2 - A CONTRIBUTION TO THE STUDY OF AFRICAN CASSAVA MOSAIC DISEASE

Jean Dubern, Virologist
ORSTOM, Abidjan, Ivory Coast

The various reports so far published on the Cassava Mosaic Disease have been concerned with symptomatology, epidemiology, transmission by Aleyrodidae, varietal selection and treatment by heat. No paper has given sufficient information on the nature of the agent (s) responsible for the disease. This is the subject of this paper.

The study of transmission was undertaken. No attempt at transmitting the disease by mechanical means was successful. On the other hand, the attempts to transmit the disease by dodder (Cuscuta subinclusa) were successful. The present study is based on transmission from a single cassava variety. This variety, Agba Baoulé, is particularly susceptible to the disease: mosaic, deformation, and stunted growth reminiscent of the symptoms of "witches'broom" (Fig. 1).

The diseased cassava plants of this variety were obtained by cuttings. Seeds planted in insect-proof shelters, provided disease-free seedlings which were used when they were 6 to 10 weeks old. Young stems of dodder were deposited at the axil of healthy seedlings.

The experimental procedure is as follows :

> The stems of Cuscuta subinclusa that had grown on diseased cassava plants for three weeks, while still intertwined on these plants, were hung on the stems of the healthy plants and left for three weeks. They were then detatched from the plants. Six to eight weeks after contact with dodder that had developed on the diseased cassava, a yellowing was observed on Vinca rosea, Petunia violacaea, Capsicum frutescens and Capsicum annum. Control plants that had been in contact with healthy dodder, though showing chlorotic symptoms due to the parasitic effect of the dodder, showed no yellowing and quickly regained normal appearance once the parasite was cut away.

Four to six weeks later, the new leaves that appeared on the peppers (Capsicum frutescens) showed peculiar characteristics (Fig. 2). They were very small, with thick laminae and ribs bulging at the upper surface. The internodes were short, dwarfing was common and plants showed a beginning of the "witches'broom" phenomenon. Many flowers dropped and a few were deformed. Symptoms were transmitted from pepper to pepper by dodder, but could neither be transmitted mechanically nor by plant lice. The transmission percentage was quite high: about 20 to 25%. Some weeks later the symptoms grew blurred and plants became normal. If a plant is cut however the young shoots exhibit deformations, showing that the agent is still present.

Experiments on cassava to cassava transmission produced less definite results. There was a low percentage of success, about 1 to 2%, and the affected seedlings did not always express the symptoms of diseased plants in natural settings.

No symptoms were obtained when transmission was attempted from healthy cassava that had been grown from seeds.

Fig. 1 (upper left). Agba Baoule variety showing mosaic, deformation ans stunted growth.

Fig. 2a (upper right). Pepper exhibiting extremely small leaves.

Fig. 2b (lower). Pepper with typical leaf deformation.

An anatomical study was conducted with the aid of an electron microscope, under both visible and ultraviolet light. Cutting of samples was done by hand, with thickness varying from 30 to 70 u.

In the phloem of cassava plants attacked by mosaic, some necrosis could be noticed (Fig. 3) when visible light was used. Some fluorescent, abnormal, yellow spots were seen (Fig. 4) under ultraviolet light. A histochemical coloration using the Feulgen method (Fig. 5) revealed that some cells of the phloem were very rich in nucleic matter.

Fluorescent spots and necroisis could also be observed in the inner bark of old stems of deformed pepper.

Thanks to Doctors Pfeiffer and Gigot of the Plant Viruses Laboratory, and to Professor Hirth in Strasbourg, we were able to carry out an ultrastructural study.

An observation of raw extracts and more or less clarified and concentrated suspensions of cassava after coloration with uranyl acetate revealed the presence of a number of rod-like particles about 12.5 nm in diameter with widely varying lengths. These particles were observed both in extracts from diseased plants and in extracts from healthy plants. Whatever these particles were, it did not seem that they responsible for the disease.;

No other virus-like particle was observed.

A study of sap-carried material was started recently with fragments of diseased Capsicum annum. Samples were put into 5 % glutaraldehyde for four hours at ambient temperatures; placed in 1 % osmium tetroxyde for 1 hour at 40° C and finally dehydrated in alcohol before being put into araldite.

The first extra-fine slices made revealed in some phloem cells quite large inclusions of between 50 and 200 nm. Small corpuscles of about 50 nm, either in free forms or linked in chains, can also be observed. Other inclusions are filament shaped with digits and of varying lengths. No particle had any feature that was characteristic of a virus. On the other hand, they resembled those observed in plants attacked by mycoplasmic diseases, and, even, mycoplasms isolated from vertebrates.

We have not yet done a study of healthy pepper, but non author has ever referred to such structures in healthy plants.

Actual results are fragmentary. Hence, the transmission of a pathogenic agent was made from cassava attacked by mosaïc disease to pepper. In diseased pepper, mycoplasm-like particles not usually found in healthy plants were observed. Work is continuing in an attempt to define the role of this mycoplasm-type agent in cassava disease.

SUMMARY : A pathogenic agent was transmitted to different herbaceous plants from African Mosaic diseased cassava seedlings. In one of them, Capsicum annum, mycoplasm-like particles were observed.

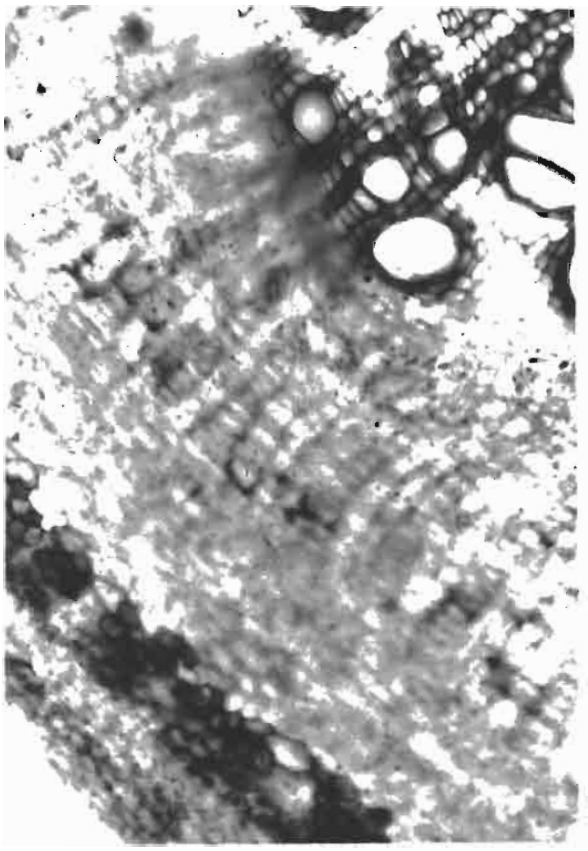

Fig. 3. Necrosis in phloem tissue of diseased cassava plant under natural light.

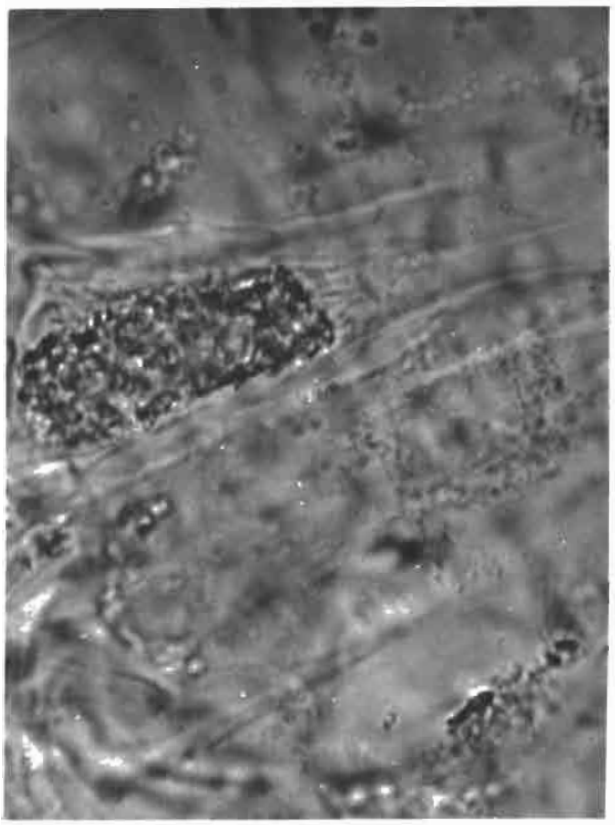

Fig. 4. A histochemical coloration using the Feulgen method reveals some phloem cells rich in nucleic acids.

Fig. 5. Mycoplasma-like organisms in Pepper phloem tissue imbedded in Epon-Araldite, visible under electron microscope.

Jean DUBERN
Louise GIVORD

COMPTE-RENDU DU SEMINAIRE SUR LA MOSAIQUE DU MANIOC

International Institute
of Tropical Agriculture
1 et 12 Decembre 1972

OFFICE DE LA RECHERCHE SCIENTIFIQUE ET TECHNIQUE OUTRE-MER

CENTRE D'ADIOPODOUME - COTE D'IVOIRE

B.P.20 - ABIDJAN

OFFICE DE LA RECHERCHE SCIENTIFIQUE ET TECHNIQUE OUTRE-MER
CENTRE D'ADIOPODOUME

Laboratoire de Virologie

COMPTE RENDU DU SEMINAIRE SUR LA MOSAIQUE DU MANIOC

International Institute of Tropical Agriculture

Ibadan - Nigeria

11 et 12 décembre 1972

par

Jean DUBERN et Louise GIVORD

Janvier 1974