and more distinctive to the mind of the observer as a larger number of individual plants comes under his observation. This is markedly true in the case of the three species of oranges which have been considered.



## EXPLANATION OF PLATES.

### PLATE I.

Fig. 1—Root of Trifoliolate orange showing the type of branching.

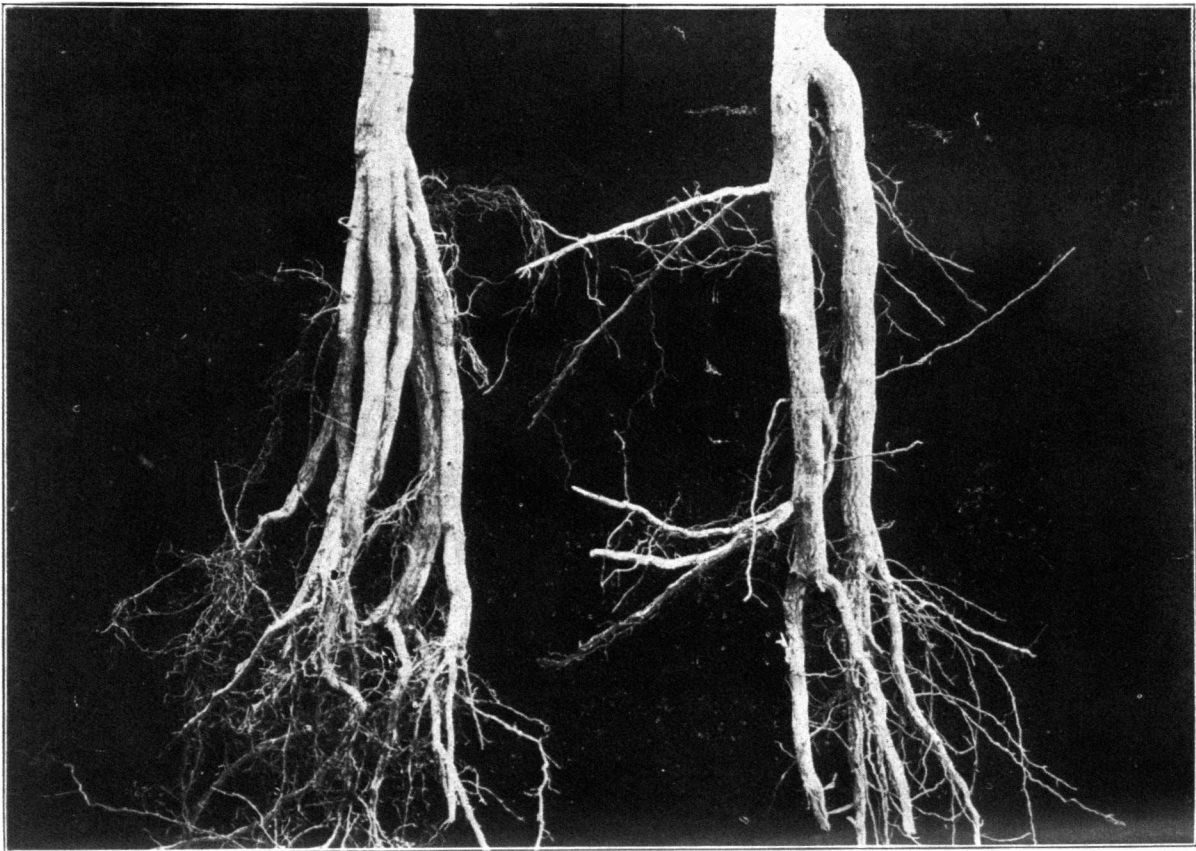
Fig. 2—Root of the Yuzu orange showing the type of branching.

### PLATE II.

Figs. 3-4-5.—Microphotographs of sectors of orange roots, Trifoliolate, Yuza and Sour, respectively.

### PLATE III.

Diagrams representing the arrangement of parts in entire cross sections of the roots of the three species of oranges: Fig 6 of the Trifoliolate, Fig. 7 of the Yuzu and Fig. 8 of the Sour. In each case (a) represents the corky portion of the outer bark, (b) a group of bast fibers, (c) a xylem vessel, (d) the cambium, (e) a medullary ray, (f) the central area.

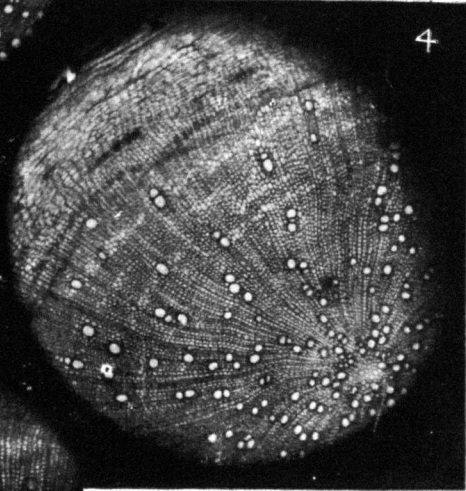
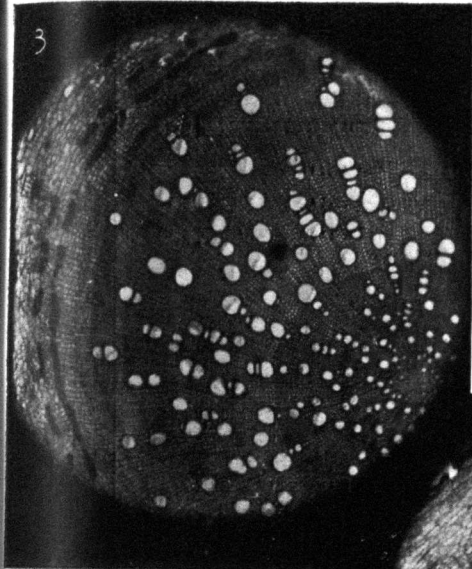


(a)

(b)

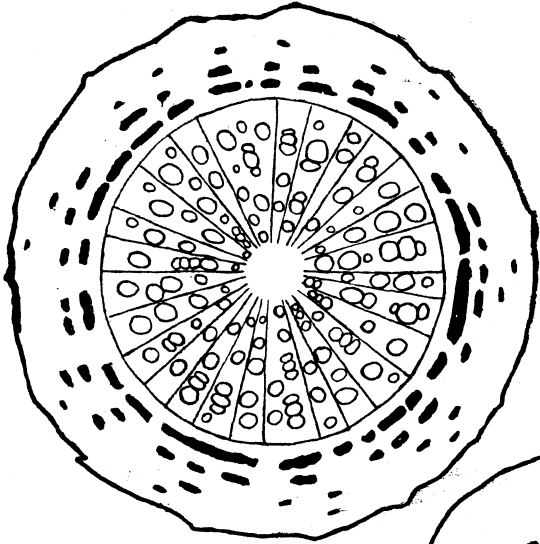
PLATE I

PLATE II

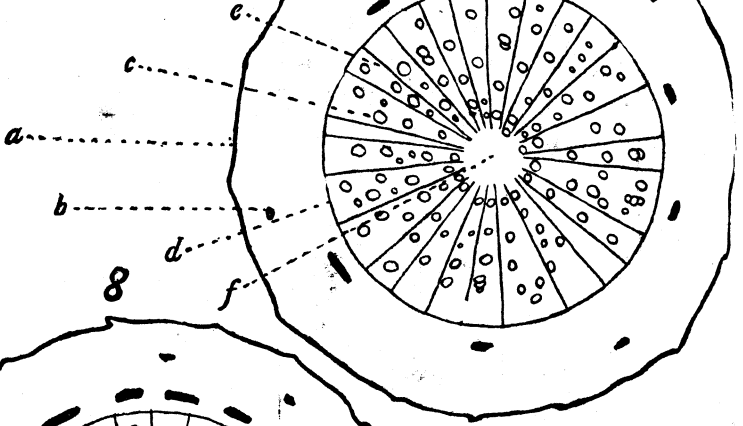


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PLATE III.



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