Transmission of Leprosis by Grafting

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ABSTRACT. This paper reports the efficiency of the top-cleft-graft method for leprosis transmission. Green shoots of Pera sweet orange showing leprosis symptoms were disinfected from mites and insects, cut into pieces of 5-10 cm long, and inserted on potted, healthy receptor seedlings of Caipira sweet orange. After 4.5 to 13 months, leprosis transmission could be observed from diseased donor to healthy receptor tissues. Of 50 grafted shoots, 7 lived, 5 of which transmitted leprosis. The top cleft-graft method showed a higher percentage of leprosis transmission than leaf implantation and bark patch insertion previously described.

Several authors believed that leprosis was caused by a toxin injected by mites when feeding on citrus leaves, fruits, and stems. Fawcett (5) and Bitancourt (2) suggested the possibility that leprosis might be caused by a local virus. This hypothesis was strengthened by later findings (6, 8). Knorr (7) reported that leprosis could be transmitted by the insertion of a piece of bark tissue from plants that had leprosis, into the bark of healthy potted plants. Chagas and Rossetti (3, 4) successfully transmitted leprosis when they inserted leaf tissue with symptoms of leprosis into the bark of healthy potted plants. These results reinforce the assumption that the disease is caused by a transmissible infectious agent.

In this paper, the effectiveness of cleft-graft transmission of leprosis is reported in comparison with other methods previously described.

MATERIALS AND METHODS

Green shoots of Pera sweet orange exhibiting typical chlorotic lesions of leprosis without tissue necrosis of the stems were field collected, and cut into pieces from 5 to 10 cm long. These pieces were cleaned with 70% alcohol and examined under a stereomicroscope to ensure that they were free of insects or mites. Then these donor shoots were properly prepared with a sterilized blade for top-cleft-grafting (1) and inserted on potted healthy receptor plants of Caipira sweet orange. The grafts were taped and each plant was covered with a plastic bag to form a moist chamber for at least 10 days. Fifty such grafted plants were kept at 25C in a glasshouse where they were periodically sprayed with miticides. Seventeen check plants were similarly treated after being grafted with apparently healthy clean shoots from affected orchards.

RESULTS

After 4.5 to 13 months, 7 of 50 grafted shoots remained alive and 5 of those showed leprosis symptoms on the receptor tissue adjacent to the affected donor tissue. Of 17 check grafts, 7 were alive and showed no symptoms. Fig. 1 illustrates the stem of a receptor plant with the first symptoms 4.5 months after grafting. Ten months later the symptoms had progressed downward affecting the stem of the same plant (fig. 2). No symptoms were observed on leaves or on other parts of the stems, except for the grafting region. Table 1 shows the results obtained by this method compared with those previously reported by the authors.

DISCUSSION

Although several attempts to

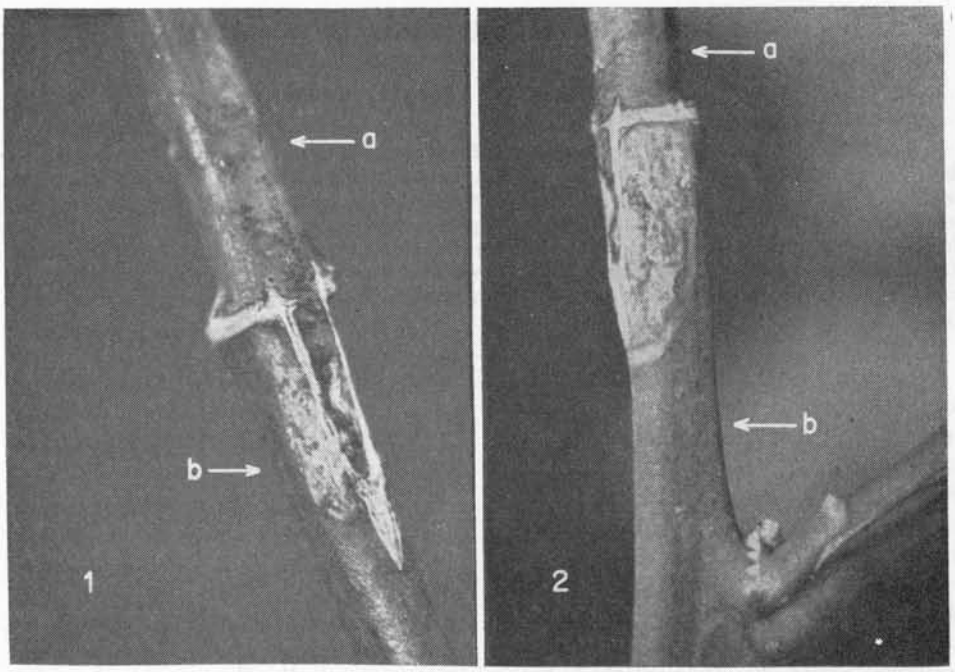

Fig. 1. Top, cleft graft of leprosis infected stem onto healthy potted sweet orange seedling showing transmission 4.5 months after grafting: a—donor stem; b—receptor.
Fig. 2. Same plant as in fig. 1, 10 months after grafting.

transmit leprosis mechanically to indicator hosts have failed (Chagas, unpublished data) the disease has been repeatedly transmitted by grafting diseased tissue onto healthy potted plants. The first evidence in the latter case was reported by Knorr (7). He inserted leprosis-affected bark patches into healthy immature green shoots from which similar sized patches of cortical healthy tissue had been

TABLE 1
TRANSMISSION OF LEPROSIS BY CLEFT-GRAFTING AND LEAF TISSUE IMPLEMENTATION

Experiment	No. surviving/ total no. grafts	No. transmitted/ no. surviving	No. transmitted/ total no.	Transmission* (%)
Graft: cleft with lesions	7/50	5/7	5/50	10
Graft: cleft control	7/17	0/7	0/17	0
Implantation: leaf tissue with lesion	1/50	1/1	1/50	2†
Implantation: leaf tissue (control)	0/40	0/40	0/40	0†

* Considering the total cleft-grafts or leaf implantations made.
† Chagas and Rossetti (3).

removed. Of 193 grafted patches, 11 induced symptoms on receptor plants. Knorr (7) considered the possibility of a toxin diffusion from the diseased to the healthy tissue. Further evidence of leprosis transmission by grafting was reported (3) using the implantation of mite-free affected leaf-pieces under the bark of immature green stems of healthy plants. Transmission rate was low, in that case, only 1 of 50 grafts (2%) survived and transmitted leprosis (table 1).

The present results confirm that leprosis can be transmitted by grafting and that the top-cleft-graft method gives higher transmission than the other grafting methods that have been used. All of these grafting experiments reinforce the possibility that leprosis is caused by a virus. Since electron microscopy revealed the presence of virus-like particles similar to the rhabdoviruses in leprotis-affected tissues (6) such possibility can be considered more plausible.

For a better knowledge of the disease and its etiology the higher transmission rate through the cleft-graft method is desirable so that electron microscopic observations can be made during the grafting experiments to verify the presence or absence of virus particles.

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