

# CTA

THE INTERNATIONAL SEMINAR ON



# AFRICAN CASSAVA MOSAIC DISEASE AND ITS CONTROL



YAMOOUSSOUKRO, COTE D'IVOIRE  
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**CTA**

TECHNICAL CENTRE FOR AGRICULTURAL AND RURAL  
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**IAPC**

INTER-AFRICAN PHYTOSANITARY COUNCIL  
YAOUNDE, CAMEROUN



**AFRICAN CASSAVA  
MOSAIC DISEASE  
AND  
ITS CONTROL**

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#### SEMINAR PROGRAMME

**THE INTERNATIONAL SEMINAR  
ON  
AFRICAN CASSAVA MOSAIC DISEASE AND ITS CONTROL**

**INTRODUCTION**

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The international seminar on **African Cassava Mosaic Disease and its Control** was organized, under the auspices of the Ministry of Scientific Research, Côte d'Ivoire, by the following research institutes and organizations:

<b>CTA</b>	<b>Technical Centre for Agricultural and Rural Cooperation</b>
<b>FAO</b>	<b>UN Food and Agriculture Organization</b>
<b>ORSTOM</b>	<b>Institut français de recherche scientifique pour le développement en coopération</b>
<b>IITA</b>	<b>International Institute for Tropical Agriculture</b>
<b>IAPC</b>	<b>Inter-African Phytosanitary Council (OAU)</b>

The initiative was taken by FAO who wished the international scientific community working in Africa to debate this subject. CTA, an EEC-ACP institute created under the Lomé Convention, whose most important function is the dissemination of scientific and technical information, was the main financier. Lastly, ORSTOM, a French research institute which has devoted considerable research effort to this subject in Côte d'Ivoire, undertook the organization and hosted the seminar.

**THEME OF THE SEMINAR**

African Cassava Mosaic Disease (ACMD) is one of the most important diseases of cassava, which is the most important food crop in tropical Africa, in terms of tonnage. ACMD is a viral disease whose pathogen agent is a geminivirus transmitted on the one hand by the whitefly, and on the other hand by man himself as he plants diseased cuttings. While cassava originates from America, the disease is typically African and it has progressively invaded all cassava crops on the African continent. Taking into account this crop's variability on the African continent and sanitation conditions under which it is grown, the impact of the viral disease upon production is very hard to estimate. Nevertheless, the calculated crop losses are always high and may reach around 40%.

## **OBJECTIVES OF THE SEMINAR**

In spite of studies undertaken in several parts of the African continent and in several research institutes, ACMD remains a major problem of cassava and is a severe constraint to agricultural development of all the producing countries in Africa.

It consequently became necessary to gather all available knowledge to try and re-examine this viral disease, the final aim being to solve this problem, or at least to reduce its impact upon cassava.

The objectives of the seminar are threefold:

- to review the situation on ACMD in all countries concerned,
- to review existing scientific knowledge on ACMD and draw up an inventory of available tolerant plant germplasm,
- to highlight control methods, research programmes and action programmes for ACMD control.

## **National representatives**

Since Cassava is cultivated in the whole of tropical and subtropical Africa, it was decided that all countries interested should participate and consequently one representative was invited from each of these countries. The list of participating countries is as follows: Angola, Benin, Burkina Faso, Burundi, Cameroon, Congo, Côte d'Ivoire, Guinée, Guinée Bissau, Ghana, Kenya, Liberia, Madagascar, Malawi, Mozambique, Nigeria, Uganda, Central African Republic, Rwanda, Senegal, Sierra Leone, Tanzania, Togo and Zaïre. All national representatives were asked to answer a set of 36 questions about the cassava crop, viral diseases in general and particularly ACMD. It is essential to know to what extent African countries experience and are familiar with this viral disease and to know whether they would be ready to undertake actions to try and eradicate it.

## **Participants**

Participants at this seminar who are mainly research scientists come from different research institutes and from different African or European countries, but they all have been or still are involved in research on cassava. These researchers work in different scientific disciplines: virology, genetics, plant-breeding, entomology, bioclimatology and agronomy. Their task is to state the current level of scientific knowledge about this subject and to identify existing inadequacies. In addition, some of them have been asked by the organizing committee to present a synopsis on a subject close to ACMD, for instance: "cassava



bacterial blight", or on a general subject, related to the disease; for instance: "viral sanitation of potato in France". This is meant to bring together all information or ideas related to the control of the disease. Finally, some researchers who study other aspects of the cassava crop in Côte d'Ivoire and regularly encounter the disease will present their own experience.

#### **Representatives of national and international organizations**

One objective of the seminar was to develop new programmes of action and research on ACMD. The following international research, extension, development agencies or donor organizations concerned with the problem, were therefore present.

##### **Research organizations:**

**CIAT, CIRAD, GTZ, IITA, ODA, ORSTOM.....**

##### **Development agencies:**

**EC, IDRC, CTA, FAC, FAO, FED, OUA, USAID.**

As usual in seminars and symposia, one of the main objectives here is to disseminate information as widely as possible. Researchers and extension workers need to know the opinion of African peoples who are the first to be concerned with the disease; their experience with this disease and whether research should be intensified or not. The people of Africa need to be informed of the current position of research and of what they are likely to expect of it in the near or more distant future.

Finally, researchers should exchange results and ideas and organizations should be informed in order to be able to take the right decisions.

# **SESSION A**

## **CURRENT SITUATION**

Chairman

Dr. ASSOUMOU M'BA

Rapporteurs

Dr. HAINNAUX

Dr. VAN DER GRAAFF

## AFRICAN CASSAVA MOSAIC DISEASE AND ITS CONTROL

Guthrie, E.J., Ceres,  
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### INTRODUCTION

Cassava is grown in 39 African countries; the production of 56 million metric tons grown on 7.5 million hectares represents 43% of the total world production (FAO, 1985) and is a major food source for some 300 million people.

Almost certainly the greatest single cause of crop loss is African Cassava Mosaic Disease (ACMD). There is therefore little room for doubt as to the importance of this seminar.

### THE HOST

Cassava (Manihot esculenta Crantz) is indigenous to tropical America, where it has been cultivated for about four thousand years (Sauer, 1951). It is not known in the wild state, except as escapes from cultivation. De Candolle (1886) and Vavilov (1951) considered the area of origin to be eastern Brazil, while Sauer (1952) favoured the Venezuelan savannas. Rogers (1963), on the basis of extensive collections in South America, postulated at least two geographical centres of speciation, one in West and South Mexico and Guatemala, the other in North-East Brazil. There is evidence that the cultivars have hybridised with the native species in these areas to form a number of complexes. Species apparently closely related to M. esculenta include M. carthaginensis, M. gualanensis, M. aesculifolia, M. palmata, M. tweediana and M. saxicola. From tropical America, cassava has spread to tropical and sub-tropical regions in many parts of the world and is now widely grown in Africa, India, Indonesia, Madagascar, Malaysia, the Philippines and Thailand.

It would seem that cassava was introduced into Africa by the Portuguese in the late 16th century via Sao Tomé and Fernando Po in the Gulf of Benin and to Warri and the Congo river. Its early spread was slow and it was of little importance elsewhere before the 19th century. It was not known in Nigeria north of the Niger and Benue rivers before World War I (Jones, 1959). Cassava became a major food crop on the coastal plains of Ghana at the beginning of the 19th century and reached Ashanti and the north during the early 1930s (Doku, 1969).

On the East coast, cassava was introduced to the island of Reunion in 1736 and thence to Madagascar. It was reported in Zanzibar in 1799 but was unimportant in inland East Africa before 1850, except around Lake Tanganyika, which it had reached by spread from the West. Speke and Grant reported that it was not present further north than 4 18'S in 1862 (Grant, 1875), but Stanley (1878) noted it in Uganda in 1878. The area planted to cassava has increased considerably over the past 50 years and more cassava is grown in Africa than in any other continent.

Cassava is predominantly grown on small farms for domestic or local consumption as fresh tubers or dried derivatives and increasingly to supply the needs of urban centres. It may also be used for animal feed and production of industrial alcohol, while the foliage forms a high protein food for humans and livestock (Coursey, 1983; Lutaladio, 1983).

#### THE DISEASE

African Cassava Mosaic Disease is unknown in South America, although similar symptoms are produced by an unrelated virus, Cassava Common Mosaic Virus. This has rod-shaped particles 495 x 15 nm and belongs to the potex group (Costa & Kitajima, 1972).

ACMD was first described by Warburg (1894) under the name "Krauselkrankheit". It has since been reported throughout Africa and Madagascar, Indian Ocean islands Zanzibar, Pemba and Seychelles, India and Java (Bock & Harrison, 1985).

In East Africa the disease was not reported to cause serious losses until the 1920s; it was first reported in Uganda as "curly leaf" by Hall (1928) and as "mosaic" by Martin (1928). In West Africa, mosaic was first recorded in coastal areas of Nigeria, Sierra Leone (Deighton, 1927) and Ghana; it was observed in Ibadan (Nigeria) in 1929 and spread northwards as far as 10 10'N by 1945. It was present in all northern provinces by 1963. In Ghana the disease had reached Kumasi by 1934 and was severe and widely distributed in that area in 1936 (Golding, 1936, Doku, 1969).

ACMD is undoubtedly the most important disease of cassava in Africa. Padiwick (1956) estimated the overall loss of yield to be 11%, while losses in individual cultivars have been reported to range from 20 to 95% (Beck & Chant, 1958, and Briant & Johns, 1940, in Nigeria and Zanzibar respectively).

Bock and Guthrie (1978) reported losses of 70% in a moderately resistant hybrid and 86% in a susceptible cultivar in trials at the Kenya coast. Later trials in which the yields of healthy plants were compared with those established with infected cuttings showed yield losses ranging from 44 to 71% in West Kenya and 69 to 86% at the coast (Bock, 1983). The incidence of the disease is often extremely high; a survey of 20 farms in Ghana revealed an average level of ACMD of 96% (Walker, Heydon & Guthrie, 1985), many plantings were 100% infected. Similarly, Bock (1983) reported the incidence of ACMD to be more than 80% in some districts of Kenya, approaching 100% in some small holdings. Many workers have reported the disease to be more serious in coastal areas than at

higher and drier elevations, but in the light of these figures it would seem that the figure given by Padiwick may now be a serious underestimate.

#### THE PATHOGEN

The viral etiology of ACMD was first suggested by Zimmermann (1906), although some workers subsequently ascribed the disease to a visible parasite (Kufferath & Ghesquière, 1932; Strong & Shattuck, 1930). Storey, working at Amani, Tanzania during the 1930s, demonstrated that the disease was definitely caused by a virus and studied its transmission by the whitefly Bemisia tabaci, previously reported by Ghesquière (1932) and confirmed by Storey (1934, 1937) and Golding, 1936 in Tanzania and Nigeria respectively). African Cassava Mosaic Virus (ACMV) is transmitted by B. tabaci in the persistent manner; the minimum acquisition access period is 3.5 hours, minimum latent period 8 hours and minimum inoculation access period 10 minutes (Chant, 1958; Dubern, 1979). Storey also reported the existence of a second virus disease, Brown Streak, which was particularly prevalent at low altitudes (Storey, 1936).

Storey and his co-workers reported the existence of strains of the virus, differing in severity of symptoms and studied the transmission by grafting, previously reported by other workers (Deighton, 1927; Pascalet, 1932; Zimmermann, 1906). He was unable to transmit the virus by mechanical inoculation, although this had previously been claimed by Hedin (1931), Kufferath & Ghesquière (1932) and Lefèvre (1935). Successful mechanical transmission was reported by Bock and Guthrie (1978) and Bock and Woods (1983). Storey was able to show that the whitefly vector could only infect young leaves of shoot tips and not mature leaves; no infection was obtained when he used leaves below the third above the youngest mature leaf. He reported that the virus showed a marked tendency to move down a shoot, but did not necessarily infect branches en route; the virus is not always fully systemic. Experiments in which plants were infected with a mild strain and subsequently with a severe strain showed that mild strain infection did not protect against the severe strain (Storey & Nichols, 1938). Dubern (1979) was unable to transmit ACMV from cassava or other hosts by means of dodder (Cuscuta gronovii and C. subinclusa).

In addition to the cassava host, ACMV has been isolated in Kenya from Jatropha multifida (Euphorbiaceae) and from Hewittia sublobata (Convolvulaceae), which is a widespread component in bush and grassland, especially at the coast. It is likely that H. sublobata is of significance in view of the fact that cassava acquired the ACMV virus after its arrival in Africa (Bock *et al.*, 1981). In Nigeria, Laportea aestuans (Urticaceae) is suspected to be a natural host of ACMV (Anon., 1979).

Cassava plants infected with ACMV were examined by Kitajima and Costa (1964), but their investigations failed to demonstrate the presence of virus particles. Menon and Raychaudhuri (1970) reported the presence of ACMV in Kerala, India, and suggested that

cucumber, which they infected with cassava mosaic by means of whiteflies, might be a source of secondary infection.

Isolation of ACMV was first accomplished by Bock, Guthrie and Meredith, working at the Kenya Agricultural Research Institute (Bock *et al.*, 1978). Partially purified preparations from West Kenya material contained a geminivirus, but Bock and his co-workers were unable to obtain virus particles from coastal material. The etiology of the virus therefore remained in doubt and the geminivirus was named Cassava Latent Virus (CLV), as it seemed that another virus might be involved in ACMD. It was subsequently possible to isolate the geminivirus from coastal material using a different isolation host (Bock *et al.*, 1981). It was then apparent that the earlier difficulties in isolation of the virus from the coast were due to strain differences and that CLV should correctly be referred to as ACMV. The existence of different strains in East and West Kenya may reflect two distinct paths of entry of cassava into Kenya, one across Africa from the west and the other into coastal regions from the offshore islands (Bock & Harrison, 1985).

More recently, ACMV from India has been shown to be serologically distinguishable from both Kenya strains (Bock, Shanta & Malathi, unpublished). Sequeira and Harrison (1982) have described the Angola Defective Isolate, which is apparently defective for particle production (Robinson *et al.*, 1984). ACMV has also been isolated by workers in other countries including the Ivory Coast (Walter, 1980) and Nigeria (Igwegbe, 1980).

The morphology of ACMV particles is typical of the geminiviruses. They contain circular, single-stranded DNA (Harrison *et al.*, 1977) and are closely serologically related to bean golden mosaic, squash leaf curl and euphorbia mosaic viruses (Bock & Harrison, 1985).

#### CONTROL OF ACMD

Two main methods of control of ACMD have been advocated and investigated by workers in many African countries: breeding and sanitation.

Storey (1936) noted that the incidence of ACMD was less in up-country areas of Tanzania than at the coast and stated that "it is perfectly feasible ..... to establish healthy plots and to maintain them, by inspection and roguing, practically disease-free". He did not pursue this possibility, however, but undertook an extensive and protracted programme of breeding for resistance which continued for over 20 years (Nichols, 1947; Jennings, 1957; Doughty, 1958) until it was terminated in East Africa in 1957. It was based on crosses made between a wide collection of M. esculenta cultivars and three tree species of Manihot: M. glaziovii (Ceara rubber), which produced the best combination of yield, quality and disease resistance, M. dichotoma and M. catingea (Jennings & Hershey, 1985). Seed from this programme was subsequently sent for use in many African countries (Beck, 1982) and in particular provided the main source of resistance used in

the breeding programme which was begun at IITA, Ibadan, Nigeria in 1971. This has now provided effective resistance for 25 years, despite high inoculum pressure at many locations. Deficiencies in yield and quality have been remedied by crosses with material from South America and India. Material from the IITA programme has been distributed to more than 20 countries including Nigeria, Sierra Leone, Gabon, Zaire, Zanzibar and India, where it has provided acceptable resistance (Hahn et al., 1980).

Some of the earliest work on control of ACMD by sanitation was carried out in Uganda. A programme to establish plots of healthy plants in each chiefdom failed because it relied on highly susceptible local varieties which rapidly became reinfected (Hansford in Tothill, 1940). This was followed in 1941 by a much more effective programme, based on the Serere Experiment Station and described by Jameson (1964) in a paper which seems to have been largely overlooked. A famine in Teso district prompted a plan based on multiplication and distribution of the local cultivar Binti Misi and followed by the use of elite material on an "advancing front" basis, using material obtained from Storey's programme at Amani and involving regular roguing of infected plants. This work gave successful results over a 10 year period; so much so that it led to an increase in the incidence of kwashiorkor by reducing the protein/calorie ratio in the local diet.

Bock and Guthrie (Bock, 1983) conducted a series of trials in Kenya over the period 1974-1981 to study the behaviour of healthy plantings of "improved" material from the Amani programme and also local farmers' cultivars. They found the rate of reinfection to be low, normally less than 2% over a 12 month period at sites on the Kenya coast and in West Kenya. They concluded that sanitation gave effective control of ACMD under Kenyan conditions, where the principal vector was not Bemisia but man, by using infected cuttings for plantings.

In contrast, cassava in a trial conducted in 1980 by De Bruijn (pers. comm.) at a hot, dry site in the Machakos district of Kenya rapidly became diseased, although identical planting material at two other sites behaved as expected. This breakdown can probably be attributed to the lack of alternative food sources for the local whitefly population around the trial site.

The epidemiology of ACMD has recently been studied in depth by workers in the Ivory Coast (Fauquet et al., 1986) who have made extensive studies of the behaviour of the whitefly vector and the influence of climatic factors. Results in up-country areas support the findings of Bock and Guthrie in Kenya; reinfection rates in coastal regions are higher and show the limitations of the sanitation approach under these conditions. Their work has also demonstrated the existence of vector resistance, as yet unexploited.

Two techniques which are of relevance in cassava research are the use of high temperatures, in the range 35-39 C (Chant, 1959), and meristem tip culture (Kaiser & Louie, 1982) for the production of

virus-free material from infected plants. Use may also be made of the fact that infection with ACMV is not always fully systemic (Storey & Nichols, 1938).

#### THE FUTURE

This review shows that a most impressive body of information on cassava improvement has been developed by many workers in many countries over many years. By contrast, despite massive losses from ACMV in African countries where cassava is a major food source, remarkably little has been attempted to tackle the problem on a practical field scale. There is little doubt that the available technology could be used to great effect.

This seminar offers the unique and exciting possibility of putting the situation to rights; it is up to us to ensure that it does so.

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GENERAL SUMMARY OF NATIONAL REPORTS  
ON  
AFRICAN CASSAVA MOSAIC AND ITS CONTROL

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INTRODUCTION

This document results from an analysis and synthesis of the various national reports on:

- the importance of cassava growing in comparison with other food crops;
- constraints on the production of cassava;
- the impact of pests and diseases, mainly cassava mosaic, on the cultivation and production of cassava;
- national or regional strategies for controlling African Cassava Mosaic Disease (ACMD).

Though some countries did not fill in the questionnaire, those national reports that have been received cover reasonably well the three large growing regions concerned (Western Africa, Central Africa, and Eastern and Southern Africa). We believe this synthesis reflects the overall situation in Africa.

THE IMPORTANCE OF CASSAVA FARMING

Though in some countries cassava ranks behind such foodstuffs as rice, sorghum, and yams, in many others (the Congo, Central African Republic, and Zaire) it constitutes the staple food of the large majority of the population. In Burundi, for example, cultivation of tubers and roots, especially cassava, is increasing while that of leguminous plants and cereals is falling. Production per hectare is 5 to 10 tonnes. This low yield is compensated for by the area of land under cultivation and by the large number of farmers.

In the majority of producer countries, cassava farming remains mainly traditional, although there are large industrial production

units in Nigeria and Liberia. In the traditional setting, cassava is usually grown together with maize, peanuts, rice, beans. Cassava is grown everywhere when climatic conditions are favourable. It grows throughout the year, but yields are higher when the farming cycle is well matched with the seasonal cycle.

Each country has a collection of cassava clones ranging from 10 to more than 100 cultivars; countries such as Malawi, the Congo, Zaire, and the Ivory Coast have more than 100. Cassava is propagated mainly vegetatively. Propagation via seeds is done only in research centres.

#### CONSTRAINTS ON CASSAVA PRODUCTION

The constraints on cassava production are mostly related to the traditional cultivation methods which are still predominant in most countries growing this crop.

These include:

1. The structure and function of the cropping systems, in which cassava generally comes at the end of a rotation.
2. The use of unproductive farming techniques and a lack of inputs.
3. A strong preference for traditional varieties that are low-yielding and more susceptible to disease and pests.
4. The rapid decrease in soil fertility as a result of poor management.
5. Disease and pests.
6. The lack of a stable and sufficiently profitable market.

#### FOOD PROCESSING AND CASSAVA CONSUMPTION

The tuber is the part of cassava that is eaten most, but in Central Africa (Burundi, the Congo, the Central African Republic, and Zaire), and to a lesser extent in West Africa (Liberia), the leaves are consumed, and constitute an important source of protein.

There is a multitude of traditional techniques, used to transform cassava into more or less storable local food products. Chikwangué or fofou is the most representative product of Central Africa, while gari, cassava flour and attiéké are seen most often in Western Africa. The products with the longest storage life are gari and cassava flour, which are the most suitable for export. Nowhere is cassava an official export item.

## DISEASES AND PESTS OF CASSAVA

Cassava is susceptible to the following main diseases: ACMD, by far the most widespread; vascular bacteriosis; anthracnosis; and to a lesser extent cercosporiosis. The most often reported pests are the mealybug (Phenacoccus manihoti), the green spider mite (Mononychellus spp.), and the stinking locust (Zonocercus variegatus). Aulacode has also been reported.

## THE SIGNIFICANCE OF AFRICAN CASSAVA MOSAIC DISEASE

This disease attacks all cultivars, though the level of susceptibility varies from one cultivar to another. In each of the producer countries, the presence of ACMD is related to the density of the cassava population and to the climatic characteristics. The wettest regions are most heavily affected. Mosaic attacks are much more prominent on young plants than on old ones. It is generally agreed that ACMD causes 20 to 80% loss of yield.

The vector of the mosaic virus is the whitefly (Bemisia tabaci), which multiplies rapidly at the start of the rainy season and disappears in the dry season. This fly is found in all the cassava-growing areas. The cassava plant is not the only host of the whitefly, which also colonizes the leaves of cotton, tobacco, and sweet potato. The presence of whiteflies has also been reported on fruit trees in the Central African Republic. It is important to stress that, though the vector of the mosaic is the whitefly, the persistence of the disease in all the areas of cassava production is also attributable to the farming practices of the peasant farmers, who make no distinction at planting time between healthy and infected plant stocks.

## CONTROL STRATEGIES

Strategies for controlling the disease would include:

- Establishing and reinforcing existing programmes to investigate the etiology and epidemiology of the virus infection, necessarily complemented by the production of resistant varieties. For this purpose, investigations are under way in some countries (Zaire, Nigeria, the Congo, Liberia, Kenya, the Ivory Coast, Togo), but the scope differs from one country to another. Many countries would therefore like to strengthen their research activities.
- Establishment and implementation of a development programme to propagate resistant varieties and secure their widespread acceptance by the peasant farmers. A good many countries (Liberia, Burundi, Rwanda, Nigeria, Benin, Malawi, and Zaire) already have such development programmes, but activities vary according to the local facilities. Such programmes should be reinforced where they exist, and created where they do not exist.
- The introduction of mosaic-resistant clones.

The International Institute for Tropical Agriculture (IITA) has vast experience in this field and is the preferred consultant for almost all the producer countries of Africa except the Congo and the Ivory Coast which are at present introducing bacteriosis-resistant varieties.

#### CONCLUSION

Cassava production per unit area remains very low in Africa. The absolute necessity to reach self-sufficiency in food production in Africa requires that the yield per hectare be improved substantially. This objective can be reached through adjustment and careful attention to effective farming techniques and above all the use of high-performing cultivars which are appropriate to the environment and resistant to the majority of pests that threaten cassava.

All the producer countries have understood this and are aware of the effect of mosaic disease on cassava production. They all wish to participate in a programme of action to control the disease. They would also like to see the already existing infrastructure and national strategies strengthened..

**AFRICAN CASSAVA MOSAIC IN THE CONGO,  
ITS IMPORTANCE, DISTRIBUTION  
AND METHODS OF CONTROL**

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Cassava (Manihot esculenta Crantz), a woody Euphorbiaceae, is the staple foodstuff of the Congo. Its starchy tubers, the main source of carbohydrate, are eaten in various forms: raw for the sweet varieties and, for both sweet and bitter varieties, as cassava flour, fofou, and also after fermentation and cooking in the form of cassava bread, chikwangué. The chopped green leaves are used in the preparation of a vegetable sauce, saka-saka, which is a considerable source of protein (FAO, 1970).

Cassava originated in South America and was introduced to the Congo delta in the 16th century by the Portuguese (Jones, 1959), who then spread it to India and Indonesia. It is at present the most important food crop in this country, from the point of view of both the area under cultivation and the tonnage produced.

Table 1. Crop Production in the Congo.

Crop	Total area under cultivation (hectares)	Total production (tonnes)
Cassava	91,140	628,400
Maize	13,910	11,003
Groundnuts	19,750	113,863
Rice	2,350	2,826
Potatoes	1,287	6,435

Source: Department of rural economy.

Cassava is an adaptable plant; it is found from the Sudan-Sahel zone (500 mm of rain) to the heart of the equatorial zone (more than 5000 mm). It is a perennial shrub from 2 to 5 m high, generally with three branches and with tubers that are usable until they are up to three years old (Massala, 1984).



Cassava has enormous potential (it can produce up to 80 t/ha), but has unfortunately not been fully exploited. In 1983 worldwide production fell by 2%, but decreased in Africa by 7% because of widespread drought, unfavourable planting conditions, and above all attack by insects and disease (Revue Afrique, 1984). In the Congo, there has been declining production since 1976 (DGRST, 1982). The limiting factors include the use of low-yield varieties, traditional farming techniques, and biological factors such as pests, bacteria, fungi and viruses (Makambila 1980; Boher and Daniel, 1981; Daniel et al., 1981; Fabres and Boussiengue, 1981; N'kouka et al., 1981).

Cassava is grown in the Congo in two ways: by peasant farmers, who plant small areas using traditional techniques, often growing cassava alongside other crops, and as an industry, with mechanized production (Mantsoumba state farm). A multidisciplinary team of researchers from various institutions (DGRST, ORSTOM, M.NGOUABI University) has set up a national programme to improve the cultivation of cassava in order to achieve food self-sufficiency. In this context we undertook an investigation of African Cassava Mosaic.

Regardless of the style of the farming, cassava is propagated by cuttings. This practice favours the propagation of cassava diseases, including viral diseases such as African Cassava Mosaic. The latter appeared in Africa at the end of the last century (Warburg, 1894) and spread very rapidly. The disease manifests itself mainly in the morphology of the leaf. The severity of symptoms depends on the variety, and furthermore is not the same for all the leaves of a given plant. Cours (1951) proposed a classification of the severity of the mosaic from 0 (healthy plant) to 5 (leaf-blade reduced to one-tenth, a branch with short internodes, and usually death after several months). The fall in yield ranges from 5 to 95% and depends on the variety used, on the time and type of infection, and on climatic factors. The viral origin of the disease involves a geminivirus called African Cassava Mosaic Virus (ACMV) (Bock and Woords, 1983).

The mosaic is transmissible by cuttings, grafts, and insects of the genus Bemisia: Aleyrodidae (Storey and Nichols, 1938; Dubern, 1979).

Here we present the results of an evaluation of the severity and distribution of the disease in the Congo. In addition, a method of curing local varieties by heat-treatment and in vitro culture of plant tissues has been developed.

#### CURRENT DATA ON ACMV

The symptoms observed during our investigations are the same as those described in published reports: an often severe mosaic covering 20 to 100% of the leaf blade, deformations of the leaves, reduction of the vegetative apparatus, and short internodes, leading sometimes to stunting of the plant. On plants attacked

very young, the leaf-blades disappear and sometimes the plant dies (Storey, 1936; Alagianagalinga and Ramakrishana, 1966; Fauquet et al., 1980).

So far, we have carried out surveys in the three main cassava-growing regions, around Cuvette in the north of the country, and Bouenza and Pool in the south. In addition, we have looked at the collections of the Centre de Recherches Agronomiques de Loudima (CRLAL).

All the observations so far show that the disease is present throughout the country. The severity of symptoms differs from one cultivar to another. The index of severity of symptoms lies between 0 and 5 in the forest zone, and from 0 to 3 in the savanna zone (see Cours' scale). Three-month-old plants express the symptoms most strongly and a phenomenon of regression of the disease with the age may be observed.

Nevertheless, we have found cultivars that are tolerant to ACMV: Doumi, Lepae, and Nionguili in the Cuvette region, and Kousakanandi in the Pool region. Cuttings taken from plants with no symptoms of mosaic in the field and planted in the laboratory have sometimes shown symptoms at the beginning, which then became muted, suggesting some tolerance towards the mosaic by the plant.

#### ATTEMPTS TO OBTAIN AND MULTIPLY HEALTHY CASSAVA PLANTS BY IN VITRO PLANT TISSUE CULTURE

In collaboration with the ORSTOM phytopathology laboratory, we have developed the in vitro culture of plants. The objective was to obtain healthy plants from the virus-infected local cultivars and then to rapidly multiply them and others imported from the International Institute for Tropical Agriculture (IITA), for example, in order to set up a stock of cuttings to popularize them.

Two techniques were used:

- in vitro culture from microcuttings;
- in vitro culture of meristems, combined with heat-treatment.

The in vitro culture of plant tissues (microcuttings or meristems) appears today to be the best means to obtain healthy plants from diseased ones (Féréol, 1978).

Within 2-3 months, a plant can be regenerated that can then be multiplied or transferred to a greenhouse.

The regeneration in vitro of a complete plant from a meristem implies the use of a medium containing several phytohormones: Benzyl amino purine (BAP), alpha naphthalene acetic acid (NAA), giberellic acid (Kantha et al., 1975; Féréol, 1978; Tilquin, 1978; Mabanza, 1980). For the in vitro multiplication, a basal medium without phytohormones or containing only NAA is sufficient. For a better development of the roots, a medium containing indole-3-butyric acid is necessary (Smith et al., 1986).

On the one hand, we have obtained from one meristem the regeneration of entire plants without change of medium. On the other hand, following two or three successive transfers, we have obtained two to four small plantlets from one meristem, giving a callus with two to four shoots. This second method is important, since it allows us to obtain more plants from a single meristem.

Heat treatment stimulates the growth of the stems of cassava plants. It should be stressed that the combination of the heat-treatment and the in vitro culture of meristems make it possible to cure and multiply cassava plants (Féréol, 1978; Kartha, 1975).

The application of these two techniques depends on the dexterity of the person performing the experiment and on factors such as the size of meristems, the effect of phytohormones, the effect of culture media.

Small plants obtained by in vitro culture, then transferred to pots and sheltered in a cage for 14 to 21 days, show no sign of the disease, but we are far from being able to assert that these plants are virus-free. As a matter of fact, the symptoms may actually be masked in in vitro culture (Féréol, 1978).

Only the use of virological monitoring can detect a possible virus. Sanitary monitoring after eradication of the virus could be performed with the Elisa serological method.

#### CONCLUSION AND DISCUSSION

The symptoms observed on cassava suggest the presence of ACMV in the Congo. This diagnosis must be confirmed by the identification and characterization of the pathogenic agent. Cassava in the Congo probably has a wide genetic diversity, and the number of morphologically distinguishable cultivars is large. In each small field three to six different cultivars are to be found. Unfortunately some of these cultivars are very susceptible to African cassava mosaic, and therefore represent a source of permanent contamination for nearby cultivars.

Contaminated cuttings are the main means of propagation of the disease. In addition, the farmers pay no attention to the mosaic and do not select the material they plant, which leads to a high level of contamination with the disease.

Moreover, the whitefly present on the plants may be vectors for the disease (Storey and Nichols, 1938; Chant, 1958; Dubern, 1979).

Compared with the economic effect of other diseases (bacteriosis and mealy-bug) the impact of ACMV seems negligible, but a prevention of the severe losses of yield reported elsewhere is important. In addition, it is necessary to safeguard the quality and quantity of the leaf cover, as for the part which is eaten.

The introduction of tolerant or resistant cultivars derived from the selection programmes performed in other countries is still not satisfactory. These varieties are not readily accepted by the

Congolese farmers because of a heavy branching and leaves that are not good to eat. Action is required to improve local plant stocks, including:

- breeding of cultivars,
- educating farmers on how to choose cuttings,
- improving farming techniques and mastering various agronomic parameters,
- furthering of the production of healthy plants for large-scale multiplication by techniques of in vitro culture and heat treatment.

The rate of recontamination in the field remains to be determined.

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IMPACT OF AFRICAN CASSAVA MOSAIC  
ON THE GROWTH AND YIELD OF CASSAVA

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The impact of a disease on the cultivation of a crop is proportional to the effect of the pathogen on the host plant, multiplied by the number of plants actually affected. In the case of African Cassava Mosaic (ACM), the magnitude of the latter factor is known with certainty, since practically all cassava plants grown in Africa are virus-infected. This fact was unanimously reaffirmed in our investigation covering the 25 African producing countries participating in this seminar on African Cassava Mosaic. The effect of the pathogen agent on the host plant, however, is much more difficult to define since the many articles on ACM give highly variable estimates of reduction in yield, ranging from 5 to 95% (Briant & Johns, 1940; Beck & Chant, 1958). This wide variation undoubtedly reflects a wide variability of cassava itself, but also widespread ignorance of the exact impact of this virus infection upon the production of cassava at the level of a single plant and therefore at the level of the whole African continent. Here we summarize the information available at present about the impact of ACM on the growth and yield of cassava.

INFLUENCE OF ACM ON THE DEVELOPMENT OF CASSAVA

The histological effect of ACM has been relatively well investigated in the past, and the disorganization of the virus-infected tissues has been demonstrated. The cribrovascular clusters are reduced in size and their differentiation is disturbed (Pascalet, 1932; Chant & Beck, 1959; Dubern, 1976). There is also an effect on the chloroplast and metabolic activity of the cells, producing a general disturbance of the diseased plant, with a decrease in essential metabolites such as carbon and nitrogen, and conversely an increased respiratory and peroxidase activity in the tissues attacked (Beck & Chant, 1958; Chant *et al.*, 1971; Ayanru & Sharma, 1982).

## SYMPTOMATOLOGY OF THE DISEASE

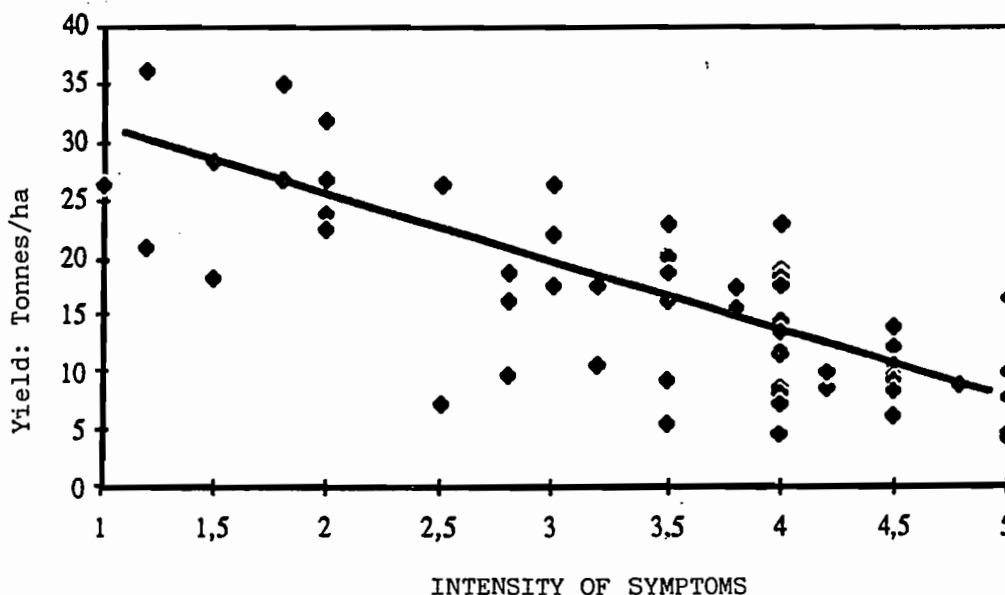
The most obvious manifestation of this viral disease is the expression of the characteristic symptoms that have given it its name. These symptoms are highly variable, ranging from a very slight, barely perceptible mosaic, to total stunting of the plant and the virtual disappearance of the leaves' limb. Several authors have established scales of severity of symptoms. We chose the one proposed by Cours (1951), ranging from 0 for no symptoms up to 5 for a plant with leaves reduced to veins. Using this scale, we quantified symptoms by assigning a score to each leaf and by calculating an index of severity of symptoms (ISS).

This ISS is specific for a given clone, but it changes with time. After the planting it increases progressively, to reach a maximum 60 days later; then it levels off and declines or disappears, depending on the clone and the time of year. The ISS of a cassava leaf does not change further once the leaf is completely open, but the scores for the leaves on a single stem are extremely variable. It is very important to standardize a method for quantifying symptoms, both to evaluate objectively the effect of the disease on a given plant and to estimate the progress of the disease with time.

## RELATIONSHIP BETWEEN SYMPTOMS AND PRODUCTION

There have been programmes to select cassava for resistance to ACM since 1938 (Storey & Nichols, 1938), and it has always been considered that there is a priori a close relationship between the intensity of the symptoms and the loss of production. It has often been shown that, considering all varieties together, there is a relationship between the intensity of the symptoms and the cassava production (Cours, 1951; Vandevenne, 1975; Mahungu, 1984). In a collection from the Ivory Coast in 1969 (Fig. 1), the mean harvest was 29 tonnes of tubers per hectare for clones with a mean ISS of 1, and 9 tonnes per hectare for clones with a mean ISS of 5. This result, which in other respects is also very astonishing, shows that the variability which ACM introduces into cassava production is far higher than the clonal variability of production potential. However, one must bear in mind that this result does not offer a means of evaluating the losses of yield due to ACM, but, rather, shows the amount of impact ACM can have on the production of cassava roots.

Figure 1. Yield of cassava varieties according to the intensity of the symptoms.



In addition, Mahungu (1984) has shown that the production, in the case of a susceptible variety, was proportional to the percentage of infected leaves. It seems therefore that ACM may be a very important factor limiting the production of cassava, by amounts proportional to the degree of the attack, as evaluated using the ISS.

However, the postulate "no symptoms - no losses" has not yet been confirmed. Various authors in East Africa and Nigeria have demonstrated that even varieties considered to be resistant could show losses of the order of 24 to 78% (Terry & Hahn, 1980; Seiff, 1982; Bock, 1983). The relationship between symptoms and losses of yield therefore needs to be clarified: this is one of our research objectives for future.

#### RELATIONSHIP BETWEEN MODE OF CONTAMINATION AND LOSS OF YIELD

We have investigated the losses of yield of a moderately susceptible clone such as CB and have shown that any given clone may lose from 0 to 77% of its production, depending on the mode and time of contamination and on the plant's environment.

A cassava plant obtained from contaminated cuttings (i.e. contaminated before being planted) loses much more production (55 to 77%) than one contaminated by whitefly (i.e. after being planted), even if the contamination takes place early (35 to 60%). If the contamination by vector occurs more than 100 days after the



planting, there is no further effect on production (Fargette et al., 1987). This is a general phenomenon for plant viral diseases: the earlier the contamination the greater the effect. Note that these results show the beneficial effect that could be produced by a phytosanitary control method. The simple act of planting healthy cuttings could increase the production of cassava of an African country by at least 50%).

#### RELATIONSHIP BETWEEN YIELD AND ENVIRONMENT

One virus-infected cassava plant, isolated in a field of healthy cassava, will produce 70% less than its neighbours, whereas if all the plants are diseased the loss will be only 33%. There is competition among the plants, so that the weakest, virus-infected, are weakened further by the strongest plants.

In fact, to evaluate the actual impact of ACM on the cassava yield, the yields of healthy plots must be compared with those of virus-infected plots. This is very difficult to carry out, because healthy plant material is very rare and because, in most situations, healthy plots become recontaminated with time.

#### RELATIONSHIP BETWEEN YIELD AND GROWTH

One possibility for studying the influence of ACM on the yield of cassava would be to consider such markers of growth as the height of the plants, the basal diameter of the stems, the number of leaves, the number of apices, the wet or dry weight of matter, etc., and to correlate these with production. But for the moment nothing is known of the relationships between these various markers. We are trying at present to define these relationships between these various markers. We are trying at present to define these relationships in a set of relatively resistant clones, and it seems that though certain factors are clearly correlated with one another, such as the diameter and the height of the stems, others are not. In addition, it appears that not all the clones manifest the same susceptibility to ACM. The impact of the disease may sometimes affect one criterion for growth and not others. It would be interesting to determine these relationships in a collection of cassava clones, and to see if there are any nondestructive markers of growth.

#### CONCLUSION

The impact of ACM on the growth and yield of cassava is difficult to evaluate. However, some points are clear:

- The number of infected plants on the African continent is enormous, so that even if the impact on one infected plant is low, the impact on cassava culture as a whole must be considerable.
- The available information, though it is not very specific, shows us that this impact is in fact very great.

- There are relationships between the symptomatology and the yield, at least in a collection of cassava clones: the more pronounced the symptoms, the greater the loss of yield.
- For a clone such as CB, known to be tolerant (Vandevenne, 1975), considerable losses may be recorded, which in certain cases may reach 70%.
- The "symptoms-yield" postulate is not always confirmed, and consequently selections made solely on the basis of symptoms need to be complemented by specific confirmation.
- The mode of contamination of cuttings is fundamental to the effect of ACM on yield. Plants derived from healthy cuttings produce a 50% increase in harvest.

Consequently, multiple factors are involved in determining the losses of yield due to ACM, but it is certain that this effect is enormous, of the order of 30% for a tolerant clone. Across the African continent, which produces 50 million tonnes of cassava, this represents a loss of some 15 to 20 millions of tonnes of dry matter. ACM is therefore a plague, and every effort must be made to reduce its impact.

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## STRATEGIES FOR CONTROLLING AFRICAN CASSAVA MOSAIC VIRUS

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### INTRODUCTION

African Cassava Mosaic Virus (ACMV) is now prevalent in many parts of Africa and induces very serious losses. The virus and its whitefly vector Bemisia tabaci have been studied for many years in East and West Africa and much attention has been given to possible control measures. This paper considers the various strategies that have been or could be adopted and the possibilities for their use on a suitably large scale.

Four important features must be considered in assessing the need and justification for control measures against a virus disease:

1. The severity of the damage caused, as determined by the virulence of the prevalent strains of virus and the sensitivity of the varieties grown.
2. The proportion of plants which become infected.
3. The stage of growth when infection occurs.
4. The economics of the crop in relation to the cost of control measures and the implications of an increase in overall production.

On the basis of these criteria there is an overwhelming case for controlling ACMV, with the important provision that the measures used must be simple, inexpensive and within the limited capacity of the farmers concerned. The measures used against ACMV and B. tabaci should also be fully compatible with those being used against other pests and diseases and with recommended cropping practices.

### POSSIBLE CONTROL MEASURES

There are three possible approaches to decreasing the losses due to viruses and they can be adopted singly or in combination:

1. By decreasing the proportion of plants that become infected.
2. By delaying infection to such a late stage of crop growth that the loss of yield becomes insignificant.
3. By decreasing the severity of the damage sustained after infection has occurred.

These objectives can be achieved in diverse ways and the main possibilities are set out in the following sections.

#### SANITATION: VIRUS-FREE PLANTING MATERIAL

Cassava is a vegetatively-propagated crop and a basic approach to control is to use virus-free cuttings for all new plantings. The potential benefits are considerable as healthy cuttings establish better and grow more rapidly than infected ones. Yields are also greater, even if the plants become infected later in the growing season due to an influx of infective vectors from outside sources (Fargette et al., 1987).

The feasibility and effectiveness of this approach to control is dependent on the rate at which virus-free material becomes infected, and on the availability of adequate stocks of the varieties required at prices farmers can afford. Herein lie the difficulties, as African farmers have limited access to improved planting material of any type and even fewer can obtain virus-free cuttings. Moreover, even if healthy stocks become available in quantity, there may be problems due to rapid infection by whiteflies. Unless spread is very slow, it is necessary to introduce healthy cuttings at frequent intervals or to adopt very stringent selection procedures to ensure that only healthy plants are used to provide cuttings for further plantings. These are serious limitations and add to the difficulty of developing a simple set of guidelines for farmers to adopt.

There are no technical difficulties in producing basic stocks of cassava cuttings free of ACMV. This has been done simply by careful selection amongst the stocks already available, or by exploiting the recovery phenomenon referred to as "reversion" (Fauquet, this volume), or by meristem-tip/heat therapy. Once basic ACMV-free stocks have been produced, they can be multiplied quickly to produce the large quantities required. Initially this can, if necessary, be done in insect-proof structures to provide a favourable year-round environment and to avoid infection by whiteflies. However, stocks for general distribution cannot be produced in great quantity without using large outdoor sites. The plants are then at risk and it is essential to identify suitable propagation sites where there is little risk of infection by incoming whitefly vectors.

The performance of healthy cuttings when grown in areas of cassava production depends on their inherent susceptibility to infection and on the overall "infection pressure" to which they are exposed. Whiteflies are numerous and the winged adults are very active for much of the year in the lowland rain forest areas of Ivory Coast and other regions of intensive cassava production in West Africa.

Sources of infection are prevalent and plantings of all but the most resistant varieties are totally infected within a few months of exposure (Fargette, this volume). Spread is much less rapid in Kenya and other parts of East Africa where cassava is grown less intensively and where plant growth and whitefly populations are restricted by long dry periods (Bock & Robertson, this volume).

These findings explain why much greater use has been made of virus-free planting material in East than in West Africa. Storey (1936) noted that farmers in parts of Tanzania obtained cuttings from the mountainous areas, where there was a low incidence of ACMV. There have since been official schemes for the propagation and release of virus-free material in Zanzibar, Uganda, Tanzania and Malawi. However, these schemes have not been sustained for sufficiently long periods or on a suitably large scale to obtain the full benefits of this approach to control. Many farmers continue to use infected cuttings, either through ignorance or because healthy material is not available in the quantity required at prices they can afford. This emphasises the need for improved procedures to ensure that farmers utilize virus-free material and also have access to improved varieties with resistance to ACMV and to other pests and diseases. There is obviously great scope for new initiatives not only in East but also in West Africa now that ACMV-resistant varieties are becoming available (Jennings & Rossel, this volume).

#### SANITATION: ROGUING

A possible method of maintaining or even improving the health of cassava plantings is to inspect them regularly to ensure that all plants with symptoms of disease are removed at an early stage and before they have acted as foci for further spread. Such roguing procedures have been widely advocated as a means of controlling ACMV and they undoubtedly have a role to play. However, roguing is not always effective and in some circumstances may be totally inappropriate.

Roguing is most effective in areas where the planting material being used is largely free of ACMV and where there are low rates of spread by whiteflies. This is the situation in large areas of eastern Africa, where early roguing is recommended as growth begins soon after planting. Plots are most readily accessible at this time and symptoms tend to be particularly conspicuous. Moreover, infected cuttings are easily removed and they can be replaced by healthy ones or by other crop plants with little loss of yield.

Roguing later in the growing season is much more difficult, especially with multi-stemmed varieties that form very dense stands. Vacancies are not easily filled to that there is a loss of crop and symptoms may be masked by the effects of drought, mealybugs, spider mites or foliar pathogens. Nevertheless, a late inspection just before crop maturity is advocated in India to mark infected plants so that they can be harvested but not used to provide cuttings for further plantings (Malathi, this volume).

There is limited scope for roguing in the many parts of West Africa where stocks are substantially infected with ACMV and where there is such rapid spread by whiteflies from outside sources that frequent treatment is necessary and many plants have to be destroyed. Roguing only becomes feasible in these circumstances once healthy stocks of suitably resistant varieties become available on an adequately large scale. However, roguing is likely to be unnecessary if varieties are developed that are virtually immune to infection, or so tolerant that they sustain little damage after infection has occurred.

These considerations explain why the effectiveness of roguing is related to varietal susceptibilities and to overall infection pressure. They also account for the success of roguing as used in crop improvement programmes in Malawi, Tanzania, Uganda and Zanzibar (e.g. Childs, 1957; Jameson, 1964), but not in the main cassava-producing areas of West Africa. However, roguing is an irksome and unpopular measure that has not been widely adopted anywhere. Farmers are understandably reluctant to remove actively growing plants that contribute to yield and they cannot obtain the full benefits of roguing unless it is also practised by their neighbours and preferably throughout the whole locality. This suggests that it will not be easy for extension agents to persuade farmers to change current attitudes. The most immediate application of roguing is likely to be in operating official schemes for producing healthy planting material and in large-scale commercial farms or plantations under strong central management.

#### VIRUS RESISTANT VARIETIES

The use of varieties with resistance to or tolerance of infection has obvious advantages in seeking to decrease the losses due to viruses.

This was appreciated by early workers on ACMV in East Africa who screened local and introduced varieties of cassava (Mannihot utilissima) and intra-species crosses between cassava varieties. They were found to be insufficiently resistant to infection and attention turned to hybrids between M. utilissima and other species, of which Ceara rubber (M. glaziovii) proved to be the most important (Nichols, 1947). There was no evidence of immunity to infection, but M. utilissima x M. glaziovii hybrids had considerable resistance. This was also apparent in progenies obtained by back crossing hybrids to cassava.

Resistance was assessed at first by exposing batches of plants to infection and recording when symptoms developed. It later became apparent that selections that were resistant to infection tended to develop inconspicuous symptoms that were sometimes ephemeral and restricted to parts of only one or two shoots (Jennings, this volume). This led to a revision of recording procedures to take account of both the incidence and the intensity of symptoms. Some of the varieties selected in this way proved suitable for East African conditions and were released to farmers (Doughty, 1958). Seeds was also sent to Nigeria in the 1950s and clone 58308 which originated from this material has since featured prominently in

resistance breeding at the International Institute for Tropical Agriculture (IITA), Ibadan.

The most comprehensive cassava breeding programme in Africa is now based at IITA where response to virus infection is based on a scale ranging from no symptoms (1) to severe mosaic and distortion (5). A difficulty with this approach is that the incidence of infection and symptom severity are not clearly distinguished and symptomless plants could have escaped infection or they could be extremely tolerant. Consequently, a low average score could be recorded because a few plants become infected and develop severe symptoms or because many succumb but are only slightly affected.

The most promising of the IITA selections are resistant to infection, develop inconspicuous symptoms when infected and may eventually recover (Rossel, this volume). Several varieties with these characteristics have been released in Nigeria and neighbouring countries (Hahn *et al.*, 1980). Clones and seed stocks have also been distributed for use in various national breeding programmes. However, there is limited information available on the performance of the virus-resistant varieties released by IITA and on the extent to which they are being grown by farmers. A recent survey in Nigeria established that IITA varieties including some that are resistant to ACMV account for approximately 20% of the cassava area in Ondo State (IITA, 1986). The results of an even more recent questionnaire (this volume) suggest that IITA varieties are less widely grown elsewhere except in Benin, where they account for over 60% of plantings. Uptake has been limited in many other countries and several problems are listed by respondents. They include the serious shortage of planting material and of information on overall performance and suitability. There are also complaints about the flavour or bitterness of the roots, growth habit and lack of adaptation to local conditions. Moreover, only some countries have the resources to handle the seed material distributed for local evaluation and selection.

These are serious constraints that will not be easily resolved and there is an obvious need to make greater use of the material already available and to develop additional virus-resistant varieties with the required season, growth characteristics, adaptability and flavour. The general use of resistant varieties is an essential component of any disease control strategy in areas of high infection pressure. It then becomes possible to exploit the benefits of virus-free planting material and roguing in all areas, including some where this would otherwise be totally ineffective. There is also scope for using varieties that are so resistant to or tolerant of infection that roguing becomes optional or entirely unnecessary.

#### CROPPING PRACTICES AND CROPPING DISPOSITION

Planting dates, cropping practices and crop disposition can have an important influence on vector populations and virus spread. This has been demonstrated in studies on ACMV, although only limited information is available and additional work is required before farmers can be given definitive advice on the most



appropriate practices to adopt. Even then there are likely to be great difficulties in implementation due to the small size of many farms. The limited availability of suitable sites imposes serious constraints on plot size, shape and disposition. Moreover, there is frequently a need to produce a continuous supply of food for family or local consumption or for processing. This facilitates virus spread by leading to successive plantings in close proximity and in overlapping sequence. These difficulties are less acute on large commercial farms or at official establishments where the aim is to produce virus-free cuttings for distribution to farmers. In these rather exceptional circumstances, much can be done by implementing practices that decrease spread and facilitate control.

#### Planting date

Cassava is easily established from cuttings and there is usually some latitude in the choice of planting dates. The aim should be to avoid exposing plants to serious risk of infection when they are at a young and highly vulnerable stage of growth.

In the coastal districts of Kenya the main spread of ACMV occurs between mid-May and mid-July, during the early rains (Bock & Robertson, this volume). There are likely to be advantages to planting later in the year, provided that crop establishment and subsequent growth are not seriously impaired. This is also true for the forest areas of West Africa, where spread is greatest from March to July and least from August to November during the latter part of the rainy season (Fargette, this volume). Comparable information is required from other areas because changes in planting date can be introduced readily and at no great inconvenience or expense.

#### Plant spacing

There is evidence from Ivory Coast that the incidence of ACMV expressed as a percentage of the total stand is greater at low than at high plant density (Fargette *et al.*, this volume). This provides grounds for suggesting that all farmers should be advised to establish uniformly dense stands. However, additional evidence is required from other areas and on the implications of inter-planting cassava with other crops, as commonly practised in many parts of Africa.

#### Plot size and shape

A feature of experiments in the forest areas of Ivory Coast is that whitefly numbers and virus incidence are greatest in the outermost rows of plantings and especially those orientated across the direction of the prevailing south westerly wind (Fargette *et al.*, this volume). This suggests that there are likely to be advantages in planting in large, compact blocks and in orientating elongated plots along rather than across the prevailing wind direction, so as to decrease the proportion of plants in the most vulnerable peripheral areas. It may also be appropriate to discard the outermost rows in propagation plots being used to raise virus-free planting material. An alternative approach could be to adopt wind-breaks or grow barrier rows of

another crop to intercept incoming vectors and restrict virus spread, but little attention has been given to this possibility.

#### Crop disposition

Recent experiments in Ivory Coast have demonstrated spread over considerable distances by wind-borne adult whiteflies and emphasised the importance of older sources of infection located upwind (Fargette *et al.*, 1985). Young plantings are at greatest risk where there are upwind sources nearby and there is little likelihood of spread where the nearest sources are downwind and remote. Spread is also likely to be restricted by establishing sequential plantings in an upwind direction and not downwind and by orientating plantings so as to decrease the length of the interface across which spread is most likely to occur.

The opportunity of utilizing this information is obviously influenced by the patterns of land use, cassava production and land ownership. It is greatest in areas where cassava is not widely grown and there is considerable separation between plantings and also in atypical situations where cassava is established in large uniform blocks of similar age.

#### Vector control by insecticides or other means

A possible method of controlling arthropod-borne viruses is by using pesticides or other means of decreasing vector populations. Little progress has been made in controlling ACMV in this way and there is no immediate prospect of introducing safe insecticides or other procedures as a routine component of disease control strategies.

#### Insecticides

The insecticides currently available are inappropriate because the main spread of ACMV is into and not within crops and it is unlikely that incoming vectors can be killed before they have had an opportunity to transmit. Moreover, experience with cotton and other crops is that whiteflies are less readily killed than their natural enemies. This can lead to a resurgence of whitefly populations soon after treatment and the benefits are short-lived. Further limitations of the use of insecticides are that few farmers can afford chemicals or sprayers and there are likely to be hazards in using toxic chemicals on such a widely grown food crop as cassava that is frequently grown in mixed stands with maize and grain legumes.

#### Natural enemies

There are limited opportunities for biological control of whiteflies by means of natural enemies and it is likely that the main effort on these lines will continue to be against the green spider mite and cassava mealybug. As recently introduced pests they are more amenable to biological control by exotic parasites than B. tabaci.

## Vector-resistant varieties

A possible long-term approach to decreasing whitefly populations and virus spread is to breed varieties that are resistant to B. tabaci. Recent studies have identified sources of resistance and shown that resistance to vector and virus are independent attributes that can be combined in the same genotypes to give a very successful combination of features (Fauquet, this volume). Such material is likely to play an important role in future disease control strategies once suitable varieties have been developed and become widely grown.

## Mild strain protection

A possible means of alleviating the effects of virus infection is by prior inoculation with a mild strain of the same virus. The ability of mild strains to protect plants from the effects of closely related virulent ones has long been recognized. Nevertheless, virologists have shown considerable reluctance to adopt this approach to control, despite some recent successful application (Fulton, 1986). No attention has been given to the possibility of utilizing mild strains of ACMV, although they could be useful in areas where rapid spread occurs and suitably resistant varieties are not available. This suggests the need for an extensive programme of research and development to determine the feasibility of using mild strains of ACMV and the circumstances in which they are most likely to be effective.

## INTEGRATION OF CONTROL MEASURES

ACMV is a serious constraint on cassava production in many parts of Africa and there are great benefits to be gained from introducing effective control measures on a suitably large scale. However, only limited and intermittent progress has been made, even though ACMV has been studied for many years and much information is available on the various possible means of control.

There is no immediate prospect of using insecticides, biological control, mild strain protection or truly tolerant varieties and the basic approach to control in all regions involves virus-resistant varieties, crop sanitation and crop deployment. Virus-free planting material of varieties with adequate levels of resistance should be grown at suitable sites where there is no serious risk of infection and where roging can be practised effectively and inexpensively. Unfortunately, an integrated approach to control along these lines is often totally unrealistic. Acceptable varieties are not always available, or they are released in insufficient quantity, or the minimum degree of resistance required has not been determined. Furthermore, farmers may be unwilling to rogue or they have not been taught to do so. They seldom have much latitude in choice of site because of the limited land available and they have little or no control over the activities of their neighbours who may retain severely diseased plantings in very close proximity.

Despite these constraints, the available evidence suggests that ACMV can be controlled effectively in large areas of eastern Africa by releasing virus-free material of existing varieties for all new plantings and by persuading farmers to rogue effectively. Experience in Uganda, Tanzania, Kenya, Zanzibar and Malawi suggests that there are no intrinsic difficulties in adopting such an approach, provided that suitably resistant varieties are used. This suggests that the main problems likely to be encountered will relate to the production and distribution of cuttings on the huge scale required and with the need to change the current practices of vast numbers of peasant farmers.

Conditions are very different in the main cassava-producing areas of West Africa, where there are limited prospects of control by adopting virus-free planting material and roguing with the usual varieties that are now generally available. However, such measures may be effective on large commercial farms or at special propagation sites where it is possible to plant big areas synchronously and to adopt a suitable disposition of plantings with at least some degree of isolation. These circumstances are exceptional and control on typically small farms where plantings are grown in overlapping sequence and in close proximity is largely dependent on the introduction of highly-resistant varieties of the type now being developed at IITA.

The most appropriate strategy to adopt in the important cassava-producing areas of Zaire, Congo and other countries of Central Africa is unclear. ACMV is prevalent in these areas, but whether this is due mainly to the dissemination of infected planting material or to spread by whiteflies, has not been determined. Thus, the scope for using virus-free planting material and the way it should be deployed is uncertain.

## CONCLUSIONS

Much could be achieved by utilizing the existing information on ACMV and research has in some respects outstripped the ability to apply the results obtained. However, the control measures now advocated depend upon the availability of virus-free clones of suitably resistant varieties and on the willingness of farmers to utilize them effectively. This emphasises the importance of developing additional varieties and the means to introduce them on an adequately large scale. There is also a need for further information on the epidemiology and control of ACMV, especially in the many parts of Africa where no previous studies have been undertaken.

It is uncertain whether the present approach to control is appropriate in all circumstances. In some of the worst-affected areas, the adoption of mild strain protection or tolerant varieties may be a more satisfactory alternative than sanitation. Whatever the strategies advocated it will be necessary for the extension services to introduce greatly improved procedures to ensure that the measures advocated are widely adopted.

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# **SESSION B**

## **THE FACTORS**

Chairman  
Dr. FARGETTE  
Rapporteurs  
Dr. IGWEGBE  
Prof. FOUABI

PROPERTIES AND GEOGRAPHICAL VARIATION  
OF GEMINIVIRUS ISOLATES  
FROM MOSAIC AFFECTED CASSAVA

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Research on the cause of cassava mosaic disease in Africa can be divided into three phases. In the first phase (1894-1974) the disease was described and its causal agent shown to be transmissible by grafting and by the whitefly Bemisia tabaci (Storey & Nichols, 1938). The second phase began in 1975 with the transmission of virus isolates from cassava by inoculation of sap to herbaceous test plants such as Nicotiana clevelandii and N. benthamiana (Bock, 1975; Bock et al., 1978). At first it was not clear whether these isolates were the cause of cassava mosaic and they were named cassava latent virus. Later they were transmitted back to cassava, in which they caused mosaic (Bock & Woods, 1983), and it is now recommended that they should be called African Cassava Mosaic Virus (ACMV). The sap transmissibility of these isolates enabled ACMV to be further studied and characterised. The virus particles were purified, shown to be geminate and to contain circular single-stranded DNA (Harrison et al., 1977); virus antiserum was prepared and used for diagnosis and for investigating virus relationships; and the intracellular localisation of virus antigen (Sequeira & Harrison, 1982) and ultrastructural effects of infection (Horvat & Verhoyen, 1981) were determined. In the third phase, which began in 1983, the structure and bipartite nature of the virus genome were established by sequencing the two kinds of DNA molecule obtained from virus particles (Stanley & Gay, 1983). This has led to further molecular biological studies of gene function and expression, of virus replication and of variation both among geminiviruses from different hosts and among different geminivirus isolates from cassava.

MAIN CHARACTERISTICS OF ACMV

In addition to Manihot esculenta, the host range of ACMV includes several other euphorbiaceous species, especially Jatropha multifida, and six Manihot species which were infected with a West African isolate in transmission tests with whiteflies. Suspected natural hosts also include Hewittia sublobata (Convolvulaceae) in coastal Kenya and Laportea aestuans (Urticaceae) in Nigeria, but

the virus was not transmitted from these species to cassava and further tests are needed to determine their host status unequivocally. Several solanaceous plants can be infected by inoculation with sap, especially species in the genera Nicotiana and Datura. N. benthamiana develops chlorotic local lesions followed by severe systemic leaf curling and stunting with or without yellow blotches. It is the best source of virus particles for purification. D. stramonium develops chlorotic and necrotic local lesions when inoculated with some isolates, followed by systemic veinbanding and leaf distortion, and can be used for local-lesion assays. Infectivity is moderately unstable in sap, being lost in a few days at room temperature or in 10 minutes at 55 C (Bock & Harrison, 1985).

The virus particles are best purified from tissue by a method that involves extracting sap in buffer which contains a reducing agent, plus chloroform, followed by precipitation with polyethylene glycol, differential centrifugation and one cycle of sedimentation in sucrose density gradients. The particles are geminate in shape, measure about 30 x 20 nm with a waist at the mid-point of the long axis, and have a sedimentation coefficient of 76 S. They contain a protein of mol. wt about 30,000 (perhaps 110 molecules per particle) and one molecule of circular single-stranded DNA of mol. wt about 0,92 x 10<sup>6</sup>. In leaf tissue, the virus particles accumulate mainly in the nuclei of phloem parenchyma and companion cells, but also in some nuclei of cortical, epidermal and other cells. Abnormalities induced in nuclei include granular inclusions which are strongly stained by the immunogold method using antibody to virus particles (Roberts & Harrison, 1987). Rings which are cross-sections of hollow spheres of fibrillar material are also found in some infected nuclei.

Natural spread of ACMV depends on the whitefly B. tabaci, which is known to be a vector in several West African countries, coastal Kenya, India and elsewhere, but other Bemisia species have not been tested sufficiently to rule out the possibility that they too can transmit ACMV. Individuals of B. tabaci require at least 3.5 hours on infected plants to acquire ACMV, there is a latent period of at least 8 hours in the insect, and at least 10 minutes is needed for virus inoculation to healthy plants (Chant, 1958; Dubern, 1979). ACMV can be retained by infective whiteflies for about 9 days, and is retained through the moult, showing that it is carried internally. However, it is not passed through the eggs of whiteflies to progeny insects. In cassava, the virus is not seed-borne but is maintained and distributed by man in vegetatively propagated planting material.

The properties of ACMV, notably its particle shape, genome of circular single-stranded DNA, its whitefly vector and its ultrastructural effects, make it a typical member of the geminivirus group.

#### GENOME STRUCTURE AND GENE FUNCTION

The genome of ACMV consists of two circular molecules of single-stranded DNA which have similar sizes (DNA-1, 2779 nt; DNA-2 2724 nt) and known sequences, which are different except for a shared



sequence of about 200 nt, known as the common region (Stanley & Gay, 1983). Both DNA-1 and DNA-2 have genes in plus-sense (the form in virus particles) and minus-sense (the complementary form) molecules. Plus-sense DNA-1 contains the particle protein gene at the 3'-side of the common region and minus-sense DNA-1 contains three genes at the other side of the common regions, the largest of which codes for a protein of 40Kd that may be involved in replication of the virus DNA. The functions of the other two genes are not known. DNA-2 carries one gene in each sense, and one or both of these seem to be involved in the cell-to-cell movement of the virus in plants (Stanley, 1983; Townsend *et al.*, 1986). Infected plants contain polyadenylated viral RNA transcripts of five sizes, which can be assigned tentatively to the six open reading frames coding for proteins of 15, 16, 29, 30, 34 and 40Kd (Townsend *et al.*, 1985). Further details of the molecular biology of ACMV are summarised by Stanley (1985). In its genome properties, ACMV resembles three other geminiviruses which are transmissible both by whiteflies and by inoculation with sap, namely bean golden mosaic, mung bean yellow mosaic and tomato golden mosaic, all of which have bipartite genomes organised as in ACMV. These virus genomes contrast with those of leafhopper-transmitted geminiviruses, which are monopartite and consist of a circular single-stranded DNA molecule of about 3000 nt, and are organised differently from the ACMV genome (Harrison, 1985; Stanley, 1985).

#### RELATIONSHIPS WITH OTHER GEMINIVIRUSES AND AMONG GEMINIVIRUS ISOLATES FROM CASSAVA

The type strain of ACMV from western Kenya (ACMV-T) is serologically related to several other geminiviruses, all of which have whitefly vectors (Roberts *et al.*, 1984). These relationships are easily demonstrated by Elisa using polyclonal antibody to ACMV-T, or by immunosorbent electron microscopy using a range of polyclonal antisera. In contrast, ACMV is serologically unrelated to geminiviruses which are known or suspected to have leafhopper vectors and which are mostly not serologically related to one another. More recent work has shown that all the whitefly-transmitted geminiviruses tested to date are serologically related to ACMV, even though they may have no hosts in common and may occur at widely separated locations in Africa, India, the Middle-East, South-East Asia, or North-, Central- or South-America. The discovery of these relationships, together with the facts that all these viruses are transmitted by the same whitefly species, *B. tabaci*, whereas different leafhopper-transmitted geminiviruses have different leafhopper vectors and are mostly serologically unrelated to one another, suggest that the virus particle protein plays a key role in vector specificity (Roberts *et al.*, 1984).

When the nucleotide sequences of different whitefly-transmitted geminiviruses are compared, many similarities are found in the coding sequences, especially in the gene for the virus particle protein, whereas there is less homology in other genes, and little or none in the non-coding sequences of different viruses, except that a small part of the common region is similar in all geminiviruses (Hamilton *et al.*, 1984; Harrison, 1985). One result of these genome homologies is that complementary DNA (cDNA) probes

sequence of about 200 nt, known as the common region (Stanley & Gay, 1983). Both DNA-1 and DNA-2 have genes in plus-sense (the form in virus particles) and minus-sense (the complementary form) molecules. Plus-sense DNA-1 contains the particle protein gene at the 3'-side of the common region and minus-sense DNA-1 contains three genes at the other side of the common regions, the largest of which codes for a protein of 40Kd that may be involved in replication of the virus DNA. The functions of the other two genes are not known. DNA-2 carries one gene in each sense, and one or both of these seem to be involved in the cell-to-cell movement of the virus in plants (Stanley, 1983; Townsend *et al.*, 1986). Infected plants contain polyadenylated viral RNA transcripts of five sizes, which can be assigned tentatively to the six open reading frames coding for proteins of 15, 16, 29, 30, 34 and 40Kd (Townsend *et al.*, 1985). Further details of the molecular biology of ACMV are summarised by Stanley (1985). In its genome properties, ACMV resembles three other geminiviruses which are transmissible both by whiteflies and by inoculation with sap, namely bean golden mosaic, mung bean yellow mosaic and tomato golden mosaic, all of which have bipartite genomes organised as in ACMV. These virus genomes contrast with those of leafhopper-transmitted geminiviruses, which are monopartite and consist of a circular single-stranded DNA molecule of about 3000 nt, and are organised differently from the ACMV genome (Harrison, 1985; Stanley, 1985).

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migrated slightly slower in polyacrylamide gels than did that of ACMV-T. In Elisa, an Indian isolate was detected readily by its own antibody or by ACMV-T antibody, but ACMV-T was detected better by its homologous antibody than by that to ICMV. Thus ICMV and ACMV-T are serologically distinguishable using polyclonal antibody and the relative ease of detection of each virus by the heterologous antibody probably reflects the different homologous titres of the two antisera used in these tests (Lennon, Aiton & Harrison, 1987).

#### VIRUS DETECTION AND DIAGNOSIS

Geminivirus infection of cassava usually results in the production of characteristic leaf symptoms. However, complications can arise with cassava genotypes that do not react typically, with plants that are infected with a complex of two or more viruses, or that have chimaeric abnormalities resembling cassava mosaic. In addition, it may be necessary in epidemiological work to test other plant species, or even vector whiteflies, to determine whether they are carrying ACMV. For all these reasons, a definitive and sensitive test for ACMV strains is required. However, reactions with polyclonal ACMV antisera or with cDNA probes for DNA-1 of ACMV are not adequate to identify the virus because these reagents will detect many other whitefly-transmitted geminiviruses in addition. Tests with probes for the common regions of ACMV DNA may be satisfactory in principle, but it is not clear whether they are sensitive enough, and at present they require radioactive reagents which would not be readily available in many countries. The most satisfactory diagnostic procedure now available is Elisa using monoclonal antibodies to ACMV (Thomas *et al.*, 1986), a method which has given promising results in preliminary tests at Dundee and in the Ivory Coast. Monoclonal antibodies can be selected that will detect only isolates in Group A, or only those in Groups A and B, so that isolates in these two groups can be detected and distinguished (Harrison *et al.*, 1986). As yet no monoclonal antibody is available that will distinguish Group C isolates from other whitefly-transmitted geminiviruses, but Elisa using polyclonal antibody to a Group C isolate is available and partially effective. Elisa seems suitable for detecting isolates in all three groups in infected cassava, providing that young symptom-bearing leaves are used. However, monoclonal antibodies specific for Group C isolates are still needed and work is in progress to produce them.

#### CONCLUSIONS

The main conclusions to be drawn from this survey on the virus isolates that cause cassava mosaic in Africa and Asia are as follows:

1. Cassava mosaic diseases in Africa and the Indian sub-continent are caused by whitefly-transmitted geminivirus isolates and are quite distinct from superficially similar diseases found in South America.

2. No field host other than Manihot spp. and J. multifida has been identified unequivocally.
3. Geminivirus isolates from cassava fall into three clusters which have different distributions, Group A isolates occurring in West Africa and Western Kenya, Group B in East Africa including coastal Kenya, and Group C in India and Sri Lanka. Group C isolates may be best considered a separate virus.
4. Each group of isolates must be studied separately to determine its ecology and epidemiology, and its ability to infect cassava genotypes which have been bred for resistance to isolates in another group.
5. Methods are needed to detect all isolates and to distinguish those in different groups. Elisa using monoclonal antibodies is the best such method currently available for Group A and Group B isolates.

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THE ETIOLOGY OF  
CASSAVA MOSAIC IN NIGERIA

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SUMMARY

The geminivirus earlier isolated from diseased cassava (Manihot esculenta Crantz) and Ceara-rubber (M. glaziovii Müll. Arg.) at IITA by using Nicotiana benthamiana as a test plant, was consistently isolated from CMD-affected cassava from all ecological zones of Nigeria. Similar viruses were isolated from Sechium edule (Jacq.) Swartz (Fam. Cucurbitaceae) and Laportea aestuans (L.) Chew (Fam. Urticaceae), the latter a common weed that may represent an original, natural host of the geminivirus involved in cassava mosaic in Africa.

The same geminivirus was isolated from diseased cassava when N. clevelandii and N. clevelandii x glutinosa were used as test plants. With N. benthamiana the virus was isolated only from tips of diseased cassava shoots, but not from the shoots' mature leaves. Isolation attempts were unsuccessful when symptomless shoot tips on otherwise diseased cassava plants were used to prepare inoculum.

Isolates of the virus from cassava, obtained either by inoculation to N. benthamiana or N. clevelandii x glutinosa, were transmitted back to cassava by sap-inoculation, but only at low frequency. This strengthens earlier reports that the geminivirus isolated from CM-affected cassava in East Africa may represent the causal agent of CMD. Low infection incidence obtained in attempts to transmit the virus back to cassava is attributed, in part, to the virus' poor translocation within this host.

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Several improved varieties developed at IITA are available for distribution and adaptive testing in Africa. All have been virus-tested to ensure that they are free from CMD. They are being distributed only in tissue culture form to comply with plant quarantine requirements for international transfer on the continent.

## INTRODUCTION

For many years research on CMD in Africa, after pioneering work on its etiology by Storey & Nichols (1930), and Chant (1958), involving detailed vector transmission studies, did not lead to significant further information in support of the supposed virus etiology of the disease.

Whitefly-transmitted diseases, however, are generally considered to be of virus nature though usually not sap-transmissible. Indeed, with CMD, except for one report (Bock, 1978) sap-transmission was never achieved. In Brazil, however, sap-transmission was obtained with difficulty for Euphorbia mosaic (Costa & Bennett, 1950). Later this virus was also transmitted to certain test plant species and to and from other natural hosts, including french beans (Phaseolus vulgaris) (Costa, 1965). In general, sap-transmission of whitefly-transmitted disease has proven to be very difficult, except for a whitefly-transmitted disease of cucumber in Israel (cucumber vein yellowing) (Cohen & Nitzany, 1960), and cowpea mild mottle virus (Brunt and Kenten, 1973) both of which are readily sap-transmissible.

Another exception to the rule proved to be the 'golden mosaic' disease of french beans in Central and South America (Bird et al., 1975). In some cases, high sap-transmission rates were obtained, particularly when a number of conditions, including choice of the right test plant, were met (Goodman et al., 1977). This might explain why, in earlier attempts, no sap-transmission was achieved with a golden mosaic of french beans in Brazil (Costa, 1965). In general, the type of virus source plant and the choice of test plants used in the transmission experiments appeared to be among the most important factors governing successful sap-transmission of the agents.

Bock and Guthrie (1976) at the Kenya Agricultural Research Institute (KARI), Nairobi, Kenya, were the first to achieve and report on sap-transmission of an agent from CMD-affected cassava to N. clevelandii. From this test plant, it was further transmitted to various other solanaceous test plants. Attempts to transmit the isolated virus back to cassava, in order to elucidate the etiology of CMD, were initially unsuccessful. It was concluded that only a latent virus (CLV) and not the virus which caused CMD was involved (Bock et al., 1978); however, attempts to re-isolate the virus from inoculated cassava, in order to satisfy the criteria of latency, were apparently not made.

Further evidence, in support of the hypothesis of involvement of a latent virus, was thought to be provided by findings indicating that the agent could not be isolated from all CMD-affected cassava in Kenya. In particular, further isolation attempts with

materials originating from the region east of the Rift Valley were unsuccessful (Bock et al., 1978).

Later, Bock and Guthrie (1978) reported sap-transmission of CMD from cassava to cassava, when cassava test plants were used that had been established from cuttings of 2 highly susceptible, South American clones. To date, sap-transmission from cassava to cassava has not been confirmed elsewhere.

At IITA, in 1978 (IITA, 1979; Huttinga & Rossel, unpublished), a geminivirus was isolated from CMD-affected cassava by means of sap-inoculation of Nicotiana benthamiana. Prominent and characteristic symptoms developed in this test plant within 7-10 days after inoculation. From N. benthamiana, the virus was further transmitted to a number of other solanaceous test plants, including N. glutinosa, N. tabacum "Samsun NN", "White Burley", and Xanthii "n.c.", N. clevelandii x glutinosa, as well as to N. clevelandii, first used successfully for isolation of a geminivirus from CMD-affected cassava by Bock et al. (1976) (IITA, 1979; Rossel & Huttinga, unpublished). Similar or identical virus isolates were obtained at IITA from Manihot glaziovii. The latter represents a common, weedy remnant of earlier cultivation as a latex crop in the south-western region of Nigeria. It often shows virus disease symptoms which are very similar to CMD in cassava.

Also, among the population of the common weed, Laportea aestuans (Laportea aestuans (L.) Chew, formerly: Fluerya aestuans L.) virus infected plants were found from which in routine inoculations to N. benthamiana virus isolates were obtained that looked identical to isolates from CMD-infected cassava and Manihot glaziovii.

Recently also, virus isolates very similar to those from diseased cassava as far as reaction on N. benthamiana is concerned, were obtained from Secchium edule (Jacq.) Swartz (Fam. Cucurbitaceae), plants which at IITA often show severe virus-like disease symptoms (Thottappilly, unpublished).

Walter (1980), at ORSTOM in Adiopodoumé, Ivory Coast, also reported sap-transmission of geminiviruses from virus diseased cassava, and Manihot glaziovii, by using N. benthamiana as a test plant. As at IITA, his attempts to transmit the virus isolated from cassava back to cassava failed. Only Bock and Woods (1983), in Kenya, reported transmission back to cassava from N. benthamiana of the virus earlier referred to as cassava latent virus (CLV). Consequently, they concluded that CLV is in fact the virus causing CMD. Apparent difficulties in earlier attempts to transmit the virus back to cassava were attributed to low infectivity of the isolates, as success in such transmission was only achieved if semi-purified, and concentrated preparations from N. benthamiana were used.

The studies reported here were undertaken in an attempt to contribute to a better understanding of the etiology of CMD in Africa, and in Nigeria in particular.



## MATERIALS AND METHODS

Diseased cassava materials were obtained from IITA's experimental fields at its main station, at Ibadan, as well as from farmers' fields in various regions of Nigeria. During surveys, cuttings were taken from apparently diseased plants which were planted in pots and grown out in an insect-proof greenhouse.

Diseased M. glaziovii samples were taken from the natural vegetation of this weedy Manihot species at IITA, and elsewhere in the region. Diseased L. aestuans and Sechium edule plants were from IITA's site. Obviously virus-infected plants of the common weed species L. aestuans were only found twice to date. All mechanical transmission tests were carried out using a 0.01 M phosphate buffer containing 0.001 M cysteine to which mercaptoethanol (0.1%) was added just before inoculation. Carborundum (600 mesh) was used as an abrasive.

Mortars, pestles and buffer were pre-cooled and used at a temperature of about 0 C. All test plants used were grown and inoculations performed in an insect-proof greenhouse developed at IITA and providing a temperature regime which is approaching ambient conditions throughout the day, (Rossel, 1982). Healthy cassava test plants used were grown from seeds collected from the highly susceptible, locally improved, Nigerian cultivars, '60444' and from '60506', as well as from seeds that originated from South America. L. aestuans seedlings used as test plants were grown from seeds obtained from the only two infected plants of this weed species found so far. Manihot glaziovii seedlings used as test plants were grown from seeds obtained from plants found commonly growing as a weed at forest edges near IITA.

## PURIFICATION, ELECTRONMICROSCOPY AND SEROLOGY

Batches of 100 g systemically affected N. benthamiana plants, harvested 12-15 days after inoculation, were homogenized in a blender using 300 ml 0.1 M sodium citrate buffer, pH 7.0 containing 0.004 M EDTA, 0.1% thioglycolic acid and 0.1% Na-sulphite.

The homogenate was mixed with 100 ml chloroform and the emulsion was broken by centrifuging at 7,700 g for 10 minutes. To the aqueous layer was added 6% PEG (6000) and 1% NaCl and after stirring for 1 hr at 4 C, the precipitate was collected by centrifugation in the Sorvall GSA rotor for 15 minutes at 10,000 r.p.m. The pellet was resuspended in 0.01 M citrate buffer, pH 7.0 with 0.004 M EDTA, centrifuged for 10 minutes at 10,000 r.p.m. in the Sorvall SS-33 rotor and the supernatant again centrifuged for 3 hours at 78,000 g. The pellet was resuspended in 0.005 M citrate buffer, pH 7.4, with 0.004 M EDTA and centrifuged for 10 minutes at 10,000 r.p.m.

The supernatant was then layered on 10-40% linear sucrose gradients in 0.005 M citrate buffer, pH 7.4 with 0.004 M EDTA and the gradients were centrifuged for 4 hr at 25,000 r.p.m. in a Beckman SW 27 rotor. Virus fractions were collected, diluted in buffer and centrifuged for 4 hr at 78,000 g. The pellets were

resuspended in 0.005 M citrate buffer, pH 7.4 with 0.004 M EDTA and centrifuged for 10 min at 10,000 r.p.m. The virus preparations obtained were examined under a Philips EM-201 C electron microscope after staining with 2% sodium phosphothungstate (pH 7.0). Numerous geminate virus particles were seen. They were also used for infectivity studies, and for serology (IITA, 1983; Van Lent, unpublished; IITA, 1985; Thottappilly, unpublished).

Purified preparations in agar-gel diffusion tests reacted strongly with an antiserum to cassava latent virus (CLV) (Antiserum to CLV kindly provided by Dr. K.R. Bock), later renamed African Cassava Mosaic Virus (ACMV) (Bock and Woods, 1983).

Specific antisera with titres of 1/128-1/256 (in agar-gel diffusion tests) were prepared with virus isolates obtained at IITA. Antisera reacted with purified virus, as well as with crude juice of infected N. benthamiana (IITA, 1983; Van Lent, unpublished; IITA, 1985, Thottappilly, unpublished). Enzyme-linked immunosorbent assay (Elisa), developed with an antiserum obtained at IITA, is routinely used for indexing of new, improved varieties that are to be shipped from IITA as in vitro cultures to collaborators in several countries in Africa. In cassava, the virus could be detected by this means up to dilutions of 1/625-1/3,125 (IITA, 1985, Thottappilly, unpublished).

No indications have been obtained to date that other virus diseases, or types of the virus that are serologically undetectable with this antiserum (Sequeira and Harrison, 1983) occur in cassava in Nigeria.

#### SYMPTOMS IN TEST PLANTS

When using the "broad-spectrum-virus-susceptible" test plant, N. benthamiana, in sap-transmission experiments with diseased cassava samples from plants showing typical symptoms of CMD, 100% of the inoculated test plants usually developed severe disease symptoms within 2-3 weeks after inoculation (IITA, 1979; Huttinga & Rossel, unpublished). Symptoms initially consist of a striking epinasty and twisting of the newly developing leaves. This usually sets in at about 7 days after inoculation. Symptoms in N. benthamiana at later stages are quite severe and best described as a combination of leaf curl, "little-leaf" and yellow mottle.

N. clevelandii, N. glutinosa, N. clevelandii x glutinosa, N. tabacum vars. White Burley, "Samsun NN", and "Xanthii n.c", on inoculation from infected N. benthamiana, show the same type of characteristic epinasty and twisting not earlier seen with any other virus under study at IITA; however, in later stages, plants show less symptoms. In addition to N. benthamiana, inoculations of various Nicotiana species with samples taken from diseased cassava resulted in infection of N. clevelandii and N. clevelandii x glutinosa only.

## EXPERIMENTS ON ETIOLOGY AND GEOGRAPHICAL DISTRIBUTION

- I. In a typical experiment, 20 field samples, consisting of 10 growing tips from healthy-looking plants, randomly collected from IITA's cassava germplasm collection were inoculated to 5 N. benthamiana each. With only a few exceptions, each N. benthamiana plant in the series inoculated with samples taken from diseased plants developed characteristic and severe symptoms, whereas none of the test plants became infected in any of the series inoculated with materials from healthy-looking plants.
- IIa. In another typical experiment, 14 shoot-tip samples were taken from a newly established multiplication field of one of IITA's improved, moderately CMD-resistant cassava clones ("4(2)1425"). Of these 14 samples, 10 represented shoot tips from symptomless apical or lateral branches on otherwise symptomatic plants. Four represented shoot tips from still actively symptom-developing branches. With the four diseased shoot tips, transmission was obtained in all four series, whereby effectively all N. benthamiana plants developed prominent and characteristic symptoms. In contrast with the latter, only in one series of the 10 inoculated with healthy-looking shoot tips taken from otherwise diseased plants, two N. benthamiana plants, of the five inoculated, developed the characteristic symptoms.
- IIb. Similarly, in an attempt to isolate ACMV from cassava plants which had been subjected to 9 weeks of heat treatment (45 C) in a controlled environment cabinet, no transmission was obtained in each of three series of 10 N. benthamiana inoculated with growing tips of three plants treated in this manner. No CMD symptoms were observed in the upper leaves on the plants treated in this manner by the time the growing tips were collected for inoculation.
- III. Parallel to the last experiment, young, healthy-looking shoots, taken from plants of the same clone ("4(2)1425") which at least initially had all been affected by CMD, as concluded from symptoms on the lower parts of these plants, were planted in the greenhouse and monitored for further disease expression. New plants were established from five of such young shoots, which each measured approximately 20 cm of length.

Four of the five plants thus obtained remained symptomless throughout the period of observation (9 months); whereas one of these plants developed symptoms in subsequent new growth; however, in the first three consecutive leaves only. Additional new leaves developing on this plant all remained symptomless. After 9 months of observation all five plants were cut back, cloned and new cutting plants established from them. The growing tips of all five plants were tested by inoculation on N. benthamiana. All proved to be virus-free, as concluded from absence of symptoms in the test plants.

All plants obtained from cuttings taken from the four cassava plants that had not developed any symptomatic leaves remained symptomless. Only three cuttings (all taken from one of the five plants which had originally developed three symptomatic leaves) developed symptoms after sprouting. These three cuttings were the ones taken from the basal part of the stem of this plant. This proves that the virus tends to be non-systemic in cassava, and that cassava shows a strong tendency to grow away from infection.

The cut-back mother-plants were also kept for further observation of their subsequent new growth. The sprouting mother plants (all developing two or three new shoots: one shoot out of three on one plant, two shoots out of three on a second plant, and two shoots out of two on a third plant) developed symptoms on varying numbers of new leaves. Of the remaining two plants all shoots remained symptomless. This shows that the virus may stay "dormant" in some dormant buds and that it may be absent from others on the same cuttings.

IV. In an experiment aimed at obtaining an impression of the geographical distribution of CMD in Nigeria, with particular regard to the possibility of isolating a gemini-virus from diseased plants by means of N. benthamiana, cuttings were taken from diseased plants grown in various ecological zones of Nigeria. Samples from typically diseased cassava were collected from the southernmost part of the country (Port Harcourt region), with a humid lowland tropical climate, up to as far north as the region around Kano with a typical Sudan-savanna climate. In all cases, with inoculum taken from diseased shoots that had developed on the greenhouse-planted cuttings, transmission was obtained to N. benthamiana and characteristic symptoms developed in every plant of each series.

V. Attempts were also made to isolate the virus from older leaves taken from diseased plants of the highly susceptible cultivar, "60444" which usually shows severe and typical symptoms of CMD on every leaf. In this experiment, no transmission was obtained to N. benthamiana with leaves that were fully expanded at a position approximately 10 cm below the apex.

The virus was readily isolated from apex pieces from those plants, consisting of the upper 1 cm, including the young, unexpanded leaves; however, from the young, still expanding and immature leaves in the apex region (see Table 1), this was only achieved with difficulty.

VI. In routine inoculations from virus-diseased weeds to N. benthamiana, inoculation from two separate L. aestuans plants resulted in development of characteristic, "CMD-like" symptoms which were indistinguishable from those obtained with diseased cassava materials. The two L. aestuans isolates. in further host range studies, were

transmitted to the same solanaceous test plants as cassava isolates and symptoms observed were identical.

All attempts to transmit the isolates from L. aestuans, as well as isolates from cassava, to seedlings of L. aestuans, have failed to date.

Similarly, no transmission back to Sechium edule was obtained with isolates obtained from this Cucurbitaceous species.

- VII. In an experiment where 10 diseased and 10 healthy-looking samples, randomly collected from "CMD-resistant", improved cassava clones (e.g. TMS 30573, TMS 50395, TMS 30001) were tested on N. benthamiana, characteristic symptoms developed in series inoculated with samples taken from shoots with actively developing symptoms. These results were similar to those obtained earlier with samples from CMD-susceptible clones.
- VIII. The geminivirus isolated from CMD-affected cassava was transmitted back to cassava, though at low frequency (IITA, 1984, 1985; Rossel, unpublished; Rossel and Thottappilly, 1984, 1985). Best results were obtained with seedling populations of South American origin (see Table 2). One seedling clone (No. 207) was selected, that has consistently shown approximately 10% infection incidence on inoculation with crude juice from infected N. benthamiana. This seedling clone has been grown in IITA's virology greenhouses, under strictly insect-proof conditions only. Upon request, it can be made available to interested researchers as in vitro cultures.
- IX. Strong differences were observed among Nicotiana species when compared for susceptibility and/or sensitivity to the geminivirus isolated from cassava by means of N. benthamiana (see Table 3).
- X. When tested with an antiserum to a cassava-isolate, the isolates obtained from M. glaziovii, L. aestuans and Sechium edule all proved serologically closely related to this virus as concluded from curved, and confluent (specific) precipitation bands observed in agar-gel diffusion tests with purified and concentrated virus preparations obtained from isolates from these species at IITA. Spur formation was only observed with isolates from Sechium edule.

Table 1. Influence of leaf position on transmissibility of geminivirus from CMD-affected cassava to N. benthamiana.

Source of inoculum	No. of plants inoculated	No. of plants infected
Growing tips	a. 5	a. 5 out of 5
Young, unexpanded leaves	a. 5	a. 1 out of 5
Appr. 2 cm below apex	b. 5	b. 2 out of 5
Fully expanded leaves	a. 5	a. 0 out of 5
Appr. 10 cm below apex	b. 5	b. 0 out of 5

Table 2. Summary of results of inoculations from N. benthamiana infected with geminivirus from CMD-affected cassava 1), back to cassava.

Origin of cassava seedlings used as test plants	Number of seedlings inoculated	Number of seedlings infected 2)
"60444" (local cultivar	20	0
"60444"	18	0
"60506 O.P."3) (local cultivar)	15	0
"60506 O.P."	20	2
"60506 O.P."	20	0
"60506 O.P."	50	1
"Latin America" (mixture of various sources)	57	7

- 1) Virus isolate originally obtained by inoculation of N. clevelandii x glutinosa.
- 2) All plants developing symptoms positive in back-tests on N. benthamiana.
- 3) Seed obtained from open-pollinated plants.

Table 3. Comparison of Nicotiana species as regards relative efficiency of becoming infected when inoculated with geminivirus from cassava, maintained in N. benthamiana.

		Inoculated/Infected
15	<u>N. benthamiana</u>	15/15
20	<u>N. tabacum</u> "Samsun NN"	15/15
5	<u>N. glutinosa</u>	5/5
10	<u>N. tabacum</u> "Xanthii n.c."	17/20
10	<u>N. clevelandii</u> x <u>glutinosa</u>	4/10
5	<u>N. clevelandii</u>	2/5
5	<u>N. tabacum</u> "White Burley"	1/10
5	<u>N. megalosiphon</u>	0/5
5	<u>N. rustica</u>	0/5

\* Inoculum prepared from infected N. benthamiana.

## CONTROL

Earlier studies at IITA by Terry on the epidemiology of CMD (IITA, 1978, 1979, 1980; Terry, unpublished) have shown that the infection pressure of this virus disease at Ibadan is so high that, insusceptible varieties, infection incidence approaches 100% within 3 months of planting in May, during the early growing season.

In resistant cultivars, however, e.g. "30395", infection does occur, though typically does not exceed a certain low percentage (appr. 20% in case of "30395"). In spite of vegetative propagation and high infection pressure, infection percentage of resistant cultivars stays at low levels. Taking results of earlier-mentioned cloning studies on plants that had obviously outgrown infection into consideration, it seems to represent an equilibrium situation resulting, on the one hand, from primary infected plants outgrowing infection (self-elimination) and, on the other hand, from reinfection of a certain, low, number of plants. Thus, only a fraction of the cuttings taken from infected plants of resistant clones result in primary infected plants in any newly planted crop. Such roguing in these varieties is not necessary. In fact, it seems to be a futile effort, as reinfection in such clones, unlike in susceptible clones, takes place to a very limited extent only. Furthermore, many plants that initially show symptoms, outgrow the disease. Low reinfection rates are thought to be a manifestation of the resistance mechanism operative in such varieties.

## CONCLUDING REMARKS

Studies at IITA confirmed, like those reported from Kenya by Bock and Woods (1983), with 'cassava latent virus' (CLV), that the geminivirus isolated from CMD-affected cassava in Nigeria can be transmitted back to cassava, thereby causing symptoms indistinguishable from CMD as observed in the field. Although transmission back to cassava was obtained with some difficulty, it was obtained by inoculating the cassava test plants with crude juice of N. benthamiana. This finding leaves little doubt as to the disease-inciting nature of CLV, especially because, in our case, transmission was obtained to seedlings obtained from true seed grown under strictly insect-proof conditions. Furthermore, transmission was obtained by sap-inoculation with crude juice of infected N. benthamiana. The seed was obtained from cultivars of South American, as well as local, Nigerian origin.

Successful transmission back to cassava seems to be dependant on genotype (susceptibility ?) of the cassava test plants used; however, differences in infectivity among isolates may also play a role. Bock and Woods (1983), in their transmission studies (besides performing their inoculations with semi-purified and concentrated preparations from N. benthamiana) used test plants which had been obtained through clonal propagation of a cassava cultivar of South American origin ("N Meex 55").

N. benthamiana, which had earlier proved to be a susceptible isolation host, (IITA 1979; Huttinga & Rossel, unpublished), apparently is a highly sensitive test plant with which at IITA, 100% infection incidence is readily obtained, provided inocula are prepared from materials taken from growing tips, and when optimum conditions for inoculation are adhered to. Interestingly, in all cases, with infected materials obtained from various regions of Nigeria, transmission was obtained to N. benthamiana, whereby the same characteristic symptoms developed in this test plant species. Bock and Guthrie, using N. clevelandii, did not obtain transmission to this species with infected materials originating from the coastal region of Kenya which indicated that N. clevelandii possibly differentiates between different "strains" of the virus.

Transmission was obtained only when tips from apical and lateral shoots displaying active symptoms were used as inoculum, whereas no infection was obtained with growing tips of symptomless plants or symptomless shoots on infected plants (II). This suggests a restricted occurrence of the virus in infected cassava. The latter seems to be strongly supported by the results obtained in our cloning experiment (III). It seems that the apparent poor systemic distribution of this virus in the cassava plant is in some way connected with difficulties encountered in obtaining transmission back to cassava of the geminivirus obtained from it.

Chant (1958) found that by using the whitefly vector of CMD, the disease could only be transmitted to healthy cassava plants when inoculation was performed on the youngest leaves, which seems to concur with findings reported here. The geminivirus associated with CMD apparently occurs in the growing tip region of apical and lateral shoots only, including (dormant) buds.

Sap-transmission of such viruses, which are thought to be essentially phloem-restricted, besides being governed by their presumably unstable nature, may well also be dependant on the possibility of tissues other than phloem becoming infected, from which further transportation to the phloem is thought to occur.

In conclusion it can be said that, both in East and West Africa, "Koch's postulates" have been adequately fulfilled with regard to the geminivirus isolated from CMD-affected cassava. Our studies have also shown that this geminivirus is closely related to geminiviruses occurring in, and thought to be responsible for, severe virus disease symptoms observed on M. glaziovii, L. aestuans, and Sechium edule, in Nigeria.

Whether L. aestuans may be considered as an original natural host of this virus, for which in line with the name under which the disease has been known for several decades, the name African Cassava Mosaic Virus (ACMV) has been proposed (Bock and Woods, 1983), also depends on whether this plant species originates in Africa (According to Dalziel (1937) and Uphof (1968), Laportea aestuans (L.) Chew is an African species). M. glaziovii and Sechium edule which, like cassava, are comparatively recent introductions to Africa, are certainly not.



N. benthamiana, first used as a test plant for this virus in studies of CMD at IITA in 1978 (IITA, 1979; Huttinga and Rossel, unpublished), apparently does not differentiate among possibly different types or strains of this virus in Nigeria, as found for N. clevelandii in Kenya (Bock et al., 1978). Thus, it appears that N. benthamiana is a suitable test plant for ACMV. It is being used at IITA for indexing purposes, particularly as it appears to be a very sensitive test plant for this virus. It is possibly the most sensitive of all Nicotiana spp. tested so far.

Several high-yielding varieties which combine superior agronomic characteristics with moderate to high levels of resistance to CMD, bacterial blight, anthracnose, and other important biological constraints, have been developed at IITA (Hahn, 1978; Hahn, Terry, Leuschner, Akobundo, Okali and Lal, 1979; Hahn, Terry and Leuschner, 1980). In addition to being widely grown in Nigeria already, several of these varieties have been sent to national programmes in Africa for adaptative testing in various agroecologies. Such materials have all been put in tissue culture form (IITA, 1981; Frison and Ng, unpublished; Frison, 1981), and have been virus-tested in order to comply with quarantine requirements governing the international transfer of vegetative plant parts of this crop species across the African continent.

The infection pressure of CMD in Nigeria's major growing areas in the humid and subhumid southern region is so high that, in susceptible varieties, effectively 100% infection incidence is reached within the first 3 months of planting. Therefore, genetic resistance to CMD is a prerequisite to the development of new, improved varieties.

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THE ROLE OF BEMISIA TABACI GENNADIUS IN THE  
EPIDEMIOLOGY OF ACMV IN EAST AFRICA.  
BIOLOGY, POPULATION DYNAMICS, INTERACTION WITH  
CASSAVA VARIETIES

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#### INTRODUCTION

Experimental work in Kenya (Bock, 1984) has shown that the African Cassava Mosaic Virus (ACMV) can cause yield losses in cassava reaching 80%. Yield loss is greatest when infection takes place early in the growing season, and is less important later, after the roots have formed. At all times, however, it causes loss of planting material because the planting of infected cuttings means maximum yield loss and early and rapid spread of the virus through the crop.

A major and successful programme aimed at controlling losses from ACMV by the breeding of resistant cassava varieties was begun by Dr H.H. Storey at Amani in Tanzania in 1937. This programme continued for 21 years and the results have been summarised by Doughty (1958).

In 1974, as part of an ODA Sponsored Crop Virology Project, based at KARI, Muguga, Kenya, Bock and Guthrie (1978, 1983) began a study of the epidemiology of ACMV throughout Kenya. At the Kenya coast they found a marked difference in the incidence of ACMV, which became much more important in the wetter areas to the South. The author joined the project in 1982 to study the role of the insect vector in more detail.

#### BIOLOGY AND POPULATION DYNAMICS

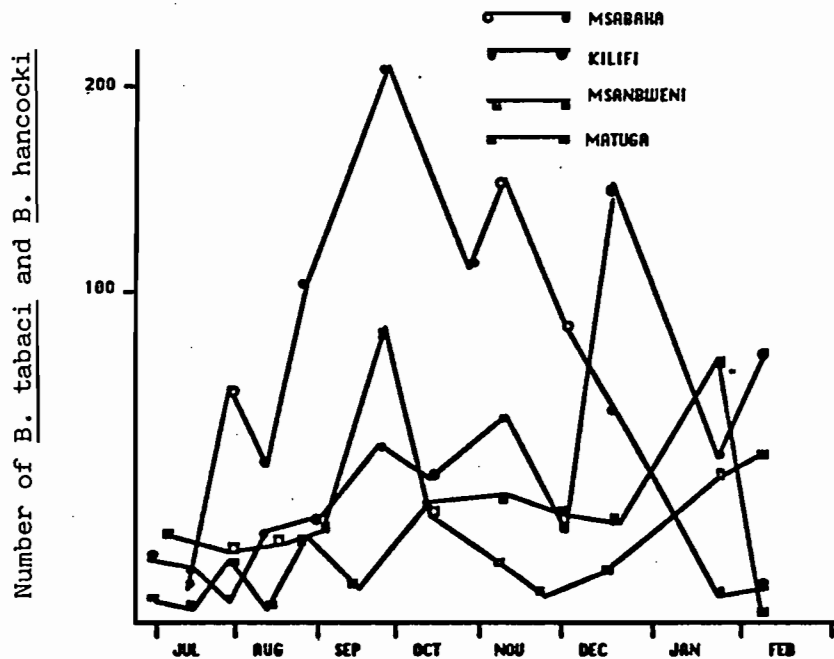
The work was done at the Kenya coast from 1982 to 1984. Because of the difficulties of identifying adult whiteflies, emphasis was placed on the counting of last instar "puparia" on the underside of leaves, which could readily be identified. The host range of the vector was investigated and 178 plant species were examined. Bemisia tabaci was found on 83 of them and while another 36 species of whitefly were found, B. tabaci and its very close relative B. hancocki Corb. were the only whiteflies recovered from

cassava. Most of the plant species were collected in the close vicinity of cassava fields and were weeds and herbs. It was noted that B. tabaci was not found in any more natural vegetation communities examined.

The presence of B. hancocki on cassava was a complication. Mound (1965) said that in Nigeria B. hancocki was not involved in the transmission of ACMV, but, considering that in some localities where ACMV infection pressure was high almost 40% of the whitefly present on cassava were B. hancocki, this fact needs to be confirmed using modern techniques.

Population studies were done by counting numbers of puparia on leaves, and also by the use of yellow sticky traps to count numbers of adults moving both outside and inside the canopy of the crop. The counting of puparia (Fig. 1) showed that there was an increase in egg laying during the main rains, in July, when new plantings or old plants began to produce new leaves.

Fig. 1. Totals of the immature stages, including eggs, of B. tabaci and B. hancocki combined at four sites on sampling dates from June 83 to Feb. 84.



This would have been caused by dispersal of adults from relatively unattractive old plants into the rapidly expanding habitat of immature leaves. At this time the incidence of ACMV was very high (Fig. 2), and this supports the finding of Storey and Nichols

(1938) that *B. tabaci* adults are able to survive successfully on mature cassava leaves, but can only transmit ACMV through immature leaves. At this early stage in new plantings, all the leaves present were susceptible to ACMV and new leaves were being produced at a rate of one every 2-3 days (Fig. 3).

Fig. 2. The incidence of ACMV in coastal districts, Kenya, 1981-84: total infections at all sites, by month.

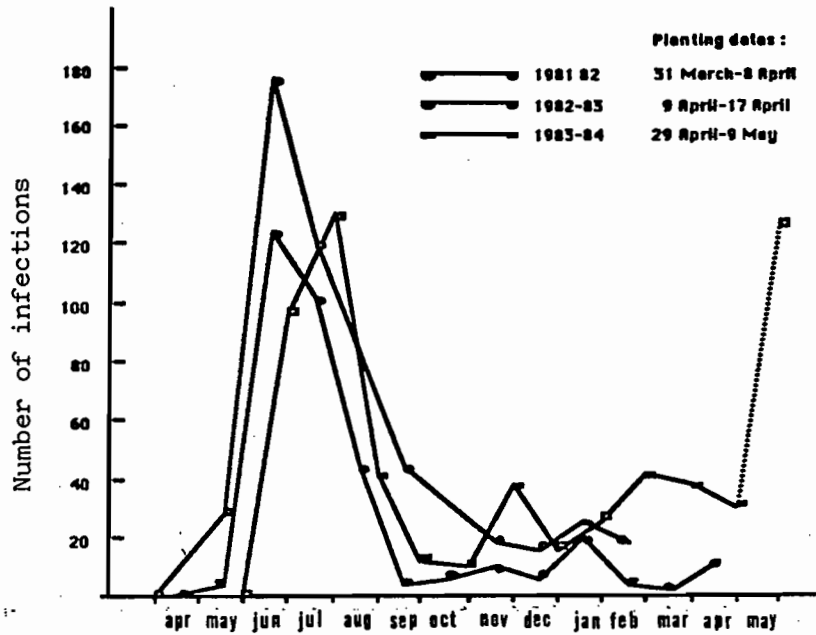
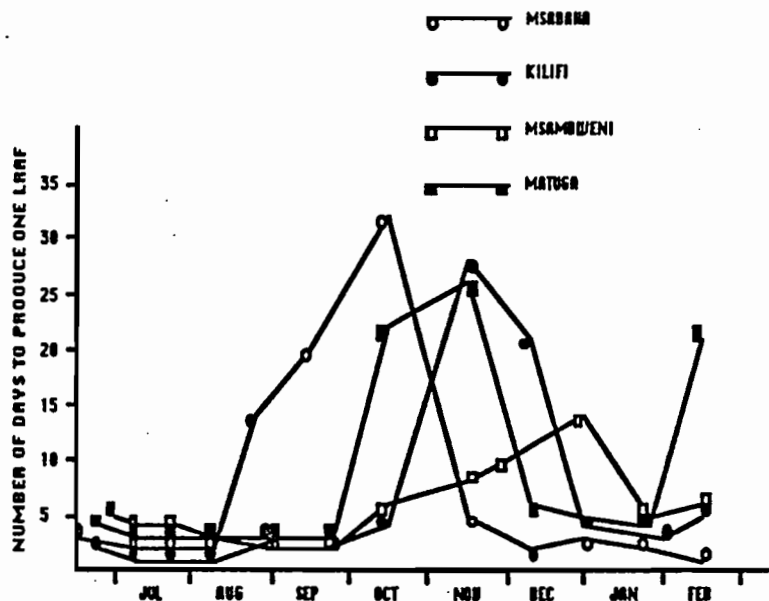
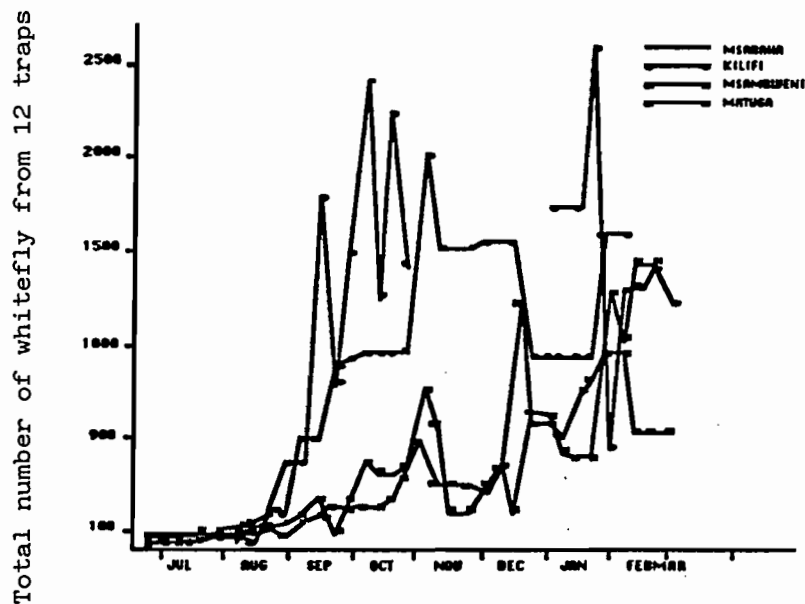


Fig. 3. The rate of growth (number of days taken to produce one leaf) of the variety 5543/156 at each of the four sites on sampling dates from June 83 to Feb. 84.



As the season progresses and the plants begin to mature, a lower proportion of the leaves are immature, and so fewer are suitable for the transmission of ACMV; also, fewer leaves are being produced at longer intervals (Fig. 3). In September and October, although ACMV incidence drops, the population of whiteflies increases rapidly (Fig. 4) helped by the dryer conditions. At the Kenya coast there is a short rains period during November and December and here again the number of immature leaves rises, and the number of days to produce a leaf is reduced (Fig. 3). The number of whitefly adults also drops (Fig. 4) because of the rainy conditions, but the incidence of ACMV rises (Fig. 2).

Fig. 4. The weekly totals of adult whiteflies recorded on yellow sticky traps at each of the four sites from June 83 to Feb. 84.



#### THE EPIDEMIOLOGY TRIALS

A series of epidemiology trials were done along the length of the Kenya coast from 1978 to 1984. The trials, which were randomised blocks with 4 replications and 12 varieties, included both local and exotic varieties, showing a wide range of susceptibility to ACMV (Table 1). Plants infected with ACMV were rogued from the trials each time they were visited, at about three weekly intervals throughout the season. In general, conditions were drier in the North and wetter in the South, with a higher incidence of ACMV in the South. The most obvious factor in this pattern was the higher rainfall in the South, but it seemed that a number of factors were involved, and that the higher rainfall meant better crops so that in the South there were more people planting more

cassava. There was a shorter distance between cassava fields and so it was easier for infected vectors to move from one field to another and, perhaps most important of all, many more farmers were planting out infected cuttings.

#### THE IMPORTANCE OF THE CASSAVA VARIETY

The epidemiology trials showed the importance of defining the cassava variety being used in any experimental work. In Kenya there exists a complete range of susceptibility to ACMV, (Table 1). Chant (1958) and Seif (1981) have demonstrated that higher rates of infection are obtained the greater the number of infected whitefly present. Relating their results to field conditions, it seems that for some varieties very few infected whiteflies are needed.

Table 1. Incidence of ACMV, southern plots, 1983/84. The number of plants rogued for ACMV from planting to harvest in the epidemiology trials, expressed as a percentage of the number of plants which grew.

Mrima <sup>+</sup>	Millani <sup>+</sup>	Msambweni Coconut N.	Msambweni Dev. Coy.	Mwabungu <sup>+</sup>	Waa	Matuga
4763 17.5	4763 27.5	4758 72.0	4772 17.5	4750 43.3	4770 33.3	4763 38.9
			4765 15.0			
4750 7.5	4748B 13.5	4750 67.5	4755 13.9	4770 33.3	4750 27.5	4758 37.3
			4760 12.5			
4770 5.0	4750 7.5	4770 61.5	4770 10.0	4763 23.3	4763 15.4	4750 36.6
			4750 10.0			
4738B 4.0	4748A 5.5	4748A 48.0	4758 9.4	KIB 18.3	KIB 9.4	4770 34.0
			4748A 7.1			
4758 2.5	4758 2.5	4748B 46.7	6332 5.5	4758 16.7	4705 3.8	4748A 20.6
			4754 5.1			
GUZO 2.5	4770 2.5	4763 45.0	4763 5.0	4748 15.0	4748A 0	4748B 6.4
			A.VAL 5.0			
4748A 0	KIB 2.5	KIB 30.7	4705 5.0	4762 10.0	4748B 0	4762 5.0
			4748B 4.3			
156 0	CHO 0	CHO 7.5	KIB 3.3	CHO 1.6	4762 0	KIB 3.3
MWA 0	GUZO 0	GUZO 5.4	4762 2.6	156 0.8	4758 0	GUZO 0
			4771 2.5			
KIB 0	27 0	156 2.2	4764 2.5	GUZO 0	CHO 0	CHO 0
			CHO 0			
CHO 0	MWA 0	MWA 0	12199 0	MWA 0	MWA 0	MWA 0
			12200 0			
34 0	34 0	27 0	MWA 0	27 0	156 0	156 0
			KAS 0			
27 0	156 0	34 0	156 0	27 0	0	27 0
			27 0			
(1) 29.4.83	29.4.83	30.4.83	9.5.83	14.5.83	16.5.83	6.5.83
(2) 16.11.83	16.11.83	21.2.84	15.5.84	17.11.83	22.2.84	14.5.84



The results indicate that there is no absolute resistance, although a few of the varieties available have still to show a single infected plant. This study has been continued during 1984-87 in a germplasm collection containing 140 entries, 70 of which are original Amani hybrids.

In Fargette et al., (1985) investigating the spread of ACMV in a cassava crop in Ivory Coast, the comment is made that the rapid spread of ACMV into healthy cassava fields differs from the effect found in Kenya, where spread was as low as 2% per year (Bock and Guthrie, 1977; Bock, 1984). If this work were repeated, it would be essential to define the varieties used and to place them on a ladder of susceptibility to get comparable results. Present knowledge would allow a choice of varieties to give 1% or nearly 100% infection in a season, assuming the presence of known numbers of infective whitefly.

#### DISCUSSION

The transmission of ACMV by B. tabaci depends entirely on the presence of infected plants and it has been observed that quite high populations of B. tabaci do very little mechanical damage to the crop and do not seem to cause appreciable loss of yield. B. tabaci only becomes important when infected plants are present and immature leaves allow the transmission of the virus. Elimination of infected plants must therefore be a primary object. The results of the epidemiology trials (Table 1) show the differences in the infection pressure between localities and also the differences in susceptibility between the varieties included. In localities where infection pressure was high it was not possible to keep susceptible varieties clean, even with regular roguing, but at these localities some varieties showed no sign of infection and the infection in many varieties was held to an acceptable level. It must be remembered that in less than six months after planting quite reasonable yields can be obtained, so plants rogued out for ACMV need not be wholly wasted. Roguing must be done regularly and rigorously in a susceptible variety, but in a resistant variety it may be done less frequently when infection is less likely. The consideration of likely infection pressure and the susceptibility of the available varieties should allow a choice to be made of suitable varieties to grow in any particular locality.

As data from epidemiology trials accumulated, it became obvious that the location of the trials was very important. The trials which were close to infected local cassava tended to suffer most from ACMV, while isolated trials suffered least. This was best illustrated by the two trials at Msambweni, which were about three kilometres apart. Msambweni Coconut Nursery was surrounded at close range by infected cassava, while Msambweni Development Company was in the middle of a coconut plantation, about one kilometre from the nearest local cassava. The difference is striking and similar results were obtained in other places.

From the information collected, it would seem that the best advice which can be given to farmers is to choose a variety of cassava with reasonable characters and as high a degree of

resistance to ACMV as possible and then to plant it as far away from other cassava as is feasible.

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MONITORING BEMISIA TABACI POPULATIONS IN CASSAVA  
FIELD COUNTS AND TRAP CATCHES

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INTRODUCTION

Apart from man himself, the whitefly Bemisia tabaci (Gennadius) (Homoptera: Aleyrodidae) is the sole known vector of the virus that causes African Cassava Mosaic Disease. This disease, recorded virtually everywhere in sub-Saharan Africa where cassava is grown, is responsible for crop losses ranging from 5-95%.

In parts of West Africa, infection levels are so high that many farmers think the appearance of the infected plant is the crop's natural condition. B. tabaci is widely distributed throughout the tropics and its pest status has focussed much attention upon it in other parts of the world and on other crops affected by it. However, its ecology in West Africa and its effect on cassava has been comparatively little studied. In an attempt to rectify this deficiency, and by so doing shed light upon the epidemiology of the disease, an intensive field study of the insect in a young cassava crop was undertaken.

This work formed part of ORSTOM's continuing programme "Etude de la Mosaïque du Manioc" at Adiopodoumé, Ivory Coast. Some of the results from this study are presented and discussed here, in particular the phenology of the adult and nymphal populations over the study period or season and aspects of the flight behaviour of the adult, as monitored by trap catches.

METHODS

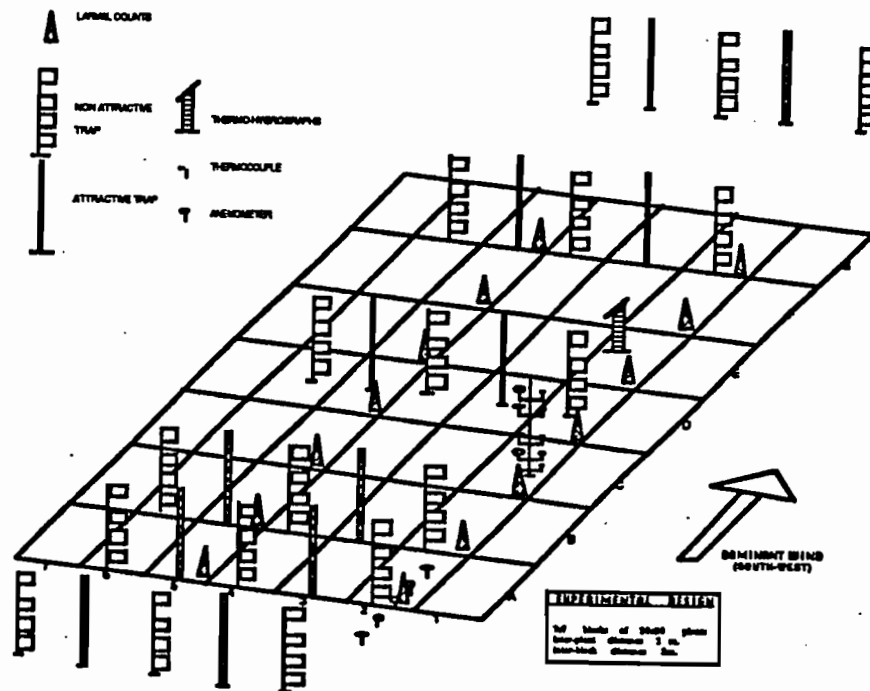
The experimental field site consisted of 1/2 ha plot divided into 7 x 7 blocks, each planted with 10 x 10 cassava plants, clone CB. The plot was aligned on a SW-NE axis and thus was orientated such that the prevailing SW wind crosses the upwind field border perpendicularly. The field was planted with virus-free cuttings on 23 Nov. 1985. Numerous other, diseased, cassava fields were located nearby, the nearest approximately 100 m to windward.

Each week between 10 December 85 and 24 April 86, adult whiteflies were counted in the field. Ten plants along the diagonal of each block were sampled, 490 plants in all. The number of adults on the undersides of the top five open leaves of one shoot were counted: earlier work had shown that the majority of the adult whiteflies were restricted to this portion (Fargette *et al.*, 1987).

The nymphs were sampled weekly also, by counting the numbers found on all the leaves of one shoot on each of 14 previously selected plants, one per block. No attempt was made to distinguish between the instars or "pupae". Several parameters of the cassava plants were measured: number of shoots per plant for those plants used in the insect counts, crop canopy height, number of new leaves per growing tip per week and number of plants showing disease symptoms.

The flight activity of adult whitefly was monitored using two trap types. One was a non-attractive sticky trap, comprising 25 cm of polyethylene held on a square wooden frame. Four such frames were mounted vertically on a bamboo pole at four heights: 0.5, 1.25, 2.0 and 3.25 m. In all, 18 such poles were used. These arrays were deployed in 3 lines of 6, orientated such that the sticky surface faced the prevailing SW wind (Fig. 1).

Fig. 1. Position of whitefly traps.



The second trap type, an attractive sticky trap, was made from a 4 m length of cylindrical PVC tubing, 10 cm diameter. Along the length of each tube, at 20 cm intervals, were stuck ten 10 cm wide yellow strips. The bottom strip was sited 25 cm from the soil surface, the top at 2.95 m. The yellow bands were marked off into eight sections corresponding to the eight major points of the compass. Over the yellow bands were affixed strips of transparent cellophane impregnated with glue. Twelve of these arrays were mounted vertically and deployed as shown in Fig. 1. The attractive sticky traps were used in two ways: to assess flight activity through the season they were set out for a 24 h period each week, while to assess flight activity within a 24 h period, the sticky strips were changed at 2-hourly intervals during daylight hours.

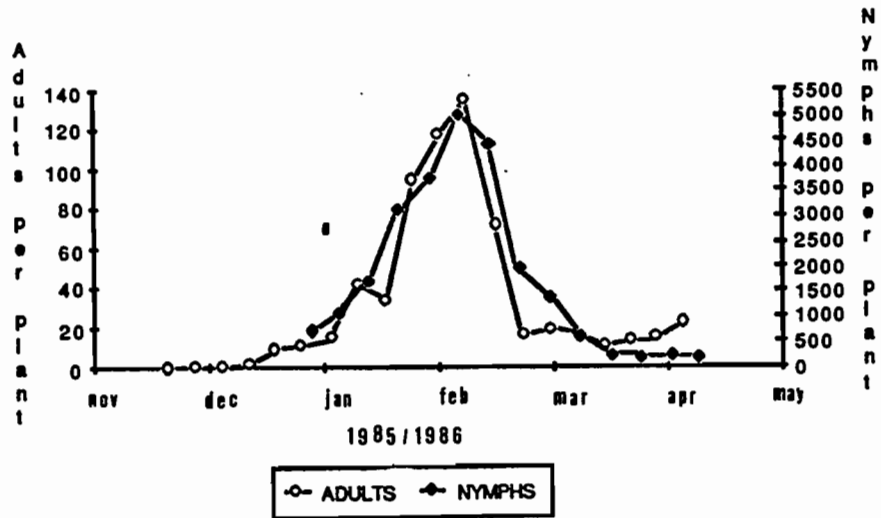
Numerous meteorological parameters were measured:

1. Nine anemometers were deployed in a vertical and an horizontal series (Fig. 1). The horizontal series were aligned on a SW/NE axis, 4 and 2 m upwind of the field, the field margin, and at 2, 4 and 25 m inside the field. Those deployed vertically were placed at heights of 50, 125, 200 and 325 cm, within the field.
2. Four thermocouples were placed within the field at height of 50, 125, 200 and 325 cm, with a fifth placed at a depth of 1 m in the soil. Data from the both sets of equipment were recorded every 2 minutes over the observational period on a data logger.
3. Three thermohygrographs were installed at heights of 50, 125 and 200 cm within the field.
4. Rainfall data was obtained from a rain-gauge located 300 m from the field plot.

## RESULTS

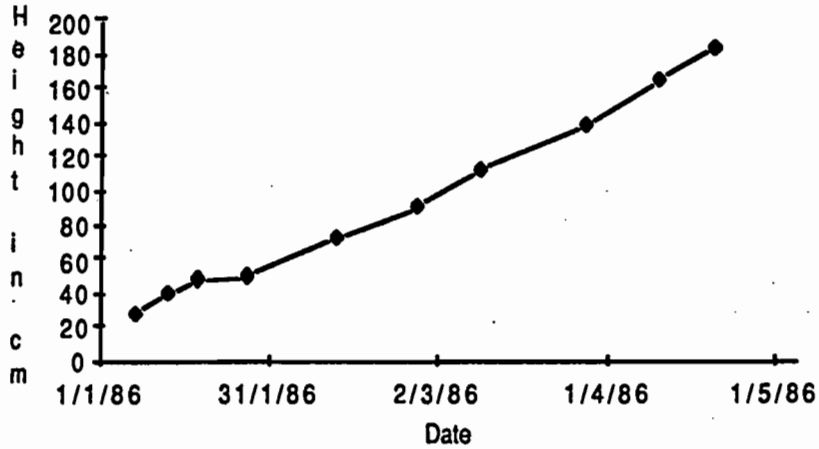
Fig. 2 shows the development of the adult and nymphal field populations over the observational period. An initial phase of slow population growth is followed by an exponential increase, which begins early January and peaks at the end of February. This is immediately followed by an even more rapid decline in early March, after which the population stabilises at a low level for the remainder of the period of observation. Also notable is the synchronicity of the adult and nymphal populations.

Fig. 2. Field count totals of adults and nymphs.



The growth of the cassava crop, as measured by plant height over the same period, displays a linear relationship (Fig. 3).

Fig. 3. Growth of cassava crop.



The distribution of adult whiteflies within the field (Fig. 4) reveals that, over much of the observational period, there is a concentration of the population towards the upwind field border.

Figure 4. Distribution of adults within field.

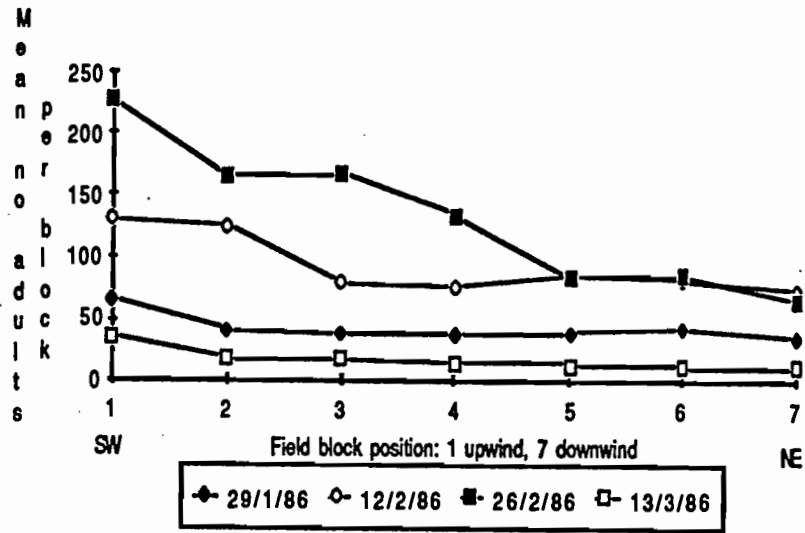
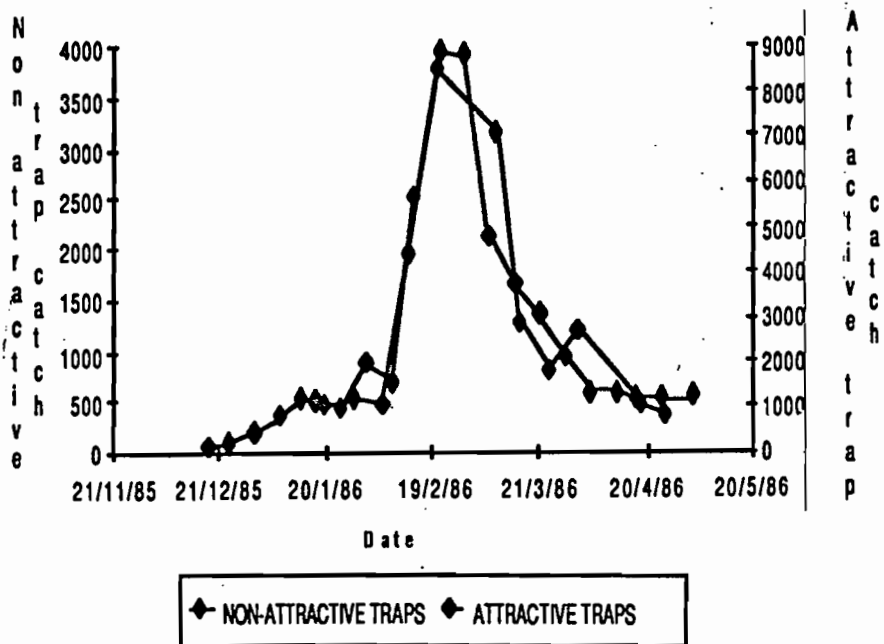


Fig. 5 shows the total catch results for both the attractive and non-attractive traps over the season. These reveal a qualitative pattern of increase and decline in numbers broadly similar to and contemporary with that shown by the field populations.

Fig. 5. Trap catch totals.



Therefore, it appears that the trap catches are largely reflecting events within the field. Unless otherwise stated data from the yellow attractive traps are used in the following, since the larger catch sizes and more detailed information permit greater analysis. However, in all cases, where applicable, results from the non-attractive traps suggest the same trends as are inferred from the yellow traps. Despite this, it is acknowledged that the behaviour-modifying effects of the attractive traps mean that inferences made from their results of the true behaviour of B. tabaci can only be tentative.

Thus, Fig. 6 shows that from 0630-1030 hrs, between 50-70% of the day's total catch is recorded (dawn c. 0600 hrs), while Figs. 7 and 8 indicate that between 65-95% of the catch within the field is recorded below canopy height and that a considerable portion of the catch is taken at 25 cm. This proportion varies between 35-80% for traps outside the field: the results are similar within the field at the start of season, but decline with time as the crop canopy increases with height.

Fig. 6. Proportion of the day's catch taken between 06.30-10.30 hrs.

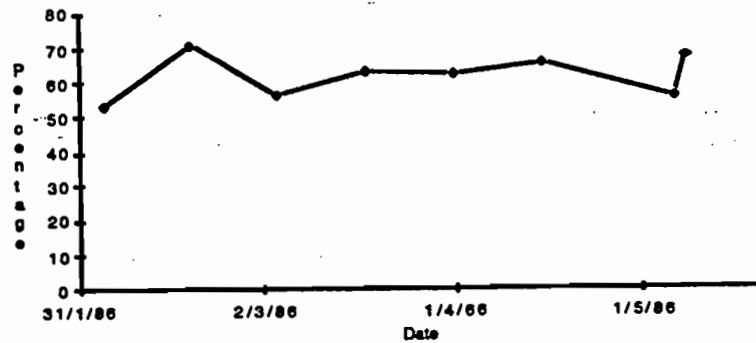


Fig. 7. Percentage of catch below canopy.

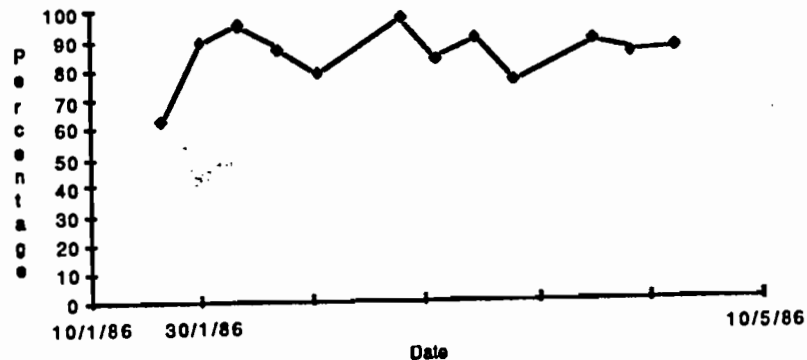
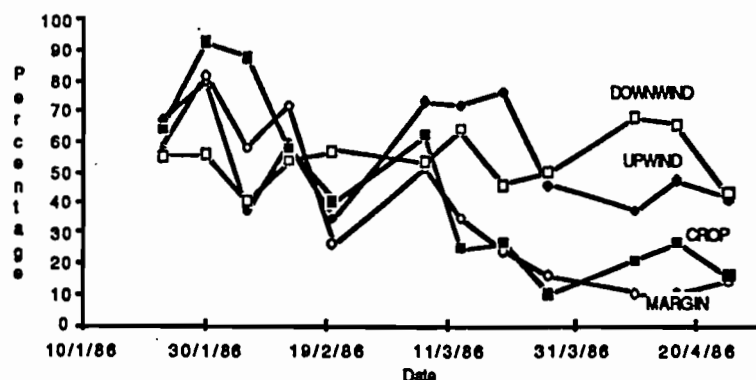




Fig. 8. Proportion of total tube catch at 25 cms at each array position.



This latter point is reinforced by Fig. 9 which displays how, as the canopy height increases, so does the level to which large catches of *B. tabaci* area recorded. Fig. 10 shows, for 2 dates, the differences in directionality of the trap catches between those caught beneath the crop canopy within the field and those caught above the canopy and outside the field. Within the canopy, higher numbers are caught on the side of the trap opposite to that of the prevailing wind, whilst above and outside the crop, relatively more are caught on the upwind side of the trap.

Figure 9a. Size of catch at different trap heights.

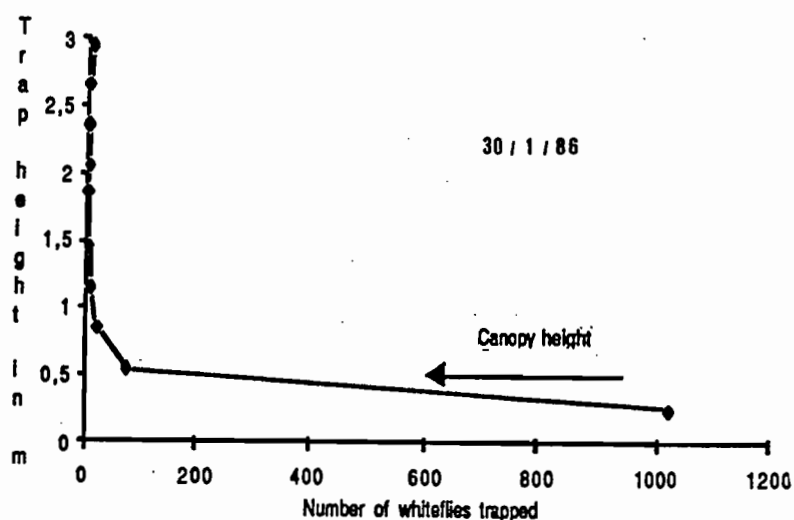


Fig. 9b. Size of catch at different trap heights.

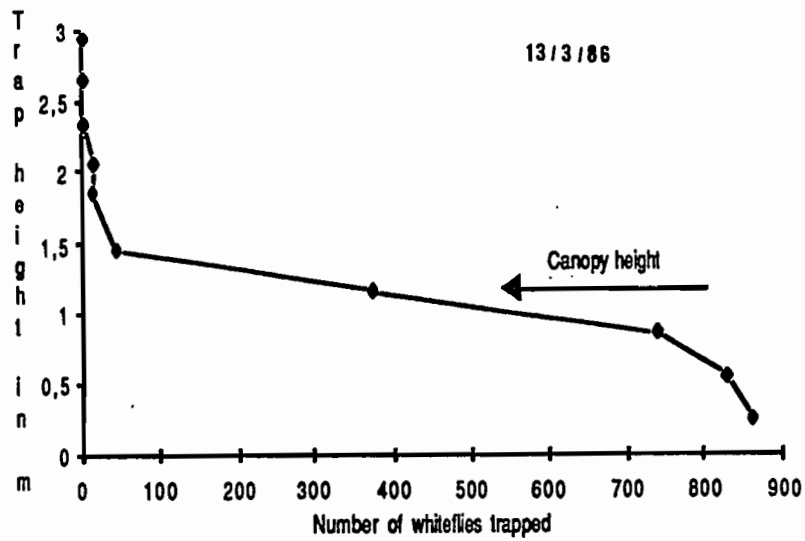
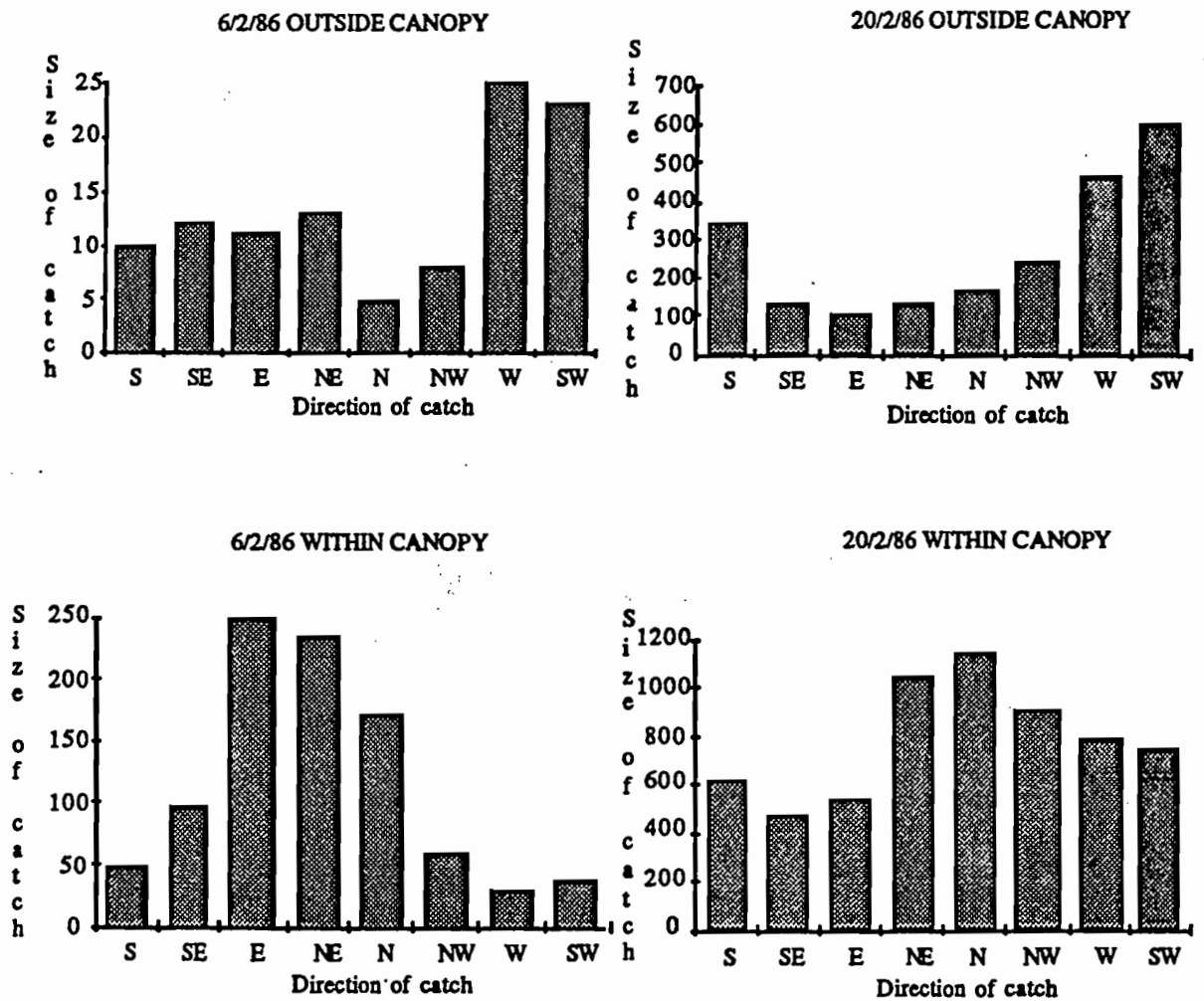


Fig. 10. Size of catch in different directions.



There is a change through the season of the relative proportions of the total catch taken at the different trap-array locations (Fig. 11). While numbers taken within and on the borders of the crop are always larger than those from outside the field, at the start of the season, and again to a lesser extent at the end, the proportions taken outside the field are higher than they are over the period of peak numbers in the field.

Figure 11. Proportion of catch at each trap position.

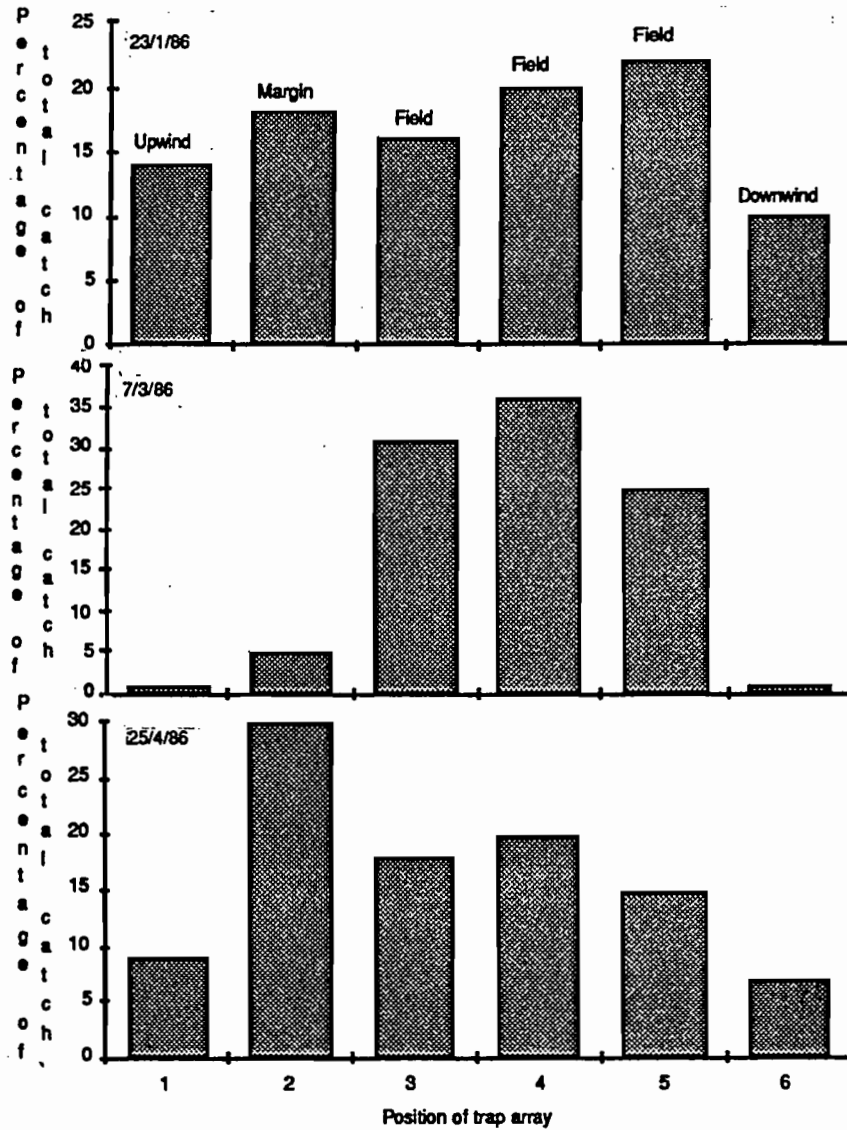
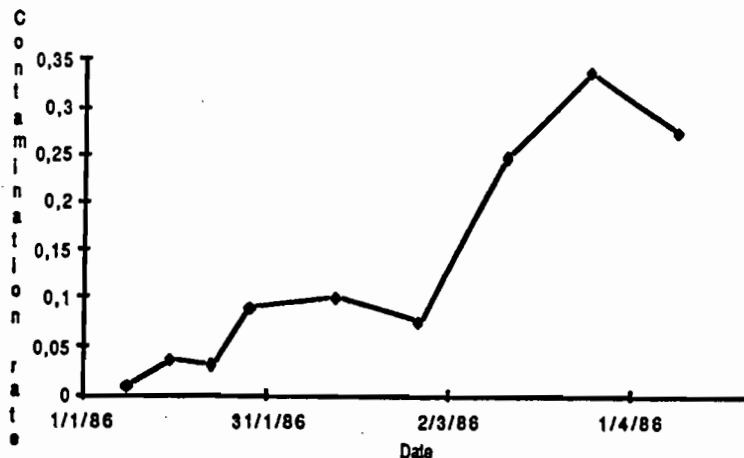


Fig. 12 shows the speed of contamination of the cassava crop with mosaic disease, where a peak comparable to the peak of the B. tabaci field populations is evident, although there is an interval of approximately four weeks between the two maxima.

Fig. 12. Speed of contamination of crop.

(% Cont. t2 - % Cont. t1)/% Cont. t1



## DISCUSSION

### Field populations

The type of growth curve shown by the B. tabaci populations in these studies is comparable to what has been found for this species on other crops in other parts of the world. Horowitz (1986) reviews these studies and identifies four stages. An early season moderate growth phase is succeeded by an exponential phase. This in turn may or may not (as here) be followed by a period of stability, before the population eventually declines.

Thus it is suggested that the pattern of events in the cassava crop was as follows. Aerial growth of crop, as the cuttings start to develop, is relatively slow until some 6-8 weeks from planting, after which there is rapid growth of the green parts of the plant in the following 6-8 weeks (Silvestre & Arraudeau, 1983, Fig. 3).

Hence, by early January the cassava was only some 25 cm high and supported only a small population of B. tabaci (the "moderate growth phase"), possibly because the crop had hitherto represented a relatively small target to migrant whitefly. However, these early colonisers were able to establish themselves and exploit the new resource and it is likely that, subsequently, local breeding

in the crop quickly swamped any continuing immigrants, although these in turn would have contributed to the succeeding exponential population increase.

From early January until the population crash in late February, at least 3 overlapping generations of whitefly developed in situ. The rate of development at average mean temperatures of approximately 27 C which prevailed over this period gives a generation time of between 2 weeks, by analogy with what is known of B. tabaci elsewhere (Gerling et al., 1986). However, direct observations on cassava under local conditions are lacking.

The explanation of the dramatic population crash at the end of February is less straightforward. Two heavy storms on 28 February and 12 March, when over 30 mm rain fell in 30 min. on both occasions, may have caused heavy mortality. This could have resulted from either direct mechanical action, or indirectly through abruptly lowered temperatures (circa 5 C) or changes in plant biochemistry, or a combination of these phenomena.

There are numerous references in the literature to heavy rain being unfavourable to B. tabaci populations (e.g. Khalifa and El-Khidir, 1964). However this cannot be the full explanation since Fargette (1985) has repeatedly observed such population crashes at Adiopodoumé in cassava aged between 3-5 months, at differing times of the year and under varying meteorological conditions, so this is not an exceptional event. It is possible that there is a deterioration in the food quality of cassava phloem at this time, and it may be relevant that at about 4-5 months old cassava reduces the amount of its resources devoted to aerial growth and the process of tuberisation begins (Silvestre & Arraudeau, 1983).

The four week delay between peak whitefly populations and peak disease contamination rate in the crop is rather faster than the average delay of about 6 weeks found by Fargette (1985) between infection with the virus and symptom expression in the field, but is not exceptional.

#### Flight behaviour

Two types of flight activity are recognised for B. tabaci by Berlinger (1986); these are short distance flights within the crop canopy representing local dispersal by, for example, newly moulted adults searching for feeding and oviposition sites (Gerling & Horowitz, 1984), and long distance flights when the whiteflies leave the crop to be caught in an air current and displaced downwind.

The stimulus for such a movement may be change in food quality of the host plant (Berlinger, loc. cit.). B. tabaci is sensitive to ultra-violet light (Mound, 1962) which would stimulate upward flight behaviour and result in departure from the crop. It is believed that whiteflies do not recognise host plants before descending from such a flight, but return to ground level and then search for suitable hosts (Gerling & Horowitz, loc. cit.). B. tabaci has long been recognised to be attracted to the colour yellow (e.g. Husein & Trehan, 1940), by which means they locate food plants. This is believed to be because yellow hues may

represent a super-normal foliage stimulus, since they reflect light at around the same wavelength as green vegetation, but at greater intensity (Prokopy and Owens, 1983). There is thus a balance between migratory flight stimulated by responsiveness to short wave radiation, and attraction to vegetation mediated by sensitivity to yellow light (Mound, 1962).

Byrne et al. (1986) consider that although B. tabaci is a poor flier, it is able to govern its flight direction to a limited extent, with most flight activity taking place just above ground level. On the basis of the formula given by Lewis and Taylor (1967), it has been calculated that B. tabaci has a flight speed of about 0.2 m/s (Yao et al., 1986), but there are no empirical observations and this figure may be rather too low. It has been noted by other authors (e.g. Gerling and Horowitz, 1984) that B. tabaci is most active in the early hours of the morning.

In general, our results are in accord with or are explicable by the foregoing. Our findings that the bulk of the trap catches were recorded at low level or within the crop canopy and usually within the first 4 hours after dawn are thought to be attributable to the relatively low wind speeds that prevail locally early in the day (0.1-0.4 m/s between 0600-0900 hours, Yao et al., 1986), at low levels and as a result of the modifying and braking effect of the crop canopy (within which wind speeds may remain below 0.4 m/s up to midday (Yao, this seminar; see also Pedgley, 1982).

This makes sense in view of the relatively low flight speed of B. tabaci; most flight activity occurs when the whitefly has most chance of actively determining its flight direction. The results shown in Fig. 10, indicating a boundary layer effect (Taylor, 1974), support this, whereas within the canopy a greater proportion of the catch is often found on the downwind side of the trap. This indicates upwind movement at low windspeeds, while above the canopy and outside the field, in stronger winds where B. tabaci is transported passively, relatively more insects are caught on the upwind side of the trap.

These observations may help to explain the concentration of whiteflies towards the upwind field border (Fig. 4), where upwind movement beneath the canopy combines with the interception of downwind immigrants on and behind the field margin.

The coincidence of the peaks of trap catch numbers and field population counts points to the bulk of the catch at this time representing local insect movements. This is supported by Fig. 11 where the relative proportions caught outside the field are higher at the beginning and again late in the season when the field populations are low.

It is suggested that these catches early and late in the season are mostly of insects undergoing long distance displacements from sources outside, probably mostly upwind of the field. This "background level" of migratory insects is relatively low when compared to the mid-season catches, but is of considerable significance in the epidemiology of ACMV.

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THE GENETIC VARIABILITY OF CASSAVA:  
ORIGIN, EVALUATION AND UTILIZATION

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Several review articles (Byrne, 1984; Hahn *et al.*, 1979; Jennings and Hersey, 1985; Roca, 1983) and a recent book (Sylvestre and Arraudeau, 1983) have been devoted to the improvement and agronomy of cassava. In this paper we therefore emphasize the theme of the genetic variability of cassava, particularly in Africa.

After a brief review of the structure of the genus Manihot and of the domestication of cassava and its dispersion in the world, we analyse:

- the origin of the genetic diversity of cassava;
- the variability of the cultivars;
- the possible applications of biotechnology to the improvement of cassava.

THE ORIGIN AND CULTIVATION OF CASSAVA

The complex of Manihot species

The genus Manihot Mill. belongs to the family Euphorbiaceae, and comprises more than a hundred species with the same chromosome number ( $2n=36$  chromosomes). Many researchers currently refer to the taxonomic classification of Rogers and Appan (1973). By multivariate analysis of the botanical characters of specimens in a herbarium, those authors classified 98 species in 19 sections, including just one cultivated species, M. esculenta Crantz.

Many wild species are sun-loving perennials, with a bushy habit and sporadic distribution: they are found in tropical savanna regions or semi-arid regions. Certain taxa readily occupy habitats disturbed by man and behave like adventitious plants of recent formation. In contrast, shrubby species of Manihot originated in the New World, with a range extending from the southern United States (Arizona) to northern Argentina. The species is divided



into more or less discontinuous geographical groups containing many wild species. Five primary zones of diversity area distinguishable (Figure 1):

1. Central America;
2. the central Brazilian plateau;
3. northeastern Brazil;
4. southwestern Brazil and Paraguay;
5. Colombia and Venezuela.

Fig. 1. Primary zones of variability of species of the genus Manihot (IBPGR, 1983).



The diversification of the genus Manihot results from several processes acting at different times. These include:

- bioclimatic changes, in particular the flora and fauna refuge zones at the end of the least period of glaciation (Vuilleumier, 1971);
- human migrations, with their trail of food-crop plants, in the pre-Colombian era (Nassar, 1978);
- genetic exchange within and between species, an active, continuous phenomenon.

The study of genetic resources and the evolutionary organization of the genus Manihot in the Americas is worth special attention. Regions of high priority were surveyed in the 1980s: Brazil (Nassar, 1980), Colombia, Venezuela, Peru, and Mexico (1982-1983). This collecting should be continued to learn more about the situation of the natural populations in relation to the ecology, and to limit the effects of genetic erosion (IBPGR proposals, 1983).

#### The domestication and dispersal of cassava

M. esculenta is a cultigen domesticated in the Americas and is unknown in the wild state. The ancestors of cultivated cassava were tuberous root plants eaten by man during their migrations in the Americas. Several ethnobotanical and archaeological studies suggest that cassava was utilized in northwestern South-America 2000 to 4000 years B.C. Recently, Ugent et al. (1986) described cassava fossils from the Casma valley in Peru, carbon-dated from 1500-1800 B.C. Furthermore, cassava seems to have been domesticated several times in different regions of the Americas (non central origin).

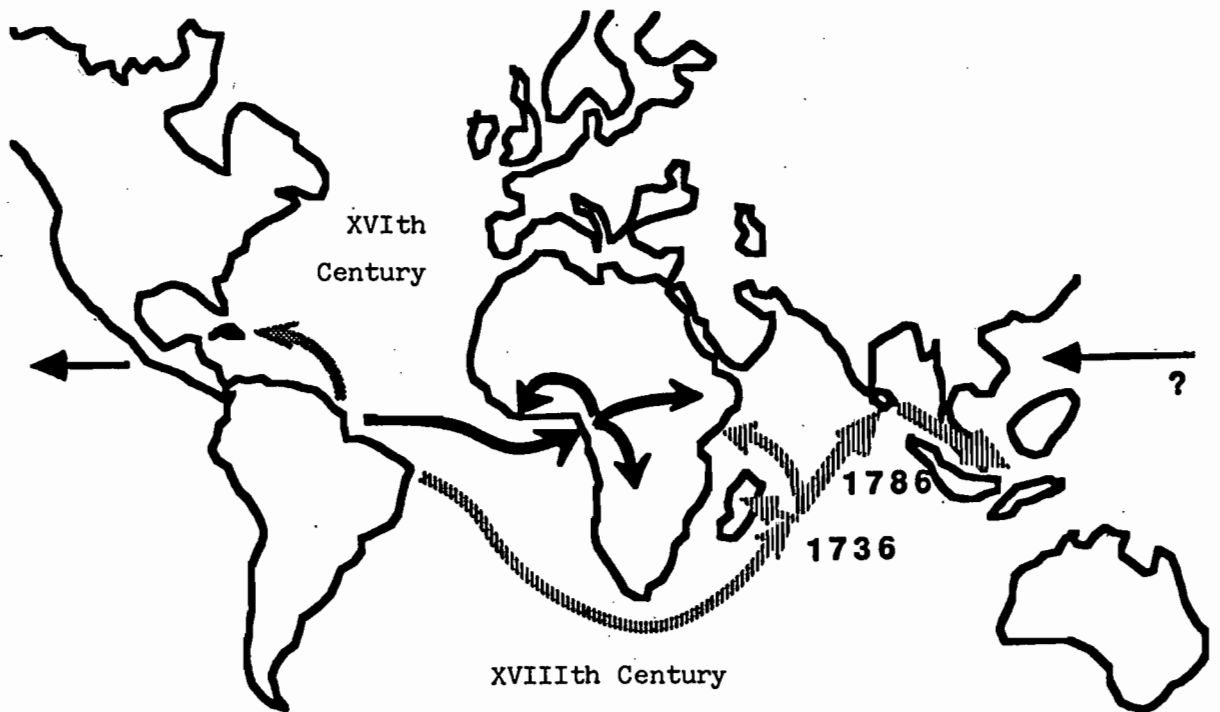
The domestication characteristics of cassava are as follows (Jennings, 1976):

- Large tuberous roots for starch storage, with a marked (Brazil, Venezuela, Colombia) or mild (Peru) bitterness.
- An erect habit with little branching; this trait is related to flowering, as domestication has resulted in fewer flowers.
- Ready multiplication by cuttings, a character favoured by the reserves stored in the cauline axes; furthermore, sexual reproduction is no longer considered (sterile plants).

The dispersal of cassava in the Americas has favoured an increase in its genetic diversity there. Like other plants originating in the New World, its introduction into Africa and Asia is historically recent, dating from after the discovery of America by Christopher Columbus.

Figure 2 summarizes the principal routes of intercontinental dispersion:

Fig. 2. Cassava dispersion.



In Africa, two main introductions are known, one in the Gulf of Guinea, in Central Africa, in the second half of the 16th century, and the other in the region of Madagascar and the eastern coast of Africa, during the 18th century. On the basis of ethnobotanical information, Kent (1969) situated the arrival of cassava in Madagascar with the migrations of populations from Eastern Africa in the 16th century. Cassava spread on the African continent in the second half of the 19th century and especially in the 20th century. The recent extension of its cultivation has been encouraged by the building up of standing food reserves (as protection against famines), and the plant's tolerance to drought and locusts. Jones's (1959) comparative study of the Congo, Guinea, and Eastern Africa clearly establishes the place of cassava in Africa.

In Asia, cassava was probably introduced directly from Mexico to the Philippines, and indirectly from the Mascarene Islands via Ceylon (1786), India (1794), and Southeast Asia.

In addition, several shrubby wild species of the genus Manihot which produce latex were introduced into Africa. The principal one was M. glaziovii from Brazil. Cross's survey in 1876 was widely publicized by Kew Gardens. At the beginning of this century, M. glaziovii was grown on about 50,000 hectares in Eastern Africa and it was utilized to a more limited extent in Western Africa (sometimes as a shade tree). The species M. dichotoma and M. piauhyensis were considered unsatisfactory for latex production.

These migrations to Africa and Asia resulted in a founder effect, with less diversity than existed in the Americas. Since the 5th century, an intense diversification has taken place locally.

#### ORIGIN OF THE VARIABILITY OF CASSAVA

The genetic variation of cassava is related to the following factors:

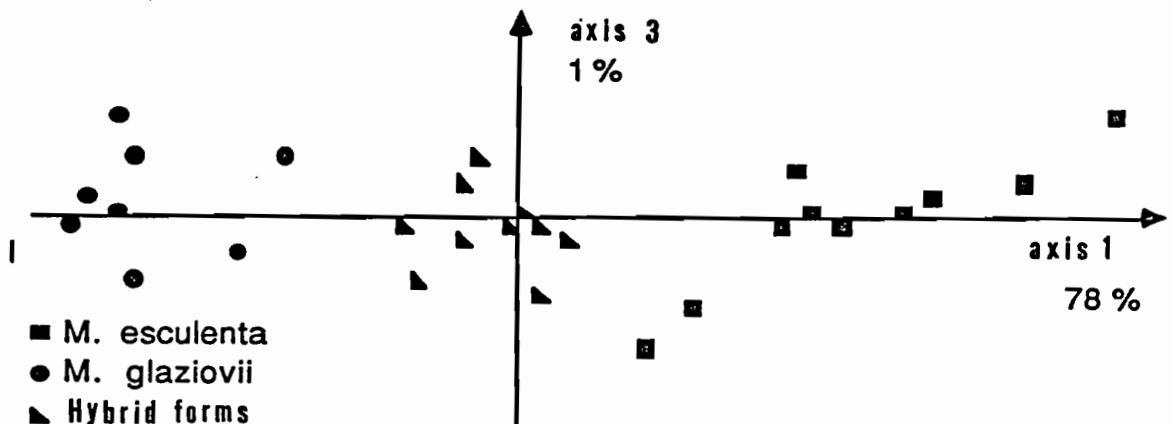
##### Active introgression processes

Wild Manihot species are often isolated from one another, but they seem to hybridize easily when they happen to come into contact. Nassar (1978) demonstrated this clearly in Brazil: M. reptans and M. alutacea crossed naturally in disturbed zones, with gradual introgression into M. reptans.

Likewise, in the regions where cassava originated, the cultivated forms can also intercross with the local wild species and give rise to various hybrid "swarms", which evolve towards adventitious forms ("weeds" in Harlan's (1971) sense of the word), colonizing the regions influenced by man.

Such genetic exchanges also exist in Africa between local cultivars and introduced wild species. In recent studies in the Ivory Coast, Lefèvre (1987) demonstrated the existence of spontaneous interspecific hybrids of M. esculenta x M. glaziovii. Their morphological characters (shape and size of the fruits, seeds, leaves and tuberization) suggest the presence of a very diversified population of hybrids (Fig. 3).

Fig. 3. Introgression between cultivated cassavas and M. glaziovii.



## A mode of allogamous reproduction developed by a plant with a polyploid structure

The traditional agriculture based on cassava (a mosaic of cultivars) favours genetic recombination between cultivars with heterozygous structure. The descendants resulting from self-pollination and natural cross-pollination are very polymorphic and the effect of inbreeding is marked.

Magoon et al. (1969) lent strength to the hypothesis of a polyploid origin of the species M. esculenta. More precisely, its segmental allotetraploid nature is in accord with the following facts:

- bivalent pairing and disomic heredity (with several cases of duplicated genes),
- a caryotype with 3 nucleolar chromosomes and one partial duplication of chromosomes affecting 6 of the 9 chromosomes of the haploid set.

The origin of this allotetraploidy remains an enigma, as all the wild species of Manihot studied so far have 36 chromosomes.

## Vegetative reproduction

Thanks to this mode of multiplication, every valuable new individual can be used to make a strain (a clone). This ancestral practice makes it possible to exploit the variability of natural reseeding depending on the selective pressures, which vary with time and over space. For example, plants from the seedbed which are free of virus are kept to renew the stock of cuttings when the cultivars in the field are very diseased. This process is eminently favourable to constant diversification and adjustment to new parasitic pressures (as in the recent example of bacteriosis in Central Africa).

## Somatic mutations

Few authors refer to this subject (Leon, 1976; Martin, 1976), in the absence of experimental proof.

In conclusion, the genetic pool of the genus Manihot evolved in its area of origin according to a pattern of disruptive selection, with increase of the diversity of the cultivated pool (human selection) and of the wild pool (adventitious forms). The result is a taxonomically complex situation linked to the evolution of wild and cultivated forms in the area where cassava originated, but also in Africa.

With reference to these evolutionary patterns, it is entirely appropriate to improve the stock by exploiting the possibilities of introgression with wild species and recombination between cultivars. This option makes it of prime importance to evaluate, and circulate the genetic resources of the genus Manihot for their use in selection. This is not a new undertaking and has already led to recognized successes in Africa from virus-resistant interspecific hybrids (Jennings, 1976). The agronomic characters

of the wild species of Manihot answer many agronomic needs (Nassar, 1986):

- resistance to viral diseases and to bacteriosis (M. glaziovii);
- a high protein content (M. oligantha);
- tolerance to drought, cold, and hydromorphic soils;
- reduction of the hydrocyanic glucoside content (M. gracilis, M. oligantha);
- prolificness (M. oligantha, M. tripartita, M. zehntneri, M. anomala).

#### DESCRIPTION OF THE DIVERSITY OF CULTIVARS

It is customary to distinguish the cultivars of cassava according to:

- their content of hydrocyanic glucoside (sweet or bitter cassava);
- the color of the root flesh (yellow flesh is often more bitter than white flesh);
- the length of the growth cycle (early, sweet cultivars, or bitter ones with a 1- to 2-year cycle).

This diversity of cultivars is often reflected in their vernacular names. Research centers that do work in cassava selection have built up collections of local or introduced cultivars (such as the IBPGR repository, 1980). The largest are held in Brazil (by Embrapa), Colombia (by CIAT, which has 2600 strains), India, Indonesia, and Nigeria.

There are two main objectives for the collections:

- to identify cultivars on the basis of morphological and physiological characters;
- to utilize them in selection as a function of their agronomic characteristics and their behaviour relative to the biotic and abiotic environment.

These descriptions are taken from the list of characteristics recommended by IBPGR (1983). Well-documented descriptive studies have been made from collections in:

- Madagascar (Cours, 1951),
- Ghana (Doku, 1966),
- Venezuela (Montaldo, 1982),

- Mexico (Galindo, 1982),
- Brazil (Costa, 1983),
- The Ivory Coast (Zoundjihekpon, 1986).

All these descriptive studies of diversity are very useful in the improvement of cassava. However, they are of limited value for describing the genomes of the cultivars, because:

- a great many characteristics are noted;
- the expression of morphological and physiological characters is very dependent on the environment, local strains of parasites, and the state of health of the collection;
- the genetic determination of characters is not known, or is not quantitative in nature.

For all these reasons, significant progress in the description of genetic variability has been made with the use of biochemical markers.

The first such methodology, put into use in the 1970s, was based on electrophoresis to determine enzyme polymorphism. Enzyme markers have the double advantage that they have simple genetic determination and selectively neutral behaviour. Its recent application to cassava is due to Zoundjihekpon (1986), CIAT (1985), and Lefèvre (1987). The latter author at present uses 10 enzyme systems revealed on starch gel: 12 loci and 28 alleles have been identified. This technique makes it possible to demonstrate:

- a simple diploid type of heredity,
- the existence of duplicated genes (phosphogluconate dehydrogenase),
- cases of fixed heterozygosity (phosphoglucoisomerase) and of interactions between loci (malate dehydrogenase).

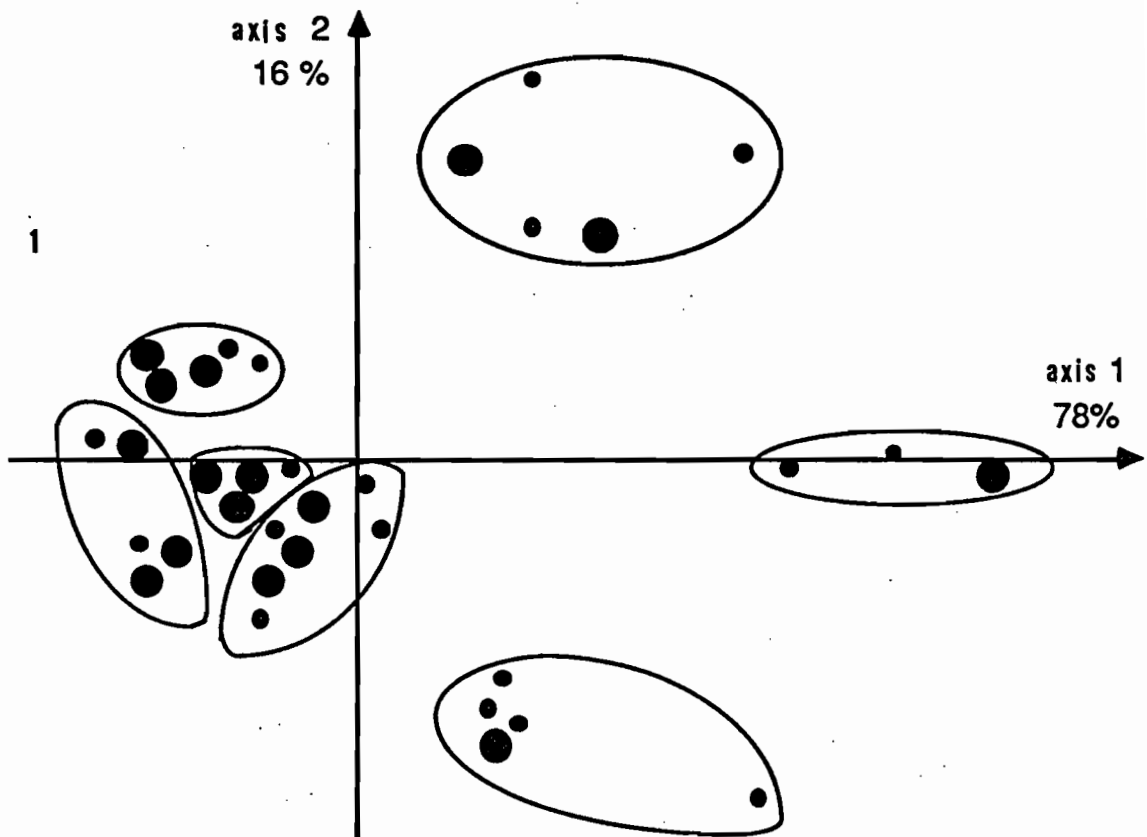
The study of a collection of 168 Ivory Coast cultivars illustrates this approach to the genetic diversity of a collection:

- the 12 loci so far known identify 78 different electrophoretic genotypes;
- 19 alleles have a frequency higher than 30%;
- the various possible allelic combinations can exist, many of them in the heterozygotic state.

By multivariate analysis (Fig. 4), it has been shown that the local cassavas of the Ivory Coast form several groups defined by particular combinations of common alleles. This structuration closely reflects their genetic relatedness, the genetic distance between groups, and the origin of intermediate groups. The improved varieties introduced in this country present the same

structuration, with several previously unreported allelic combinations.

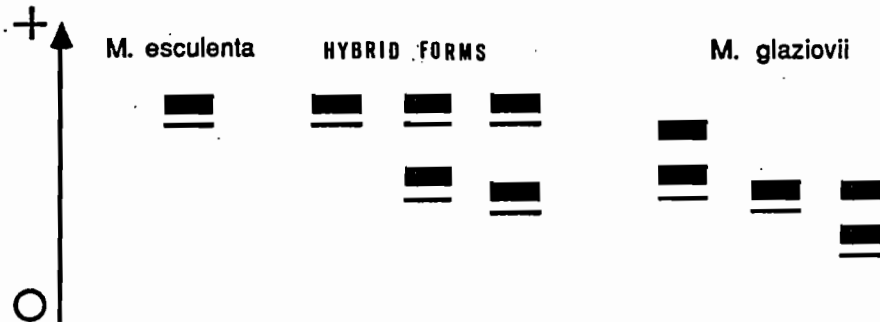
Fig. 4. Structure of enzymatic diversity of 168 Ivory Coast cultivars (AFC with 10 enzymatic alleles).





The enzymatic markers also make it possible to follow the phenomena of interspecific hybridization and introgression (Fig. 5., leucine aminopeptidase).

Fig. 5. Enzymatic markers of the introgression between cultivated cassavas and M. glaziovii, (LAP zymograms).



In the case of the spontaneous hybrids M. esculenta x M. glaziovii, one allele from each of the parent species is found at each locus. An exception, one of the hybrids, is enzymatically no different from M. esculenta and reflects a reversion to the type of the cultivated parent. Conversely, older traces of introgression can be identified in some cultivars (rare, common, or fixed alleles in M. glaziovii).

The technique of enzyme electrophoresis seems to be the main effective biochemical approach. Other chemotaxonomic markers can also be used (such as phenol polymorphism). But, particularly since the beginning of the 1980s, workers have turned to direct marking of nuclear, chloroplast, or mitochondrial genomes.

With the use of restriction enzymes and molecular probes, it is possible to locate a specific fragment of DNA and to study its intraspecific or interspecific polymorphism. For example, it is possible to distinguish the chloroplast genomes of the parents, and to obtain the origin of the cytoplasm of sexual hybrids, introgressions, and hybrids (somatic hybrids). The analysis of restriction fragments of mitochondrial DNA is a convenient tool for distinguishing the fertile or sterile male cytoplasms. The detection of a nuclear DNA fragment by hybridization with a specific probe makes it possible to reveal a polymorphism, called restriction-fragment-length polymorphism (RFLP). This can be exploited to:

- characterize a cultivar (biochemical identity card),
- mark a character of agronomic interest (if the appropriate molecular probe is available), or
- establish a detailed genetic map of a cultivated plant species.

In the case of cassava, such molecular studies on DNAs are indispensable if one wishes eventually to attempt genetic manipulations.

## BIOTECHNOLOGIES APPLICABLE TO THE IMPROVEMENT OF CASSAVA

### Sanitation

In this cultivated vegetatively reproducing species, the risks inherent in diseases caused by systemic agents transmitted through cuttings are well understood on the basis of studies by Martin and Morel. The new techniques of plant multiplication in vitro have made it possible to regain normal state of health for many vegetatively propagated species. The effectiveness of in vitro culture of meristems combined with heat treatment was established for various viral diseases of cassava in the 1970s (Kantha, 1975; CIAT, 1980; IITA). The important problem is to have reliable indexing methods available for each viral strain; decisive progress has been made in this field, associated with molecular biology (serological tests, monoclonal antibodies). The gradual degeneration of heavily virus-infected cassava significantly affects their growth, their vigour, and their production (estimated losses of 40 to 70%) and makes it difficult to assess their agronomic value. There are several hundred healthy clones at CIAT (1982). Furthermore, this technique of micropropagation of cassava makes possible a faster vegetative multiplication of healthy strains to set stocks of cuttings.

### Conservation of genetic resources

The in vitro culture of cassava satisfactorily resolves the difficulties of conservation and diffusion of genetic resources. Every selector knows the problems of maintaining several hundred cultivars in a collection of live plants in the field (diseases and parasites, climatic accidents, etc.). The culture of meristems in vitro can be used for this purpose in two ways:

- The preservation of microcuttings in slowed growth (at 20-22 C) makes storage possible for 4-5 years with periodic pricking out; 1500 cultivars are preserved in this form at CIAT.
- Meristems can be frozen if protected with cryoprotectors (such as DMSO or sugars) and then stored in liquid nitrogen at -180 C; using this technique, Kantha (1982) obtained 90% survival of meristems and 10% regeneration of plants. Such long-term preservation is still very demanding to use (in technique and regeneration).

## Culture of tissues and cells in vitro

Though the micropropagation of cassava has technically come into use, the other modes of reproduction in vitro have not been very successful so far:

- Culture in vitro of immature cassava embryos would be helpful in work on interspecific hybridization; this has not been tried.
- The regeneration of plants from tissues has been known to succeed in cassava. The first such plant was obtained by Stamp and Henshaw (1982) by somatic embryogenesis from explants of cotyledons, with large doses of auxins favouring the induction of variants. Likewise, Mabanza and Jonard (1984) found up to 95% regeneration by neof ormation of buds on callus from cotyledons from seeds at maturity.
- The regeneration of plants from cells does not yet work. The protoplasts of leaf mesophyll develop into microcolonies and microcallus, but the regeneration only occasionally goes on to the formation of cauline axes (Shahin and Shepard, 1980).
- Obtaining haploid plants, which are so useful in the analysis of genomes and the fixation of characters in the homozygous state, has not yet succeeded. Cell culture stumbles against organogenesis (with the formation of callus and root axes only) (Liu and Chen, 1978; CIAT, 1982). Other possibilities for generating haploid forms remain to be surveyed: direct culture of microspores, gynogenesis, induction of haploidy by fertilization with the pollen of another species or of irradiated pollen.

## Somaclonal variation and genetic engineering

The development of these new approaches to selection is blocked by the difficulty of regenerating whole cassava plants by somatic embryogenesis:

- Culture in vitro of various explants has demonstrated the capacity of isolated cells to regenerate whole plants, phenomena of rejuvenation during successive microcuttings, and even the loss of cellular identity so that it becomes possible to start off in new directions (variants resulting from mutation, chromosomal remodelling, genetic regulation, etc.). In cassava, this route could be exploited with pressures such as physical stress, toxins, and specific antibodies.
- The usefulness of interspecific hybridization products has been demonstrated in cassava in particular. Approaching it by somatic hybridization may open up the range of possibilities of recombination of genomes and of cytoplasms: this gives rise to more or less viable, more or less stable transgenic plants.
- Finally, plant transformation by genetic engineering has resulted in several transfers of bacterial genes and plant proteins. The transformation can be direct or by intermediary

vectors derived from plasmid T1 of Agrobacterium. There have so far been only a few projects involving the improvement of plants (TMV capsid protein, or resistance to glyphosate).

## CONCLUSIONS

This general review of the genetics of cassava sheds light on the gaps and need for research in this field:

- Knowledge of the cassava genome and of the structure of the genus Manihot is very limited. It may progress rapidly with the use of various molecular markers.
- Genetic resources have a primary influence in the improvement of cassava. Prospecting for wild forms and cultivars should thus be encouraged, as should the circulation of plant material. A decisive step was the building up of tissue culture collections of healthy microplants.
- The classic methodologies of vegetative and sexual selection remain entirely applicable to cassava.

In contrast, modern methods will be applicable to cassava only after the following points have been resolved:

- knowledge of how to regenerate normal, stable transgenic plants in vitro;
- knowledge of how to identify and isolate the genes to be transferred (probes) applicable to simple genetic characters (such as resistance);
- an understanding of how to control the expression of transferred genes;
- the availability of equipment and of personnel competent in plant biotechnology and molecular biology.

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## MICROCLIMATE OF A CASSAVA CANOPY

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### INTRODUCTION

Several researchers (Fargette, 1985; Fargette *et al.*, 1986; Van Helden and Van Halder, 1986) have already shown a close relationship between the dispersal of African cassava mosaic and the characteristics of the wind. *Bemisia tabaci*, the vector of the disease, moves slowly, so its dissemination depends closely on the wind speed and direction (Byrne, 1986; Yao *et al.*, 1986). To understand the epidemiology of this viral disease it is necessary to analyse the relationships between the dissemination of the pathogenic agent, the movements of the insect vector, and the characteristics of the wind (Yao *et al.*, 1986). Though the wind is the most important climatic parameter for the dissemination of the whitefly, the role of other parameters (temperature, humidity, etc.) is not negligible. Thus, a comprehensive investigation of the microclimate of the cassava leaf-canopy and of certain meteorological phenomena should provide better understanding of the dissemination of the insect and the dispersal of the disease.

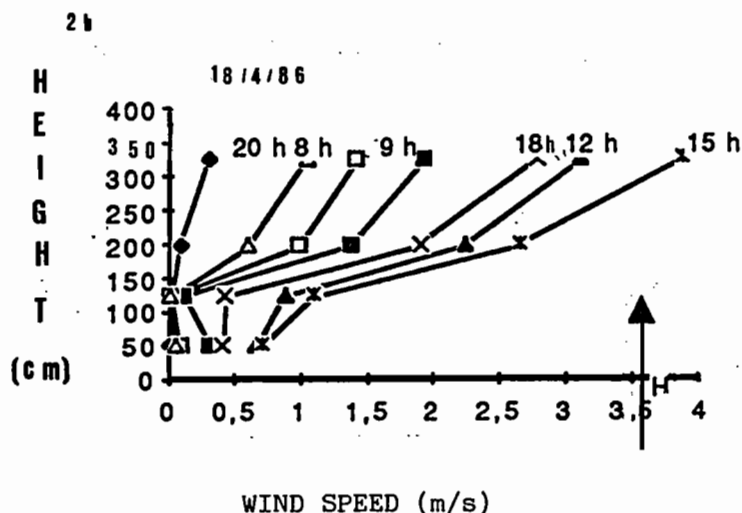
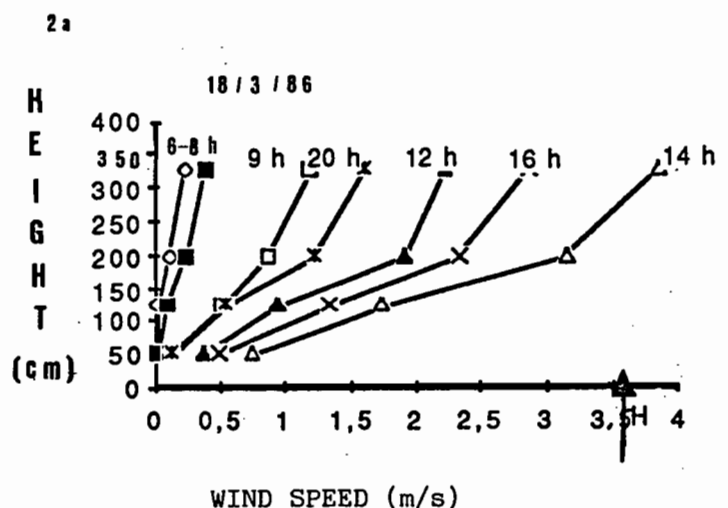
### THE WIND

#### Wind profile

The characteristics of the wind above a surface (bare soil, or plant canopy) differ greatly from those observed within a plant canopy. Above a surface the speed of the wind,  $U$ , increases rapidly and logarithmically with the height,  $Z = U^* \text{Log} [(Z-d)/Z_0] + k$  where  $d$ ,  $Z_0$ ,  $U^*$ , and  $k$  are parameters that characterize respectively the height of movement of the reference plane, the roughness of the canopy, the speed of friction, and Karman's constant (0.4). Inside a plant canopy the wind speeds are reduced, the increase in speed with height is much less, and the wind profile is not necessarily logarithmic, but is then determined mainly by the architecture of the plant (foliar index, distribution of leaves) and the characteristics of the crop (planting density) (Colville, 1968). Thus, a high foliar index or a high density will strongly reduce the wind speed in the canopy.

Figures 2a and 2b show the wind profiles in and above a cassava canopy at different times of day. When the canopy was approximately 120 cm high and the leaves were uniformly distributed at all heights, the reduction of the wind speed in the canopy was steady and was greater towards the bottom. Above the canopy the profile was logarithmic (Perrier *et al.*, 1970). For a 6-month canopy 170 cm high and with a distribution of leaves biased towards the summit after the fall of the lower leaves, the speeds are greater at the base than higher up. Above the canopy, the profile was again logarithmic. In all cases, the higher the unimpeded wind speed, the greater its reduction in the canopy. This is shown by the fact that the profiles are not parallel.

Fig. 2. Profiles of the wind in a cassava canopy.





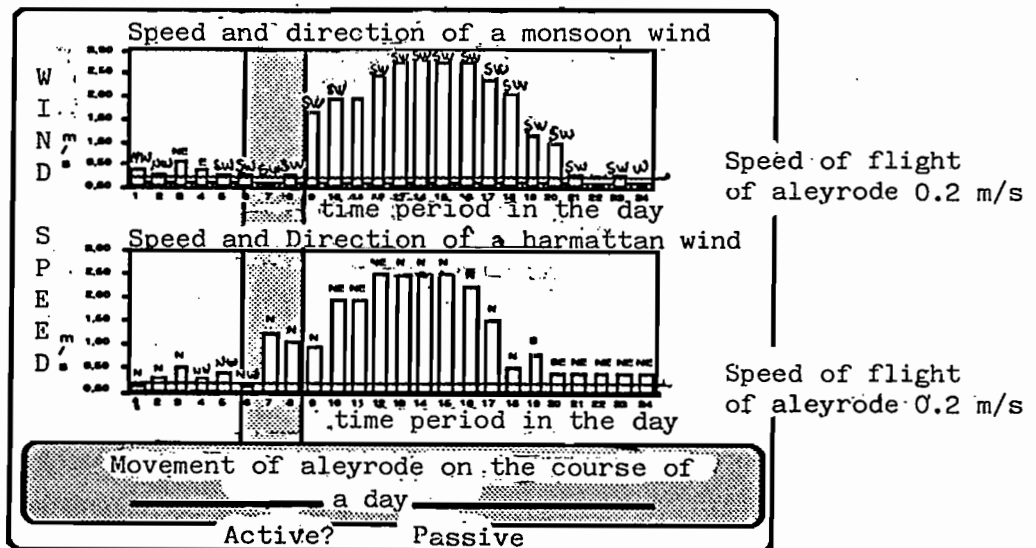
### The boundary limit and importance of the wind direction

Taylor (1958) introduced the notion of boundary layer for the flight of an insect as the layer within which the speed of the insect's flight is higher than the wind speed. The depth of this layer fluctuates from moment to moment with changes in the wind speed (Fig. 1). It is a function of the architecture of the canopy (of its foliar index in particular) and is determined also by the intrinsic flight capacity of the insect. The boundary layer and the higher layer are two zones within which the insect's movements are quite different in nature. Within the boundary layer, the insect can control its flight, move actively by itself, and reach a goal. The boundary limit depends therefore, on the insect's own speed of movement. To our knowledge, there have been no direct measurements of the speed of flight of B. tabaci. However, a general relation established between the size of insects (wingspan x body length) and their speed of flight (Lewis and Taylor, 1967) gives for B. tabaci a speed of movement of the order of 0.2 m per second.

In view of the surface wind speeds for the major part of the day (Fig. 3) and by analogy with other insects investigated (Taylor, 1974), the boundary layer must lie just a few centimetres above the ground. However, the reduction of the wind speed by the canopy may increase this boundary layer.

Above the boundary layer, on the other hand the wind speed is higher than that of the insect, and so no progress may be made against the direction of the wind; the insect is then transported passively. Its direction and speed of movement are then similar to those of the wind. Distances of the order of several kilometres per day may thus be traversed.

Fig. 3. Speed and direction of a wind at different times of the day.

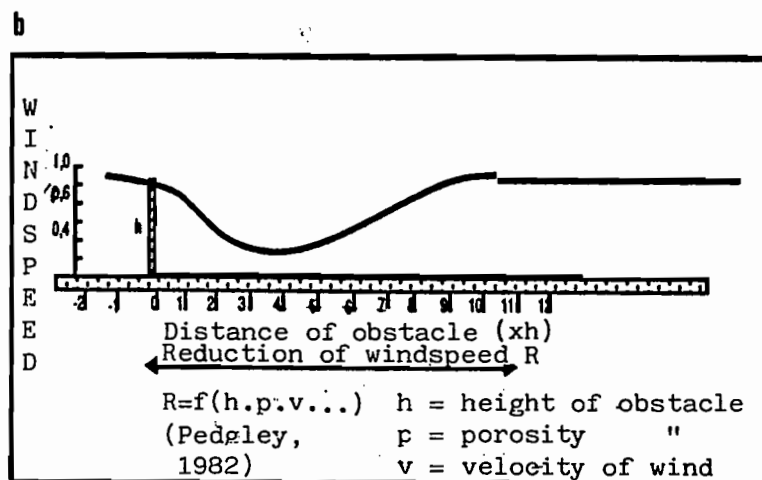
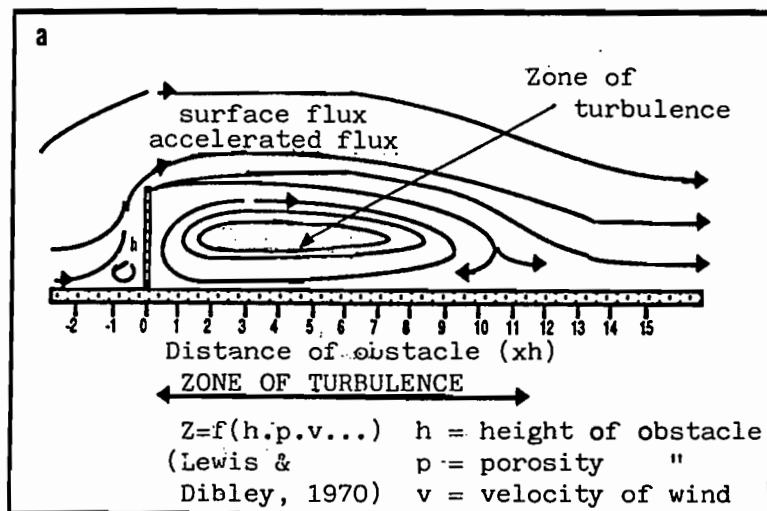


## Effect of an obstacle on the wind

### The windbreak

Any natural or artificial obstacle produces phenomena of turbulence and induces a reduction in the wind speed (Fig. 4a,b) downwind (Rosenberg, 1974) which favour the accumulation of insect particles, such as insects floating in the air (Lewis and Dibley, 1970). In fact, downwind of an obstacle or a barrier there is a bubble-like zone of calm (Fig. 4a) which, depending on the angle of incidence of the wind and the permeability of the barrier, extends 7 to 20 times the height of the obstacle. Within this bubble the wind fluctuates in all directions, but follows on average a circular movement perpendicular to the barrier around a zone of calm in its centre. The bubble is bounded at the bottom by the ground and above by the turbulent shearing layer. Also, certain insects approaching the barrier in the incident wind will be transported above and beyond this zone; others will be scattered into the interior of the "bubble" by the turbulent shearing layer and will be blown about there by an entering flux, generated near its downwind extremity.

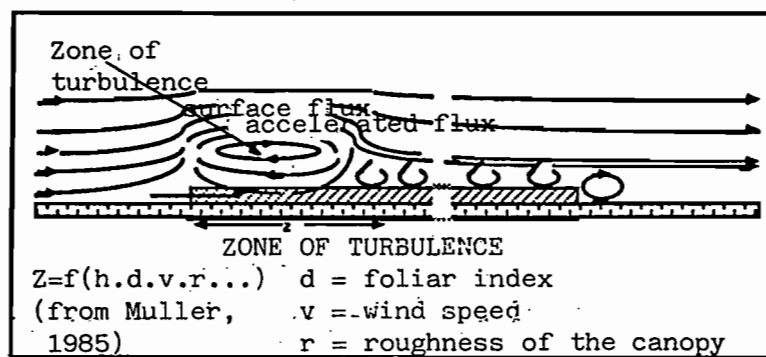
Fig. 4. Effect of an obstacle on wind direction.



## The cassava canopy

As a windbreak, the cassava canopy constitutes a natural obstacle which creates turbulence phenomena before and after the plot, and of course above the canopy. The size of the zone of turbulence (Fig. 5) will be a function of the height and the roughness of the canopy, the foliar index, and the wind speed. So the turbulence favours the landing of insects floating in the wind above the boundary layer.

Fig. 5. Effect of the canopy on the direction of the wind.



Figures 6a and 6b show that the wind speed begins to slow well before the plot. The reduction is maximal at the edge of the plot and depends on the level of measurements relative to the height of the canopy (Fig. 6b). Here again, as in the case of the vertical profile, the stronger the wind in the free air, the greater the reduction in speed. However, strong winds may change the architecture of the canopy, sometimes leading to smaller reductions at the edge (Fig. 6b).

Fig. 6. Effect of a cassava canopy on the speed of the wind.

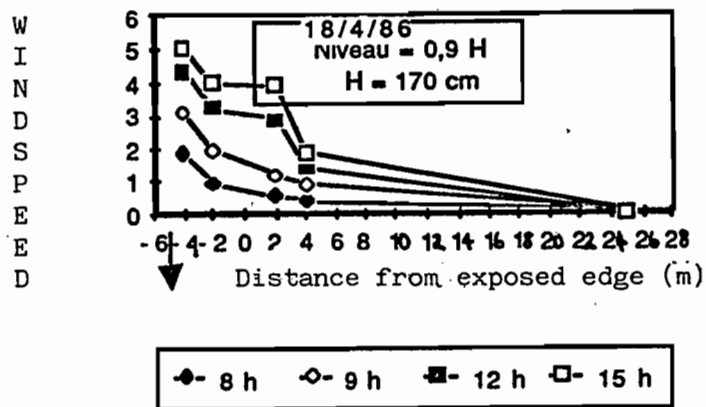
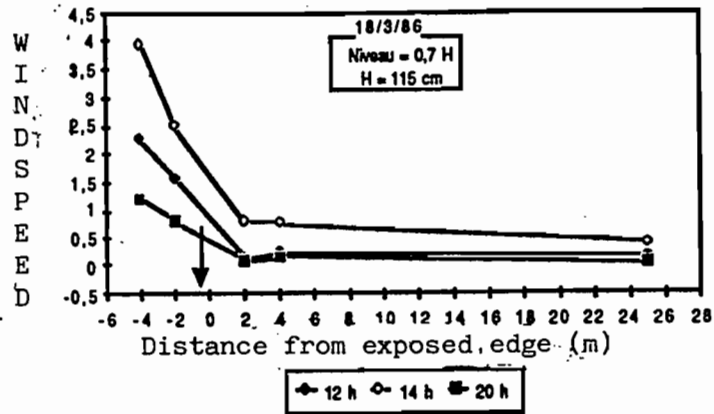
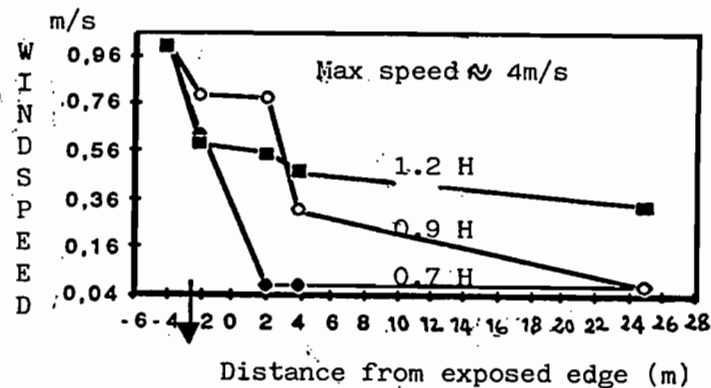


Figure 7 shows the reduction relative to the maximum wind speed. Note that above the surface of the canopy the reduction was 40%, while in the lower third of the canopy the reduction varied between 90 and 100%, depending on the maximum speed.

Fig. 7. Decrease in wind speed at different levels in a cassava canopy.



## TEMPERATURE AND HUMIDITY

Authors such as Pedgley (1982) have stressed the importance of air temperature and humidity in the activity of insects. Pedgley reported that there is a temperature threshold necessary to the flight of insects and that there is evidence that a rapid fall in relative humidity favours insect flight. Figures 8a and 8b show the general tendency of the daily changes in temperature and humidity at three levels in a cassava plot. The temperature was lowest during the night and increased during the day, reaching a maximum towards 14-15 h, and thereafter decreased. This change in the air temperature is directly related to solar radiation. In fact, the incident solar energy (the net radiation) is used in evaporation through transpiration (flux of latent heat), in heating the air (flux of sensible heat) and heating the ground (flux of conductive heat). The more unfavourable the water conditions, the greater the rise in air temperature. The difference of temperature between the three levels (at 0.5, 1.25, and 2 m) may sometimes reach 4°C. In contrast with the temperature, the relative humidity is greatest at night and least in the day, and it changes in inverse proportion to the temperature. The decrease in relative humidity during the day does not mean an actual reduction of the quantity of water vapour in the air. On the contrary, evaporation of transpired water provides additional water vapour, but the rise in temperature reduces the degree of saturation of the air more rapidly.

Fig. 8a. Change in air temperature over the day.

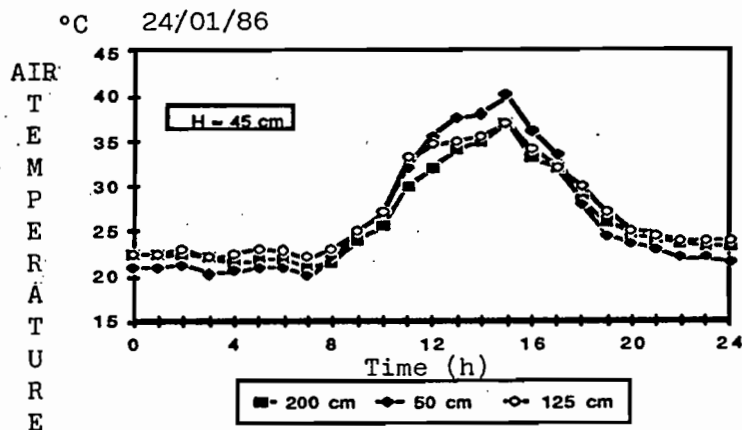
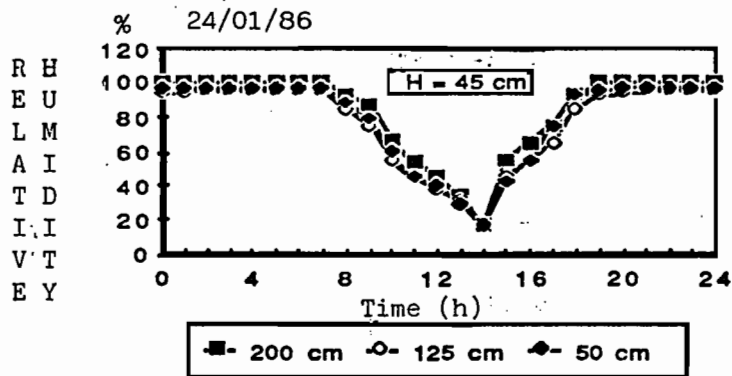


Fig. 8b. Change in relative humidity over the day.



#### IMPORTANT CLIMATIC PHENOMENA

The climatic parameters that we have just analysed are influenced by several systems of circulation: monsoon winds (southwestern trades) and harmattan winds (northeastern trades) onshore and offshore breezes and storms.

#### The intertropical front

In the intertropical zone there is an area of confluence between two masses of air. The first, which is oceanic, from the southwest, is called a monsoon but is just the trade wind from the southern hemisphere, pushed eastwards by Coriolis forces after crossing the equator. The second is dry, of continental origin, from the north east; this is the trade wind of the northern hemisphere, called the harmattan. This zone of confluence is called the intertropical convergence zone, and the line where the two trades meet is called the intertropical front (ITF). These masses of air, and consequently the ITF itself, move mainly under the influence of the pressure gradients between the anticyclone of the Azores to the North, the anticyclone of Saint Helena to the South, and the Equatorial trough. The Saharan thermal depression situated between the Azores anticyclone and the Libyan anticyclone cell plays the principal motor role. When it moves to higher latitude, it creates a monsoon force which pushes the ITF northward, and the converse when it moves down towards the equator (Eldin, 1970).

The passage of the ITF does not only lead to a change in the direction of the winds. The temperature and especially the humidity of the air undergo changes. There are also phenomena of convergence at the front level which could explain sudden accumulations of insects (Joyce, 1983). In the lower Ivory Coast, at Adiopodoumé, the period of January/February, corresponding to

the passage of the ITF, produces an unusual instability in the level of the populations of whiteflies (Fargette et al., 1986).

#### Offshore and onshore breezes

This circulation is thermal in origin and is reversible. In still air, the sun heats the earth more rapidly than it does a body of water, producing a horizontal temperature gradient. Consequently the air above the land also heats and expands more rapidly than that above the water. Because of hydrostatic conditions, the vertical pressure gradient is higher in the cool air above the water than in the hot air above the land. This means that, at a given constant level above the land and water, the pressure is higher over the land than over the water. This pressure gradient (approximately 1mb/50 km) creates (high up) a slight movement of air, from the land (B) towards the sea (C). The convergence in C increases the pressure, which produces subsidence from C towards D, in response to the hydrostatic disequilibrium and to the flow of air from D towards A. The divergence near B leads to a reduction of pressure producing a flow of air from A towards B. This process forms an onshore breeze. An offshore breeze is formed by the opposite process.

The offshore breeze is not as strong as the onshore breeze, either in speed or in height, since there is no source of energy (at night) to transport the circulation to greater heights (Atkinson, 1981). Note that the appearance of the breeze usually coincides with a change of wind speed and direction. The onshore breeze produces a fall in temperature and a rise in the relative humidity. The wind speed can reach 10 m/s. In the lower Ivory Coast an onshore breeze is often confused with a monsoon. An offshore breeze has a much lower speed (less than 2 m/s (Atkinson, 1981)) and produces less change in temperature and humidity. The depth of the breezes varies between 100 and 1000 m. The returning current has not been much investigated, but it can vary between 500 and 3000 m and can reach 7 m/s. The distance covered by the breezes usually varies, between 20 and 50 km on average, but can reach 300 km in the tropical zones.

#### Storms

The advection of cool maritime air onto hot continental air is liable to induce storms. Storms are accompanied by the emission of masses of cool wet air, which move away from the center of the storm, creating a front with the drier masses of surrounding air. Storm winds are of short duration but often very violent, reaching as much as 30 m/s (Estienne and Godard, 1970). The passage of a storm is accompanied by a change in the direction of the wind and an increase in the wind speed, with an increase in turbulence. The temperature of the air falls rapidly (Fig. 10a), whereas the relative humidity increases (Fig. 10b). Storms have a mechanical effect on all particles and insects floating in the air of even resting on leaves, because of their turbulent winds and extremely heavy rains, which can reach 70 mm/h. These strong rains and low temperatures could cause the death of larvae and even of insects. The storms can indirectly favour the development of the canopy by provision of rain. This increases the boundary layer of flight of the whitefly and allows a larger accumulation of insects.

Fig. 10a. The effect of a storm on air temperature.

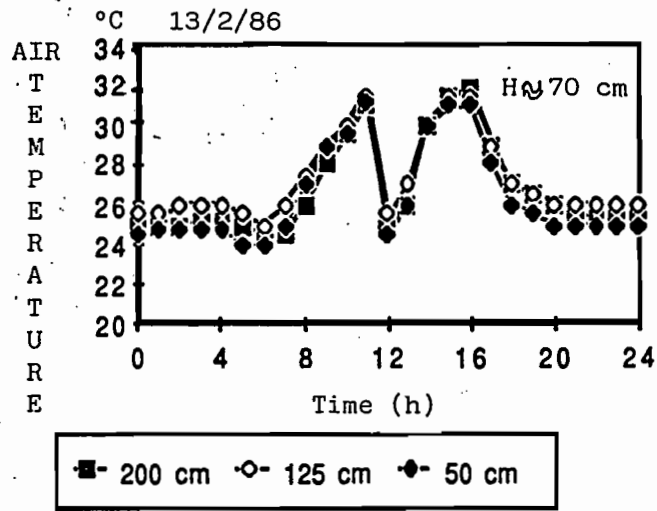
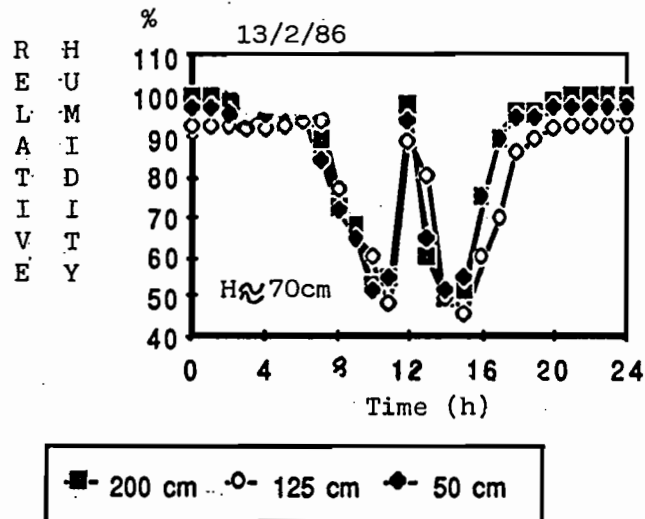


Fig. 10b. The effect of a storm on relative humidity.

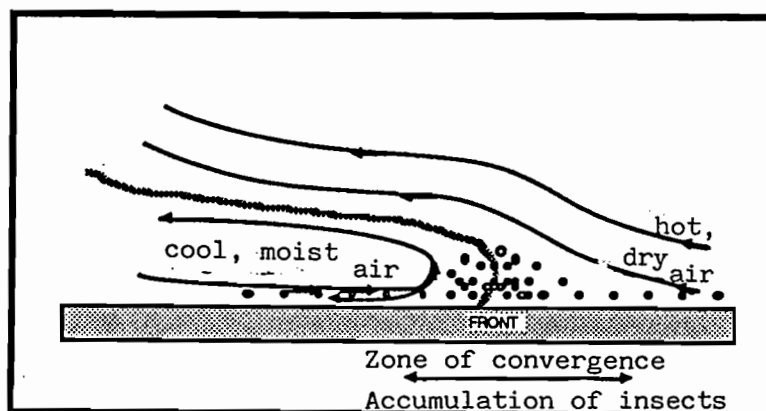




## Mechanism of accumulation at the front

Pedgley (1982) proposed a mechanism of accumulation of insects at fronts, which may apply not only to the ITF, but also to other types of front such as those observed with offshore breezes and storm winds. Insects are transported here and there in the front by a wet, relatively cool southwesterly wind in one direction, and a dry, hot northeasterly one in the other. These fluxes of insects converge in the front, then tend to be carried up by the divergent ascendant currents. If we accept that the insects then orient their flight towards the low layers, in order to stay in the zone of convergence because of the particular conditions which prevail there (a pocket of calm, humidity, and favourable temperature) or in order to avoid an unfavourable zone (turbulent movement, etc.), there then occurs an accumulation of insects at the front (Fig. 11). This mechanism proposed by Pedgley (1982) is presented only as a possibility, not as a certainty. In particular it is not known how strongly these movements of divergence at the front can pull the insects along and to what extent the whiteflies are capable of resisting them. Joyce (1973), however, thought this was one of the main phenomena explaining the dispersal and the fluctuations of populations of whiteflies.

Fig. 11. Likely mechanism of insect accumulation at the front level (after Pedgley, 1983).



## CONCLUSIONS

The reduction of the wind speed (which can reach 90%) by the canopy of the cassava and the phenomena of turbulence at the edge of a plot and above the canopy, must allow a larger accumulation of whiteflies. The speed and direction of the wind are only important outside the boundary layer of the insect's flight.

The interaction between the temperature, relative humidity, and wind speed in the course of a day and the height of the canopy should allow an insect much more activity at the beginning and the

end of the day. The passage of the ITF, of onshore or offshore breezes, or of storms could explain changes in the whitefly populations in a canopy of cassava.

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## VIRUS/VECTOR/PLANT RELATIONSHIPS

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### INTRODUCTION

The relationships between African Cassava Mosaic Virus (ACMV), the symptoms, the vector, and the host plant were investigated at the plant level. The concentration of virus was evaluated by the Elisa test, the intensity of the symptoms was recorded on a scale from 0 to 5, the size of the whitefly populations was estimated, and the growth of the cassava plant was taken into account by observing the size of the leaflets. The role of wild species as a reservoir of virus was also investigated.

### RESULTS

#### Cassava

##### - Localization of the virus in the leaves:

Leaves in stage 2, displaying symptoms of intensity 1 and 2, were sampled. The yellow and green parts of each leaf were detached separately and the extracts, sometimes clarified, were tested by the Elisa method to estimate their virus content. Without treatment with chloroform, ACMV was detected in the yellow parts of nine out of thirteen samples. The four other samples gave optical densities slightly higher than background level, but the significance of these results is uncertain. However, after clarification, the virus was then clearly detected in all the samples from yellow areas. The observed optical densities lay between the background reaction and 0.27 for the untreated extracts, and from 0.24 to 0.54 for the treated ones.

##### - Intensity of the symptoms and virus content:

A relationship exists between the intensity of the symptoms and the optical density of the unclarified extracts, the leaf having been sampled at stage 2. ACMV was present in most leaves with symptoms, even of class 1. The virus could also be sometimes detected, among those leaves which did not show external signs. There was a close relationship ( $r = 0.98$ )

between the intensity of the symptoms and the mean optical density of the samples of each class. Within each class, however, the observed optical density was very dispersed, and there was an important overlapping of classes.

### Evolution

The development of symptoms on the leaves, from the "apex" stage until the fall of the leaf, were observed on 25 diseased cassava plants, naturally infected in the field. The symptoms were clear and could be classified according to Cours's scale once the leaflets reached a mean length of 9 cm. Thereafter the symptoms did not change any further, though the leaf continued to grow. The correlation calculated between the intensity of the symptoms when first observed and then 20 days later reached 0.96. However, along a given stem, the intensity of the symptoms varied greatly, and two successive leaves might have intensities ranging from 0 to 4.

### Age of the leaf and virus content

The measurements of optical density were performed on the extracts from leaves sampled in the same conditions on a given stem:

- each of the first 12 leaves was tested separately.
- leaves from 1 to 4, then leaf 6, then every 4th leaf up to the 34th was also tested separately.

In all the plants the virus was detected in the extracts of the first three leaves (Leaf 1 = "first" leaf = stage 1: leaf having, on average, leaflet length of 12 cm). In three out of eight cases no virus was detected until the 4th leaf, and in one case not until the 6th. The highest optical density was obtained, in six out of eight cases, for leaf 1, in one case for leaf 2, and in another case for leaf 3. The level of virus was always highest in the younger leaves.

The growth of the leaf was estimated as a function of the length of the largest leaflet, a measurement closely related to the leaf's surface area. The maximum length was generally reached at stage 4 of the leaf, but sometimes also at stages 3 or 5. The most rapid rate of growth came between stages 1 and 2. With leaves before stage 1, we could sometimes detect the virus by the Elisa method, sometimes even with young leaves showing no apparent symptom of mosaic disease.

- Susceptibility to infection

The susceptibility of cassava leaves of various ages was estimated by Storey and Nichols. They placed groups of 100 whiteflies on leaves of increasing age and observed the number of plants showing symptoms afterwards. They observed that young, growing leaves were more susceptible to the disease than "adult" leaves, and that the oldest leaves were no longer susceptible.

## Identification of the vector

The great majority of puparia identified belonged to the species Bemisia tabaci. A few B. hancocki were sometimes seen.

## Distribution on the cassava plants

On 25 cassava plants we counted adult whiteflies and the number of larvae present on each leaf, from the apex to the oldest leaves. The count was made on plants aged 4, 5, or 6 months. The great majority of the whiteflies were found on growing leaves (numbers 2 and 3) and their number decreased with the age of the leaf. No whiteflies were observed on leaves 9 to 25. The distribution of the larvae followed the distribution of the adults, but shifted slightly.

## Virus-carrying ability

The virus-carrying power of the whiteflies in the fields of virus-infected cassava was evaluated periodically by collecting groups of 50 whiteflies and placing them on test plants. The percentage of whiteflies which transmitted the virus was always very low (0.45 on average), with the 90% confidence interval between 0.1 and 1.6%. The percentage varied little from one sample to another (from 0 to 1.7). The level of transmission observed depended also on the variety of plant used in the test; the highest level reached was observed with the susceptible variety H58. A very approximate upper-limit estimate of the level of virus-carrying flies was obtained, in the field, by comparing the size of the whitefly populations and the amount of contamination observed afterwards. This crude approach indicates that the virus-carrying power of the whiteflies present on cassava is, at the most, a few per cent.

## Adventitious plants

Several tens of plant species currently encountered in and around cassava fields were tested by Elisa. The presence of whiteflies was looked for. The results obtained were not unequivocal, given the presence of artefactual reactions with several species. All the same, the results of the Elisa tests, of the symptomatology, and the mechanical transmission test all agree and confirm what has already been published: two euphorbiaceae closely related to M. esculenta, M. glaziovii, and Jatropha multifida, and one Convolvulaceae, Hewitia sublobata, are reservoirs for ACMV. It appears, however, that the number of wild species that actually function as reservoirs is low and that their epidemiological role is limited, considering their limited diffusion and the small number of whiteflies that they harbour.

## DISCUSSION

A reservoir plays an effective role in the propagation of a pathogen, if the vector is present, if it acquires the pathogen and if it then inoculates other plants. The location of most B. tabaci on young, growing leaves favours at the same time the acquisition, the inoculation, and therefore the propagation of the

pathogen agent in the field, because of the high content of virus in young leaves and of their greater susceptibility to infection. This distribution of the vector and of the pathogen agent favours the spread of the disease. However, the virus-carrying power of the whiteflies present on cassava is much lower than that of other geminiviruses, particularly cowpea golden mosaic virus, where 70% of individuals are vectors.

In order to evaluate the actual role of a species as a source of infection, its range, the proportion of plants infected, their virus content, and the presence and abundance of the vector all must be taken into account. Two euphorbiacea, J. multifida and M. glaziovii, may well play a role in the epidemiology of the disease, because of their virus content and of the presence of B. tabaci, sometimes in large numbers. However, J. multifida is used as an ornamental plant in urban environments and its role as a source of infection in the field seems doubtful. On the contrary, M. glaziovii is present in the vegetation, although sparsely. We monitored the recontamination by B. tabaci of fields planted with healthy cassava cuttings, which was bordered at several points by groups of diseased M. glaziovii harbouring whiteflies. No focus of infection from these wild cassavas was observed. The absence of propagation of the disease from these glaziovii indicates that the role of M. glaziovii in ACMV field spread is probably very limited.

The virus content, the presence of the vector, and the large areas under cultivation make M. esculenta the principal reservoir of virus all year round and the most dangerous potential source of infection by whiteflies, especially in regions of heavy cassava culture. To assess the extent of transmission by whiteflies, it is necessary to investigate the recontamination of fields planted with healthy cuttings. This has been done, at the field and region levels, and is included in the communications which follow.

DISTRIBUTION AND SPREAD  
OF AFRICAN CASSAVA MOSAIC  
IN A CASSAVA FIELD

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INTRODUCTION

Our investigation of the spatial development of African Cassava Mosaic aims to characterize and understand the factors that govern the development of the disease at the field level. The distribution of the disease in a field is rarely completely random or totally homogeneous. Most of the time, the characteristics are related to the position of the sources of infection, the movements of the insect vector, and the ecological site of the field. The disease is characterized by some regions where the frequency of infection is high and others in which it is low, and investigation should reveal the gradients thus created. The observed infection gradients (or disease gradients) may have two different bases. They may be due to variations, across the field, of factors such as soil, vegetation, or microclimate; these are called environmental gradients. Or they may be due to spatial variations in the quantity of inoculum. These are called dispersal gradients.

These two types of gradients are closely related to the various movements of the whitefly and to climatic conditions. Consequently, the results concerning the dispersal of the disease have to be related to the knowledge available about the microclimate in a cassava field and the movements of Bemisia tabaci; information about these has been presented earlier. A model of dispersal is proposed here.

At the same time, we have sought to distinguish the contamination arising from outside the field from that arising within it. Distinguishing between these two types of contamination is essential for the establishment of methods to control the disease.



## RESULTS

### Trapping of whiteflies

The trapping was done in a field of 1 ha in area from September to December 1982 inclusive. During this whole period, there was a prevailing wind from the southwest; this prevails throughout most of the year. The spatial distribution of the captures in water traps during the first two months of cultivation (October-November 1982) was mapped. The distribution was not homogeneous within the field: there were more captures along the south and west edges. During the third month of cultivation (December 1982) the traps were no longer placed in the center of the blocks, but at the junctions of the paths. The greatest number of captures were, again, recorded in the southwest section of the field.

### Distribution of the disease within the fields

Contamination of each block of one hundred plants in a 1-ha cassava field after 3 months of cultivation was measured. Certain characteristics of the disease's distribution can be seen. The infection was not uniformly distributed throughout the field; instead, the incidence of the disease was higher along the south and west edges than along the north and east edges. This particular distribution shows up as a curved contamination gradient along the diagonal running southwest. This gradient occurred in all the fields investigated, despite very different surfaces and exposure conditions.

### Dispersal of the disease from the focus

The figure shows the dispersal of the disease five and a half, and six and a half months after planting, upwind and downwind of a source consisting of 50 plants (F 50) derived from diseased cuttings. The dispersal of the disease was noticeable on both dates, but its range remained very limited, not more than a few metres. It shows up as a dispersal gradient. Unlike the gradients investigated previously (where there was no direct contact with a defined source of inoculum), the dispersal of the disease took place in all directions. It nevertheless seems that there was a more marked dispersal in the direction facing into the wind.

### The environmental gradients

The environmental gradients are related to the passive movement of the whitefly. The whiteflies carried by the wind gathered preferentially on the first edges of the plot that they came to, that is, the upwind edges. Several hypotheses have been proposed to account for the mechanism of this accumulation. For example, it could be related to phenomena of turbulence and/or reduction of wind speed at the edges of the field. Whatever the underlying mechanism, this distribution of the vector results in a greater contamination in the upwind part of the field, and the environmental gradients thus appear. Many observations based on the captures in insect traps remote from all sources of whiteflies suggest that distances of several kilometres may be covered in this way.

## Dispersal gradients

Dispersal gradients, by contrast, are related to the presence of well-defined sources of diseased plants. With African Cassava Mosaic, these gradients are limited in extent, do not exceed several metres, and do not show any particular orientation relative to the wind. It cannot be excluded, however, that in certain conditions this local dispersal faces the prevailing wind. This limited dispersal is probably the consequence of active movements of the whiteflies within the boundary layer, which seem to take place more obviously against the prevailing wind, but does not exceed several metres, whatever their direction.

The dispersal of the disease from foci of diseased plants shows that cassava plants within the plot do indeed contribute to its contamination. In the following experiment, we sought to quantify the fraction of the contamination that was external to the field (primary spread) and the fraction due to interior sources, consisting of plants that became diseased with the passage of time (secondary spread). We therefore followed the contamination in fields from which the diseased cassava plants were rogued as soon as the symptoms appeared. We have no evidence to suggest that the cassava plants were the source of the virus before the appearance of symptoms. Even if that was the case, the systematic roguing of the virus-infected plants should reduce to a minimum any secondary spread. So, the disease progress curves in this type of plot reflect mainly the contamination arising from outside the field. This spread is important and results from the influx of virus-carrying whiteflies throughout the year. Detailed analysis of the inoculum pressure and the factors that govern its fluctuations will be given in a subsequent communication ("Temporal development of African Cassava Mosaic").

We compared the kinetics of contamination in plots where a 4% source of inoculum was available, with fields which had none. It was clear that initial sources did contribute to the contamination, since the disease progress curves in the two conditions differed. The difference was limited, however, to not more than a few per cent.

## Development of the disease and distribution of host plants

It is known that, as with many viral diseases, the distribution of the host plants influences the development of the disease. We investigated the influence of the planting density on the incidence. It appears that the plots in which the density was lowest were contaminated the most rapidly, whereas those in which the density was greatest had the lowest incidence of the disease. These differences in the spread of the infection may be related to differences in the size or behaviour of the insect vector.

## CONCLUSION

These results show the close dependence that exists between the dispersal of the disease and the movements of the whiteflies which themselves are largely dependent on the characteristics of the wind. They emphasize the usefulness of an interdisciplinary

approach to the epidemiology of a viral disease. However, we stress that the main objective of this investigation was to understand the mechanisms that govern the progress of the disease, on the scale of the individual field. These basic relationships between virus, vector, plant, and environment are probably valid whatever the agronomic situation. In practice, however, the fields used in the investigation were homogeneous and were large in area. Judging from the responses to the questionnaires by representatives of each country, we see that in the great majority of cases cassava is grown in very small plots, and in association with other crops. We have therefore sought to establish how the disease behaves when cassava is grown with other plants. We chose the combination of cassava and maize, with various densities and various dates of planting of the maize. The infectious processes in the various combinations did not appear fundamentally different from that in cassava grown on its own. A major study of how the disease behaves in the conditions of peasant farms remains to be carried out, but a number of methodological obstacles must be overcome first.

In practice, the heavy primary contamination which exists at Adiopodoumé makes it somewhat uncertain whether healthy fields can be maintained with the current varieties. Though such agricultural methods as eliminating diseased plants, changing the density of planting, or trying to achieve some isolation may reduce the incidence of the disease, the inoculum pressure is such that this control strategy remains risky with the varieties in use now. This situation has been confirmed in the coastal region of the Ivory Coast. It does not seem impossible, however, to be able to maintain healthy fields in such regions so long as varieties are used that are highly resistant in the field (see the accompanying paper "The resistance of cassava to African Cassava Mosaic"). This situation is not, however, characteristic of the entire Ivory Coast. These are regions where healthy fields of relatively susceptible varieties can be maintained healthy because of the low inoculum pressure there (see "Development of the disease on a regional scale").

**SESSION C**

**EPIDEMIOLOGY**

Chairman  
Dr. ROSSEL  
Rapporteurs  
Dr. FRISON  
Dr. NOLT

AUTOMATIC MAPPING OF THE SPREAD  
OF  
AFRICAN CASSAVA MOSAIC VIRUS

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INTRODUCTION

Modern statistical methods make it possible to describe and structurally analyse spatial variables more effectively than methods involving only means and variances (soil type, spread of disease, extent of pest attacks). These techniques also enable isovalues to be used for mapping purposes with respect to a given variable.

These modern methods, developed during the 1960s (Matheron, 1963, 1965; Krige, 1966) were perfected by geostatisticians who use them extensively. They have now been successfully applied to the field of agronomy by soil scientists at IRAT-CIRAD and at ORSTOM, by agroforestry specialists at CTFT-CIRAD, plant virologists at ORSTOM and entomologists and phytopathologists at IRHO-CIRAD.

THE CONCEPT OF REGIONALIZED VARIABLES

A variable is "regionalized" when its values are dependent on its position in space (Matheron, 1965). Moreover, it is commonly accepted that there exists a dependence between two closely linked samples which determines correlations between values measured in certain zones. This indicates that there is a structure within the area explored.

A basic sample (Matheron, 1965) clearly shows what is meant by this concept. Let us say that two series of measurements taken for a given variable at regular intervals along 1 row in the field give this following sequences:

A: 1-2-3-4-5-6-5-4-3-2-1,  
B: 1-4-3-6-1-5-3-4-2-5-2.

Sequence A is characterized by a well defined symmetric structure whilst that of sequence B, if there is one, is difficult to determine and very irregular. Nonetheless, these two series of 11 measurements have the same mean and variance. This shows that it is impossible to describe spatial variable distribution using these traditional concepts only.

A method is therefore required which simultaneously analyses localization, continuity, anisotropy and the transitive character of such a variable.

The analysis tool is the semi-variogram, defined for any distance  $h$  as the mean of the squares of deviates of values determined by the variable at all points separated from each other by  $h$ .

$$G(h) = 1/2E [F(x+h) - F(x)]^2$$

It can be seen that the semi-variogram and covariance correspond through the relationship:

$$G(h) = C(0) - C(h); \text{ or } C(0) = V.$$

At great distances, correlations between points can no longer be ascertained, with  $G(h)$  tending toward maximum value  $C(0)$ . The value  $a$  of  $h$ , corresponding to this maximum, is called the semi-variogram range. The behaviour of function  $G(h)$  with  $h < a$  characterizes the degree of regularity of the regionalized variable.

To exploit data on a simple basis, a semi-variogram constructed for any  $h$  by the following mean is generally sufficient:

$$G(h) = (1/2 N_h) [F(x_i + h) - F(x_i)]^2,$$

where  $N_h$  = the number of couples  $(x_i, x_{i+h})$ .

Different types of semi-variograms can be observed which can be adjusted to certain traditional models.

Fig. 1. Semi-variogram characteristic of the nugget effect. Phytophthora at Assinie (cumulative rate of losses).

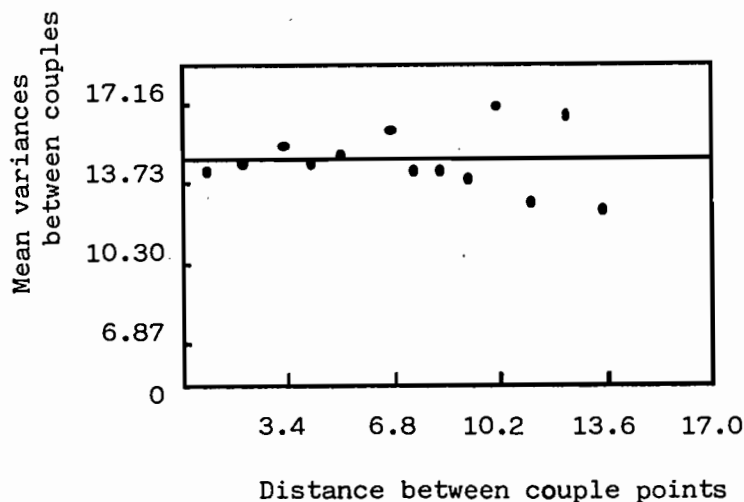


Fig. 2. Semi-variogram characteristic of the linear model.  
African Cassava Mosaic Virus. ORSTOM, Adiopodoumé.

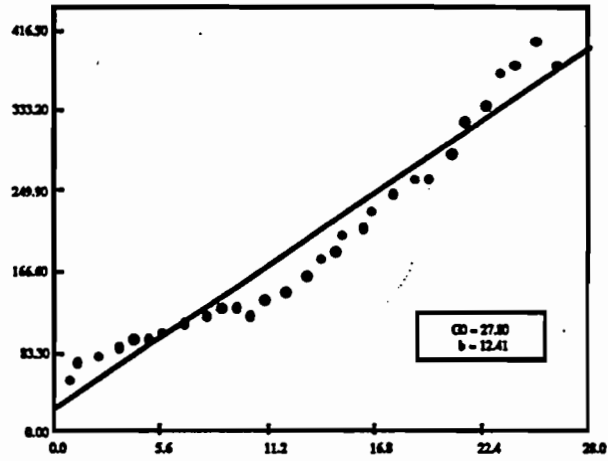


Fig. 3. Semi-variogram characteristic of the Gaussian model (simulation).

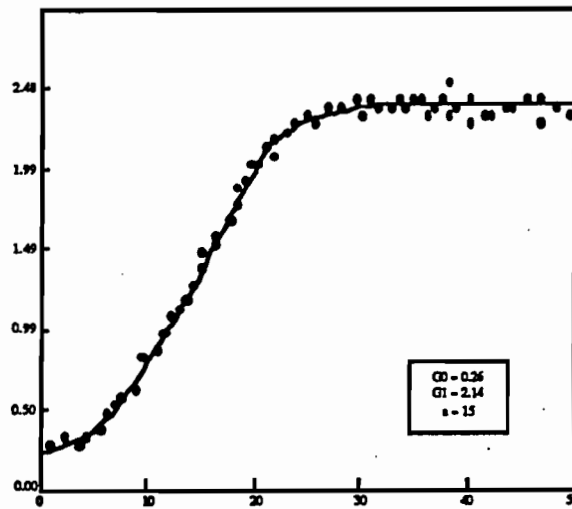
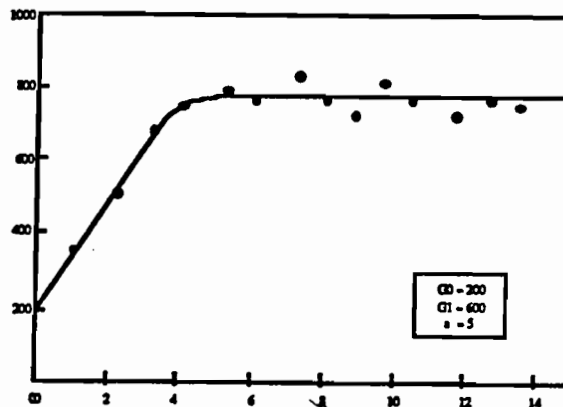


Fig. 4. Semi-variogram characteristic of the spherical model.  
Thickness of top soil-silt. Hole Farm, Norfolk.



If the correlations are null, the semi-variogram immediately takes on its maximum value (this is strictly a random phenomenon), which is represented by a flat semi-variogram (Fig. 1): we are dealing with a pure nugget effect.

This nugget effect, dependent on a microstructure, is generally always superimposed on other structures, though the semi-variogram observed can be adjusted to the following theoretical models (GO = nugget effect):

- the linear model, without a plateau (Fig. 2)

$$G(h) = GO + bh;$$

- the Gaussian model, whose practical range is  $a' = \text{sqr}(3a)$  (Fig. 3)

$$G(h) = GO + G1 [1 - \exp(-h^2/a^2)];$$

- the spherical model, whose range is  $a$  (Fig. 4)

$$\begin{aligned} G(h) &= GO + G1 [3/2(h/a) - 1/2(h/a)^3] && \text{for } h \leq a, \\ G(h) &= GO + G1 && \text{for } h > a; \end{aligned}$$

- the exponential model, whose practical range is  $a' = 3a$  (Fig. 5)

$$G(h) = GO + G1 [1 - \exp(-h/a)].$$

Simulation studies have shown that the evolution of pest attacks or diseases can be followed by observing the semi-variogram corresponding to the simulated control dates.

Fig. 5. Semi-variogram characteristic of the exponential model (simulation).

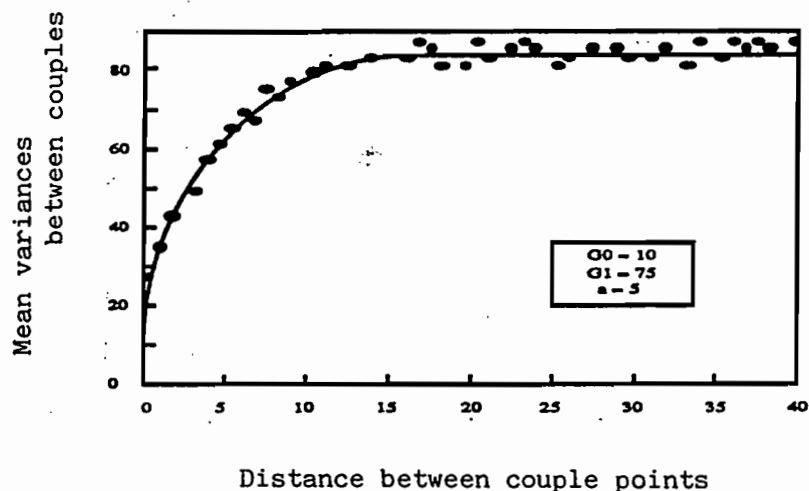
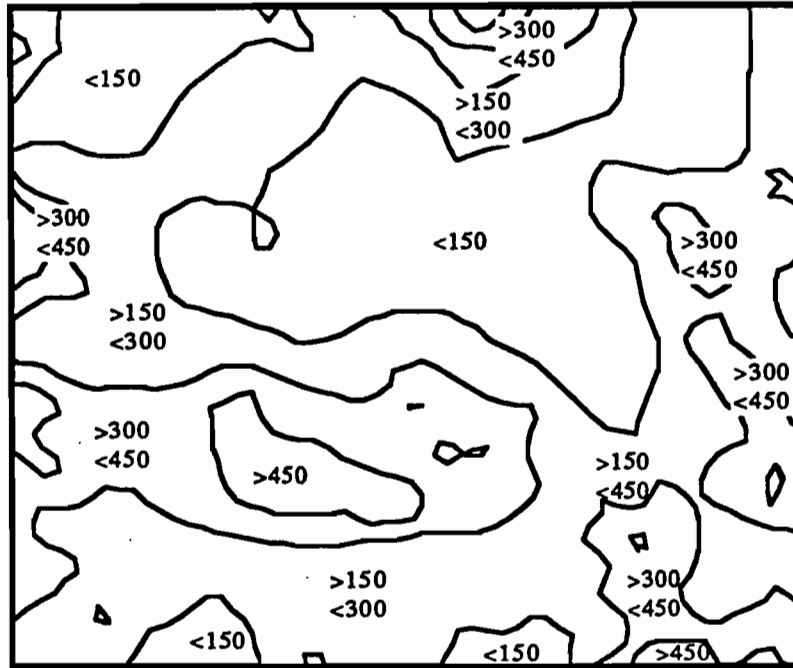




Fig. 6. Isovalue mapping of a density variable.



#### KRIGING

Kriging is a method involving local estimates of variable values at all points of a geographical zone, based on the theory of regionalized variables, which takes semi-variograms into account. This method was primarily developed by Matheron (1963, 1965) and Krige (1966) to estimate mineral resources for the exploitation of mines, for which it is used more and more frequently.

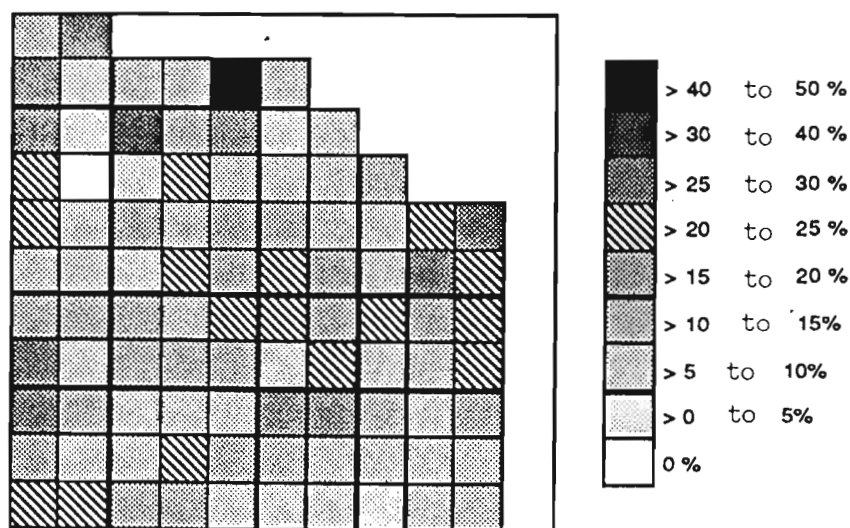
This method is termed unbiased because, unlike other simple methods, it plots the mean and variance of the phenomenon, restores their value measured at sample points and ensures a minimum estimate variance.

It is not worthwhile using a sample point located at a distance greater than the semi-variogram range to estimate the value  $F(x_0)$  at any one point.

This method can also plot isovalue maps or contour lines (Fig. 6); whilst other graphic methods enable mapping of experimental plots (Fig. 7) whatever their shape or arrangement, or by plant. It is absolutely obligatory that the variable  $F(x)$  has a variable of density character (weight, thickness, rate of attack, population density, etc.).

This technique is very useful for defining pest attack zones and for studying plant disease propagation characteristics.

Fig. 7. Diagram of the spread of Phytophthora affecting coconut at Assinie. Mapping of the cumulative loss rate per subplot (3 years after the first outbreak).

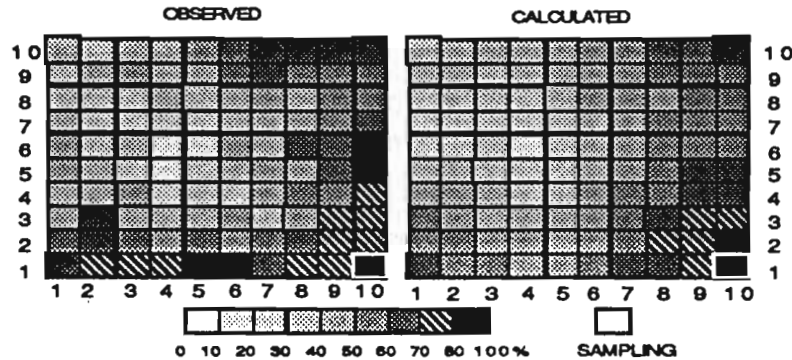


#### APPLICATION TO AFRICAN CASSAVA MOSAIC VIRUS

In this particular study, the contamination is essentially a primary contamination, coming from outside the field, following the direction of the prevailing wind and with a border effect as found in field experiments. The experimental semi-variogram which characterizes this case of contamination is the linear model, shown for the first time on ACMV and then on Bud Rot affecting oil palm tree in Peru and Ecuador. The studies (comparing the calculated values obtained from a given sample and from a hmax distance from which the  $x_i$  values are considered to have no more influence upon this  $x_0$  calculation), show that in the case of ACMV, a sample of 7% (7 blocks of 25 or 100 plants in a trial of 50 to 100 blocks) and a hmax distance near 5 blocks (25 to 50 meters) give the best estimates.

Figure 8 shows the results obtained with the automatic mapping of a cassava field of 1 ha, 6 months after planting, with a sampling of 7%. The correlation coefficient between the observed and the calculated values is 0.81. Nevertheless, the knowledge of the existence border effect, particular to the spread of the ACMV disease, implies that a structured sample collection rather than a random sample collection should be chosen.

Fig. 8. Comparison between observed spreading of ACMV on a cassava field of 1 hectare, and one obtained by kriging with a sampling of 7% (6 months after planting).



The study of temporal evolution of the semi-variograms shows that we pass progressively, as a result of propagation, from a typical nugget effect to the linear model without plateau.

#### CONCLUSION

Entomologists, phytopathologists, and phytovirologists presently have at their disposal a mathematical tool enabling the structural analysis of the spatial variables with which they work.

These methods also make it possible to draw up isovalue maps from samples for a given variable; this is known as kriging.

We successfully used semi-variogram analysis to follow the epidemiology of ACMV in Ivory Coast. These techniques make it possible to reduce field controls by a factor of 14, by sampling 7% of the sub plots, in order to map the distribution of ACMV on experimental plots at ORSTOM in Ivory Coast, while correctly giving the necessary structural information needed to study the spread of the ACMV viral disease in the experimental trials.

Readers interested in these analysis and calculation techniques can refer to the reference works given in the bibliography for additional information.

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## TEMPORAL DEVELOPMENT OF AFRICAN CASSAVA MOSAIC

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### INTRODUCTION

The development over time of a viral disease depends on many factors. Among those investigated in the case of the African Cassava Mosaic are the place and the date of planting, the clone used, and the situation in the field.

#### Factors that affect the development of the disease

Completely different epidemics may develop on different sites, even if they are very close to each other. For example, the degree of contamination of initially healthy fields was much lower in Toumodi (200 km north of Abidjan) (2%) than in Tontonou (36%), which is only a few kilometres away from Toumodi, and lower than in Adiopodoumé (20 km west of Abidjan) (62%) (see the accompanying paper "Epidemiology of cassava mosaic on a regional scale in the Ivory Coast").

In a given field, the incidence of the disease depends on its position in the field. The recontamination was much lower at the center of the field and near the downwind edges (18 and 34%, respectively) than at the upwind edges (89%) (see the accompanying paper "Distribution and spread of African Cassava Mosaic in a cassava field").

The clones showed a high variability of resistance in the field. For example, a very low incidence of the disease was observed in a hybrid of M. esculenta and M. glaziovii (18%), whereas strong contaminations were observed among the local clones (89%) (see the accompanying paper "Resistance of cassava to African Cassava Mosaic").

In a given site, for a similar exposure and a given clone, the incidence of the disease depended broadly on the planting date: it was low in October (12%), high in April (44%), and very high in December (89%). Our investigation of this fluctuation throughout the year is the focus of this communication.

## Annual fluctuation of the inoculum pressure

From 1981 to 1986, an area of 0.1 ha was planted with cassava each month. Every week samples were taken, the incidence of the disease was determined, and the infected cassava plants were rogued. An index of inoculum pressure was calculated from the increase in incidence of the disease in the cassava plots from the second to the third month. The whitefly populations were evaluated by weekly samplings. Leaf growth was monitored by the Laboratory of Agronomy (work of M. Raillaç), and the leaf area index (LAI) was calculated 60 to 90 days after the planting. Detailed climatic data are available for the whole of this period. The curves of progression of the spread of ACMV differ from month to month and the "classic" adjustments may not be applied to the whole set of contamination curves. A high contamination despite the roguing of diseased plants indicates that there was an influx of virus-bearing whiteflies all year long in the fields. This situation differs from that in Toumodi, where the level of contamination registered was low.

According to the results obtained over 5 years, it appears that an annual fluctuation occurred in each of the following variables:

**Inoculum pressure:** High from March to July, low from August to November. Obviously, this fluctuation of the inoculum pressure can be observed only if virus-free planting material is available and if it is planted on different dates. Such conditions are generally not fulfilled in Africa, where the vast majority of plant material is virus-infected. So, it is not surprising that this fluctuation in inoculum pressure was not often mentioned in the national questionnaires.

**Whitefly population:** High from February to June, low from July to October. This fluctuation in the populations of aleurodes throughout the year, linked with climatic data, was often noted in the national questionnaires. The relationship between the peaks in the whitefly population and the rainy season and, conversely, the absence of whiteflies in the dry season has been reported several times.

**Cassava leaf growth:** High from February to May, low from June to September. It is recognized that certain periods favour the growth of cassava.

**Temperature:** Although the variation was not great, the highest temperatures were observed from February to May and the lowest from June to October.

## Virus/vector/plant/environment relationships

We have investigated the relationships between the virus, the vector, the plant, and the climatic conditions of the environment. With appropriate shifts in time, there are close relationships between rate of contamination, size of the aleurode populations, and climatic data. On the basis of the relationships mentioned above (bearing in mind that it is just one possible explanation), a simple series of events can be envisaged that takes account of

the relationships between the environment, the vector, and the contamination. The climatic conditions, in particular the temperature, determine the size of the populations of whiteflies, which acquire and then transmit the virus. This would determine the contamination rate in the cassava fields. This outline, in view of the complexity of the relations between the different actors in the epidemic, must be only a simplification of reality. However, it must be stressed that the data obtained in greenhouses relating to the duration of development from egg to adult (approximately 3 weeks at 26 C) and the latent period of the pathogen agent in the plant (approximately 4 to 6 weeks) are compatible with the proposed diagram.

#### Prediction of the development of the disease

In order to test the value of the relationships shown, we have sought to use them predictively. The descriptive value of our model is high. The predictive value of the model is also high.

The predictive value of a model is evaluated by comparing calculated values or "predictions" with observed values which were not used in the construction of the model. Two types of prediction are possible. From the mean climatic conditions of Adiopodoumé, one may establish a curve of "typical" contamination and define the probable periods of high and low inoculum pressures. Given the variability of the climatic conditions from year to year, this prediction only approximates the real situation. Conversely, it is possible to predict the rate of contamination 2 months in advance, with a high degree of certainty. Such a predictive model has limits. Though this model accounts for and appropriately predicts the speed of contamination at Adiopodoumé with good precision, it may apply only in the conditions tested and therefore may not automatically be extended to other places than Adiopodoumé.

#### CONCLUSIONS

There is therefore an annual and seasonal fluctuation of the inoculum pressure. On the other hand, there are highly resistant varieties in the field. We are at present testing some of them, but it now appears that it is possible to maintain, at Adiopodoumé, and therefore in regions of strong inoculum pressure, several successive years of observation of the fields where the contamination is of the order of several per cent. It is tempting to try to recommend planting dates based on the knowledge of this fluctuation, using specially chosen varieties. It seems that such recommendations would make sense only subject to two conditions: first that the dates recommended be compatible with the agronomic requirements for cultivation; and secondly, that this planting guarantees the farmer that the cassava plants derived from healthy cuttings will give enough healthy plants each year to provide a sufficient stock for replanting the following year (it is probably not possible to provide healthy material to the farmer every year, for obvious reasons of cost). Fluctuations from year to year make such guarantee impossible. This uncertainty is the main weakness of such control methods. No one can guarantee the farmer that the recommended variety, combined with the recommended agricultural

methods, will enable him to overcome the disease with any degree of permanence. This problem is particularly complex, and crucial, and it constitutes one of the main concerns of this colloquium: how viable are such control methods, in biological, economical, and human terms?



SOME ASPECTS OF THE EPIDEMIOLOGY OF  
AFRICAN CASSAVA MOSAIC VIRUS  
IN COASTAL DISTRICTS OF KENYA

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INTRODUCTION

Bock & Guthrie (1982) and Bock (1983) reported a low rate of reinfection of African Cassava Mosaic Virus (ACMV) in mosaic-free plots and maintained that the disease could be effectively controlled in Kenya by using mosaic-free planting material. They demonstrated that rate of spread of ACMV into ACMV-free plots over a five year period of experimentation was consistently low (less than 2%), irrespective of cultivar type, size of plot (0.02-1.00 ha), location (on agricultural research stations and in farmers' fields), and annual or regional climatic differences (Coast and Western Kenya, 1974-1980).

Observations on the incidence of ACMV among newly introduced Brazilian cultivars revealed that they were highly susceptible, but that, in spite of this, incidence apparently varied with locality. It seemed possible that these varieties, used together with local Kenya cultivars and resistant hybrids, offered a means of studying the epidemiology of ACMV in different climatic zones and in different situations within those zones.

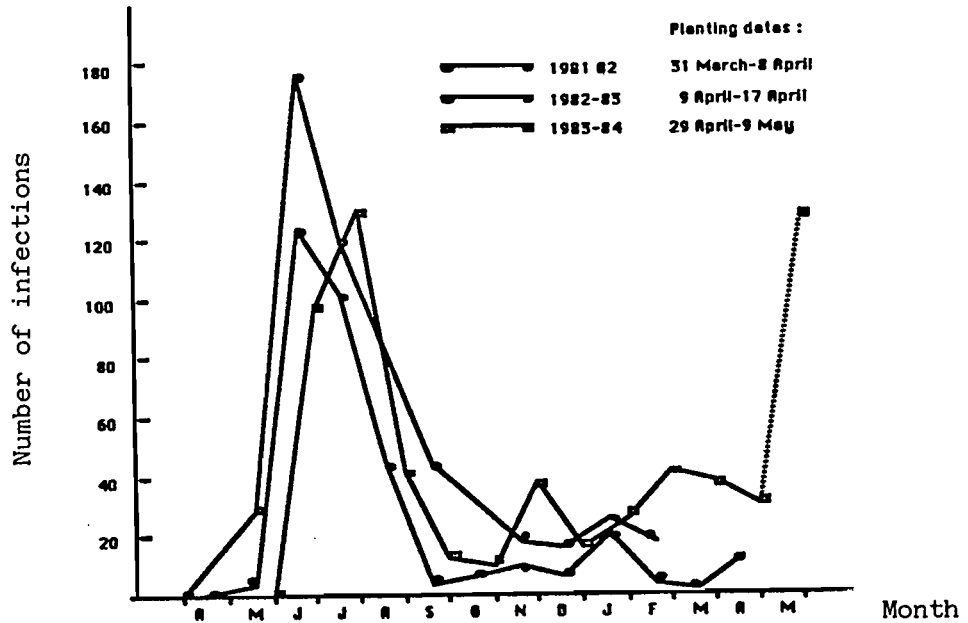
EXPERIMENTATION

During three years of study (1981-1984), sites were selected in a north-south alignment between Magarini Settlement Scheme in the North and Kiruki in the South, a distance of about 200 km. Experimental design of these standard epidemiology plots consisted of randomized plants. Incidence of ACMV was compared among Brazilian cassavas (several cultivars), local Kenyan cultivars (Kibandameno, Mwakazanga, Chokorokote) and resistant hybrids (46106/27 and 5543/156). Infected plants were rogued at each successive recording date. All experiments were planted at the onset of the main rains (April) and were terminated after about 12 months. The results of these studies have been reported in detail (Bock, 1984; Robertson, 1985). They are summarised in Figs. 1 and 2 and in Table 1.

**ANNUAL FLUCTUATIONS IN INCIDENCE**

Fig. 1 illustrates that a single major period of infection occurred in May, June and July, with a marked decline in infection rate in August, September and October. This was consistent over three years. A second less pronounced peak occurred later in the season, and seemed also to be associated with the rains which occur variously and somewhat less predictably between September and December.

Fig. 1. The incidence of African cassava mosaic virus in coastal districts, Kenya, 1981-82, 1982-83, 1983-84: total infections at all sites, by months.



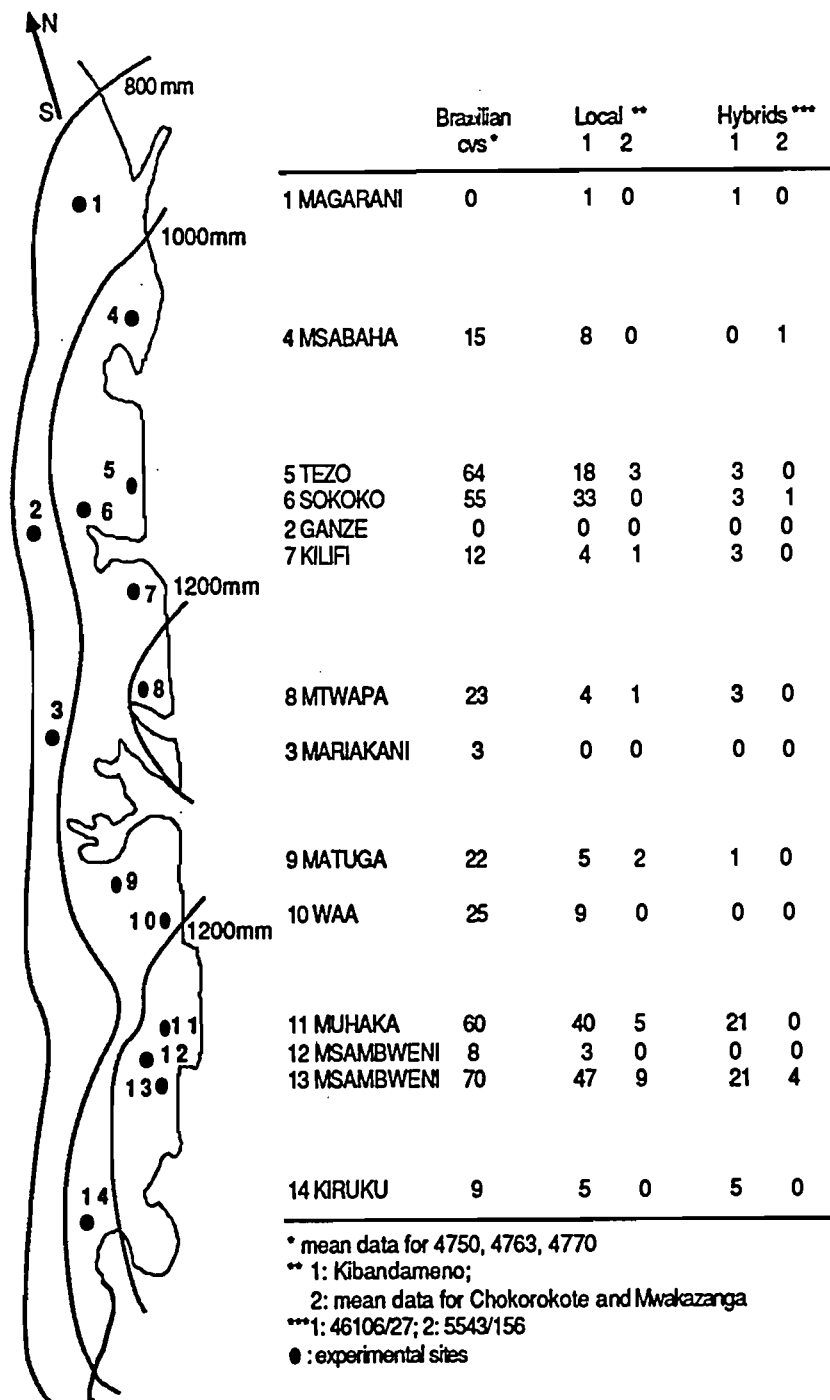
Robertson (1985) studied whitefly populations in cassava plots and found no correlation between numbers of vector and incidence of disease. He also conducted preliminary studies on rate of leaf production and growth and his data afford a probable explanation for the paradox. Rate of leaf production and increase in leaf size is rapid until August; there is then a marked decrease during October, November and December. Storey & Nichols (1938) found that transmission of virus occurred only through immature leaves. Probability of infection might therefore be expected to be highest during periods of rapid growth, and least when growth is retarded.

**INCIDENCE IN RELATION TO LOCALITY**

Fig. 2 and Table 1 summarise variation in incidence of ACMV in relation to locality in highly susceptible (Brazilian entries 4750, 4770, 4763), moderately resistant (Kibandameno and 46106/27)

and highly resistant (5543/156, Chokorokote and Mwakazanga) cultivars. Several salient features concerning the epidemiology of ACMV in coastal districts of Kenya may be deduced from these data, in conjunction with details of sites.

Fig. 2 Mean incidence (%) of African Cassava Mosaic Virus in Brazilian, local and hybrid cultivars in relation to distribution of sites and approximate mean annual rainfall isohyets (mm), Coast Province, Kenya, 1981-84.



Source: Bock, K.R. African cassava mosaic in Kenya. Proceedings of the AOUSTRIC Symposium on virus diseases in Africa. Nairobi, Kenya, 31 May-7 June, 1976 .

Table 1. Incidence of African Cassava Mosaic Virus (% plants infected) on different cassava cultivars at different localities, Coast Province, Kenya, 1981-1984.

Sites	4750	4770	4763	KIBa	27	CHO	MWA	156
1. Magarini 81-82	0	0	0	3	3	0	0	0
Magarini 82-83	0	0	0	0	0	0	0	0
Magarini 83-84	0	0	0	0	0	0	0	0
2. Ganze 83-84	0	0	0	0	0	0	0	0
3. Mariakani 82-83	3	0	0	0	0	0	0	0
Mariakani 83-84	0	11	2	0	0	0	0	0
4. Msabaha 81-82	23	15	0	5	0	0	0	0
Msabaha 82-83	10	8	3	8	0	0	0	0
Msabaha 83-84	20	25	28	10	0	0	0	0
5. Tezo 81-82	68	53	70	18	3	5	0	0
6. Sokoke 83-84	56	50	58	33	3	0	0	1
7. Kilifi 82-83	16	35	20	8	5	0	0	0
Kilifi 83-84	3	0	0	0	0	0	0	0
8. Mtwapa 81-82	38	30	15	5	8	3	0	0
Mtwapa 82-83	3	5	8	3	0	0	0	0
Mtwapa 83-84	13	21	13	3	0	0	0	0
9. Matuga 81-82	33	13	30	9	3	5	0	0
Matuga 82-83	3	5	8	3	0	0	0	0
Matuga 83-84	37	34	39	3	0	0	0	0
10. Waa 83-84	28	33	15	9	0	0	0	0
11. Muhaka 82-83	78	57	46	40	21	0	5	0
12. Msambweni 83-84	10	10	5	3	0	0	0	0
13. Msambweni 81-82	100	100	100	72	64	20	23	10
Msambweni 82-83	50	45	57	38	0	0	0	0
Msambweni 83-84	68	62	45	31	0	8	0	2
14. Kiruki 82-83	20	5	3	5	5	0	0	0

a KIB = Kibandameno; 27 = 46106/27; CHO = Chokorokote;  
MWA = Mwakazanga; 156 = 5543/156.

Source: Bock, K.R. African Cassava Mosaic in Kenya. Proceedings of the OAU/STRC Symposium on Virus Diseases in Africa. Nairobi, Kenya, 31 May - 7 June, 1986.

## DISPERSAL OF ACMV

ACMV incidence in highly susceptible cassavas was either absent or extremely low in sites located between the 800 mm and 1000 m isohyets. Although these were more or less isolated from areas of dense cassava cultivation, the south-east trade wind, which blows steadily throughout the period when infection rate is highest (Fig. 1), traverses many hundreds of small plots of infected cassava which occur between the coast line and inland sites. It appears, therefore, that long-range dispersal of ACMV proceeds only at a very low rate, and seems to be of no significance in the progress of seasonal infections.

In contrast, short range dispersal was apparent at several sites, of which Msambweni site 13 (Fig. 2) was typical. This site was more or less enclosed, the prevailing wind blowing through and across about 1.5 km of a heavily tread area of long-standing habitation, with many small patches of 1-3 year-old cassava with a very high incidence of ACMV. The nearest infected cassava was cultivated 30 m upwind. Average ACMV incidence in susceptible varieties over the three year period was 70 per cent. Sites 5 (Tezo), 6 (Sokokke) and 11 (Muhaka), where incidences of 50-60% were recorded, were similar in their proximity to infected plants. Msambweni site 12 was less than 3 km from site 13, but was surrounded on three sides by a coconut plantation and isolated from infected cassava by about 1 km; incidence over a 12-month period was 8 per cent.

We may therefore deduce that, in Kenya, effective dispersal of ACMV leading to comparatively high incidence of disease in susceptible cassava is restricted to short distances, measurable in tens of metres. Long range dispersal, over distances greater than a kilometre, proceeds only at low rates.

## CONCLUSIONS

It seems evident from the data that, with the exception of those sites where test material was grown in close proximity to sources of infection, infection pressure in Kenya does not seem to be high. Average annual infection rates of highly susceptible varieties at all other sites varied between 9 and 25 per cent: rates of infection of local Kenyan varieties and resistant hybrids were either consistently very low, or infection did not occur.

The results confirm that, in Kenya, ACMV can be successfully avoided by using mosaic free planting material of cultivars which contain a measure of resistance. The conclusion remains consistent with the suggestion that in Kenya man is the principal vector of ACMV, by means of his inadvertant use of infected cuttings as planting material. However, this assertion must now be modified to exclude situations where cassava is exposed to massive short range dispersal of infective vectors.

Successful field control of ACMV is thus seen to involve the use of mosaic-free planting material as a first prerequisite, but strategies should also take into account varietal susceptibility and both regional and local factors governing incidence.

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EPIDEMIOLOGY OF  
AFRICAN CASSAVA MOSAIC  
ON A REGIONAL SCALE IN THE IVORY COAST

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African Cassava Mosaic Virus (ACMV) is transmitted in two different ways:

- by the aleurode Bemisia tabaci,
- by man, when he plants contaminated cuttings.

Tests on the east coast of Kenya have shown that farmers play the main role in the spread of the disease, and the insect vector only a minor one (Bock, 1983). These conclusions, based on the results of epidemiological investigations in East Africa, contrast with those of workers in West Africa, where the dynamics of contamination of healthy cassava are extremely rapid and therefore seem to indicate that the spread by the vector is more effective (Leuschner, 1977; Fargette *et al.*, 1985). In order to define the exact role of the vector in various ecological conditions, and in the context of comparative epidemiological investigations, we have performed a multi-site experiment at a regional level in the Ivory Coast. To do this, we took into consideration the dynamics of contamination of healthy cassava plants, the vector populations, the environmental situations in the fields, and the growth of the plants.

#### MULTI-SITE TRIALS. PLANTING MATERIAL

Most of the trials involved planting with the clone CB (a moderately susceptible clone originating in the Congo), and the several other cassava clones used included H58 (a very susceptible clone from Madagascar) and BR (Bonoua Rouge, a resistant clone from the Ivory Coast).

#### GEOGRAPHICAL LOCATION

The experiments were done in two very different places in the Ivory Coast:

- the first is in the south of the country, in the forest region, where there are two rainy seasons and an annual precipitation of 2000 mm;
- the second lies in the savanna region, in the centre of the country, where there is only one rainy season and an annual precipitation of 1000 mm.

#### EXPERIMENTS

In the forest region, we worked for one year with only one clone (CB), but in various environmental conditions.

In the savanna region, we compared clones H58 and BR in two different environments, also for one year.

Finally, we compared the two regions with each other, by monitoring the reinfestation of the fields planted with several clones for several years, or at different planting dates for a single clone.

In each region, the areas of the fields ranged from 0.06 to 1 ha, but they were all oriented in the direction of the prevailing wind, in order to obtain a homogeneous contamination of the plots (Fargette et al., 1985).

#### VARIABLES MEASURED

Every month for 9 months, we recorded the contamination of the plants, the vector population, and the plant growth. The whitefly populations were estimated by counting the adults directly on the apical leaves of 25 plants per plot. The growth of the plant was evaluated by measuring the diameter and the height of the main stem of 25 plants per plot.

The percentages of contamination and of whitefly populations were analysed by comparing the cumulative numbers. We also compared the cumulative numbers of whitefly per plant and the cumulative percentages of contaminated plants per plot, in order to obtain the apparent transmission power (ATP) of the whiteflies, as a function of time and region.

#### COMPARISON OF FOREST AND SAVANNA REGIONS

Regardless of year and clone, the contamination was always higher in the forest than in the savanna. The overall percentage of contamination for all clones and years ranged from 10 to 88% in the forest, whereas in the savanna it ranged from 1 to 20% (Table 1).



Table 1. Percentage of contamination of clones in forest and savanna regions.

Clone		BR	H57	CB	TA49	H58	BB
Forest	1982	32	45	82	--	88	81
Forest	1983	10	25	74	67	84	69
Forest	1984	--	--	49	--	--	
Savanna	1982	3	3	1	--	5	20
Savanna	1983	1	2	3	1	2	7
Savanna	1984	--	--	4	--	--	

In the same way, the cassava plots planted at various dates, in the same year, were more contaminated in the forest than in the savanna: from 42 to 91% in the forest and from 4 to 43% in the savanna for the same period, March - July 1984 (Table 2).

Table 2. Percentage of contamination of cassava plots in forest and savanna regions.

Date of Planting	March	April	May	June	July
Forest 1984	91	58	49	42	50
Savanna 1984	4	43	11	4	12

#### COMPARISON OF TWO SAVANNA SITES

We compared the levels of contamination of two different clones, H58 and BR, in the savanna region (Table 3). In one case the fields were so situated that there were no diseased cassava plants upwind, and in the second case the fields were in the middle of a large plantation of diseased cassavas. In the second case, the contamination was 25 times as great as in the first case for clone BR, and 40 times as great for clone H58. The number of whiteflies was always higher in the most contaminated site, but the difference was not proportional to the level of contamination.

Table 3. Levels of contamination of two clones at different sites.

Clone	Number of whiteflies per plant		
	Savanna 1	Savanna 2	Forest
Clone BR	2.4	9.5	3.0
Clone H58	3.7	9.2	4.3

## COMPARISON OF FOREST SITES

Five different 0.06-ha fields were planted with clone CB, in the forest region, along a North-South axis, starting at the seashore (field 1) and ending 10 km inland (field 5). Each site differed both in the environment provided by nearby cassavas and in the area of diseased cassava which had already been crossed by the prevailing southwest wind before it reached the particular experimental field.

A sixth field planted at the ORSTOM research station was used as a reference (field 6). The highest contamination was recorded in fields 2 and 5, and the lowest in field 1. Field 1 harboured the largest whitefly population, and fields 3 and 4 harboured the smallest. The ATP was similar in all the fields, including the reference field, except field 1, where it was approximately a tenth as great. The differences in plant growth in the fields did not account for these differences in contamination.

## DISCUSSION

Differences between the dynamics of the contamination of cassava fields were very variable within a single region and between the different regions. Neither the climatic conditions nor the plant growth could account for the level of contamination. In some sites, there was a good correlation between the number of whiteflies and the level of contamination (Leuschner, 1977; Fargette *et al.*, same publication). However, these were not linked from site to site or from region to region. In comparing the ATPs we found two radically opposite situations:

- 1) field 1 in the forest with an ATP of 300 and field 1 in the savanna with an ATP of 1000,
- 2) in all the other situations the ATPs ranged between 40 and 80.

The fields with high ATPs had no diseased cassava upwind, whereas those with low ATPs were surrounded by fields of virus-infected cassava. These results support the hypothesis that cassava is a reservoir both of ACMV and of its vector *B. tabaci*. As the wind passed over fields of virus-infected cassava, it picked up the viruleferous whiteflies, which then landed in the healthy experimental plots. These whiteflies were more numerous and more viruleferous when they came from virus-infected cassavas upwind of the healthy plots.

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**SESSION D**

**SANITATION**

Chairman  
Dr. DIOMANDE  
Rapporteurs  
Dr. ATCHAM  
Dr. SAUTI

## TISSUE CULTURE AND CASSAVA SANITATION

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### INTRODUCTION

Cassava is affected by a wide range of pathogens including fungi, bacteria, viruses and mycoplasma. Many insect, mite and nematode species are cassava pests and parasites. These organisms can cause considerable losses and among them African Cassava Mosaic is one of the most important. A majority of these organisms is confined to specific geographical zones, i.e. continents or ecological regions within continents (Lozano, 1977).

The transfer of plant materials on a global scale, either for further improvement or for conservation of genetic resources, involves possible risk of widespread distribution of plant pathogens and plant parasites.

International exchange of germplasm of cassava has, until recently, officially been limited to true seed for fear of introducing vegetatively transmitted pathogens, particularly viruses. Although much progress has been made in genetic improvement in some countries based upon seed, the inability to move clones across national boundaries does limit further progress and precludes comparison among clones developed in various national and international programs. International exchange of germplasm is essential for further improvement in root and tuber crops. Exchange of materials has to be done without endangering the agriculture in the countries concerned.

### PRODUCTION OF VIRUS FREE MATERIAL

Advances in tissue culture have produced techniques for eliminating viruses from vegetatively propagated crops. A combination of thermotherapy and meristem tip culture has proven to be successful in eliminating viruses from many different crops (Quak, 1972).

Distribution of virus-free material in tissue culture form has the advantage, if sufficient care is taken, of assuring that the material is also free from most other pathogens and pests and is therefore the ideal way for exchanging germplasm of vegetatively propagated crops.

The methodology for production of virus-free material of cassava through thermotherapy and meristem tip culture has been available for several years (Kantha and Gamborg, 1975; Kaiser and Teemba, 1979; Frison, 1981).

This methodology was proposed to and accepted by the Inter African Phytosanitary Council for international movement of cassava clonal material in Africa. Improved varieties from the International Institute of Tropical Agriculture have been produced virus free, and distributed to many countries in Africa (IITA, 1982; Frison, 1981).

Recent advances in virology have produced techniques which make the indexing of cassava even more reliable (Mohanraj and Narayanasamy, 1984; Robinson et al., 1984)

#### GERMPLASM PRESERVATION

Germplasm collections of vegetatively propagated crops, which are important for breeding programmes, are traditionally maintained as living collections and have to be cultivated in the field during the growing season and losses during the storage between two growing seasons.

Maintenance costs of vegetatively propagated crops are high due to large inputs of labor and land. The maintenance of a collection in tissue culture reduces the risks of losing material and prevents recontamination of virus-free clones (Frison, 1981).

#### RAPID MULTIPLICATION

Another asset of the tissue culture technique is that the rate of multiplication of cassava can be increased considerably by using in vitro rapid multiplication. At IITA, the in vitro single node cutting technique has given satisfactory results (Frison, 1981).

#### CONCLUSION

Disease-free cassava material can now be produced through tissue culture techniques, safely maintained in collection, rapidly multiplied upon request and easily transported as tissue cultures.

In order to avoid widespread distribution of diseases and pests, international exchange of cassava clone material should only be done in the form of tissue culture material.

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VIRAL SANITATION OF POTATOES IN FRANCE:  
TECHNIQUES, ORGANIZATION AND REGULATION  
OF PLANT PRODUCTION

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It has long been known that one of the factors which most strongly affect the vigor and yield of a potato crop is the quality of the plants, and in particular their degree of contamination by various viruses. According to Reestman (1970), the serious symptoms induced by PVY and PLRV produce an average reduction of yield of the order of 50% or even more in susceptible varieties.

There are no conveniently applicable preventative or curative methods for controlling these viruses directly. The only way to control and limit their spread, aside from obtaining resistant varieties, is to produce healthy material and to multiply it up free from contamination, in order to obtain plants for use in sowing the potato crops.

In France, such sanitary breeding was begun in 1921 and has been officially regulated since 1934. Since then, techniques of production have been much improved thanks to the experience acquired over several decades and a better knowledge of the biology of the vector aphids and of how the virus is transmitted. These improvements have led to the establishment of the present scheme and organization of production in France, which is described here.

#### THE BASIC PRINCIPLES OF SANITARY SELECTION

First, like Madec et al. (1974), we summarize some characteristics of the viral diseases of the potato and their incidence in this vegetative multiplied plant, to provide a better understanding of the methodology used in the production of plants.

The diseases considered are very contagious, and are transmitted easily from plant to plant, either by direct plant to plant contact or indirectly by man and his agricultural implements, or else by the intermediary of living vectors such as aphids and in



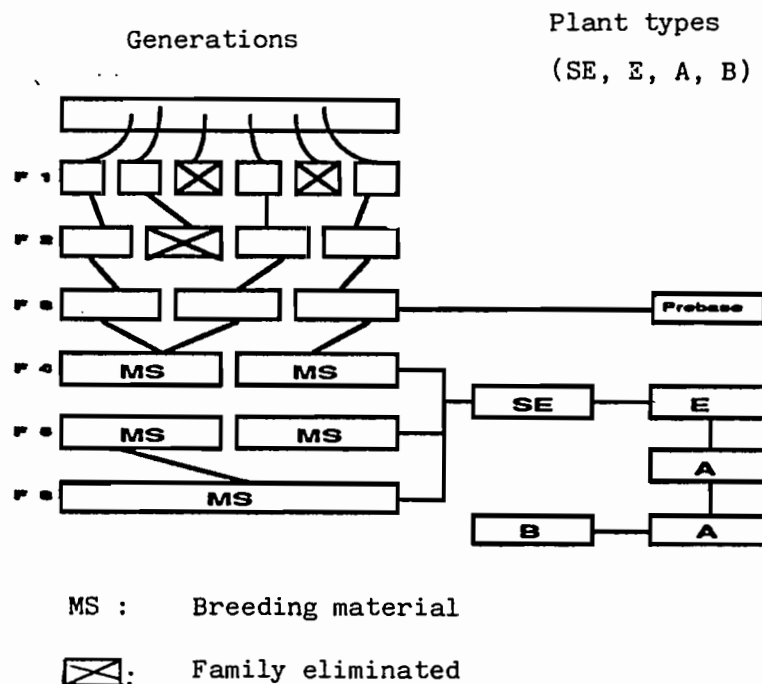
certain cases nematodes. They display a wide variability in expression of the symptoms, depending on the virus present, the variety and stage of the plant, and the climatic conditions. As visual inspection is often insufficient to detect infection, laboratory techniques of various degrees of sophistication must be used. In addition, in the large majority of cases, diseased tubers do not show any symptoms distinguishing them from healthy tubers.

Viral diseases are generalized diseases which affect all the organs and tissues. The embryonic tissues in seeds and the apical meristems, however, fairly often escape infection (Morel and Martin, 1955). The viral diseases are passed on vegetatively therefore from generation to generation and may be considered virtually hereditary. The result is that all batches of potatoes multiplied vegetatively without special precautions tend to gradually decline in health during successive generations of multiplication.

Aside from laboratory techniques, such as regeneration from meristem cultures or heat-treatment, no direct curative method is known that is practical for everyday use. The only means of controlling these diseases therefore remains the production of healthy plants. Such a programme tends necessarily to comprise two stages:

1. Obtaining and maintaining very healthy basal or selection stocks that are practically free from virus diseases and from diseases transmissible by the tuber. These stocks may take the form of plants, tubers, or cuttings.
2. The multiplication of this material on a large scale, in order to produce large quantities of plants for sowing potato crops.

Fig. 1: Clonal selection



Obtaining and maintaining healthy original stocks necessitates the establishment of reliable diagnostic methods that are sensitive and specific for the virus diseases: serological tests, passage on differential hosts, electron-microscope observations etc., with repetition of tests from time to time. Heat-treatment or meristem cultures may also be used, to cure varieties that are chronic carriers of certain viruses. Thus, the method envisaged by Morel and Martin (1955) has been used in France to cure several varieties, such as Belle de Fontenay, Saucisse, and Eersteling.

This material known to be healthy is then multiplied in open field and is subjected to clonal selection. This method, illustrated in Figure 1, consists of isolating the descendants of each tuber, cutting, or original plant. The individual descendants thus obtained constitute families. The original tuber and the clump of potatoes it produces constitute the FO family. The descendants of each FO constitutes the F1 and so on. If a single diseased plant is present in the F1 or the F2 family, that family is eliminated. In subsequent generations, a very low percentage of sick plants may be permitted.

This method has proved to be particularly effective for:

- the elimination of contact-transmissible viruses such as PVX, or viruses that produce only slight symptoms, such as PVX or PVM, and the tuber-transmissible bacterial diseases,
- The maintenance of varietal conformity, as aberrant types are easily eliminated as soon as they appear.

As successive multiplication of the healthy material take place in open field, new viral contaminations must obviously be avoided for as long as possible. To do this, it is possible to act on the two most important factors of contamination, the vectors and the sources of inoculum and to limit the number of generations of multiplication.

### Vectors

A very important element of success is to choose a region unfavourable to aphids in which to do the growing. In France, the Atlantic maritime or nordic regions (Brittany and the north of France) and the regions of high altitude are the most favourable.

Early destruction of the haulms protects the crops from the strong summer proliferation of aphids and thus limits the level of contamination.

The control of the insect vectors must be envisaged. Treatment with aphicides is particularly effective against the dissemination of PLRV, and mineral oils against that of PVY.

### Sources of inoculum

Here the aim is to decrease the amount of diseased plants as sources of contamination within or near crops intended for the production of plants. This may be done by:

- early, continuous elimination of virus-infected plants in the growing crops. The method is much more effective if this is carried out early, before the aphids can fly.
- the creation of protected zones where only crops intended for the production of plants may be grown is a very good solution. But for a great many reasons, such zones are often very difficult to create.

As regards how long multiplication is continued, every generation adds some risk of contamination. Limiting the number of generations and going back to the sources regularly gives the best guarantee of good results of the sanitary measures.

#### THE SYSTEM AND ORGANIZATION OF PLANT PRODUCTION IN FRANCE

The production of potato plants in France has been a highly organized profession from the beginning. A producers' organization includes all the producers in a given geographic area. The Fédération Nationale des Producteurs de Plants de Pommes de terre (National Federation of Potato Plant Producers) includes all the producers' organizations. Only the producers' organizations in the most favourable settings, in Brittany to be precise, carry out the clonal selection. The others limit themselves to multiplying the resulting high-quality plants for one or two generations.

The production and maintenance of original stocks, and the first and sometimes the second multiplication are performed in two specialized stations belonging to the producers and located in Brittany. Several years ago, the original stocks consisted of tubers grown in isolation frames after having been indexed and tested for the absence of viral diseases. At present, particular use is being made of cuttings grown in vitro on an agar-containing nutrient medium and stored at low temperatures (5 to 6°C) in a 16-hour light period.

These cuttings are multiplied to produce FO plants according to the method proposed by Nozeran et al. (1977) as modified by Madec et al. (1979). The original cutting is divided into fragments comprising a node and a leaf, which are transferred onto fresh medium. The plants derived from these are in turn divided after 5 to 6 weeks of growth and so on until the required number of plants is obtained. The microplants are then taken out of the tubes, grown in a greenhouse, and then grown in insect-proof shelters. This micropropagation is particularly useful in that it makes it possible to obtain a given quantity of plants in fewer generations, and hence with less risk of contamination.

The F2 and F3 generations are produced by specialist producers, under the supervision of the producers' organizations. These generations are closely monitored by specialist technicians and are subject to serological and biological laboratory tests during the growth and after the harvest.

Each family may remain distinct for up to 6 years, but usually, for practical reasons, the F3 harvests from a given variety and a given place of production are mixed and distributed to the producers belonging to that particular producers' organization. These establishments may then reproduce material up to the 6th year to obtain plants of "SE" quality. After that, they must replace it with younger stocks or be content to multiply SE plants to obtain class-E plants in the first year and class-A plants in the next two years.

#### CONTROL AND CERTIFICATION

Current legislation in France prohibits any sale of plants which have not been officially certified. Certification is performed by the Service Officiel de Contrôle et de Certification des semences et plants (SOC). Production, testing, and certification procedures for the plants are defined in a technical regulation (Règlement Technique, Anon., 1984) issued by the Secretary of Agriculture.

The SOC exercises its powers either directly, or by checking that producers and technicians at all the stages of production, of treatment after the harvest, and of commercial distribution, have followed the rules. Certification is made after these checks. The plants are then classified into four categories: SE and E basal plants, and certified A and B plants. Only clonally selected material can be classed as SE. A class-E rating may then be given to the first reproduction of SE plants. Certified A and B plants are those obtained after not more than two multiplications from class-E plants.

To classify the plants, account is also taken of their origin, of their varietal purity, of their degree of contamination by virus, of their degree of infestation by bacterial and cryptogamic diseases, and of their physiological condition. This last characteristic influences their germinative power and is greatly affected by conditions of production and storage (Perennec and Madec, 1980).

Tests of these various characteristics are made at several levels before planting, during growth, after the destruction of the haulms, during storage, and before sale. The growing crops are subject to two or three control visits by specialist technicians, during which the state of health and the isolation are confirmed. After the destruction of the foliage, which must be carried out on dates fixed annually by the SOC, a sample of tubers is collected from the cultures and treated with Rindite, a germination activator, so they can be planted immediately and observed before the autumn. This is the preculture test, performed in a greenhouse or in an open field. The maximum percentages of viroses allowed for the various categories of plants are shown in Table 1.

After the harvest, the tubers intended for use as plants must be free of certain diseases (verrucose gall, bacterial wilt, vascular bacteriosis, nematode cysts), to satisfy the tolerance

norms fixed by certain diseases attached to the tubers and calibrated according to the current regulations. Only after having satisfied all these criteria can they be certified by the issuance of an inspection tag. This must accompany every sack which, in addition, has to be hermetically sealed.

Table 1. Norms for classification by plant viruses

	Categories of plants (1)	SE	E	A	B
1. In culture	% virosis	0.25	0.33	1	3
2. In preculture	% virosis	1	2	4	10

(1) maximum % of the total of the recorded marks

#### RESULTS : THE IMPORTANCE OF FRENCH PLANT PRODUCTION

The area of land devoted to the production of potatoes in France has remained unchanged in recent years. In 1985, it was 15,200 ha, 8300 in Brittany, 6100 in the North, and 800 in the central region. 3463 farmers were involved in this production, and the monitoring and certification were provided by 80 technicians.

2.8% of the material grown was rejected. The remainder was distributed as follows: 3% was kept for the production of prebasal plants, 6.4% as SE plants, 10% as class E, 79.6% as class A and 1% as class B. The total harvested tonnage was 363,485 tonnes, of which 220,000 were certified and sold, mostly on the national market. Approximately 50,000 tonnes were exported. Today, this production is easily enough to cover France's annual requirements for potato plants of very good quality.

#### CONCLUSION

The organized and regulated production of potato plants in France has made it possible to control the viruses of this plant effectively. But this is an expensive method, since it uses very elaborate laboratory techniques for the detection of the viruses and is very demanding of labour and surveillance personnel. It is also sometimes subject to uncertainties, since sanitary accidents are always a danger, particularly following unusual climatic conditions. The more continental the location of the growing region, the more frequent these accidents are, but they can also happen in the maritime zones. Thus in France, in 1970 and in 1976, the spread of viruses was enough to compromise the production of quality plants of certain of our better varieties for several years (Perennec, 1982).

To decrease these drawbacks and risks, the resistance of our varieties to the principal viruses must continue to be improved.

This improvement should lead to a reduction in the strong limitations and the cost of the production of plants, by lightening the burden of purification and inspection in the plant cultures, by limiting the risks of sanitary accidents, and by lessening the annual demand for plants of less frequent renewal of those in culture.

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SELECTION OF HEALTHY CASSAVA PLANTS  
OBTAINED BY REVERSION IN CASSAVA FIELDS

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We call reversion the biological phenomenon that allows a virus-infected plant to produce a symptom-free plant in the next generation. As this phenomenon is actually found in cassava, we were able to select, like Bock (1983) in Kenya, healthy plants right in virus-infected cassava fields. In addition, although this phenomenon is by nature very unstable, the limited results obtained suggest that it may be genetic, and therefore could be considered a component of the resistance of cassava.

These are still very speculative results, but they are useful enough and unusual enough to merit further attention. If our hypotheses are confirmed, this could offer a new route for the selection of plant material and a new method for controlling African Cassava Mosaic.

OBTAINING HEALTHY MATERIAL BY REVERSION

The percentage of symptom-free plants in a field of virus-infected cassava was often extremely low, of the order of 0.1 to 0.01. For some varieties of cassava, we never found a single plant without symptoms. However, despite this low percentage, in 1979 we began selecting healthy plants from six different clones: CB, Ta 49, H57, H58, BR, and BB.

This selection was based solely on visible symptoms, and was done by systematically roguing all plants presenting symptoms, however slight.

Since the goal was to multiply healthy material, the plants selected were planted in a region of the Ivory Coast with a low inoculum pressure, taking care not to put the new plants downwind of virus-infected cassava. In these conditions, the percentage of plants rogued for clone CB, which is moderately susceptible, fell progressively over 3 years, from 50% to 20% and then 5%, and then stabilized at this value.

The same thing happened with the six other clones selected, though with values that varied depending on the clones' natural susceptibility to the disease. For the clone Bonoua Blanc, for example, we observed levels of contamination ranging from 100% to 60% , and for clone 86, from 98% to 95%! Obviously the plants that were not displaying symptoms at the time of selection were nevertheless contaminated, and they therefore produced the virus-infected plants.

The fact of having symptom-free plants is obviously no guarantee of obtaining a virus-free plant; however, we can report that our entire epidemiological programme was based on this principle, and throughout our experiments we have not met with any problems that could be attributed to this cause. Every month for 6 years we have planted approximately 1000 cuttings derived from symptom-free plants, and we have not recorded any level of contamination that could not be explained in other ways.

#### GENETIC ORIGIN OF REVERSION

Reversion is a biological phenomenon which, though unstable, is probably genetic. The same virus-infected clones observed during several successive years showed highly variable percentages of reversion, ranging from 0% to 100%. On the other hand, when reversion did occur, the phenomenon was general for all the clones, with variable percentages specific for particular clones.

In a collection of 10 clones resistant to African Cassava Mosaic, the percentage of reversion in 1986 ranged from 0% to 10%. In the same year, on a single site, we found for 10 different clones a highly significant correlation of 0.79 between the two experiments (Table I).

Table I. Percentage of "reversion" (plants showing no symptoms after plantation) for 10 cassava clones, coming from East Africa, in two different trials.

CLONES	EXPERIMENT I	EXPERIMENT II
7	77	32
13	97	88
14	76	68
17	58	36
18	81	39
19	31	0
20	65	72
21	84	50
22	51	3
23	53	5



The climatic and/or biological conditions which give rise to this phenomenon are not yet known. It may depend on the "history" of the cutting during the preceding cycle, or, in contrast, on the conditions in which the cuttings were stored, or, finally, on the growing conditions in the plantation in which the phenomenon was observed. It is noteworthy that reversion phenomena have been observed following cultural accidents, such as a very severe attack of mites which practically destroyed the apical meristems; in this case, the secondary meristems started without symptoms.

#### REVERSION IS THE RESULT OF A COMPONENT OF RESISTANCE

The phenomenon is probably related to the diffusion of virus in the plant and, consequently, to the component of resistance previously called R5. When the symptoms of mosaic disease on cassava are recorded over time, for certain clones the intensity of symptoms decreases considerably and even, in some cases, falls to zero: this is reversion of the symptoms.

We have characterized this decrease in symptoms by the slope of the curves considered; this estimation is independent of the quantification of symptoms which is performed to characterize the resistance to their expression. It must be noted also that we are not talking about a general phenomenon: some varieties do not show it at all, and always produce strong symptoms. On the other hand, in the six varieties tested for their reversion ability, there may be a relation with the decrease of symptoms.

This preliminary result obviously needs to be confirmed, but it is sufficiently interesting to merit further attention.

It has been shown, furthermore, that in the case of cassava the correlation between symptoms and virus concentration is very strong, so it is possible that this decrease in the intensity of symptoms is correlated with a decrease in the concentration of viral particles in the leaves, at least for the clones considered.

It seems that there may be, in each cell of each cassava leaf, a kinetic of synthesis of viral particles that is dependent on the cell's resistance mechanisms. This equilibrium will depend, of course, on the genome of the plant, but it will also depend on the conditions of the cell: age of the leaf, particular physiological conditions, climatic conditions, etc.

In certain conditions, which remain to be defined, the concentration of viral particles reaches a critical threshold whereby the plant of the next generation derived from this cutting will remain free of virus.

If this biological phenomenon is confirmed to be truly dependent on plant genes and if we succeed in finding out in which conditions they are expressed, a new route could open up for selecting clones with a high percentage of reversion in natural conditions. This would be a new method, self-regulated and independent of any technology, for controlling African Cassava Mosaic.

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AFRICAN CASSAVA MOSAIC  
AMONG FARMERS OF THE LOWER IVORY COAST

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Along with yams, cassava is the principal food crop in the lower Ivory Coast. In addition, it provides a useful income to growers living within 100 km from Abidjan. Between 1984 and 1986, an agricultural and economic investigation into the farming practices was conducted relating to food crop, to find out what the technical and economic constraints are and to account for the low cassava yields (10 t/ha on average).

To compare the various growth situations, it was necessary to estimate the effect of African Cassava Mosaic (ACM), which affects all plantations of sweet cassava. Some bitter cassava strains, however, show far fewer symptoms of mosaic; these are grown by certain ethnic groups, for example, the Adioukrou near Dabou.

On the basis of monthly observations of farmers' plots and some experimental tests, three series of questions were initially asked about this endemic disease:

1. What causes the viral disease: The vectors involved, the role of farming techniques and/of the environment, and most important, the role of the peasant farmers themselves when they use any empirical means of control.
2. Its effects on plant growth. In a context of low yields, is mosaic a major constraint ?
3. A control programme: The need for it, how to set it up, and what associated measures may be needed. In the lower Ivory Coast, cassava has specific roles in varied systems of agricultural production.

AFRICAN MOSAIC IN THE LOWER IVORY COAST

The analysis is limited to the varietal group Bonoua Rouge and to three villages between 30 and 90 km from Abidjan, one to the west and the other two to the east, all situated on sandy soils in an equatorial climate with two rainy seasons. In this ecological

context, the agricultural situations are quite different, but the levels of mosaic are similar and the village sample can give a representative view of the region.

Table 1. Some systems of agricultural production in three villages of the lower Ivory Coast.

Village	Ethnic group	Typical structure	crops	Surface covered
Béniakré	Baoulé	10 ha reduced family (pioneer)	coffee, cocoa yam-cassava-fallow	7 ha 3 ha
Djimini-Koffikro				
- Land owners	Numerous	20 ha extended family	oil palm yam-cassava-fallow tenanted and other	5 ha 3 ha 11 ha
- Landless farmers	Dioula Burkinabé	3 ha reduced family	cassava-sweet potato or pineapple-cassava	3 ha
Songon-Agban-Attie				
	Attie	20 ha reduced family (pioneer)	coffee-cocoa cassava-fallow	10 ha 10 ha

Three types of observations on peasant plots were made: 1) observation of small patches (40 m<sup>2</sup>); 2) experimental testing of local cultivars with non-infested cuttings (provided by the laboratory of virology of ORSTOM at Adiopodoumé); 3) observation of pairs of small patches comparing a clean control with the peasant weeding practices. The cassava plants, in all cases, were recorded individually each month. The objective of the experimental test was partly methodological and the 6 blocks, in 3 villages, were measured just once. In order not to overlook possible effects and to prepare more sensitive experiments, we chose a risk threshold of 8%. It was not possible to check for any possible clonal difference between local cuttings and virus-free ones. Because of these restrictions, we consider the results of this test only as hypotheses.

#### The scale of symptoms

Because we do not have adequate information about the actual mechanism of ACM, and also because our objective was primarily agronomic, we worked on the leaf symptoms, which are liable to

produce losses of yield directly (other processes are probably also involved). In addition, it should be simple to record ACM in the peasant setting, and as the yield is obtained per plant (and not per stem), we opted for an overall marking scheme in which the percentage of nongreen surface of the leaves (for which photosynthesis was affected) was estimated. The scores ranged from 1 to 10; 100% represents level 3 on Cours's scale. The subjective character of this marking scheme was reduced, though not eliminated, by regular visits from a reference investigator, to check the work of the observers.

With large cassava plants, this overall marking scheme was no longer sensitive enough to record the effects of different growing conditions. We recorded in addition the last 10 leaves, after examining each stem. This criterion turned out to be pertinent either to the characterization of the overall evolution of the mosaic or to the detection of what determines it.

#### Pattern of development of the mosaic

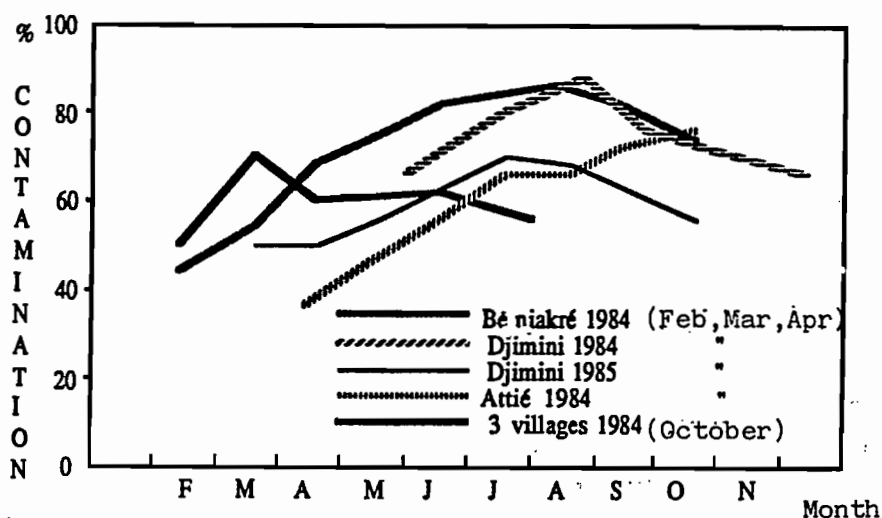
The contamination of the crop arises from cuttings. The farmers were not recorded as performing any empirical selection, except that they eliminated weak stems as they weeded. The need for many cuttings at planting time leads to indiscriminate use, to the point where during the cassava "boom" of 1984 several plots had to be replanted. From the first month, the cassava showed a mean level of 40% to 50%, whatever the date of planting. The variability between plants within a plot and between plots was large, whereas certain villages (Djimini for example) showed less initial variability of ACM.

Table 2. Mean coefficient of variation of ACMV in the first month (%).

<u>Between plants</u> <u>in a patch (40 m<sup>2</sup>)</u>		<u>Between groups of 4 plants</u> <u>within a plot</u>
Béniakré, - beginning of 1984	58	42
Attié - beginning of 1984	54	41
Djimini - beginning of 1984	33	33
Djimini - beginning of 1985	28	-

The patterns of development of the symptoms varied from one village to another, depending on the year and date of planting, showing that the expression of ACM was clearly related to the growing conditions (Fig.1). In two villages, ACM stabilized and even subsided slightly, from the 6th month. The mean maximum lay around 80% in the various situations (which does not signify an actual 80% loss of photosynthetic capacity, since our scale is above all comparative and relative).

Fig. 1: Evolution of the symptoms in the lower Ivory Coast



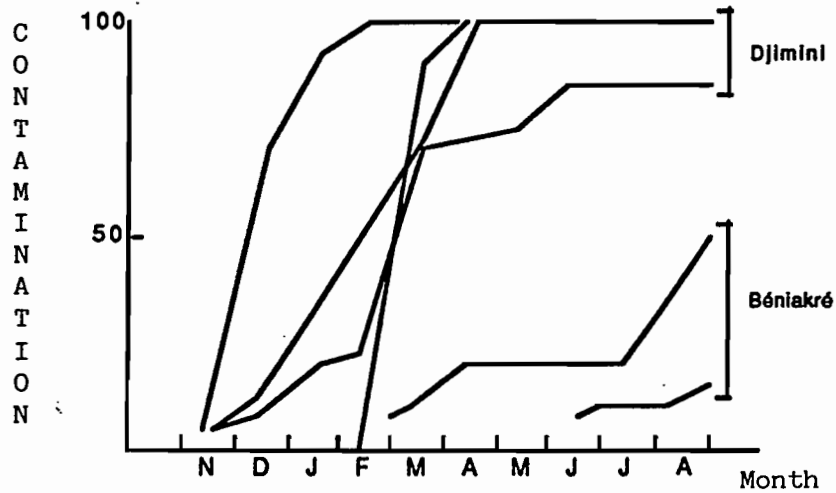
Some causes involved

Though contamination occurs mostly through cuttings, other vectors are likely to play a role. After analyzing the latter, we will discuss whether the expression of symptoms was related to certain growing conditions:

- Contamination of healthy cutting

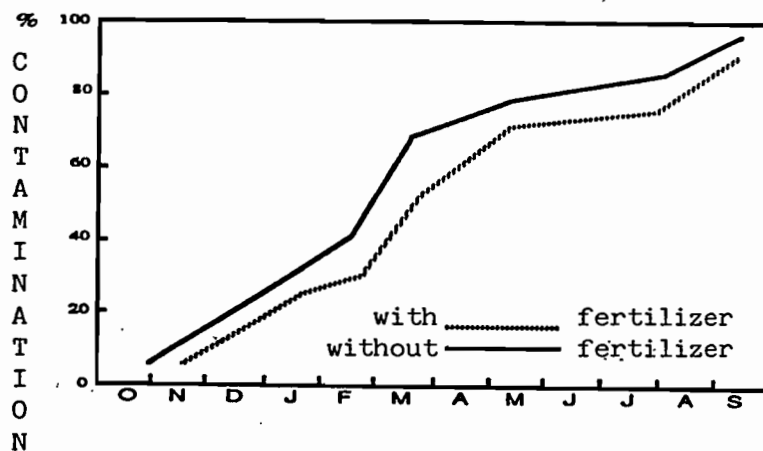
Although issued only from a single experimental test, the results are convincing: virus-free cuttings, planted in contaminated plots, showed dynamics of contamination that differed among villages (Fig. 2). It seems that the outcome may be related to the level of subdivision of the land and to the relative amount of cassava in the village: Béniakré, where contamination was slow, has mainly coffee and cocoa fields, broken by a few clearings for food crops; Djimini is much more open, with wide diversification (trees, food crops, and pineapples); Attié has the largest amount of cassava (although one case is hardly significant). There are several limitations applicable to these hypotheses: the contamination is measured by the appearance of symptoms, perhaps related to different growing conditions. The village effect is not independent of the climatic effect.

Fig. 2: Changes in the level of contaminated plants between blocks and villages.



For the plants whose virus disease is derived from secondary contamination, the level of symptoms is clearly lower: 30% versus 60% for the 6th and 7th month. It seems that the NK fertilizer had no effect on the expression of the symptoms, because the shift on the figure appeared before the application.

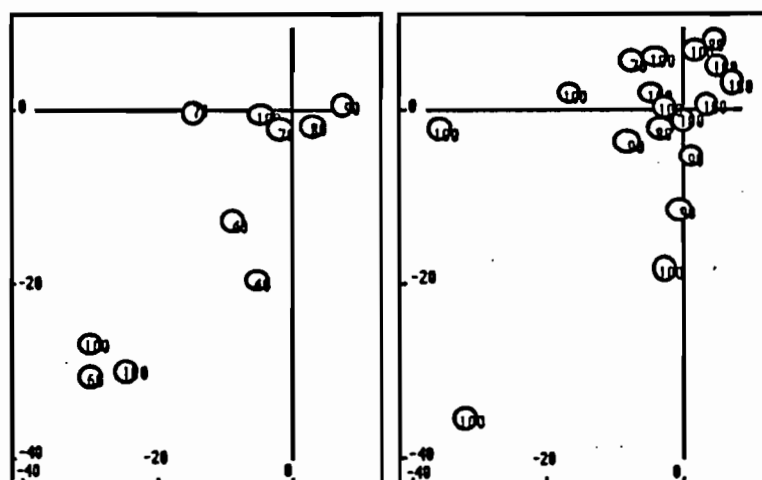
Fig. 3: Changes in the level of contaminated stalks (mean of the 3 villages).



- Weeds and ACM

The expression of ACM might depend on the conditions in which the plant population is growing. Two examples are analysed from the tests in the peasant environment: the effect of the NK fertilizer (in the following section) and the effect of a lot of weeds on the expression of the mosaic. From pairs of cassava patches, we chose the most unfavourable situations (infestations 80% and height of the weeds > 30cm). To avoid the poor sensitivity of the criterion "overall mosaic of the plant", we distinguished plantings less than 3 months old from those more than 3 months old (Fig. 4). However, in neither case did the pattern of the changes seem related to the unfavourable weed conditions (the different situations did not diverge significantly from the bisector).

Fig. 4: Changes in the symptoms of ACM with heavy weed infestations. a. First 3 months. b. Following months.



Each number represents the level of infestation of a weed-infected patch compared with a clean control. The displacement between mosaic in the control and that in the patch is given by the X-axis position for the month M, versus the month M + 1 on the Y-axis.

## THE CONSEQUENCES OF AFRICAN CASSAVA MOSAIC

### Effects of ACM on the harvest

In the experimental test and for the treatments without fertilizer, the non-infected cuttings doubles the yield on fresh roots (14 to 27 t/ha). With fertilizer, the gain was not more than 74%. In contrast with the NK fertilizer, which acts preferentially on the above-ground parts, the mosaic acted preferentially on the weight of the roots (Table 3).



Table 3. Analysis of variance of the experimental test.

Treatment	Level of significance (risks)			
	<0.001	<0.01	<0.05	<0.08
Mosaic	Number of tubers (-)	% Abnormal plants (+)	Dry matter, roots (-)	
NK fertilizer			Dry weight, above ground (+)	Total dry weight (+) Number of tubers (+)
Interaction	-	-	-	-

The mean weight of a tuber did not differ detectably with the different treatments (approximately 0.16 kg dry weight), so yield was strongly correlated with the number of tubers. The level of plant destruction also did not differ, but there was a higher percentage of atypical plants in the case of virus-infected cuttings: these were vigorous plants (above-ground part > 1000 g wet weight), but without any tubers, detracting from the linear regression between weight of roots and weight of above-ground parts. This correlation would be even more marked if it had integrated the plants with many roots and reduced tuber formation. Some cases of root rot were observed, but they were not attributable to a particular treatment. In this test, the fertilizer did not significantly affect the weight of the roots. Nevertheless, it produced a non-negligible mean gain, of 53 and 32% respectively, depending on whether the cuttings used were the local or the virus-free ones.

Table 4. Action of fertilizer on the cuttings.

Fertilizer	Local cuttings		Virus-free cuttings	
	without fertilizer	With NK fertilizer	Without fertilizer	With NK fertilizer
% Atypical plants	7	16	0	0
Root dry wt/plant (kg)*	0.53	0.81	1.07	1.41
Above-ground dry wt/plant (kg)*	0.58	0.91	0.86	1.39
Number of tubers**	2.8	4.3	6.8	8.2

\* all the plants present at the harvest

\*\* on 5 blocks

The use of diverse growing conditions made it possible to contrast the common functioning better, in particular to weight the effects of the treatments. Two blocks showed a clear effect of the mosaic on the weight of the above-ground parts, or, rather in one village there were marked interactions between fertilizer and mosaic.

ACMV: Planting and harvest

As the cassava grows continuously, and its phases of development induce hardly any break in growth, it is legitimate both to monitor the effect of ACM from the first month of planting and to analyse how this period explains certain components of the yield. As the cycle lasts 12 months, the low correlations (>0.4) will be preserved initially.

The farmers' virus-infected cuttings produced taller plants in the first month, a phenomenon which fell off thereafter (Table 5). These plants branched sooner. From the second month, the mosaic decreased the leaf cover on the ground.

Table 5. Analysis of variance (2 fixed, crossed factors).

(Risk)	Mosaic	Fertilizer	Interaction
1st month: height	0.006 (+)	-	-
2nd month: area	0.02 (-)	-	-
3rd month: height	-	0.04 (+)	-
3rd month: competition	0.03 (-)	0.015 (+)	-
3rd month: % branched plants	0.04 (+)	0.003 (+)	-

In addition, the components of the yield were correlated with the state of growth at 3 months. Thus, the dry weight of tubers at 12 months was positively correlated both with an estimator of the leaf cover on the ground ( $r = 0.65$ ) and with the depth of fallen leaves ( $r = 0.70$ ). The following figure summarizes some elements that may determine the plant's growth, which are then specified by the multiple regressions.

Fig. 5: Factors affecting plant growth, specified by multiple regressions.

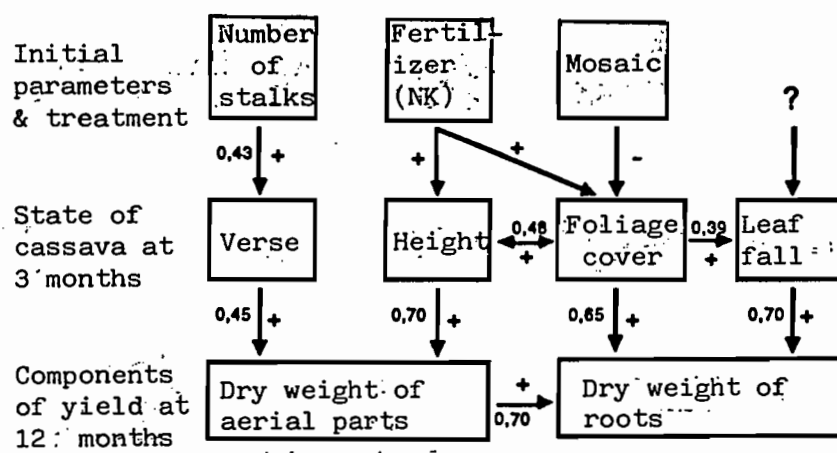


Table 6. Approach of correlations between treatments, plantings, and the components of yield, on the basis of experimental tests.

Equations	Correlation		Risks	
	r	Total	Partial	Partial
Dry wt. roots (kg) = 0.014 Comp3 (%) + 0.019 LF 3 (cm) + 0.29	0.81	0.048	0.004	0.001
Dry wt. above ground (kg) = 0.011 Height3 (cm) + 0.445 T 3 - 0.76	0.78	0.059	0.0001	0.006
Leaf fall (cm) = 0.21 Area3 (dm2) + 14.9 T 3 + 0.21	0.53	0.22	0.041	0.027
B1-3 = 0.45 Height3 + 0.52 M3 (%) - 55.9	0.61	-	0.013	0.007

Note: Dry wt. of roots and above-ground parts at 12 months; and Comp, competition (leaf-cover index), leaf fall (LF), height, toppling (T) (scored from 0 to 0.5)% of branched plants (B), area and mosaic (M) at 3 months.

At 3 months, the percentage of branched plants was not much correlated with the height nor with the mosaic ( $r = 0.33$  and  $0.59$ ). It was, however, clearly related to the two of them at a given height, the effect of the mosaic interacted with branching (there might be a clonal difference, but none of the treatments showed a simple regression of height versus percentage of branched plants). The weight of roots at 12 months was clearly correlated with the leaf cover on the ground (expressed as a criterion of competition) and, more surprisingly, with the amount of fallen leaves. In summary, the absence of mosaic in a cutting is translated mainly into a greater ground area from the first month. With no change in the weight of the parts, this greater effectiveness of light capture shows up in the root yield.

As for the components of yield, we did not see any interaction between mosaic and fertilizer on the criteria of cassava planting. Nevertheless, to eliminate partially a strong initial variability between plants, we calculated the difference between the mosaic at the 3rd and the 1st months. It turned out that fertilizer had a significant effect (risk = 4%), which could signify that the mosaic is expressed more in the presence of fertilizer.

#### AFRICAN CASSAVA MOSAIC AND FARMERS

Although the experiment lasted only one year and covered only one particular period, the gain of nearly 100% provided by the virus-free cuttings gives an idea of the impact of this disease. It would be necessary to clarify this relation further, first to specify the actual gain, according to years and dates of planting, provided by the non-infected cuttings. In this respect, it is also necessary to evaluate the real effect of ACM for the sake of knowledge (and therefore of improvement). In fact, there is a possibility that there is some varietal difference within the Bonoua Rouge group, and it would be desirable to be able to work in a non-experimental setting with a genetically characterized and virus-free cassavas. Finally, it is not enough to have shown a gain for a given year if the increase will not persist without a longer fallow period. Therefore, it is advisable to determine it in terms of the cropping system, and particularly when cassava follows a fallow period. If the gain were to be confirmed experimentally, it would then be possible to go on to analyse the conditions for setting up a possible programme of combative action.

At present, no structural shortage of cassava is recorded in Abidjan, though there are substantial fluctuations in supply and price. Controlling ACM would serve mainly to free farmland and to lower the price to the consumer. To achieve these objectives, it will be necessary to ensure a clear gain to the producer, despite foreseeable reduction in the price. In addition, the release of land will be an opportunity that is put to use differently by the various systems of production in the lower Ivory Coast. One must not forget that two nearby villages, even two contiguous stretches

of farmland for some villages (Djimini for example) operate differently, under different constraints. Cassava plays different roles there, depending on whether it is all consumed in the village, or is sold, either the total production or just the surplus.

This release of land will be felt in different ways, depending on the pressure to diversify, which combines two opposing factors:

i) the systems of production in which the food crop is imposed;  
ii) those for which cassava is a sideline. In the first case, we have the indigenous Adioukrou, whose social system is largely based on cassava monoculture. There are also landless farmers who have only temporary access to a plot, often planted with cassava, which will be sold. In the second case, the small and medium planters of Béniakré may respond circumstantially to a rise in price, but to the detriment of their small reserve of fallow land.

In the Congo, the virus may even represent an advantage, since the consumption of the young cassava leaves shows that there is a preference for virus-infected leaves. Whatever programme of combative action is envisaged, it is necessary to avoid marginalizing the most specialized and most fragile systems of production; a prudent, gradual action is necessary to allow each of them to adapt themselves. In addition, the provision of healthy cuttings may lend itself to the organization of profitable cassava production, for which a knowledge of differentiated and original systems of agriculture is essential.

VASCULAR BACTERIOSIS OF CASSAVA:  
SUMMARY AND OUTLOOK

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BRAZZAVILLE, CONGO

INTRODUCTION

Cassava (Manihot esculenta Crantz) is an important source of calories for the 500 million inhabitants of developing countries. Its consumption is constantly increasing (by 2.5 to 3% per year), so that the population dependent on cassava for their food will have doubled by the end of this century (Cock, 1985). It is adaptable to cultivation with little investment and low technology, and in a wide diversity of soils and climatic conditions. At present, cassava is the most reliable and cheapest source of carbohydrate in tropical regions. However, many biological constraints combine to limit its production on the African continent, including vascular bacteriosis, African cassava mosaic, the mealy bug and, more recently, mites. Here we are concerned with vascular bacteriosis.

The causal agent of this disease is a bacterium, Xanthomonas campestris pathovar manihotis which is responsible for a serious loss of production in Africa, Latin America, and Asia. Of the various cassava diseases, bacteriosis may be the one that has caused the most severe damage during the last two decades. At present, it is known as one of the most important factors limiting production in all regions where cassava is grown (Lozano, 1986).

Africa, which produces 38% of the world's cassava, on 53% of the worldwide land area used for its cultivation, has suffered a 7% loss of production due in part to drought, but due especially to the appearance of new biological constraints, including bacteriosis. Thus in 1983 these constraints caused a 30% loss of yield in Mozambique, 25% in the Central African Republic, and 15% in the Congo. These losses of production have produced a major decrease in the amounts available for sale, and a strong rise in price.

This situation is particularly worrying in developing countries where governments are encouraging producers to re-establish local food-crop production in order to limit the importation of basic foodstuffs.

## GEOGRAPHIC DISTRIBUTION AND IMPORTANCE

Bacteriosis of cassava was first reported in Latin America. On the African continent, one report referred to a similar disease (a leaf attack), in Uganda in 1937 (Hansford, 1937) and then in Malawi in 1949, whose causal agent was identified as Xanthomonas cassava (Wiehe and Dowson, 1953). Likewise, in Madagascar and Java, Bourriquet (1946) identified a bacterium causing angular leaf spots and leaf blight (Bacterium robici Bourriquet). In none of these descriptions was there any reference to the vascular presence of the parasite, from which it is assumed that the present bacteriosis was probably introduced from Latin America into Africa and likewise into Asia.

The disease in its present form, that is, the vascular form, is endemic in regions where cassava is grown in many countries of Asia (China, Thailand, Taiwan, Indonesia), of Latin America (Colombia, Venezuela, Brazil) and of Africa (the Ivory Coast, Nigeria, Cameroon, the Congo, Zaire, Kenya, South Africa).

In the particular case of Africa, the major epidemic appeared in the 1970s (Nigeria, 1972; Cameroon, 1974; Zaire, 1975; the Congo, 1976; Uganda, 1976; Rwanda and the Central African Republic, 1977). Vascular bacteriosis is now endemic in all the regions of cassava cultivation in Africa.

## EFFECT

When the disease is in an epidemic phase and the environmental conditions are favourable, losses of tuber yield can range from 80 to 90% (for example in the Congo in 1976 and 1978, and in Zaire, 1973-1978). Generally, the losses of yield are lower (30%) in regions where the disease is endemic. However, the slight but progressive contamination of the plant material for re-planting induces major losses of yield after 3 or 4 growing cycles without any spectacular attacks ever taking place. Such losses have been observed in farmers' plantations and even more in industrial plantations where cassava is a monoculture or where only few cultivars are grown.

The damage to cassava is reflected not only in a loss of yield in tubers, but also in a lowered starch content, a destruction of the leaves (an important source of protein for the populations of Central Africa) and a contamination of the plant material intended for the production of cuttings. Since the 1980s, there have been no reports of a serious outbreak of bacteriosis in either Africa or South America.

## SYMPTOMS

Vascular bacteriosis of cassava appears in the rainy season and is characterized by a diversity of symptoms:

- angular leaf spots;

- leaf blight with toxin production;
- wilting of the leaves;
- lesions on stems, with production of exudate;
- defoliation of the branches;
- desiccation of the shoot apices.

This combination of symptoms is unique among the syndromes produced by phytopathogenic bacteria.

In the case of plants derived from contaminated cuttings, the disease is characterized by a rapid wilting of young shoots, by producing galls on the nonlignified stalks. The desiccation of the young shoots is rapid.

In initially healthy plants, the beginning of the disease is characterized by the appearance of angular, vein-delimited spots, sometimes combined with leaf blight. These foliar lesions multiply, then the leaves wither and drop off, so that the branch is defoliated. Simultaneously, on the young stalks, necroses appear with production of exudates, which then evolve into cankerous lesions. The underlying tissues, particularly the vascular tissues, are blackish-brown. This deterioration may extend to the pith. Histological sections show the presence of the parasite in the vessels.

In the last stage of the disease, the aerial part of the plant is destroyed. At the limits of the necrosis, the plants rapidly put out shoots, which then in turn display the symptoms of the disease. These young shoots are very susceptible to the disease during the rainy season and at the resumption of growth (at the end of the dry season). They maintain an important secondary inoculum in the plantations. It is important to note that the old stems, adapted to the production of cuttings, are often apparently healthy despite the vascular presence of the bacterium. One also observes lesions on fruit and the presence of the bacterium in the seeds, the flowers and the pollen grains (Daniel et al. 1985; Elango et al. 1981).

#### EPIDEMIOLOGY

The cycle of vascular bacteriosis is characterized by the alternation of two phases:

- A parasitic phase in the rainy season, with
  1. epiphytic multiplication of the bacterium on above-ground organs;
  2. penetration and multiplication of *X. c.pv. manihotis* in the tissues of the host;
  3. induction of symptoms and dissemination of the pathogens and contamination of new organs.



- A survival phase in the dry season, with:

1. absence of developing symptoms;
2. epiphytic survival of the bacterium on the canopy with low levels of populations;
3. survival in the vascular tissues of the stems and seeds;
4. storage in dry debris on the surface of the ground.

In the field, the alternation between these two phases which correspond to rainy and dry seasons enables us to account for the continuing presence of bacteria throughout the growth cycle.

The use of infected cuttings is generally the cause of the appearance of the disease in new plantings. Vascular bacteriosis of cassava was certainly introduced in this manner from Latin America into Africa and Asia (Lozano, 1986). The quantitative importance of the inoculum in the development of the disease is a function of the interaction of several environmental variables with the level of inoculation of the pathogen in the cuttings and the frequency of their contamination.

The pathogen agent has been detected in a number of insects, which may play a role in disseminating the disease over short distances. The actual numbers of the pathogen surviving epiphytically on adventitia (Elango, 1981) as a source of inoculum, has not been determined. The pathogen agent survives only poorly in the soil. However, the bacterium remains viable in the host tissues, in dry exudates, and on the soil surface in dry debris.

The traditional idea concerning bacterial diseases is that the lesions (leaf spots, necroses on stems) are a source of primary inoculum. Thus, epidemics appear when this inoculum is dispersed by rain drops and by the wind blowing from diseased leaves towards the healthy leaves of nearby plants—conditions which come together in the rainy season. These mechanisms ensure the redistribution of the pathogen within the canopy and its dissemination within the plantation. However, in dry sunny weather, epiphytic bacteria have been found in the troposphere surrounding the canopy of diseased plants. The dispersal of these populations and their deposition on the healthy organs could be quantitatively important in disseminating the bacteria in dry conditions (Hirano *et al.*, 1983).

Vascular bacteriosis is much more widespread in regions of savanna and/of savanna/forest transition than in forest regions. The lower incidence of the bacteriosis in forest regions may be attributable to the absence of dry and rainy seasons which favours the development of the epidemic (preservation of the inoculum in debris) and also by the small range of temperature variation (Lozano, 1986).

## THE PATHOGEN AGENT

The bacterium responsible for vascular bacteriosis of cassava is Xanthomonas campestris pathovar manihotis. It is classified as Xanthomonas, whose morphological and biochemical characteristics it possesses, but it is taxonomically distinct on the basis of its pathogenicity for Manihot species and its lack of pigmentation.

The bacterium penetrates the plant via the stomata and wounds the limb tissue. The pathogen invades and destroys the spongy mesophyll, then penetrates the vascular system, and in this way systematically colonizes the plant. The pathogen migrates in the plant via the vessels of the xylem, and sometimes the phloem. In old lignified stems the bacterium is preserved in the vascular tissues which often look undamaged. The vascular presence of the bacterium distinguished X. c.pv. manihotis from X.c.pv. cassavae.

At present, there is little information available on the variability of this bacterium. Many authors suggest the existence of a variability of strains of X. c.pv. manihot in vivo (Bradbury, 1975, 1977; Maraite et al. 1981; Alve and Takatsu, 1984), but this contested by others (Elango and Lozano, 1981). The results of investigations in vitro are also contradictory. Robbs et al. (1972), Elango et al. (1981) and Manico et al. (1984) did not observe any variability. Lozano et al. (1974), and Maraite et al. (1981) noted differences in the use of different sources of carbon and in the susceptibility to certain antibiotics. Thus Lozano (1974) has proposed a classification of the strains of X. c.pv. manihot into four biovars on the basis of the differential use of a range of carbon sources.

This variability has not always been related to any difference in pathogenicity of the various strains. Pathogenicity refers both to the ability of the agent to induce the disease (virulence) and to the intensity of the disease (aggressivity). Most authors observed a difference in the pathogenicities of the strains investigated (Bradbury, 1975, 1977; Maraite et al. 1981; Alve and Takatsu, 1984). Bradbury (1977) even reported some temporal variability in the pathogenicities of strains, since certain resistant cultivars became susceptible.

Grousson and Boher (1986) confirmed the existence of such a variability, and were able to classify the strains into four groups as a function of their aggressivity:

- Group 1: strains very aggressive on leaves to moderately aggressive on stems.
- Group 2: strains aggressive on leaves and on stems.
- Group 3: strains moderately to weakly aggressive on leaves and moderately to very aggressive on stems.
- Group 4: strains weakly aggressive on leaves and on stems.

These investigations reported a link between the aggressivity and the geographic origin of the strains, and an effect of the date of isolation. Thus it is possible to suppose that the strains have become more aggressive in the past ten years. This hypothesis, already proposed by Bradbury (1977) might explain the increased severity of the disease in Africa.

This variability in the strains has been confirmed by the investigation of the different stages of the infectious process. The study was done with plants obtained in vitro from meristems (cloning of the plant material, with monitoring of the stage of health) and from isolates of various aggressivities.

Thus, in the case of aggressive strains, we observed:

- The adsorption of the pathogen onto the cells of the limb tissue (the first stage of the infection process) was stronger than in the case of weakly aggressive strains.
- The installation, multiplication, and epiphytic reproduction of the bacterium took place faster and with larger populations.
- The penetration and multiplication in the tissues was rapid (4 days) and the internal populations were at a high level, leading to the rapid appearance of the symptoms (8 to 10 days). The vascular system was routinely colonized.
- The agglutination titre of the pathogen by lectin extracts from the leaves was lower than for weakly aggressive or avirulent strains.

In the case of resistant cultivars and of very aggressive strains, the multiplication and epiphytic preservation of the bacterium were still observed, but internal multiplication was inhibited. This observation indicates that a resistant cultivar may be a source of inoculation of aggressive strains.

Our results indicate also that the resistance of cassava to infection occurs at the level of the leaf, by limiting the adsorption, penetration, and especially multiplication of the pathogen in the tissues. It is at this level that lectins may be involved, by agglutinating the bacteria in the tissues and by limiting their multiplication.

These various results still do not allow the separation of strains of X. c.pv. manihot into races or pathotypes on the basis of testing the pathogenicity on a differential range of cultivars of cassava, or of plants of the genus Manihot possessing different resistance genes. On the other hand, little is known about the variability of the pathogen in nature. Notably, no one yet knows how such a diversity might be related to the epidemiology. This aspect is of major importance for varietal selection.

As a matter of fact, some authors (Brinkerhoff, 1970; Rao et al. 1971) pointed out that growing a resistant cultivar alters the selection pressure and favours the most aggressive strains. This

can be put side by side with Van Der Planck's hypothesis (1968) stating that the level of aggressivity of a pathogen agent is just sufficient for its maintenance in a host.

It is therefore necessary to adapt selection schemes to the appearance of strains whose virulence and aggressivity may be different. From this point of view, to avoid the selection of a very aggressive strain adapted to a particular cultivar, it is necessary to inoculate a sample of strains representative from the region where selection is operating and to preserve a certain diversity of cultivars, as monoculture leads to the selection of strains which are aggressive for the particular cultivar.

#### CONTROL METHODS

The present state of knowledge about this disease makes it possible to envisage an integrated means of control by combining techniques of cultivation, varietal resistance, biological methods of control, and sanitary measures.

In the Congo, a plan for an integrated attack on the disease is being evaluated. It includes:

- Production of healthy cuttings:
  1. by the multiplication in stocks of cuttings of plant material from disease-free (forest) zones;
  2. by the multiplication of improved plants derived from culture in vitro.
- Careful supervision of hygiene in the stocks of cuttings:
  1. by chemical control, spraying copper compounds (Bordeaux mixture, copper oxyacetate);
  2. by biological control, using microbial antagonists;
  3. by timing the planting to correspond to the period when the environmental factors are unfavourable to the outbreak of the disease and are favourable to the establishment of the plantation (at the end of the rainy season).
- Selection for varietal resistance: such selection must be performed in a region where the disease is endemic and must aim at stabilizing yields by integrating the other constraints on production into the selection plans. Lozano et al. (1983) indicated that the cultivars considered to be resistant must be evaluated over four growing-cycles. The final evaluation of resistance must take into account the yield, the production of cuttings, and the quality of the planting material intended for vegetative multiplication.
- The introduction of foreign varieties in the form of plants grown in vitro.

## CONCLUSION

By combining classic sanitation techniques with varietal selection into a programme to control bacteriosis, it should be possible to limit the effect of this disease. However, in this integrated pest management action, it is necessary to take account of other constraints in order to stabilize the yields.

In the specific case of vascular bacteriosis, the great variability of the causal agent implies that cassava planting is not safe from new epidemics induced by the appearance of aggressive strains.

This risk means that future investigations will have to aim for a better evaluation of the variability of the pathogen agent and of its effect on the epidemiology of the disease.

The combination of *X. c.pv. manihotis* and cassava constitutes an excellent model for fundamental investigation of the plant-bacterium interactions. The knowledge acquired (about the relation of aggressivity to the presence of plasmids, the differential adsorption of the strains as a function of their aggressivity, and mechanisms of resistance), combined with tissue culture, allow us to consider investigations on the plant's defence mechanisms, and maybe the use of genetic engineering.

Sufficient basic data are now available to allow investigations of the type already performed for Rhizobium, Agrobacterium, and Pseudomonas solanacearum to be undertaken.

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**SESSION E**

**BREEDING**

Chairman  
Dr. VAN DER GRAAFF  
Rapporteurs  
Dr. KONATE  
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HOST-VIRUS RELATIONSHIPS  
OF RESISTANT CASSAVA AND ACMV  
SOME IMPLICATIONS  
FOR  
BREEDING AND DISEASE CONTROL

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EARLY WORK ON MOSAIC DISEASE IN EAST AFRICA

Before discussing host-virus relationships in plants that are resistant to ACMV, I would like to describe the early East African work which gave rise to much of today's resistant cassava germplasm. Storey & Nichols (1938) started the work in 1935. They concluded that the level of resistance present in Manihot esculenta was not adequate for East Africa, though they obtained enhanced resistance by crossings between cultivars of diverse origins. In 1937 they turned to interspecific crosses and later concentrated on hybrids between cassava and the tree species M. glaziovii (Ceara rubber). The task was to retain the resistance of M. glaziovii while edible roots were restored by repeated backcrossings to cassava. This work proceeded with difficulty during the war years; the first backcross was raised in 1940, the second in 1943 and the third in 1946 and 1947 (Nichols, 1947). I joined the team in 1950, by which time third backcross hybrids with promising resistance and good quality roots were being tested on farmer's plots in Tanzania, Kenya and Uganda. One of the best of them was 46106/27, which I shall discuss below.

M. glaziovii is not immune from ACMV, and so resistance was assessed in field trials in which healthy material under test was planted adjacent to diseased infectors. In resistant genotypes only a low percentage of plants showed symptoms, usually mild or transient, and only after prolonged exposure to infection. But when healthy material was planted in new areas, notably on the coast of Tanzania and Kenya, some of the resistance was said to "break down", because more plants became diseased, even though the planting material came from the same healthy stocks. I was therefore given the task of breeding for higher resistance, and of studying host-virus relationships to try and explain this variable behaviour.

Selections with improved resistance were obtained by intercrossing among the third backcross hybrids (Jennings, 1957).



This is not surprising, because genes for resistance are recessive (Hahn et al., 1980; Jennings, 1978). Seeds from some of them were sent to Nigeria and subsequent generations gave rise to the main source of resistance used in the IITA breeding programme (Beck, 1962).

#### HOST-VIRUS RELATIONSHIPS

In our trials, we invariably found that susceptible genotypes became diseased within 1-2 months of planting in October-November, while about 25% of plants of resistant genotypes showed symptoms for the first time in the following April or May. To investigate the virus status of the remaining symptom-free plants, cuttings from all parts of 10-month old plants were planted in August 1954 and 1955 and the first growth cut back twice to promote symptom expression.

The results (Table 1) show that virus occurred in cuttings from 28% of the symptom-free plants and were in two categories. In 14% of the symptom-free plants tested, only cuttings from the base of the plant gave symptom-bearing plants and all the others gave symptom-free plants. Moreover, more diseased plants came from a basal cutting (B1) than from the one immediately above it (B2) and even fewer came from the third cutting from the base (B3) (Table 2). In a similar experiment with older plants (16 months), only 12% of the symptom-free plants gave symptom-bearing cuttings and in 4% the symptoms were limited to basal cuttings; no virus was detected in 45% of the plant that had shown transient symptoms and in a further 36% of the latter, virus was detected only in basal cuttings. Thus there were many more infected plants than those which had shown symptoms.

These experiments suggested an hypothesis for the relationship between ACMV and a resistant host: following infection, which can occur only at stem apices, the virus often moved downwards without inducing symptoms or inducing only transient symptoms. It left the terminal parts apparently virus-free. Some evidence from Nigeria supports this conclusion (IITA, 1980). The lower incidence of infection in cuttings from the symptom-free 16 month old plants compared to the 10 month plants suggested that all the virus had passed from the stems to the roots in the older plants. In the older symptom-bearing plants there was little evidence of systemic invasion or of upward movement of virus in the second season's growth. Thus by the end of the first season, about 25% of the exposed plants had developed symptoms, often mild or transient, about 28% of the symptom-free plants contained virus in their stems while the health status of the remainder could only be guessed. Circumstantial evidence suggested that the virus could have become limited to the roots. Alternatively, some plants may have escaped infection. It was concluded that an ability to localise virus within infected plants was a component of resistance. But, so long as some infected plants fail to develop symptoms, we do not know when a plant is not infected and cannot form an opinion on whether there is resistance to infection itself.

Table 1. Detection of ACMV in symptom-free plants exposed to infection for 10 months and in symptom-free and symptom-bearing plants exposed for 16 months (Jennings, 1960a).

No. of plants studied	No. of plants with field symptoms			No of symptom-free plants whose cuttings were healthy or infected		
	1st year	2nd year	All healthy	Infected Base B1 B2 or B3	one stem	various parts
(a) 10-month old plants, 282 symptom-free and 6 with transient symptoms						
133(154)	0	-	95(71%)*	21(16%)	0	17(13%)
149(1955)	0	-	107(72%)	19(13%)	0	23(15%)
282(total)	0	-	202(72%)	40(14%)	0	40(14%)
6(1954) 6 Transient		-	1(17%)		0	1(17%)
(b) 16-month old plants, 25 symptom-free in each year, 22 with symptoms in the first year only and 29 with symptoms in both years.						
25(1955)	0	0	22(88%)	1(4%)	0	2(8%)
22(1955)	22	0	10(45%)	8(36%)	4(18%)	0
29(1955)	29	29	0		0	15(52%)
14(48%)						
* In some instances symptoms appeared in new growth from stumps (c. 5cm) whose cuttings all produced symptom-free growth.						

Table 2. Distribution of virus in cuttings from symptom-free plants found to be infected after 10 months exposure to infection.

Level of cuttings	38 symptom-free plants found infected in 1954		42 symptom-free plants found infected in 1955	
	Cuttings tested	% virus detected	Cuttings tested	% virus detected
Basal (B1)	51	55	81	38
1st above basal (B2)	55	27	97	22
2nd above basal (B3)	57	18	147	11
Mid-upper*	50	24	284	9
Uppermost*	74	16	314	9

\* The uppermost cuttings were the most terminal from mature wood and the mid-upper cuttings were from the base of the branches from which the uppermost cuttings were taken.

The result accords with observations on farmers' plots where the incidence of symptom-bearing and their symptom intensity are commonly increased by the practice of plucking stem tips for use as a leaf vegetable ("kisamvu") and by grazing goats.

#### SYMPTOM INTENSITY AND SYMPTOM EXPRESSION IN PLANTS INFECTED AFTER PLANTING

In the East African trials, symptom intensity after infection in susceptible genotypes increased progressively as the plants grew, while in resistant genotypes the symptoms usually became reduced to a mild chlorosis with no leaf distortion or disappeared altogether in March after five months of growth. There was then a slight increase in symptom intensity in April and May. This was also the time when symptoms first appeared in most of the plants which developed them (about 25%). It was clearly a time when the resistance mechanism was under stress. Tipping to promote symptom expression nearly doubled the number of symptom-bearing plants when it was done in April, was less effective when done in May and had no effect when done in June. Possibly the large increase in the vector population associated with the April onset of the rains influenced both the symptom intensity of the symptom bearing-plants and the proportion of plants bearing symptoms. Failure to increase the number of symptom bearing plants by tipping in June was probably caused by the movement of April-inoculated virus away from the stem apices and out of range of the treatment effects (Jennings, 1960a).

#### EFFECTS OF ACMV ON PLANTS ESTABLISHED FROM INFECTED CUTTINGS

Although it is difficult to envisage that yield is seriously affected by the mild symptoms that resistant plants develop when they become infected after planting, there is no doubt that the consequences of planting diseased cuttings have serious consequences, even in resistant cultivars. Typical losses reported by Bock (1983) for East Africa range from 60 to 80%. Chant et al. (1971) estimated that ACMV reduced the leaf area by 24% and the photosynthetic activity of leaves by 23%. It also increased respiration rate and peroxidase activity. They found abnormal differentiation of palisade cells in infected leaves. Another failure of differentiation is the almost total lack of articulated laticiferous cells in diseased leaves: the fibrous masses and small crystals which these contain in healthy tissues are found instead in sieve tubes and partially block them (Murant et al., 1973). Stem tissues have not been studied, but the consequences would be serious if their main conducting tissues were similarly affected. Ekandem (1964) found that ACMV suppressed tuberous root formation; this is another failure of differentiation, because tuberous roots are differentiated in the first six months of growth and subsequent yield increases are due to increases in their size (Beck, 1960).

## ESTIMATES OF YIELD LOSSES CAUSED BY ACMV

Yield losses from ACMV infection differ for different cultivars. In the East African work, Nichols (see Jennings, 1960b) recorded a yield loss of 70% in 12-month plants of a local cultivar but only 35% in a susceptible M. glaziovii hybrid. However, Storey & Nichols (1938) rejected the option of selecting for this kind of tolerance. It is interesting to speculate that the different procedures for selection used in East Africa and at IITA might have different consequences on the selection of this kind of tolerance: in East Africa the breeding material was raised in isolation and all assessments of yield were done on plants raised from healthy planting material; at IITA, breeding progenies are exposed to infection as soon as the seedlings germinate and a high proportion become infected as young plants, whose yield potential is later assessed in plots established from infected material. The latter procedure would be expected to favour a form of tolerance which permits plants to give a high yield when diseased. Considerable yield improvement has indeed been achieved at IITA by the introduction of South American germplasm, but Hahn *et al* (1980) found that yield in a wide range of material was a remarkably close function of the severity of ACMV ( $r=0.98$ ). Nevertheless, the yield reductions attributed to ACMV infection in West Africa are lower than those reported for East Africa: Terry & Hahn (1980) reported a 32% yield loss in the resistant hybrid TS30395, and Ekandem (1964) reported no significant yield reduction in two out of five local cultivars studied, and losses ranging from 14.5 to 67% depending on symptom intensity in another.

Experience in Tanzania emphasised the considerable effect of stress conditions on the benefits to be derived from healthy planting material. Effects on plant establishment were a major factor. Thus Childs (1957) reported that healthy 46106/27, a third backcross M. glaziovii hybrid, yielded eight times as much as an infected local cultivar when planted on flat sandy soil but less than twice as much when planted on more fertile ridged soil (Table 3). Clearly, comparisons of cultivars for yield reduction due to ACMV must be made at several sites chosen to represent all situations.

Table 3. Establishment and yield in trials on the Tanzania coast of 46106/27, a moderately resistant *M. glaziovii* hybrid planted with healthy cuttings, and of the two local cultivars "Fungamkia" (moderately resistant) and "Gide" (susceptible), planted with diseased cuttings (Childs, 1957).

Cultivar	46106/27	Fungamkia	Gide
Red soils ridged	85(5)	49(8)	59(6)
Red soils flat	77(5)	43(8)	41(6)
Sandy soils ridged	86(7)	41(3)	37(4)
Sandy soils flat	51(7)	32(3)	17(4)

Yield (lbs)	Per	Per*	Per	Per	Per	Per
	plant	unit area	plant	unit area	plant	unit area
Red soils ridged	6.3	535	5.2	255	5.0	295
Red soils flat	4.3	331	3.1	133	2.8	115
Sandy soils ridged	5.0	430	2.3	94	2.8	104
Sandy soils flat	3.3	168	1.9	61	1.2	20

\* Yield per unit area is estimated from the product of yield per plant and % establishment (no. of sites in brackets).

#### CONCLUSIONS

1. An ability to localise virus in basal parts of the plant is a component of host resistance to ACMV.
2. It is unwise to speculate on the occurrence of resistance to infection, because it is usually not possible to distinguish symptom-free infected plants from uninfected plants.
3. Cuttings from symptom-free plants exposed to infection are liable to give some symptom-bearing plants. Roguing is therefore necessary to control the health of planting material, but it is liable to be minimal for resistant cultivars and excessive in susceptible ones.
4. Abnormal differentiation within the plant probably contributes to the low yield associated with plants raised from infected planting material. More assessment is needed of the consequences of the reduced differentiation of laticiferous cells and of tuberous roots.
5. Selection for yield in breeding material that is exposed to infection from the time of seed germination may give selections with an ability to tolerate disease as well as an ability to limit symptom expression. The evidence is equivocal, because both kinds of tolerance are influenced by growing conditions, but both kinds of tolerance can be recognised among cultivars.

6. Although newly bred cassava cultivars have much improved yield potential, it is likely that ACMV reduces their yield potential significantly, and that their potential could be realized more fully if the health of the planting material was managed appropriately.

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## AFRICAN CASSAVA MOSAIC DISEASE

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African Cassava Mosaic Disease is an important cause of loss of yield. The production of clones presenting as few symptoms as possible has therefore from the beginning been a prime objective of programmes for varietal improvement. The disease has been studied as regard its effects on the development of the host and consequently on yield, more to obtain results applicable to development than to understand the inner mechanisms of its transmission and development.

The main studies were carried out in Madagascar, and were based on detailed observations (Cours, 1950) which made it possible later on to effectively orient the programme to find productive clones. Studies in Africa were more modest (Vandevenne, 1980; Pouzet, 1984). They were complemented by work on in vitro cultures (Féréol, 1978).

The results enabled to confirm the presence of sources of tolerance-resistance and consequently to improve the cultivated clones. Nevertheless, the complex interferences of the relationships host plant-vector-parasite in relation with the environment, as well as a probable lack of knowledge, have limited the meaning of those results.

### THE STUDIES AND THEIR RESULTS

The observations of G. Cours in the 1940s, and more detailed work afterwards in various sites, served as the basis for the programme of varietal improvements, and its results.

These led to the following conclusions:

- The virus appears only on young leaves.
- It can be transmitted apart from propagation by cuttings - either by insects or by grafting.
- It is not transmitted by seeds.

- There are very clear and reproducible differences in the degree of susceptibility of the different clones.
- Manihot glaziovii and M. pringlei are two sources of resistance (see Table I).

Table 1. Effect of cassava mosaic disease on M. glaziovii and traditional clones (observations covering several years).

	Apparent contamination	Intensity
<u>M. glaziovii</u>	0	0
Local clones	30 - 100%	1 - 5

- The diseased leaves of a given clone contain more dry matter than do the healthy leaves and the wood of affected plants contains much more nitrogenous material than does that of healthy plants. This is also true of resins and starch (Table 2.).

Table 2. Influence of cassava mosaic disease on the chemical composition of the plant.

		Healthy Cassava	Diseased Cassava (intensity 3-4)
Leaves	Ash	1.90	2.94
Wood	Nitrogen	0.65 - 2.27	2.43 - 7.24
Stripped	Nitrogen	0.34 - 0.73	0.49 - 1.48
Root	Cellulose	0.87 - 1.76	0.45 - 0.95

- The disease is characterized by five degrees of virulence, each corresponding to a distinctive state of the affected plant.
- There is sometimes an onset of infection followed by a disappearance of the symptoms.
- There is a great variability in susceptibility, linked with the variability of the environmental conditions.
- Resistance is not stable.
- Just one part of the plant may show symptoms while another part appears healthy.
- Attacks are more severe in hot, dry years.
- Yield falls only when the degree of intensity reaches level 3 or 4.

The C/N ratio of the infected leaves is higher than in leaves with no apparent symptoms.



Crosses give the following results:

- Susceptible x susceptible always gives susceptible plants.
- Resistant x resistant usually gives resistant plants.

These conclusions were subsequently corroborated in West Africa and served very largely as the basis, or at least as factors for reflection, in the programmes in these regions. Thus in 1980, in Benin and the Ivory Coast, clones tolerant to the mosaic were cited, resulting at least in part from work done in Madagascar.

The effect of the mosaic on yield has been quantified on the basis of results covering several years. In central Ivory Coast, the mosaic affects yield only starting at level 3 to 3.5:

Mosaic level:	1-1.5	2.5-3	3.5-4	4.5-5
Yield (t/ha, wet wt.)	24.7	24.1	16.4	9.1

The same consequences have been observed in Madagascar:

Mosaic level:	1-1.5	2.5-3	3.5-4	4.5-5
Yield (t/ha, wet wt.)	36.4	34.2	23.7	12.9

A noticeable progress in the creation of new varieties has been observed over the years.

In Madagascar, particularly, clones such as H-57, H-58, H-62 and H-64, created in the middle and late 1970s, are more tolerant than older clones such as H-34 or H-43. In moderately infected humid regions, yields in two years of 50 to 70 t/ha in trials have often been achieved, versus 20 to 35 t/ha in one year. In severely affected dry regions, yields of 15 to 25 t/ha in a year are usual. These are means for several years.

During the 1980s these same clones, notably H-57 and H-58, showed mosaic levels comparable to or lower than those in the local sample CB in the Ivory Coast.

Finally, Féréol's studies (1978) have shown the value of tissue cultures in obtaining healthy plant material. These studies showed that treatment at given temperatures and times, 37°C for 15 days followed by 40°C for 30 days, then 37°C for 45 days, result in the complete disappearance of symptoms on one node cuttings originated from diseased plants (6 clones, including H-58, H-60, and CB).

These results make it possible to draw conclusions regarding both traditional cultivation techniques and selection programmes.

#### THE CONSEQUENCES FOR CULTURE AND BREEDING

Past studies and observations make it possible to recommend simple techniques that tend to limit the mosaic disease:

- Take cuttings only from healthy plants.

- Reduce the level of nitrogen in fertilizer and increase that of potassium.
- Grow improved clones that show at least a level of resistance compatible with a profitable level of production.

Certainly these techniques are not easy for the farmer to follow. But well-managed stocks of cuttings are an imperative basis if they are to be even partially effective. They are nevertheless only palliative measures, requiring vigilance because of the instability of resistance.

Varietal selection must consequently play a key role in holding the disease down to an economically tolerable level. This point is important, since it seems that a minor attack - up to level 2-2.5 - has only a minor impact on yield, and consequently a very moderate contamination is quite acceptable.

This results in two complementary breeding strategies.

One is aimed at achieving total immunity.

It is surely not unrealistic, in view of the probable potential in other species of the genus Manihot. The whole range of more than 90 species is a huge field for investigation in this respect.

The other strategy is aimed at achieving a good level of tolerance.

The experience in Madagascar has shown that:

- the great interclonal variability in M. esculenta still offers wide scope for exploitation,
- species such as M. glaziovii and M. pringlei (or considered as such in Madagascar) have shown their effectiveness in crosses. A simple technique using a combination of direct crosses and back crosses is then recommended. M. melanobasis and M. saxicola can be considered for this route.

During the breeding process, which should be carried out in naturally infected setting, the only technique is to eliminate the susceptible clones in several successive cycles. In this regard, there is a problem of evaluating and quantifying the effects of the mosaic disease as reliably and precisely as possible. The level above which it begins to have consequences is difficult to define.

- overall (for the whole of an ecology) and precisely (for a given clone)
- it is changeable, fleeting, and sometimes localized, as regards a single plant. Reliable, effective screening methods are therefore necessary to achieve valid, coherent results.

## FUTURE PROSPECTS

They are of three kinds:

- A better understanding of the mechanisms of infection and of the effects of the environment, climate and soil, are essential factors to improve the effectiveness of the breeding process. This must go together with precise genetic studies, which are also essential but which are difficult because of the heterozygosity of the clones.
- The judicious use of methods of varietal improvement, based on the understanding just mentioned, will make it possible to choose parent stocks rationally, on the basis of genetically determined resistance, and at the same time to choose a method for its appropriateness with heterozygous material.
- The use of species other than M. esculenta is a route which offers certain resources, though it poses a technical problem which can always be nevertheless resolved by appropriate methods, including tissues culture.

## CONCLUSION

In the matter of varietal improvement, the evaluation of breedings takes on a particular importance. Effective screening methods in a greenhouse or a shelter are a valuable contribution, though they are not essential for effective selection. Indeed, in Madagascar, selection sites in the field have shown that progress can be achieved, and that cultivars which are tolerant in one region are usually tolerant in another. Clones showing no attacks above level 2.5, combined with simple growing techniques, have shown over several years yields which are useful to farmers. Nevertheless, in many countries the improved clones gain popularity only slowly and with difficulty. Hence, there is a need for an integrated approach to the matter of varietal improvement, combining tolerance with other necessary qualities such as adaption to the environment and the needs of growers and consumers.

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THE RESISTANCE OF CASSAVA TO  
AFRICAN CASSAVA MOSAIC

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The first programme of selection of cassava (Manihot esculenta) for resistance to African Cassava Mosaic Virus (ACMV) was carried out by Storey in East Africa in 1938 (Nichols, 1947). Initially, he produced intraspecific hybrids, using African clones and a Javanese clone (F279), which led to the creation of the hybrid 37244E. Then he created intraspecific hybrids, in particular M. esculenta x M. glazovii, followed by three backcrosses with M. esculenta, in this way selecting a resistant clone, 46106/27.

The same source of resistance was then used by Jennings in 1951 (Jennings, 1957), leading to the selection of the hybrid 5318/34. In 1958, Ekandem, working in Nigeria with seeds from this hybrid, bred a resistant hybrid, produced the clone 58308 which became the source of resistance to ACMV used in the IITA selection programme.

Hahn et al. (1980) concluded that the resistance of cassava to ACMV is:

- i) multifactorial and recessive,
- ii) A resistance to inoculation and diffusion of the virus,
- iii) not a resistance to the vector itself.

In order to test the resistance of the selected clones in comparison with local clones in the conditions of the Ivory Coast, and in order also to determine their various levels of resistance, we investigated the various components of resistance to ACMV.

We distinguished six different types of resistance:

- RF in the field,
- R1 to the vector,
- R2 to inoculation,
- R3 to multiplication of the virus,
- R4 to the symptoms,
- R5 to diffusion.

## MATERIALS AND METHODS. COLLECTION OF CLONES

In order to test a small collection of 54 clones, as representative as possible of the genetic variability of cassava, we chose clones of different geographic origins and resulting from different selection procedures. They originated from 9 different sources: The Ivory Coast, Togo, Nigeria, Central Africa, Zaire, Kenya, Madagascar, India and South America. Likewise, we have succeeded in assembling the clones described as resistant by the authors cited above and derived from either intraspecific or interspecific hybrids, selected in Kenya and Nigeria.

The experiment itself was performed in two stages: first, in 1984 we tested 28 clones, including among others the resistant ones from East Africa; then in 1985, we tested all the resistant clones, both East African and Nigerian.

## TECHNICAL EVALUATION OF RESISTANCE

The investigation was based on two principles:

- First, the variables chosen were recorded without any a priori classification of their value for describing the biological phenomenon studied.
- Then each was measured, many times if possible (1 to 25), so that the results might be as independent as possible from any climatic, agronomic, or experimental effects.

The curves representing the changes of these variables over time were reduced by transformation into one characteristic figure.

The six different types of resistance are represented by:

- RF: rough estimate of the area of the curve of the cumulative percentage of contamination over time,
- R1: cumulative number of whiteflies counted on the plants,
- R2: regression line of the changes in the cumulative number of whiteflies versus the cumulative percentage of contamination,
- R3: concentration of virus in the diseased plants (only one measurement in 1984),
- R4: intensity of the symptoms (mean of 3 different counts),
- R5: regression line of the intensity of symptoms versus time (1985 only).

Analysis: We analysed the correlations between the variables, then performed principal component analyses, hierarchical classifications, and finally multiple regressions.

## RESULTS AND DISCUSSION

A correlation matrix was established for these components of the resistance, which shows that the resistance in the field (RF) was significantly correlated with the other resistance ( $r = 0.48$  to  $0.80$ ). The most independent type of resistance was that to the vector (R1); R2, R3, and R4 were also significantly correlated with one another.

The object of the principal component analysis was to describe the cassava clones with regard to the five different components of their resistance to ACMV (R5 could not be taken into account). The results may be visualized in the form of three-dimensional diagrams representing, in the present case, 93% of the total variability. The coefficient of correlation of each type of resistance, with its three axes, ranged between 0.75 and 0.95. Axis 1 was especially represented by RF and R4, whereas axis 2 was represented only by R1, and axis 3 was more linked to R2 and R3. The same analysis done in 1985 with another collection of cassava led, except for some details, to essentially the same diagram. All the components of the resistance lay at practically the same place in the diagram, except the virus concentration, but it must be noted that there was only one estimation, which could not be done at the same time during growth in the two cases. This difference therefore probably accounts for the recorded change.

A hierarchical classification of the cassava clones according to the various type of resistance classifies them into several groups ranging from the most susceptible to the most resistant. The resistant groups contain all the hybrids from East Africa and Nigeria, but also the local clones from Kenya, two clones from India, and the clone Aipin Valenca, which was the one most widely used in the selection programmes.

The use of the multiple regressions allowed the resistance in the field (RF) to be related to all the other types of resistance, with a high level of correlation ( $r = 0.85$ ).

Consequently, in the collection of 54 clones used, RF is a good evaluator of the general resistance of cassava to ACMV, with no distinction among components of the resistance.

### TEMPORAL STABILITY OF RESISTANCE

We tested the same collection of 10 resistant clones with the same technique for evaluating resistance in the field, at different times of year, that is, subject to high inoculum pressure in April and low inoculation pressure in July. The correlation between two experiment was of the order of 0.75, which is highly significant.

Likewise, we compared the resistant in the field, during several successive years, on collections of about 30 to 50 clones of very diverse susceptibility, but at the same time of year, so they were subject to a similar inoculum pressure. The correlations obtained ranged from 0.58 to 0.69 and were highly significant.

Similarly, we compared the stability of the other components of resistance, such as R1, R3, and R4, for the same collection of 14 resistant clones between two successive years. The correlations obtained were of the order of 0.80 and were therefore significant.

#### SPATIAL STABILITY OF RESISTANCE

We compared the behaviour of a collection of 54 cassava clones in two very different regions in the Ivory Coast, one in a forest with two rainy seasons with 2000 mm of annual precipitation, and the other in a savanna region with one rainy season and 1000 mm of precipitation. In the first case, the resistance in the field for 1984 ranged from 220 to 1669, and in the second case it ranged from 0 to 168. For 1985, we obtained respectively 0 - 450 and 0 - 248 as a quantification of the resistance in the field. There is therefore some variation from one year to another, but the correlation between the two places was 0.49 and 0.46 respectively for the two years, a result significant at the 5% level.

#### DISCUSSION

Cassava has been selected for resistance to African mosaic using symptoms as the sole criterion, and in fact this component turns out to be dominant in the scheme that we have drawn up. All the resistance components that we tried to identify tend to point in the same direction. That is, the more vectors there are and the more virus-infected plants there are, the more symptoms there will be and the more virus there will be, considering all 54 clones together.

The resistance of cassava is not a single entity; there is a clearly pronounced resistance to the vector, which is practically independent of the other components. The resistance to the virus is more difficult to investigate, but it does exist and is strongly correlated with the expression of symptoms. Resistance to inoculation, which was investigated directly in the field and not in the laboratory, is sufficiently independent to suggest that it is different from the resistance in the field and particularly from the resistance to the vector. The resistance in the field, expressed as the percentage of virus-infected plants, is ultimately the best indicator of the resistance of a cassava strain. By multiple regressions, the other components of the resistance accounts for approximately 80% of the variability of resistance in the field.

The components of resistance investigated in two different collections had quite similar relationships, except for the virus concentration, which suggests that the system is relatively stable and reproducible, even though in the second year the vector populations were much lower. From one year to the other there were good correlations between the various components of resistance. It seems that there may also be a good stability of cassava's resistance in the field to African cassava mosaic, both



in time and space; we observed this in two different ecotypes and over several years.

The clones which result from selection for resistance itself obviously are classified in the group of resistant clones, but it is surprising to find in this group local clones also, such as those from the East Coast of Kenya and those from India. We did not test many South American clones, but all those tested were very susceptible. It appears therefore that cassava, or at least the clones that we studied, has certain resources of resistance. This store of resources has not yet been fully exploited, notably in respect of resistance to the vector and especially in respect of resistance to diffusion of the virus.

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## CASSAVA MOSAIC DISEASE IN INDIA

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Cassava, a major staple or subsidiary food of low income group in certain parts of India, is believed to have been introduced in 19th century. It occupies an area of 0.37 M hectares with an annual production of about 5.5 M tonnes. Compared to all the other casava growing countries, productivity of cassava is very high in India (17 t/ha). More than 80% of the area under production is in the state of Kerala, where it still enjoys the position of a secondary staple food. As industrial raw material it has also spread into other states like Tamil Nadu, and Andhra Pradesh, where the tubers are primarily used in starch based industries.

### BACKGROUND INFORMATION AND DISTRIBUTION

Cassava Mosaic Disease (CMD) was recognized as a serious threat to the cultivation of crop in Travancore (in Kerala State) as long ago as 1942, (Abraham, 1956). Alagianagalingam and Ramakrishnan (1966) described the disease as resembling African Cassava Mosaic Disease, and noted various physiological changes in the infected plants. Efforts to transmit the virus to any other indicator host or cassava through sap and insect were, however, not successful. Menon and Raychaudhury (1970) transmitted a similar disease to a cucurbitaceous host through the vector Bemisia tabaci. Narasimhan and Arjunan (1974) compared the yields of tubers of healthy and infected plants and suggested it to be as high as 90%. Central Tuber Crops Research Institute (CTCRI), Trivandrum, carried out systematic research programmes to understand the disease, focussing mainly on etiology, transmission, ecology and epidemiological aspects of the disease in India. The salient findings of these studies are discussed in the paper.

### CULTIVARS USED

Three local popular cultivars, namely, Kalikalan, Arimani and M4, and four hybrids of cassava released by CTCRI (H-226, H-2304 and H-1687) were used in all the studies. As these cultivars have

shown differential reactions to the disease (Table 1), they were included in the studies. Stem cuttings from naturally infected plants of these cultivars were maintained continuously and used. Healthy material, resulting out of meristem culture were used in epidemiological studies. Replicated field trials were performed following recommended management practices.

#### DISEASE INDEX

Symptoms in cassava are: typical mosaic pattern, either sparsely present or spread throughout lamina, leaf distortion and reduction in size. Intensity of symptoms expression varies with the season. During humid cool and rainy days (October-November) when the plant is also in active stage of growth, severe symptoms are noted. In drier months (January-April) symptoms are either masked or reduced to only very mild symptoms.

An indexing system based on foliar symptoms was developed to assess the severity of the disease. Disease index was found to be maximum from August to October. Of the cultivars studied local cv Kalikalan showed the highest disease index (Table 1).

Table 1. Disease index in Cassava cultivars

Name of cultivar	Source	Type of symptoms	Disease index
Kalikalan	Popular local cultivar	Extreme leaf curling mosaic stunted growth	3.6
Arimani	Popular local cultivar	Extreme leaf curling mosaic stunted growth	4.0
H-226	Hybrid from CTCRI	Leaf curl and mosaic	2.7
H-165	Hybrid from CTCRI	Leaf curl and mosaic	2.6
H-1687	Hybrid from CTCRI	Leaf curl and mosaic	2.6
H-2304	Hybrid from CTCRI	Leaf curl and mosaic	2.1
M4	Selections from Malaysian accessions	Uniform mosaic	1.8

Disease severity index shown by any cultivar, though consistent occasionally shows an irregular pattern. In the same cultivar in the same plant, both mild and severe symptoms could occur. Due to uneven distribution of virus it was possible to generate symptom-free plants by single node cuttings.

#### THE VIRUS

Virus isolates from Indian cultivars like Kalikalan, Arimani and hybrids are hereafter referred to as ICMV (Indian Cassava Mosaic Virus).

Of different isolates of ICMV tried, ICMV-Kalikalan and ICMV-Arimani alone were transmissible to indicator hosts. ICMV-

Kalikalan was readily transmissible to N. benthamiana and N. glutinosa. In N. benthamiana the symptoms were leaf rolling, wrinkling and stunted growth. Percentage transmission was 87. In N. glutinosa it produced leaf wrinkling, vein banding and reduction in lamina. It was not uniformly positive.

ICMV-Arimani behaved differently. It could be transmitted to N. benthamiana, N. glutinosa and N. tabacum "Xanthi". Percentage transmission was very low (12, 20 and 11 respectively). Symptoms in N. benthamiana were similar to one produced by ICMV-Kalikalan. In N. glutinosa, yellowing along the major veins appeared in the beginning, followed by leaf wrinkling and lamina reduction. In N. tabacum "Xanthi", within 48h of inoculation chlorotic lesions appeared on the inoculated leaf. It may become either systemic or remain as such, when systemic symptoms produced were mottling, extreme leaf curling, thickening of leaves and stunted growth. Attempts to transmit the virus to other indicator hosts and from indicator hosts to cassava so far have not given positive results.

Purification of ICMV from N. benthamiana and cassava cultivars, Kalikalan, Arimani and H-1687 were initiated. Parasometric particles of 18-24 nm in diameter occurring in pairs were detected. Besides characteristic pairs, particles were also detected as monomers or tetramers. (Malathi and Sreenivasan, 1983). It was not possible to obtain a good virus preparation following proven successful method (Sequeira and Harrison, 1982) in spite of using severely infected material in N. benthamiana. Semi-purified preparations from cassava cultivars were temporarily used for immunization. Antiserum was used in gel diffusion studies after cross adsorbing with healthy antigen.

There was a single precipitin line produced by antigenic preparation from all the isolates. The reaction was obtained consistently only when concentrated preparations were used in N. benthamiana. ICMV was also detected in symptom-free plants. This needs to be confirmed by using more pure antiserum.

ICMV from cvs Kalikalan, H-226, H-2304 and M4 reacted readily with antiserum of "T" and "C" strains of ACMV (Bock, Shantha and Malathi, unpublished), ICMV from hybrid H-1687 did not react. There was a spurring pattern indicating antigenic difference between ICMV and ACMV. Reactions were obtained only when concentrated preparations were used. In cv H-1687, though 150 g of leaf material was used, no reaction was obtained. However, it was later found to contain gemini viruses unlike Angolan isolates of ACMV.

#### THE VECTOR

Whitefly Bemisia tabaci has been reported to be the vector for CMD. Whiteflies collected from the diseased plants of cassava in the field, and also virus-free whiteflies reared on tobacco plants were used for insect transmission studies in vitro. In all the cases, transmission was not obtained even when large numbers of viruliferous whiteflies were used. None of the cucurbitaceous, colenaceous or leguminous hosts responded,

though Menon and Raychaudhuri (1970) had reportedly transmitted the disease from cassava to cucumber. There was no correlation between whitefly preference for feeding and incidence of disease. Cv Kalikalan, the most susceptible, is one of the least favoured.

#### TRANSMISSION

Seed transmission: More than 5000 seeds from different cultivars were sown to raise seedlings in insect proof screen house and none of the seedlings showed any symptom, indicating that it is not seed transmissible.

Graft transmission: CMD could be easily transmitted by grafting. Cultivars Kalikalan and H-165 were ideal donors. Based on symptoms expression, germplasm collection and hybrids were classified into resistant, slightly susceptible, moderately susceptible and highly susceptible. Selections, S-1315, S-2371, S-2380 were identified as resistant lines (Table 2) showing none or only sparsely distributed specks as symptoms.

Symptom expression was seen irrespective of graft establishment. It was possible to elicit symptom expression in cv Kalikalan by grafting with severely infected N. benthamiana were the contact between the tissue lasted only for 72 h.

Table 2. Graft transmission in cassava cultivars

Cultivar	No of plants infected		Symptoms
	No of grafts established		
H-165	16 / 16		Moderate
H-226	19 / 19		Moderate
M4	11 / 11		Moderate
S-2371	34 / 34		Minute flecks
S-2380	36 / 36		Minute flecks
S-1310	0 / 22		Nil
S-1315	0 / 37		Nil

#### ECOLOGY AND EPIDEMIOLOGY

Primary and secondary spreads: primary spread from one season to another is mainly due to indiscriminate use of infected stems. Lack of awareness of the disease amongst farmers have resulted in wide spread occurrence of the disease.

#### FIELD SPREAD

- (a) In different cultivars: Field trials have been conducted over a span of ten years to assess the nature of spread in different cultivars. Percentage spread was assessed based on symptom expression following new infection. Spread due to vector activity was found to be different for different

cultivars (Table 3), cv Kalikalan showed highest (52-78%). It was less, but of considerable proportion in hybrid H-226. Other hybrids H-165, H-2304 and H-1687 and cv M4 showed less than 5% spread.

Table 3. Percentage spread of CMD in cassava cultivars

Cultivar	Percentage spread
Kalikalan	52.00
H-226	14.50
H-165	3.73
H-1687	0.83
H-2304	1.00
M4	0.67

Spread was not correlated with whitefly population or with percentage inoculum provided. There was no definite pattern nor does it depend on wind direction. Rate of spread of CMD in different cultivars recorded for active growing season of September to December (Table 4) revealed that it was low in October but high in November. Maximum spread in cv Kalikalan occurs during November, irrespective of percentage inoculum given (Table 5).

Table 4. Seasonal variation of the field transmission of CMD

Cultivars	No of test plants	<u>No of plants infected</u>				Total spread	Percentage spread
		Sept.	Oct.	Nov.	Dec.		
H-165	179	0	0	0	0	0	0
H-226	179	3	0	5	1	9	5.03
H-1687	180	0	0	2	0	2	1.11
H-2304	179	0	0	0	0	0	0
M4	180	0	0	0	0	0	0
Kalikalan	160	12	2	38	0	55	34.38

Table 5. Seasonal variation and spread of CMD at different levels of inoculum in cv Kalikalan

Percentage inoculum provided	<u>Percentage spread</u>			% Spread at 6 months
	October	November	December	
0	22	23	19	53.33
5	12	38	23	64.03
10	6	26	10	47.22
25	12	27	12	56.66
50	17	23	7	78.33

- b) With different inoculum sources: Varietal susceptibility and not the source of inoculum was found to be an important factor in determining spread. Disease spread was high in susceptible cv Kalikalan, irrespective of source of inoculum (Table 6). cvs Kalikalan and H-226 had an average spread of 50% and 13.76 % respectively.

Table 6. Different inoculum sources and field transmission of CMD.

Sources	Percentage spread in test plants						Mean
	H-165	H-226	H-1687	H-2305	M4	Kalikalan	
H-165	0	3.3	0	0	0	21.4	3.9
H-226	0	10.0	3.3	0	0	43.4	8.0
H-1687	0	0	0	0	0	30.0	5.0
H-2304	0	3.3	0	0	6.6	30.7	6.2
M4	0	3.3	3.3	0	0	18.5	3.8
Kalikalan	0	10.3	0	0	0	65.3	11.3
Mean							

- c) The effect of different agroclimatic conditions on the spread of CMD was determined by conducting field trials in four locations, selected for the differences in weather and cropping pattern adopted. The centres adopted were Nagercoil and Trivandrum (low rainfall, equitable distribution of 100-150 cm) Palghat (Medium rainfall of 200 cm with heavy winds) and Salem (very low rainfall of 60 cm, but cassava grown as irrigated crop). In all the centres spread was assessed both "within" the plot and "into" the plot. Infected cuttings of Kalikalan were provided as source (10%) in trial to study the spread within the plot.

Spread was negligible in field resistant cvs H-1687, and M4, contrasting to high percentage of spread in cv Kalikalan (Table 7).

Table 7. Secondary spread of CMD at different agroclimatic conditions.

	1		2		3		3	
	Nagarcoil		Trivandrum		Palghat		Salem	
	+	-	+	-	+	-	+	-
H-226	0.26	2.77	1.74	0.46	1.89	0.13	0.82	0.74
H-1687	0.25	1.00	1.96	0.31	0.63	0.50	0.27	0.20
M4	0.25	0	0.26	0	0.38	0.13	0	0
KK	-	-	36.98	10.85	0	2.48	0	2.32
MVD.1	-	-	-	-	-	-	0	0

+ Inoculum (10%) present                      - Inoculum absent

1 Data for one year only

2 Mean of 4 years data

3 Palghat and Salem - mean of 2 years data



## YIELD LOSS

Yield loss varied with different cultivars. Highest percentage of yield loss was noticed in cv Kalikalan (Table 8). In hybrids H-1687, H-226 yield reduction was in the range of 18-25%; in hybrid H-2304 and cv M4 only less than 18%. Moderate yield reduction in hybrids due to tolerance possessed by them is markedly different from situation in Nigeria or Kenya. Hahn *et al.*, (1980) had reported 35% yield reduction even in a highly resistant line.

In cv Kalikalan where active spread occurs, yield reduction was inversely proportional to the age when infection sets in (Table 9).

Table 8. Yield loss due to CMD in different cassava cultivars

Cultivars	Mean percentage reduction over healthy (Tuber yield at ten months stage)
Kalikalan	42.40
H-165	20.00
H-226	18.10
H-1687	22.75
H-2304	16.90
M4	17.10

Table 9. Yield of cv Kalikalan in relation to age at infection.

Age at infection	Yield/plant (g)	Shootweight/plant (g)
Primary	542.3	697.0
3 months	1012.5	856.3
4 months	1656.8	1231.6
5 months	1870.9	1393.8
6 months	2111.8	1460.5
7 months	2037.3	1259.6
Healthy	1959.7	1233.9
CD (P = 0.05)	377.6	263.5

## FIELD RESISTANCE

From the perusal of the data on disease severity index, percentage spread and yield loss, field resistance of the hybrids, H-1687, H-2304, H-165 and cv M4 becomes evident. Thus the susceptible cv Kalikalan shows a high disease severity index, high percentage of spread and yield loss, H-165, H-1687, H-2304 and cv M4 show the other extreme of negligible percentage spread, low severity index and yield loss. Hybrid H-226 is in between, showing moderate susceptibility.

These field resistant cultivars are not absolutely resistant as they do get infected through grafts. Whether it is the long incubation time required for expression of symptoms or due to the viral strain is worth looking into. A resistant cultivar S-1315, which produced only mild specks when inoculated through grafts (vide - graft transmission), remained symptom-free for the past twenty years. Recently in 1984, it started showing symptoms, which appear increasingly severe.

#### CONTROL

The disease could not be controlled by adopting various control measures like hot water, hot-air treatment, insecticidal spray, alteration of population density and manurial proportions. Since active spread of disease in field resistant cultivars is low, the strategy at CTCRI consisted of identification of resistant lines, cleaning them of virus through meristem culture and adoption of the same for breeding or cultivation. Careful screening and roguing alone offer an efficient control measure.

#### FEED BACK FROM OUTREACH PROGRAMMES

Efforts to create awareness of the disease amongst farmers by conducting CMD eradication campaigns gave encouraging results. During October-November, when the expression of symptoms is optimum, farmers are advised to paint-mark the diseased stems to differentiate them from healthy ones during harvest. Reduction in incidence of disease from 100% to 53% was achieved in an area where an intensive eradication programme was undertaken.

#### DISCUSSION

Yield loss of considerable less magnitude in India is in contrast to all the other African countries. Such productivity seems to be higher in India than other cassava growing countries. Better management practices and absence of disease like cassava bacterial blight may contribute to high productivity in India. Notwithstanding yield loss measured as percentage reduction in tuber yield over the comparable control, is significantly low in indian cultivars. Two basic questions that emerge are (1) Whether the yield loss data are vitiated by the latent presence of virus in symptom-free plants with which the comparison is made? Whether symptom-free plants are genuinely virus-free? Increase in yield from meristem derived plants over normal set derived plants suggests the presence of such latent infection (Table 10). (2) Possibility of strainal variation in India, a strain quite different from that of ACMV. A kind of well adapted milder strain not causing much yield depression.

Percentage spread of the disease is low in several cultivars. In field resistant cultivars, if primary spread though vegetatively propagated stem is prevented, it is possible to maintain a symptom-free collection for more than twenty years.

Of isolates tested, only two of ICMV were transmissible to indicator hosts N. benthamiana, N. glutinosa and N. tabacum "Xanthi". The narrow host range observed is quite different from ACMV. The absence of a serological relationship with ACMV has been found by many workers. Generally it is attributed to concentration of the virus or to defective particle assembly as in Angolan isolates (Robinson *et al.*, 1984). Absence of reaction with ACMV antiserum in gel diffusion studies for ICMV-H-1687 cannot be attributed to concentration of virus as the virus could be readily seen in partially purified preparations.

Table 10. Percentage increase in tuber yield of meristem derived plants over normal ones (tuber yield at ten months stage).

	Increased percentage over normal plants					M4
	Kalikalan	H-165	H-226	H-1687	H-2304	
1984-1985	11.4	1.0	23.5	11.6	3.4	0
1985-1986	10.7	19.1	27.4	15.2	20.4	1.4
Mean	11.1	10.0	25.4	13.4	11.9	0.7

Ease with which insect transmission has been achieved in ACMV is not met with ICMV. Even susceptible cv Kalikalan, symptom expression after inoculation with viruliferous whiteflies was not seen. ACMV does not infect Cucurbita pepo (Bock and Harrison, 1985). ICMV is reportedly transmitted to cucumber (Menon and Raychaudhuri, 1970).

Bock and Harrison (1985) classified the virus isolated from Indian cultivars as Indian strain of ACMV. They assumed that cassava was introduced into India by Portuguese settlers from Africa. Burkhill states that (Abraham, 1956) it was freshly introduced from South America into India in 1840.

Subtle differences between ACMV and ICMV such as yield loss, tolerance to spread, host range, antigenic variation and geographical separation may need to be reviewed. The number of isolates and host range studied have been too narrow to draw any conclusion. Considering the ambiguity of information on the entry of cassava to India, the presence of whitefly earlier than in African countries (mound, 1983) and the enormous range of variabilities that one comes across in cassava, it is preferable to consider the virus isolates from Indian cultivars as Indian Cassava Mosaic Virus than as Indian strain of ACMV. A clear picture may emerge when more information is available.

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AFRICAN CASSAVA MOSAIC AND ITS CONTROL  
IN ZAIRE

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The importance of cassava in Zaire is shown by the area of arable soil that it occupies (approximately 50%) relative to other food crops. As a source of energy, cassava supplies 66% of the human diet, compared with 19% for maize, 8% for rice, and 7% for plantain, yam and potato (FAO, 1983).

The tuberous roots and the leaves are eaten in almost equal proportions, depending on the region and the dietary habits.

The tubers are eaten raw, fried or boiled in the case of the sweet clones, but often in the form of various preparations: chikwangue, fufu, malemba, which require prior transformation (retting, sometimes followed by drying).

Zaire is not officially known as an exporter of cassava, although substantial quantities of cossettes (root slices) and of pondu (edible leaves) frequently cross the borders of the country.

Cassava farming is entirely traditional. The mean production is 7t/ha when grown by these traditional methods and up to 25t/ha in experimental stations. There are virtually no large industrial plantations. In recent years, however, there has been increasing interest in the planting of large areas (up to 40-50 ha) by some growers and industrial organizations, whose main aim is profit rather than subsistence. In Zaire, farmers generally plant cassava in association with crops such as peanuts, maize, cowpea, beans, sesame, squash. The combination of crops depends on the dietary and financial needs of the farmer, and on the soil and climatic conditions.

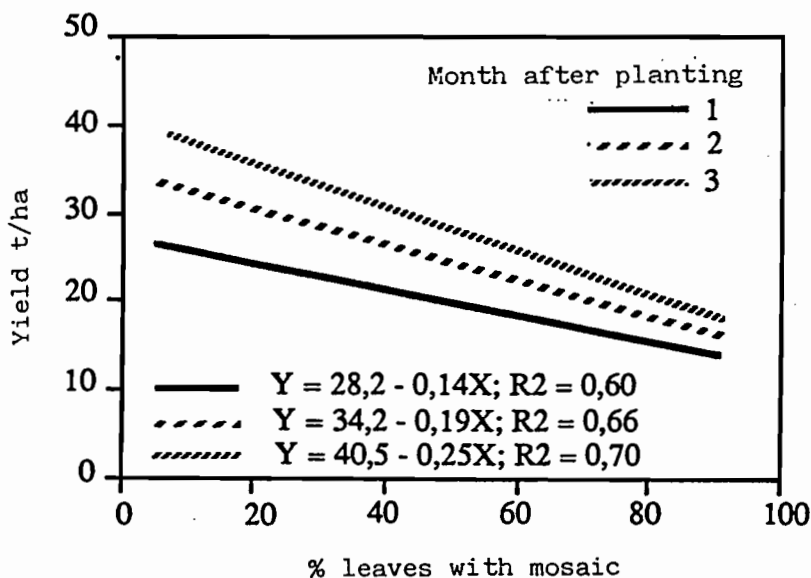
Diseases including bacteriosis, mosaic, and anthracnose, predators including the mealy-bug and the green mite, and poor crop management as regards density and time of planting, use of susceptible local varieties, choice of material for planting, choice of ground, late or inadequate weeding are the principal limitations on cassava production in the traditional system.

Cassava mosaic is endemic and present throughout Zaire. In susceptible variety it may reduce the tuber yield by as much as 60 to 70% (PRONAM, 1984). The losses of yield caused by mosaic

disease seem to depend on the intensity of attack on the leaves during the first four months of growth (Fig. 1). Its effect on the leaves seem to depend on the level of infection of the initial planting material, as scored subjectively on a scale from 0 to 5 (PRONAM, 1984) with 1=resistance apparent in the field and 5=severe mosaic with a large reduction of the leaf area.

Although mosaic disease is present everywhere in Zaire, its incidence and severity vary considerably from one geographic region to another. In the savanna region in the southeast, for example, the disease is more severe in the regions of Bas-Zaire and Bandundu. This variation may depend particularly on the highly variable soil and climatic conditions in Zaire, and on the traditional system of farming, on the lack of any choice in stocks for planting, on how long a field is used, on the viral strain, and to a lesser extent on the presence and the density of the whitefly population.

Figure 1: Relationship between percentage of mosaic infected leaves and yield in tuberous roots of cassava, PRONAM, 1984.



For production purposes, cassava is not propagated from seed in Zaire, but vegetatively. It is present everywhere in the country but in proportions that vary from one region to another depending on how important it is as the principal energy-providing foodstuff relative to rice, maize, plantains, yams, and taro. It is generally grown all year round, except in regions where the climatic conditions do not allow this.

Just as with the incidence and severity of mosaic disease, the presence and especially the density of the whitefly population varies also from one geographic region to another. In a given

region, its population density would depend on the time of year (Fig. 2), on the age of crop, since young plantations harbour more insects than old ones, and on the variety (PRONAM, 1984).

The whitefly occurs also on other crops such as cotton, onion and niébé.

Given the area of Zaire and the present level of agronomic research, it is difficult to estimate the number of clones presently grown in the national territory, or even in a given administrative region, as the number and the nature of the varieties planted frequently vary from one region to another. This is the result of exchanges of material, often practised by growers between regions and even between countries. However, the number of varieties can be estimated at a thousand.

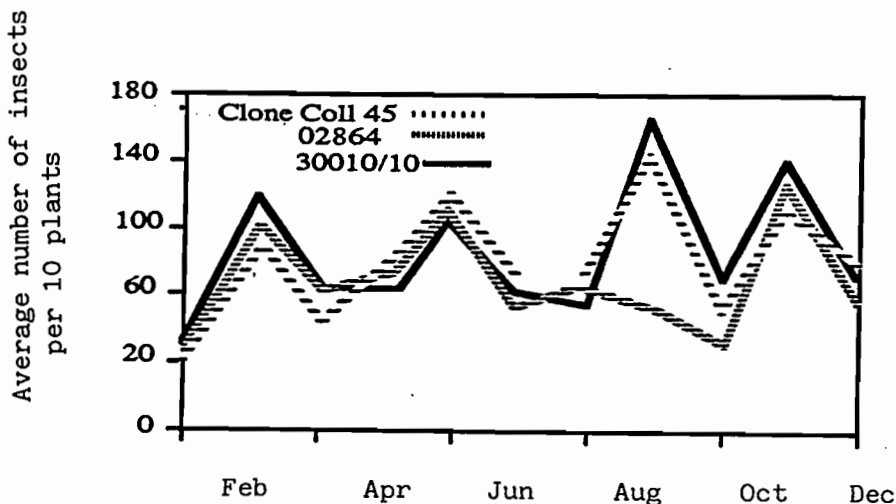
The bitter clones are more widespread and are preferred by the farmers of Zaire; they give products (chikwangué, fufu, etc.) of better quality.

Collections maintained in the country's experimental stations contain approximately 600 clones.

To create wide genetic variability, for the sake of selection, seeds and vegetative material in the form of tissue culture have been introduced into the Zaire since 1974, from the International Institute of Tropical Agriculture (IITA) and from Latin America.

From this basic population complemented with seed and vegetative material from local varieties in the country, such clones as Kinuani, F100, 30572, 02864, 4(2)0426/1, 30010/10, 30572/149, 61665/4, which are tolerant to diseases including mosaic and which give a high yield, have been selected or identified.

Figure 2: Variation of the population of whiteflies at M'vuazi, PRONAM, 1983.



Up to 1986, 3.385.413 metres of planting material of the improved clones, enough to cover approximately 1375 ha at 2500 metres of material per hectare were distributed to farmers (PRONAM, 1986). Their acceptance by the farmers, though slow, has been satisfactory and is promising, considering the strong demand recorded.

Research on cassava in Zaire was started in about 1945 by the Institut National d'Etude et de Recherche Agronomiques 'National Institute for Agronomic Study and Research'. It had reached an appreciable level when it was halted by the political and economic disturbances following Independence. Its results did not reach the majority of farmers. From 1974, following severe cassava epidemics due to bacteriosis, investigations were restarted by the creation of the Programme National Manioc 'National Cassava Programme, PRONAM', project of the Conseil Exécutif National 'National Executive Council', assisted financially by the U.S. Agency for International Development (USAID) and scientifically by the IITA.

The task of PRONAM is to improve cassava-growing in Zaire by selecting resistant high-yield varieties and to develop improved farming techniques. Working in multidisciplinary teams, PRONAM has a phytopathology section whose principal role is to assist in the selection for resistance to diseases. The selection also performs epidemiological studies and keeps records on new epidemics.

In view of cassava's place in the food supply of Zaire and the great many limitations (including mosaic) on its yield, PRONAM, though the overseeing Department, welcomes and favours participation in all programmes aimed at strengthening research and/or phytosanitary surveillance on cassava. Apart from financial assistance from the USAID and scientific assistance from the IITA, PRONAM is not cooperating officially with any other institution in its research programme.

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**IMPORTANCE, PRODUCTION AND UTILIZATION OF CASSAVA  
IN UGANDA**

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Cassava, Manihot esculenta Crantz is the most important root crop in Uganda. Among African countries, Uganda ranks fifth in the quantity produced and the area under cassava. In 1983, 3.239.000 tonnes were produced. Total production in 1986 is likely to be 30% higher. Table 1 shows the production, area and yield of cassava in the country from 1971 to 1983. The figures indicate a general increase in hectarage and a corresponding decrease in production of the crop from 1971-79, after which production and hectarage started to increase steadily. The decline can probably be attributed to the acute shortages of agricultural inputs in the country, particularly hand hoes and animal implements. The problem was even worsened by the effects of the Liberation War in 1979, which severely reduced productivity in the entire agricultural sector of the economy.

Table 1. Production area and yield of cassava in Uganda 1971/83.

Year	Production ( '000 tons)	Area ( '000 ha)	Yield (kg/ha)	Average (kg/ha)
1971	2,951	466	6,333	-
1972	2,826	373	7,576	6,265
1973	2,383	440	5,416	
1974	2,444	426	5,737	
1975	3,081	500	6,162	
1976	2,982	512	5,824	5,003
1977	2,287	540	4,235	
1978	1,589	419	3,792	
1979	0,848	143	5,930	
1980	2,072	302	6,861	
1981	3,000	310	9,677	8,124
1982	3,127	331	9,447	
1983	3,239	372	8,707	
				6,404

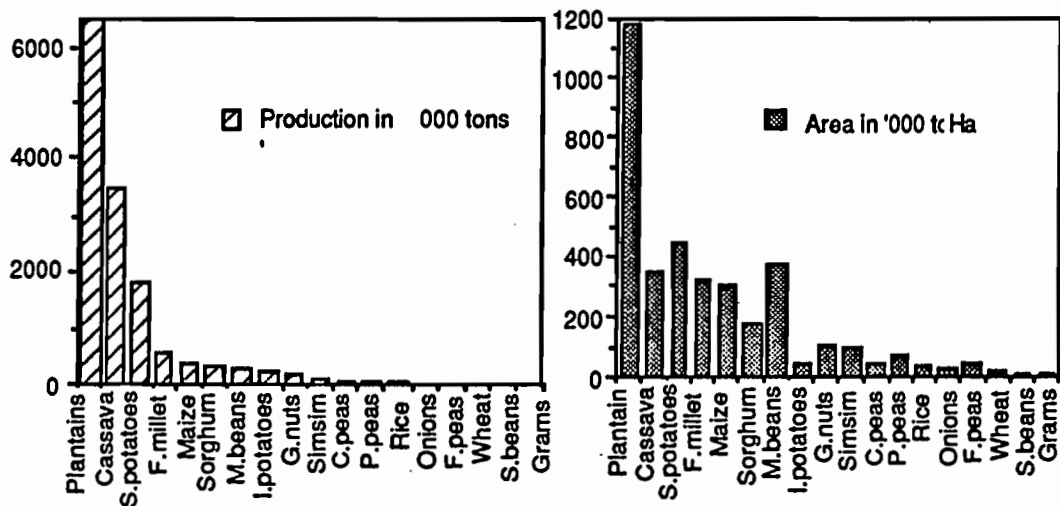
SOURCE: Dept. of Agric. Annual Report, 1983, Min. of Agric. and Forestry, Entebbe, Uganda.

After the war, production of cassava and indeed of a number of food crops, drastically increased from 848.000 tons (143.000 ha) in 1979, to 3.239.000 tons (372.000 ha) by 1983 and from a yield of 3.792 kg/ha in 1978 to 9.677 kg/ha by 1981. This drastic achievement was a result of the rigorous policies of the new government, reviving the entire agricultural sector through provision of agricultural inputs and incentives, During the same period too, Uganda experienced an economic shift. Many farmers moved away from cash crop production which for a long time has continued to earn less attractive financial returns compared to other food crops such as cassava, maize, etc. With its relatively low labour demand and high yielding ability, cassava found advantage over most agricultural crops. The crop is grown throughout the country but the greatest cassava production comes from northern (Gulu, Lira, Apach and Kitgum), eastern (Mbale, Tororo, Soroti, Kumi) and north-western (Arua and Nebbi) regions of the country. These are semi-marginal areas more prone to drought and where a decrease in rain may result in serious crop failures. In these areas, the crop commands a major role as a stable food crop. In the central (Mpigi, Mubende, Luwero, Masaka, Rakai) and western (Kasese, Bundibudgyo, Kabale, Rukungiri) regions, cassava has been largely replaced by plantains and sweet potatoes.

As shown in table 1, the total area under crop is increasing with corresponding decrease in total production as a result of continued decrease in yield/ha. This decrease in yield is due to the devastating effects of pests and diseases, notably the cassava green spider mite (GCM) *Mononycellus tanajoa* (Bondar), the cassava bacterial blight (CBB) *Xanthomonas campestris* pv. *manihotis*, the African Cassava Mosaic Disease (CMD) and the decline in soil fertility as a result of continuous cultivation.

Figure 1 shows the importance of casava with respect to the main food crops grown in the country in 1983. It can be observed that among major food crops grown in the country, cassava ranks second to plantains.

Figure 1: The importance of cassava with respect to other food crops in Uganda, 1980/83.



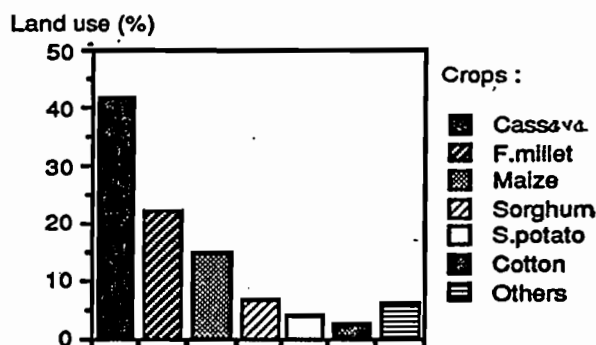
In Tororo, Kumi and Soroti districts, of eastern and northern Uganda, cassava occupies from 35-45% of total land available for crops (Figs 1.2 and 1.3) and takes 48-50% of farm family labour (Ocitti P'obwoya and Otim Nape 1986).

The crop is grown by peasant farmers on small plots varying from 0.1 ha in the Lake Victoria region to 5.0 ha in eastern, northern and north western regions. Stem cuttings about 30cm long are planted horizontally in the ground at spacing between 1m x 1 m to 1.5m x 1.5m, depending on whether the crop is grown in monoculture or intercropped. About 30% of the total cassava is planted in the first rains, 50% of it is intercropped with various annual crops such as groundnuts, beans or cowpeas, or maize, which are harvested after three to four months to leave pure stands of cassava.

Fig 1.2: Land utilization among farmers in Tororo district



Fig 1.3: Land utilization among farmers in Soroti and Kumi districts



Commercial plantations of cassava are rare, although Lira Starch Factory has a large estate (200 ha) of cassava in the northern region. Because of the subsistence nature of cassava cultivation in the country, use of fertilizers and herbicides is rare and weeding is done by hand.

Harvesting of tubers, usually done after twelve months or so, is sporadic. A few tubers for family consumption are harvested at a time. Where cassava is intended for sale, the wholer field may be harvested at once.

While according to 1981-1983 figures the per capita production of cassava in the country is about 224 kg fresh roots per person per year, making Uganda one of the greatest cassava producer in Africa, average and maximum yield/ha varied from 9.7-4.3 tons/ha and 30-20 tons/ha from 1981 to 1985 respectively. Yields at research stations, varietal trial centres and on farm trials vary from 30-55 tons/ha. This great disparity is due to the poor cassava agronomic practices followed by farmers, to declining soil fertility and to the devastating effects of pests and diseases, notably the green African Cassava Mosaic Disease. Minimum production of the cropper farmer varied from 1-0.4 tons for farmers with small (0.1 ha) plots to 48.5-21.5 tons for farmers with large (5 ha) plots from 1981-85 while maximum production varied for the two types of farmers from 3-2 tons and 150-100 tons respectively during the same period.

In Uganda, cassava is important because of its starchy tubers which can be consumed fresh or dry. Fresh cassava may be consumed steamed, mashed or as snacks according to locality and peoples' food preference.

a) Fresh cassava.

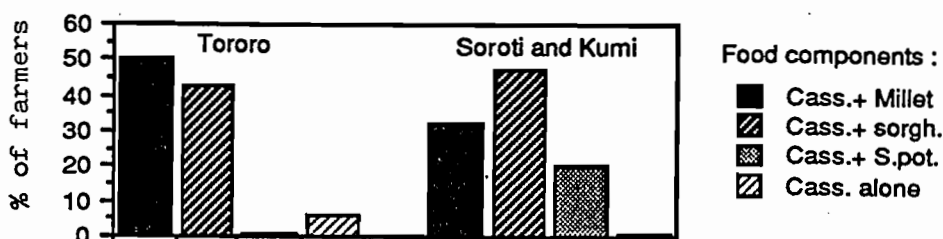
Fresh cassava tubers are peeled, sliced and/or chopped into chips, placed in water and thoroughly washed two or three times in water, placed in a saucepan or earthen pot containing little water and boiled over a fire, until they are soft and ready for eating with a sauce such as groundnut stew, boiled/cooked beans, green vegetables, meat, etc. Alternatively, fresh cassava tubers are peeled, sliced and/or chipped into smaller pieces, washed two or three times in water, placed in a saucepan or earthen pot containing enough water and boiled over fire until the chips are very soft. The soft cassava chips are mashed with occasional addition of water to achieve the desired consistency. The finished product can be eaten with any sauce. Fresh cassava, peeled or otherwise, may be roasted in fire and eaten when soft. This method is common among children.

b) Dry cassava.

Fresh cassava is peeled, chipped and sun dried for 3-5 days. The dry chips are mixed with dry sorghum or finger millet grains in ratios varying from 1:1 to 3:1 (cassava:cereal grain or vice versa) and ground into composite flour. The flour is mixed with boiling water to make a local bread known as atapa, kwon, etc. The bread is then eaten with a sauce such as beans, meat or groundnut stew. Cassava porridge can also be made for children from the

composite flour. Fig 1.4 shows the food preference among farmers in northern and eastern Uganda.

Fig 1.4: Root crops in the food system.



The flours from dry cassava chips and from sorghum or finger millet or maize are mixed, with addition of water to a wet mash which is then fermented for seven to fourteen days before roasting over fire and sun drying. The dried stuff is brewed by addition of yeast flour from pregerminated finger or bulrush millet or sorghum and water and kept in a container for three or four days when it is ready for drinking (Ajono). Drinking straws with sieves attached at the bottom are used to suck the alcohol from the stuff after water is added. After brewing the above alcohol (Waragi) escaping from the stuff boiling over the fire is collected into bottles with the help of a rubber or copper tubing passing through cold water acting as a condenser (Enguli).

Dry cassava chips are being exported in small quantities to Belgium and other EEC countries. It is expected that the quantity of this export may increase tremendously in the near future. Commercial starch is manufactured by the Lira Factory about 200 miles north of Kampala. The starch is used to meet local demand and the surplus is exported to Europe, especially to West Germany.

#### CONSTRAINTS ON CASSAVA PRODUCTION

The production of cassava has been decreasing steadily since 1980. The crop faces a number of serious challenging problems, some of which (diseases and pests) threaten the very existence of the industry.

##### a) Pests

In Uganda, cassava is attacked by many insect and mite pests: the variegated grasshopper, Zonocerus variegatus, whiteflies, Bemisia tabaci, the vector of African Cassava Mosaic Disease; the stem mussel scale Pseudaulaspis spp., cassava mite complex comprising the tetranychid mites: Mononychellus tanajoa (Bondar), Tetranychus telaris L. = T. utricae (Koch), T. cinnabarinus (boisd) and Oligonychus gossypii (Zacher). Only M. tanajoa is of economic significance to cassava. It is widespread and is

considered the most destructive pest in Africa (Nyiira, 1982). This pest was first recorded seriously attacking cassava in Uganda (by Lyon, 1973). It later spread to other parts of Africa where it is the most destructive especially during dry season (Nyiira, 1978). In Uganda, the pest caused an estimated loss of US \$ 7 million in 1974. It is always most serious during dry season when it can cause heavy defoliations of the plants (Nyiira, 1972).

Severe infestations of CGM in Uganda were reported in 1972 (Lyon, 1973). This was the first outbreak in Africa. The pest was later shown to feed and breed on a range of Manihot species, M. esculenta, M. glaziovii, Meell. Arg. M. dichotoma Ule, M. piauhyensis Ule and M. carthagensis Muel Arg. (Nyiira 1978).

Nyiira (1982) evaluated yield losses in the field due to Mononychellus tanajoa at between 17-33%, depending on whether the cassava was planted on newly opened or continuously cultivated land; he also noted that cassava grown on poor soil in dry areas, or under heavy weed competition suffered increased damage and yield losses.

Annual population trends of CGM in Uganda have been found to be associated with absence of leaves on the plants rather than the presence or absence of rain, although generally the mite population is lower during the wet season (Nyiira 1982). The long term population trend, however, appears to be influenced by natural control systems, especially depredators (Nyiira, 1978), but Nyiira (1982) stated that the factors influencing long term population changes are not known.

i) Chemical control

Dimethoate, chlorobenzilate and keithane are effective against the mites but their use in Africa is not feasible because of their cost and their adverse effects on depredators of CGM, Oligota minuta Cam., Stethorus spp. and Typhlodromus sp. (Nyiira, 1982). The subsistence nature of the crops makes application of the acaricides uneconomical. The mites also have been found to develop resistance to the acaricide very quickly, resulting in population resurgences.

ii) Biological control

Nyiira (1981) investigated the use of Entomophthora thaxteriana (Petch) on CGM and reported the possibility of attaining 90% mortality 20 days after application. However, use of this fungus for biological control is impractical since it survives in the humid conditions which discourage mite population development. Oligota minuta was identified in Trinidad as having a potential in CGM control, it was released in Kenya in 1977 and 1983 but it has not established.

b) Poor agronomic practices and lack of adoption of new technologies by farmers

Although cassava has been cultivated in the country since the

beginning of the century, the practices followed by farmers are still poor and unimproved. For instance, hill side planting coupled with inadequate soil conservation practices contribute significantly to soil erosions and decrease in soil productivity. This affects cassava root yield significantly. Adequate ground cover is recommended to farmers on mountain slopes, but very few have adopted the practice. Recommended agronomic and cultural practices such as crop rotation, intercropping, use of green covers and mulches have been scarcely adopted by the farmers. Timely planting of the crop, especially cassava at the onset of the rain, is very rare among farmers. Timely weeding and weeding frequencies are not strictly followed by farmers. The first weeding is normally done late and subsequent weeding may not be done at all. This results in poor yield.

c) Post harvest handling

Fresh cassava is a very bulky and perishable product. Because of its high water content, it deteriorates very quickly due to enzymatic action followed by mold and bacterial attack. This always leads into great tuber losses if quick action to stop it is not taken immediately. Even in drying of chips and slices, great losses can result during humid weather conditions.

d) Disease

Bacterial diseases reported in Uganda include cassava bacterial blight, Xanthomonas campestris (Pammel) Dowson pv. manihotis Berthot & Bondar) Dye (CBB); bacterial loaf spots, Xanthomonas campestris pv. cassava (Weihe & Dowson) Maraitte & Weyns; and stem gall possibly due to Agrobacterium tumefaciens (E.F. Smith & Townsend Conn).

Among virus diseases, only Cassava Mosaic Disease (CMD) occurs in the country. Cases of brown streak disease were reported in 1948 (Jameson, 1970), but the eradication campaign of 1948-49 seems to have eliminated it (Emechebe, 1976).

Fungal diseases include brown leaf spot, Cercosporidium henningsii (Ialesch). Deighton; white leaf spot, Phaeoramularia manihotis, Cercospora caribaca cif blight leafspot, Cercospora vicosae Muller and Chupp.

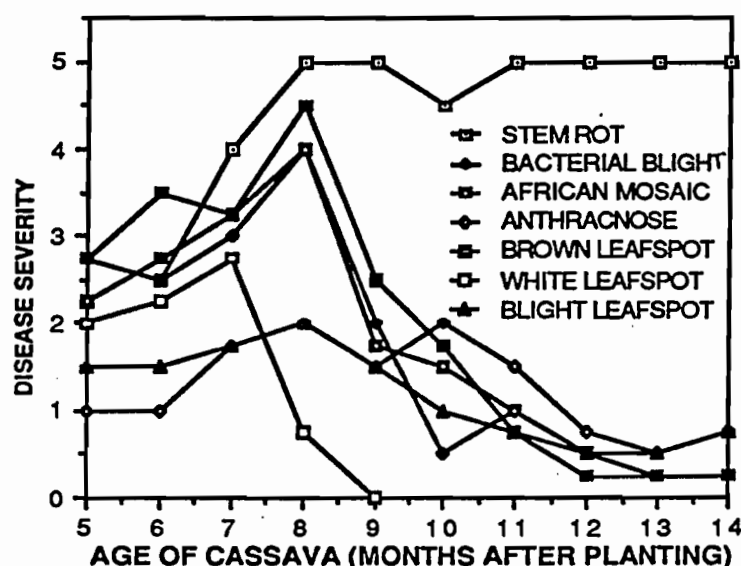
Anthraxnose, Collectotrichum gloeosporioides f. sp. manihotis Hem. (Penz.) sacc = Glomerella manihotis chev.

Botrydiplodia stem rot, Botrydiplodia theobromae Pat.; vascular wilt, Verticillium dahliae Klebahn; Phytophthora drechsleri Tucker; white root rot, Rigidoporus lignosus (klotzch) Imazoki; and Rosellinia root rot, Rosellinia necatrix Prill. (Otim Nape, 1984).

CBB and CMD and CA brown, blight and white leafspots Botrydiplodia stem rot diseases are the most serious and the most researched diseases of cassava in the country. The epidemic development of these diseases on cassava is shown in fig.2. The severity of cassava brown and blight leafspots, bacterial blight, anthracnose and the African Cassava Mosaic Disease generally

increased from onset of observation at five months, after planting. The crop reached maximum peak disease severity level at eight months after planting and thereafter declined steadily until they reached their lowest levels at thirteen months after planting. After this period, the severities of the diseases remained low until harvest, although the severities of blight leafspot and anthracnose tended to increase slightly after that period. Cassava white leafspot severity, however, increased and reached a peak at eight months after planting the crop and remained generally constant thereafter (Otim Nape, 1986).

Fig. 2: Developments of diseases of cassava with time and age of the crop.



#### THE AFRICAN CASSAVA MOSAIC DISEASE (CMD): INTRODUCTION, SPREAD AND IMPORTANCE IN UGANDA

Cassava Mosaic Disease (CMD), first recorded in East Africa by Warburg in 1938 at the coast (Storey and Nichols, 1938) was first mentioned in Uganda as "curly leaf" by Hall (1928) and as "mosaic" by Martin (1928). Cassava cultivation and mosaic disease incidence increased greatly during the outbreak of the tropical migratory locust *Locusta migratoria migratoriodes* (S and F) and during the droughts of 1939 and 1941 when lack of shipping during war time made it imperative to conserve local food supplies. These encouraged mosaic because, at those occasions, people probably dug and ate the best of their crops and used mosaic infected plants for propagation (Jameson, 1964). The disease occurs throughout the country and is most severe in Eastern, Northern and North-western regions where the poor soil conditions, the abundant and active vector populations and intensive cassava cultivation favours the disease dissemination and manifestation.



Active studies were initiated on symptoms (Hall, 1928 and Martin, 1928), transmission by planting infected stem cuttings and by the whitefly vector Bemisia tabaci (Storey and Nichols, 1938; Russel, 1958) and on yield loss due to planting CMD diseased cuttings. Field losses ranged from 87% to 55% (Hall, 1928). At that time all popular local varieties were highly susceptible to the disease. The use of resistant varieties by farmers was thought as the only solution to the problem and a breeding programme was started at the then East African Agricultural Research Institute at Amani, Tanganyika (now Tanzania) in 1933.

#### Testing for resistance

The breeding programme at Amani produced a range of resistant cassava lines (Storey, 1936; Nichols, 1947 and Jennings, 1957). Six varieties were imported into Uganda from West Indies (Hall, 1928) and many others from Amani in 1934 and subsequent years and were evaluated at Bukalasa, Serere and at three other locations in Eastern region. The trial was repeated in 1941 at Serere Research Station. Seventy local and imported clones were used. The trial was repeated in April 1943 and all clones with percentage infection of 21% or more were discarded. Sixteen most resistant clones were selected for yield evaluation.

#### Testing CMD resistant clone for yield

In June 1943, the 16 most CMD resistant clones were planted in a four times replicated yield trial and harvested in six lots at two monthly intervals starting March 1944 and ending January 1945. The highest yields were obtained at 17 months after planting. Aipin Valenca yielded highest at all locations and in all subsequent trials at Serere and elsewhere. The clones bred at Amani spread less well adapted to Uganda conditions. Of the many Amani clones evaluated, only two clones (4023-4, a Kru x Binti Misi cross and 4440-6 raised at Serere from Amani seed) yielded as well as Aipin Valenca. Later importations from Amani were restricted to seeds because of the need to exploit under local conditions the greater variability provided by seed and secondly to reduce the risk of introducing brown streak disease.

Varieties Aipin Valenca and F279 were released to farmers for growing and were gazzetted Bukalasa 8 and Bukalasa 11 respectively. There followed a series of propagation and distribution.

#### PROPAGATION AND DISTRIBUTION OF CMD RESISTANT LINES

The food shortage resulting from drought and bad harvests in 1932-34 and the limitation of CD on yield of cassava necessitated a quick programme on multiplication and distribution to farmers, Eastern Uganda, of resistant cassava varieties. The objectives of the programmes were the following:

- a) to try to reduce the rate of CMD infection and establish a crop with short term yield prospects;
- b) to follow later with elite material propagated on an "advancing front" plan.

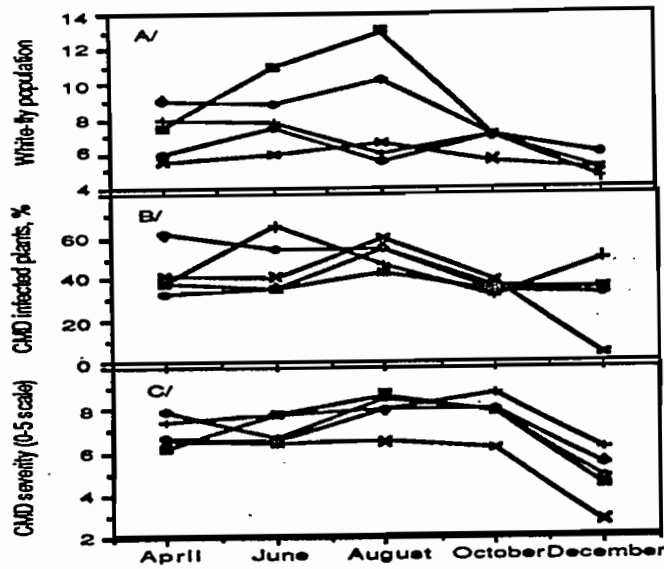
Variety Binti Minti, the only resistant variety available in quantity and in healthy conditions within a radius of 100 miles, was assembled, distributed and planted in most locations in Teso district, Eastern Uganda.

Meanwhile, the propagation of the most promising clones at Serere was giving a tenfold multiplication at intervals of six months and enabled observation plots of 20 clones to be planted in 1944 in all cassava growing districts. The plots contained 16 clones included in the yield trial plus F100, Mandesi, Mwarabu and Namazina. Due to the outbreak of the brown streak disease, all the Malindi varieties were eradicated at all centres, and at centres where the disease was well established, all plants were uprooted and replaced with clean material. At centres with only slight infection, diseased plants were rogued out, while at Serere where the roguing was not effective, all plants were uprooted and a six months quarantine period imposed in 1948-49 and clean material was supplied from Kawanda Research Station, central region. Of the twenty clones tested in observation plots, nine were selected for further multiplication and distribution and in Teso district Binti Misi, 3744E, Aipin Valenca, Msitu and F279, Eru and F100, were used.

A primary nucleus of mosaic-free material was maintained at Serere, and at the second stage of multiplication the resources at Serere were supplemented by the use of a nearby prison farm maintained by the local government. In some areas the programme involved complete uprooting of cassava in the area (e.g. Kyere 700 acres) and substituting it with new and improved resistant materials. The rapidity of the multiplication permitted a split in the advancing front plan, and another administrative unit north of the district (Achwa) was supplied with new materials. In both cases all plants with CMD plants were uprooted. By 1951 Teso district had enough planting materials to supply all needs. The district Council consolidated the result by requiring (by order) that all CMD cassava plants be pulled out.

These efforts managed to control CMD in the eastern region for decades. However, recently, CMD has become a serious problem again. Possibilities are that farmers have relaxed their observance of roguing out infected plants and intensified indiscriminate planting of CMD infected stems. There is also a possibility of breakdown in resistance by the common cassava varieties as a result of increasing buildup of CMD agent in the plant over time and of the stress offered to the plant as a result of multiple infection by other diseases and pests, notably the green spider mite, cassava bacterial blight, anthracnose, brown blight and white leafspots. Recent studies include the effect of time of planting and spacing, the effect of spacing and numbers of cassava shoots per cutting, and the effect of interplanting legumes, cereals with cassava on the African Mosaic Disease and its vector the whitefly. It was also important to understand the effect of such practices on the plant microclimate which in turn could affect the disease and the vector activity. The whitefly Bemisia spp., also common on tomatoes and egg plants, were present on cassava throughout the year (Fig. 3.1).

Fig. 3.1: The relationship between: a) Vector population, b) CMD incidence, c) CMD severity, at different planting dates and spacings.



Figures 3.1, 3.2 and 3.3 show the effect of such practices on the disease, its vector and the plant microclimate (canopy, temperature and relative humidity). Generally the CMD vector (whitefly) population, the incidence and severity of the disease significantly increased with further delay in planting cassava, reaching their highest levels on cassava planted in August, after which they decreased with further delay in planting the crop. Similarly, in both experiments, the vector (whitefly) population on cassava and the CMD severity significantly increased with decrease in cassava spacing. CMD incidence (% CMD infected plants) was not significantly affected by spacing, although it tended to increase with increase in spacing in both experiments.

Fig. 3.2: The effect of spacing and number of shoots on: a) CMD vector population, b) CMD incidence and c) CMD severity.

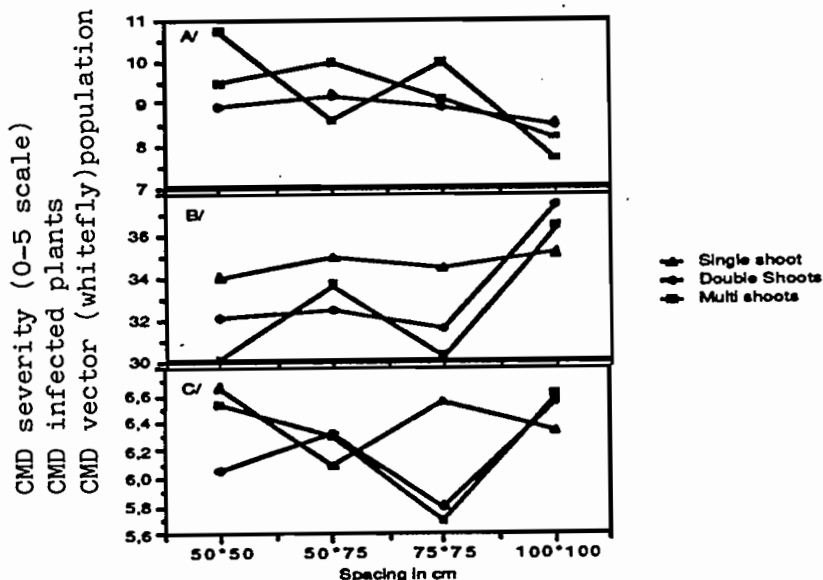
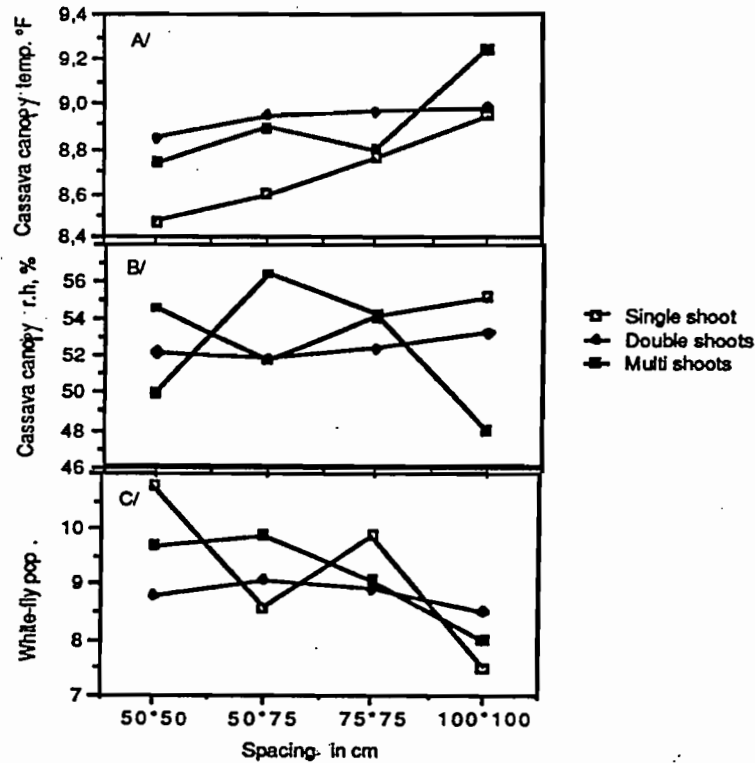


Fig. 3.3: The relationship between cassava and a) canopy temperature, b) relative humidity and c) vector population at different spacings and shoots.



The recent reappearance of CMD as a serious disease of cassava reactivated serious research efforts against the disease. The Uganda National Root Crops Improvement Programme (UNRCIMP) undertakes all research into cassava improvement and utilization. Among its major objectives are to obtain varieties which are high yielding, resistant to mosaic, other pests and diseases, while maintaining good consumer quality and also to understand the effect of agronomic and cultural practices on CMD and its vector, the whitefly.

Thousands of cassava materials (true seeds and tissue culture) of improved varieties have been received from IITA, more true seeds have also been obtained through our hybridization programme since 1981. They have undergone a series of evaluations for adaptability and desirable qualities. A number of lines and varieties have been selected for resistance to the CMD and have been advanced to uniform yield and on farm trials. It is expected that a number of resistant clones will be released to farmers for growing in the very near future.

Local germplasm collection mission was conducted throughout the

country in 1986. A total of 315 local cassava clones were collected. These are being evaluated for resistance to CMD, other diseases and pests and for desirable agronomic and consumer characteristics.

The agriculture practices and the vector (whitefly) population were, however, very negatively significantly correlated ( $r=0.65^{***}$ ) with temperature. Canopy temperature was neither significantly affected by altering cassava planting time nor by the number of cassava shoots per cutting. Similarly, canopy relative humidity was not significantly affected by any of the treatments in the two experiments.

Experiments currently going on are aimed at developing and screening cassava lines and varieties for resistance to the disease; assessing yield loss and economic importance of the disease; further understanding of the epidemiology of the disease and the effect of intercropping on the incidence and severity of CMD and its vector.

Local programmes, especially the Agricultural Extension Service, consider the African Cassava Mosaic as the most serious cassava disease in the country. Consequently, they are involved in a campaign for farmers to plant CMD-free cuttings of recommended CMD-resistant cassava varieties and to rogue any plants which subsequently show symptoms of the disease. Early planting at a spacing of 1m x 1m and leaving two or three shoots per cutting is also recommended. As a result, some farmers currently select cuttings from healthy plants. The crop is also seed-propagated but this method cannot and is not being used by farmers. True seeds are being used by research scientists for breeding purposes.

Cassava grows everywhere in the country and it is possible to plant cassava from April to October and sometimes up to mid-November, depending on the level of soil moisture. Uganda has great diversity of local cassava clones, over 286 different local cassava clones recently (Table 1), the widest diversity being in areas surrounding Lake Victoria (i.e. Bombo, Kampala and Masaka districts).

Table 2. Distribution of local cassava clones in some parts of Uganda.

Districts	Cassava clones
Soroti	11
Mbale	1
Tororo	19
Jinja	21
Bombo	44
Kampala	47
Masaka	57
Mbarara	37
Kabale	5
Fort Portal	26
Hoima	18

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In these areas, farmers prefer sweet or less bitter varieties. In north western region, farmers prefer bitter varieties. They eliminate the bitterness through fermentation before eating.

The Uganda cassava germplasm collection numbers over 350 clones. These include the African Cassava Mosaic Disease resistant clones introduced during 1940s and 1950s from Amani, Tanzania and from IITA Ibadan, Nigeria since 1981.

The IITA materials are still at the experimental stage.

Few of the Amani clones (Bukalasa 8 = Aipin Valenca and Bukalasa 11 = F279) were widely grown by farmers during the 1950s and 1960s and early 1970s. With the appearance of new and more high yielding varieties such as Ebwanateraka in the late 1970s and 1980s, most farmers abandoned Bukalasa 8 and 11 for various reasons in preference for the new varieties. Bukalasa 8 were released to farmers because of its resistance to CMD. Recent evaluations and findings indicate that it is the most susceptible to the disease. Its tubers have good consumer characteristics. Bukalasa 11 is still resistant to CMD but its great susceptibility to the cassava green spider mite and the poor storability of its tubers in the ground once matured, made it unacceptable to most farmers.

#### FUTURE PLANS FOR AFRICAN MOSAIC DISEASE IN THE COUNTRY

The Uganda Government through the Uganda National Root Crops Improvement Programme (UNRCIP) would like to see research into African Mosaic Disease reinforced and intensified. A deeper study of the disease in the country followed by an intensive control campaign are necessary. Studies into the possibility of the existence in the country of CMV strains; intensified breeding and selection for CMD resistant varieties which are widely adapted in the country; a detailed assessment of cassava yield and economic losses due to CMD infection of cassava in all ecological regions of the country are planned. A deeper study of the epidemiology of CMD; the effect of environmental factors (climatic and other biotic factors) are also planned. A programme to provide farmers with mosaic-free planting materials of high yielding varieties and to encourage them to adopt other newly developed technologies for mosaic control and other technology for increased cassava production and to train field extension staff and farmers in improved techniques in mosaic control and root crop production in general will be initiated and implemented.

Consequently, the government of Uganda would welcome any institutes, organizations and/or donors willing to collaborate/cooperate and/or fund the programme on the African Cassava Mosaic control. The government is currently looking around for any organization willing to help set up a programme for the control of the African Cassava Mosaic Disease in the country.

#### ACKNOWLEDGEMENTS

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**SESSION F**

**CONTROL STRATEGIES**

Chairman  
Dr. HARRISON  
Rapporteurs  
Dr. FISHPOOL  
Dr. M'BAYE

SESSION: CONTROL STRATEGIES

HARRISON, B.D., Chairman

FISHPOOL, L.N.C., Rapporteur

M'BAYE, A.A., Rapporteur

HARRISON, B.D.:

The session this afternoon differs from previous sessions in the sense that it is a discussion session. I shall invite a few people to introduce selected topics and we will then discuss each of these topics. I propose that we first consider the different possible ways of controlling cassava mosaic and that we then go on to define what research is still needed, if any, and how we might get that research done. Then we will discuss organizational requirements, by which I mean the arrangements that may be needed in each country, or in groups of countries, to implement the various control measures that we may decide are worthwhile.

We will therefore first discuss control strategies, and I propose that we do this, to begin with at least, on a regional basis. We will start by considering the East African situation, dealing first with the possibilities for the sanitation approach and then with the value of breeding for virus resistance. This will enable us to see which of these approaches has the greater merit, or whether they should be combined, in East Africa. Then we will go on to deal with West and Central Africa, which is a much more heterogeneous region, and to consider whether the principles that apply to East Africa are also relevant in some parts of West and Central Africa, and whether other areas of West and Central Africa need a different approach. To start this assessment, I am asking Dr. Ken Bock to lead the discussion on the sanitation approach to mosaic control in East African countries.

BOCK, K. :

The first point I wish to make is that it is not possible to divorce resistance from sanitation. I shall therefore talk about the use of sanitation in regard to resistant, or moderately resistant, varieties.

Secondly, I would like to define the region under consideration. We are in quite a strong position in Kenya, because a good many results are already available for the sanitation approach. All the evidence suggests that they would also apply to Tanzania. This

approach certainly has worked also in Uganda, and we congratulate Dr Otim-Nape on his new data; and we know, I think, that the system works in Malawi. That includes quite a chunk of Africa: Kenya, Uganda, Tanzania, Malawi, and I think that what I am going to say will hold, with reasonable certainty, for that region.

We have not yet described the results that John Guthrie and I obtained in Kenya. In plots with a central core of infected plants, surrounded by healthy, mosaic-free plants of the moderately resistant cultivar 46106-27, the greatest incidence of infection we recorded over a twelve-months period was under 10%.

Turning now to results which you had only a short time to see yesterday, we also conducted 26 separate experiments over a period of three years to assess reinfection rates under all sorts of ecological conditions. In 19 of these 26 trials there was no reinfection of resistant varieties and in another 6 the total reinfection, averaged for the 3 resistant varieties, was less than 5%. I think the figures speak for themselves, and they point very strongly to the fact that if you have good, resistant varieties that are acceptable to farmers, the solution to African Cassava Mosaic Disease is already with us in Eastern Africa. The prerequisites for success are the establishment of mosaic-free multiplication schemes and the education of farmers through the extension service, both of which are eminently possible. This certainly happens in Malawi, and if it happens in Malawi, it can happen anywhere else in Africa.

To summarise, the salient points are the availability of resistant varieties that are acceptable to farmers, the establishment of effective multiplication schemes, and the education of small farmers by the extension service.

HARRISON, B.D.:

Do you see the need for any further research to make this sanitation approach more effective?

BOCK, K.:

I think that we only scratched the surface in our work with local Kenyan varieties and that the greatest need at the present moment in eastern Africa is a sound agronomic approach to control.

The virologists must figure in this, as advisers, to ensure that virus-free stocks of the more popular cassava varieties in eastern Africa are produced. This is a comparatively easy task. These varieties should then be subjected to tests, to see what their rate of reinfection is. I think that we have to accept the fact that there are many moderately resistant varieties such as Kibandameno, that the farming community in eastern Kenya will not give up growing and similarly, in western Kenya, there are two or three moderately resistant varieties which are very popular and could be maintained indefinitely in a virus-free state in a sanitation scheme.

More work needs to be done on these casava varieties but, as I said, I think that the main need is for an agronomic thrust that would enable large quantities of healthy stocks to be multiplied, using existing knowledge, keeping close checks on the incidence of virus disease and removing any infected plants.

HARRISON, B.D.:

Before we discuss this point, I would like to hear what Dr Jennings thinks could be contributed by plant breeders.

JENNINGS, D.L.:

There are some points I should like to emphasise. If I was asked the question "Can resistance control mosaic disease using minimum sanitation methods?", I would say "Yes, with two provisos". The first is that the resistance should be of an adequate level. When a resistant variety is released, it should be produced in a virus-free state and in reasonably substantial quantities. The point I am trying to make is that, when a resistant variety is first introduced, you can expect a proportion of plants to become diseased, due to contamination from sources outside the crop, but I think it is wrong to assume that this proportion would necessarily increase progressively. What will happen is that as the area devoted to the resistant variety increases, so the inoculation pressure becomes less, because there will be fewer virus sources than before. I would expect the number of new infections eventually to go right down and approach zero. So, the answer is: "Yes, resistance alone with minimum sanitation can control mosaic in some circumstances". And I am not discouraged by the comments of Dr Otim-Nape, who says that varieties 37244E, Aipin Valenca and F279 became infected after they had been multiplied, simply because sanitation procedures had been relaxed. I say that those varieties are not resistant enough. 37244E has only moderate resistance, not quite as great as that of the first backcross material. Aipin Valenca has vector resistance but, in our trials, became infected rapidly, I suspect because whiteflies were moving on to it from contaminated material of other varieties and infecting the plants. I wonder whether vector resistance is comparable to virus resistance in this respect. Vector resistance will stop spread within plantings of Aipin Valenca and its like, but will it stop spread from outside virus sources into Aipin Valenca? The last variety you mentioned, F279, is again only moderately virus resistant. So, what I am saying is that the levels of resistance in these three varieties were not adequate.

My second proviso is that there must be enough acceptable varieties. Dr. Bock mentioned that, in Kenya, although the resistant material available has its merits, the farmers still like Kibandameno. So, we must have enough resistant varieties to supply all needs, otherwise there will be constant bombardment with virus-carrying whiteflies coming from infected plants of susceptible varieties, assuming there is no sanitation. Now breeding can put this right in due course. It is not a short-term solution, it will take perhaps about 10 years, but do not think

we should wait 10 years. We should use sanitation in the short term and resistant varieties later. So, if I was starting a breeding programme in Kenya, I would certainly breed for virus resistance, to reduce the amount of roguing that will otherwise be needed.

HARRISON, B.D.:

It is clear that the supporters of the sanitation approach for East Africa think they could do the whole job with existing varieties, whereas the breeders think they could do the whole job too. There is perhaps a difference in time scale. The conclusion that Dr Jennings ended with, that sanitation is necessary but might be replaced, either in part or totally later on, seems to be a possible formula.

Other people here may now like to make further comments, in regard to their own countries, on these two approaches.

ROBERTSON, I.A.D.:

Could I just make one comment? The breeders will probably find material that is very useful in eastern Africa, if they examine the existing germplasm collection, which I have been helping to maintain. Several accessions seem resistant to mosaic virus and some of these have root quality equal to Kibandameno, and are acceptable to the local farmers. This is the opinion of the local people who helped me do the harvest. So, rapid progress could probably be realized quite easily.

TRESH, M.:

I would be interested if we could get a comment about Uganda from Dr Otim-Nape, because I get the impression that there could be differences between the East African countries, in that Malawi and perhaps certain areas in Kenya are not very intensive cassava growing areas, whereas perhaps Uganda, or at least parts of Uganda, are. Could he also give an idea of the scale of the undertaking of the sanitation scheme in Uganda, and the degree of isolation that was necessary for the multiplication plots there?

HARRISON, B.D.:

Mr Otim-Nape, what is the scale of operations at your scheme at the moment?

OTIM-NAPE, G.W.:

In the first programme which I described, which operated during the early 1940s and 1950s, the area covered involved about two districts. Now we are covering one district, because our programme has just started, but we plan to cover the whole country eventually, as we get experience. That is why, in my

presentation, you saw that there were regional multiplication centres. These centres are to be located in various corners of the country. From the regional centres, we expect that material will flow naturally to district multiplication centres, and then to farmers. So, initially, we are covering one district, but our final aim is to cover the whole country. Regarding isolation, we have found that it may not always be necessary, if the roguing programme is rigorous; if you rogue frequently, it may not be very necessary to isolate completely the multiplication blocks, but where you intend to let the interval between roguings increase, you may need some degree of isolation.

HARRISON, B.D.:

What isolation distance is needed than?

OTIM-NAPE, G.W.:

We have succeeded in some cases with about 200 metres away from infected material, and I think this distance has worked well.

SAUTI, R.F.N.:

The story in Malawi is that after realizing the problems caused by African Cassava Mosaic Disease, and after getting information that sanitation could reduce the incidence of this disease, we embarked on the sanitation programme in collaboration with the extension staff. So we have established eight development programmes in the country, to have places where cassava multiplication programmes are initiated. In each area, we had to find out the most popular variety which was accepted by the farmers. By using various roguing systems, we end up with good material which is distributed to the farmers. We have several multiplication plots, for example in Salima for the Salima ADD. So the system is working.

But the main problem we have faced is the lack of training of the farmers and, as I said yesterday, most of the farmers have yet to be educated for roguing and so on. Otherwise, the system promises to help decrease the incidence of African Cassava Mosaic Disease, if not to eliminate it.

HARRISON, B.D.:

What scale are you working on? How many hectares of healthy material do you have in the country as a whole?

SAUTI, R.F.N.:

The scale varies from one ADD to another. For example in Salima ADD1 we have about 15 hectares, in Gapu which is in Cassandema, we have about 5 hectares of multiplication plots, so too at Gwandesesi. We have also to control cassava mealy-bug, and this is

a part of our programme. Cassava is now taken quite seriously in Malawi, and is of course the second food crop in the country.

HARRISON, B.D.:

The reason I ask these questions about areas of healthy cassava that are being grown in different countries is that, to make a real impact, you must have a large area of virus-free material producing stocks that are available to farmers. That needs a big organization. So, it is interesting to hear the progress you are making in those directions. With about 25 hectares, you already have a substantial amount of virus-free stock. How much do you think would be necessary in your country to supply the needs of farmers?

SAUTI, R,F,N,:

Just before coming, I had a discussion about this. One problem is that our planting material which is free from cassava mosaic disease has been sold to the farmers in small amounts, which is about as effective as a drop of ink in the ocean. So now we are organizing a system where selected farmers will be given virus-free material to plant the whole of their fields, which in turn will become a source of clean planting material for other farmers in the same area.

I think it is important not to aim to multiply all the virus-free material at one place, which would be the single source for all the farmers in a given area or in a country. It is better to develop a network containing many multiplication units, which may be within the same area, or may be scattered, but are relatively close to the farmers who wish to collect planting material. The officer responsible for a group of multiplication units also should make visits before the virus-free material is planted to give advice on what needs to be done.

This system recognises that most farmers are not willing to travel long distances to collect virus-free material, and ensures that the material is available at many places and can easily be taken to the farmer, who may not have the transport to go and collect it himself. We also consider the training aspect to be very important. You have to train the person who will be looking after the virus-free material in a given area. You also have to train the farmer, who has to appreciate the value of the healthy material, and to understand what benefits it will bring, so that he will continue to look after it well. I think this is an important aspect that must not be overlooked.

HARRISON, B.D.:

Dr. Perennec, have you some particular thoughts you would like to offer us in relation to running a sanitation scheme for cassava, based on your experience with potatoes?

PERENNEC, P.:

I would only like to make a comment. 60 years ago, just after World War I, virus diseases of potato were as severe as those of cassava nowadays. Two solutions to solve this problem were possible: to breed resistant varieties, or to operate a sanitation scheme. I think that if we had devoted ourselves only to breeding resistant varieties, we would still be in a very bad situation, because this approach would have resulted in little progress. However, thanks to adopting the sanitation approach, we have managed over a period of several years to decrease progressively the inoculation pressure. As a consequence, potato sanitation schemes are now successful in most European countries. Nevertheless, I think that trying to run a sanitation scheme in an environment where the inoculation pressure is very high will raise problems in the coming years. This will cause much disappointment, but we must not give it up. I emphasised in my report that sanitation is a good thing to try, but that if it can make use of resistant varieties, it might be much more effective.

HARRISON, B.D.:

Those are very relevant comments. I am one who works in a country that makes much of its living from being a sanitation area, for potato growing, and I agree with you, but we are now also moving towards getting virus-resistant varieties of potato, and we can think of that in terms of cassava as well.

MSABAHA, M.:

From our experience in specific parts of Tanzania, sanitation does work, but is subject to some limitations. To convince the small farmers of its value takes time, particularly in such a vast country. So, we thought we would go ahead with the sanitation procedures by involving a group of farmers, who start a multiplication plot, and by educating them why infected plants have to be rogued out. This is a slow process.

As a complement to this, we encourage them to plant their cassava on time. We also started to investigate the possibility of using certain cropping systems, such as inter-cropping, to decrease inoculation pressure, and certainly for the long term we would like to create a breeding programme. But I am really convinced that sanitation together with some resistance can be used for the time being. Later, I am sure, we shall come to have new material, immune to mosaic.

HARRISON, B.D.:

You have raised a new point on planting time. If one is practising a sanitation scheme, its efficiency can perhaps be increased by choosing to grow the cassava at a time when it is less likely to become infected. We have already heard the results of experiments in the Ivory Coast in which virus spread was much



decreased when the crop was planted at a certain time of the year.

In addition, I have the impression that the amount of spread of virus within fields is much less than the amount of infection resulting from virus-carrying whiteflies coming into the crop, from outside. Can we follow up these two points by suggesting a time of planting which would be the most likely to give healthy crops, and by emphasizing that it is not a good practice to plant healthy crops downwind from infected sources, be they larger or smaller sources. These are two fairly simple measures that might be adopted.

ROBERTSON, I.A.D.:

Could I just point out that in the eastern African situation, we have relatively little choice of planting date. Malawi has a six months wet season and a six months dry season. In Kenya we have long rains and short rains, but cassava planted for the short rains is very much at risk. I do not think that any of this group of countries has more than about two months in the year when they can expect to plant cassava and get successful growth.

DAHNIYA, M.:

We have a problem now in Sierra Leone, since the advent of new epidemics of mealy-bugs and mites. The recommended planting date is the beginning of the rainy season to avoid damage by cassava mites and mealy-bugs but whiteflies multiply during the rains, so the planting season which one would normally recommend for mealy-bugs and mites would become the beginning of the dry season. This is also the time when other plants are under severe stress and numbers of mealy-bugs and mites might be increasing. So, I really do not think that changing the planting date in our own situation, where we have a 6 months rainy season, and 6 months dry season, is going to help the control of Cassava Mosaic Disease. I think that altering the planting date would not work well in places where you have well defined rainy and dry seasons.

HARRISON, B.D.:

You have raised an extra point here. This is that in applying methods that help to control cassava mosaic, you may make other problems worse than they otherwise would be. In any scheme that is used, there has to be some kind of compromise to devise something that is reasonably good for minimising damage by a range of pests and diseases, if this is possible. Perhaps it was stupid to suggest that one could alter the planting date as a control measure, but this may possibly be of value in some areas.

THRESH, M.

In relation to distribution of healthy planting material, this could be combined with the distribution of natural enemies, if

the mealy-bugs and mites continue to cause problems. On another point, we have not discussed whether the farmer should be charged for healthy material, or whether it should be free. There are some fairly big logistic problems in producing healthy material, and it is a very expensive procedure. One other thing that we have not discussed, in relation to this distribution, is the likely duration of a stock. It is expected that the growers will be able to maintain a stock free from infection for 10 to 20 years by roguing, or is it going to be necessary for them to obtain fresh healthy material at fairly frequent intervals? I would also make the observation that if the same amount of effort was put into breeding resistant varieties as is being put into sanitation, the alternative solution (resistant varieties) might be possible.

BOCK, K.:

When we planted mosaic-free resistant material in Kenya, we carried this material on, very successfully, for 3 years. The annual rate of loss of plants was mostly under 5% and often under 2%, so you are removing 2 to 3 plants per hundred, which leaves more than enough planting material for the subsequent crops. Unfortunately, we did not carry the material on for 4 or 6 years, but I cannot see a problem in doing this if the scheme is operated properly. The farmers could well look after their own needs for over a decade, maybe longer.

SAUTI, R.F.N.:

I want to describe a study which was done in Malawi. In 1980, I visited an island in Lake Malawi that was isolated from the rest of the major cassava growing areas. My first impression was that almost 90% of the cassava on that island had Cassava Mosaic Disease. When I returned to the mainland, I discussed the possibility of establishing a virus-free multiplication plot on that island, and of educating the farmers. This plan was accepted. Then I told farmers on the island the effects of Cassava Mosaic Disease, in terms of yield losses. In that year, we established multiplication plots at two sites on the island. The following year (1981), the healthy material was distributed to the farmers, free of charge. When I went back two years later, I discovered that the percentage of mosaic disease on the island had decreased to about 1%.

HARRISON, B.D.:

Were the crops rogued or not?

SAUTI, R.F.N.:

They were rogued, because most of the farmers had been taught how to select healthy planting material. The percentage of mosaic on the island is still less than 1%. So the system can work, if it is done properly.

HARRISON, B.D.:

Dr. Malathi, is a sanitation scheme with your moderately resistant varieties a good approach in India?

MALATHI, V.G.:

Phytopathological methods work very well in our "outreach" programmes. The point about these programmes is that as long as you maintain a presence in an area, the farmers will always come to you for your techniques and advice, and we can definitely retain healthy stocks in fields for more than 5 years. But the moment you get out of this programme, health is not maintained. So I feel we have to pay more attention to how to run these programmes successfully and how to create permanent disease awareness among the farmers.

ROBERTSON, I.A.D.:

Taking the East African group of countries again, the logistics of distributing this material should not be excessively difficult, because all the countries in the East African group have had a long experience of introducing new cotton varieties. The cotton people have a system of multiplication and distribution. So, the kind of work that is needed has been done before, in all the East African countries, and it could be modified to deal with Cassava.

HARRISON, B.D.:

In this session, we have heard a good deal about the East African situation and although there are further points that could be discussed, I would now like to move on to consider the situation in West and Central Africa, which is a much more diverse region in some ways. Would Dr. Rossel say a few words about the breeding approach to controlling cassava mosaic?

ROSSEL, H.:

On the issue of breeding, I should now take the opportunity to point out that IITA does not have a Cassava Mosaic Virus resistance breeding programme as such. It has a cassava improvement programme, and many different constraints are taken into consideration. The importance of each constraint was carefully assessed, prior to and during the early stages of the breeding programme, and is constantly reassessed, as the need arises. I must also point out that IITA does not aim to produce finished varieties. It provides germplasm for the benefit of national programmes, either to use in their own breeding programmes, or to select from. Only in this way can national programmes come up with varieties that they think are suitable for their environment and their people. This has to be said before we discuss whether IITA is, contrary to its own aims,

resistance breeding and so attempting to remove a disease which has, of course, been identified as a major yield constraint.

I agree with Dr. Jennings that IITA tries to focus on the long term. You cannot expect a breeding programme to come up with resistant material quickly, particularly with a crop like cassava, where multiplication of stocks, particularly in the African situation may take several years. However, we should definitely continue to approach the problem, in a way that is based on ideas that can be implemented when the time comes.

I would strongly urge that, for each particular situation, it is determined whether there is scope for controlling the disease and not only whether there is scope for sanitation schemes. For instance, I can say with regard to Nigeria that there is no scope whatsoever for sanitation schemes, because in the areas where cassava is grown, and where it is a major crop, the infection pressure is so high that this approach would not achieve anything. Of course, the question still remains as to whether, with resistant varieties, it would be worth removing the 5% or 10% of infected plants, whether this would control virus spread and whether it would lead to an increase of marketable yield.

I consider that much more regional research is required, to ascertain whether the sanitation approach works. National programmes might consider this a feasible short-term solution, as far as their particular environment is concerned. I am saying this because, even in Nigeria, there are areas where the disease could be removed from the crop. But I should also say that, in those areas, cassava is not the major food crop, being only perhaps third in importance.

However, sanitation is a complex issue with many social and economic aspects, and all these things should be kept in mind. In general, I should say IITA is not against it, where it is effective, but do not forget that resistant materials are continually being developed and have many other potential benefits as well as resistance to cassava mosaic.

HARRISON, B.D.:

It is my impression that the kinds of resistance that have been introduced in the breeding programme at IITA are fairly durable. That is to say that they do not break down. Of course, if the resistance is of the recessive type, this is perhaps to be expected. Is my impression correct?

ROSSEL, H.:

As far as mosaic resistance is concerned, it has been effective for several decades, and there is no indication of any strong differential reaction in different geographical areas. We can conclude that the resistance is adequate, but not that it is unlikely to break down.

HARRISON, B.D.:

So, although it might break down in certain areas if a new virus strain appeared here, you have no evidence of this?

ROSSEL, H.:

Let me say there is no indication for virus isolates giving strong differential reactions. We have a definite indication of a differential reaction, but it is not in geographical terms alone; it is found only in a few specific cassava genotypes. For instance, out of 10 genotypes that are resistant at IITA, as Dr Mahungu will confirm, one is not at all resistant in Zaire. This is interesting, especially for scientists, but also to keep in mind. It is not important practically because only one in ten genotypes behaved in this way but it may be taken into account in the breeding strategy if such differences are expressed more strongly in far off countries, which is quite possible in spite of the fact that the sources of the resistance came originally from East Africa. It is quite possible that certain genotypes will become infected when grown in other countries. We must therefore ensure that new genotypes are tested carefully in the region where they are to be grown, before they are disseminated widely in those areas.

HARRISON, B.D.:

I think it is important to report any instance of breakdown in a supposedly resistant variety in a particular area.

I also wanted to ask you about the philosophy of the IITA programme. In the past, this has been to breed populations of plants instead of individual clones and I know that much material has been distributed to different countries as true seed. Clones can now be transported safely in tissue culture, making it possible to maintain combinations of genes which you have built up carefully over the years. So is there not a case for giving greater emphasis to clonal selection, rather than to family selection?

ROSSEL, H.:

Yes, I have emphasized that these two different breeding strategies are both subject to constant revision, and any new information available will help us to decide whether to go one way or the other. This differential reaction which I referred to is another point to be taken into account. It may even be that in other situations none of a limited number of genotypes sent for multiplication or testing may prove satisfactory. This is why the reassessment of strategies is an on-going process, based on actual findings and on testing done elsewhere.

HARRISON, B.D.:

Although some countries can do their own breeding, this is expensive, and many other countries do not have the facilities or money for their own breeding programme. These countries have to rely on the materials which people like IITA produce. For these people, I suspect that clones would be more valuable than seeds.

ROSSEL, H.:

It is not so much that the emphasis is on populations at IITA. Until four years ago, IITA was not allowed to distribute clonal material, because of the cassava mosaic issue. More recently, in conjunction with the Interafrican Phytosanitary Council, we were able to decide that we knew enough about the disease for vegetative material to be sent safely from one country to the other in Africa. That is how the programme is evolving and how it relates to clones.

HARRISON, B.D.:

Thank you for your helpful clarification. Let us hear now about possibilities for sanitation in other countries. I would like to have Dr. Fauquet's views.

FAUQUET, C.:

Most spread of African Cassava Mosaic Virus is from the forest or general environment to cassava. This means, in our opinion and according to our experiments, that the virus which infects cassava fields does not come in most part from the natural environment, but rather from other already infected cassava fields.

In the Ivory Coast, the areas of infected cassava as huge as compared to the healthy areas, which are almost non-existent. I do not know every West African country, through what I have seen in some of them and above all through the answers we have received to our questionnaire, I have the impression that this general situation prevails in this part of the continent.

We must consider the inoculation pressure of a given area. This depends both on properties of the vector and on the environment, and also on the plant material employed. We have represented differences in varietal susceptibility of cassava clones on the vertical scale. Horizontally, we have placed culture techniques such as sanitation, that might be used in each area. We always compared two regions: Toumodi, where virus spread is slow, and Adiopodoumé, where it is rapid. We think that, according to the local inoculum pressure which depends on the prevalence of infected cassava crops and on the local environment, sanitation schemes will be more successful or less successful.

I therefore think that sanitation schemes must be developed, but that we must use material with a degree of resistance likely to make them work. It is inappropriate to use sanitation methods in a cassava field if 90% of the plants must be rogued.

In my opinion, clones likely to permit success in sanitation schemes should be distributed. As this approach begins to have an impact, it may become possible, as for potato culture in Europe, to change the situation; i.e. to progress little by little to growing healthy cassava crops, at least far healthier than nowadays. In this way, we would reach the Toumodi kind of situation. It will then become possible to use clones which are more susceptible, but which have other advantage either in palatability or agronomically.

HARRISON, B.D.:

What sort of area of West or Central do you suppose might already be in the Toumodi kind of situation? I presume a large part, especially in the humid zone, is more like Adiopodoumé.

FAUQUET, C.:

What we have heard, particularly on Wednesday, when the important epidemiological studies made in Eastern Africa as well as in Western Africa (i.e. two completely different areas) were described does not allow us, in my opinion, to draw general conclusions nor clear guidelines. We thus cannot say that within a given geographical zone, on the basis of the environmental conditions, such as temperature, humidity and rainfall, sanitation schemes can be applied to such a variety in such an area: for instance an area of forest, with two rainy seasons and over a metre and a half of rainfall. This would only be speculation.

I think that before making such statements, multilocation trials should be established in other western or central African countries, so as to confirm or invalidate the impressions derived from the studies already made in East Africa or West Africa.

HARRISON, B.D.:

That is a sound point. Would anyone else like to put their point of view on these questions?

TETEVI, K.:

I think that we must be aware that cassava is not like corn or rice, in that the planting material of cassava is not bought. This implies that when improved and healthy cassava material is available for distribution, one must make enough effort to ensure a free distribution. Also, when a farmer is asked to come to a given place to pick up cassava cuttings, he must be convinced that this material corresponds exactly to what he wants: for example, sweet cassava suitable for making "foufou", with not too low

branches, thus allowing mixed cropping. If farmers are convinced, they will come to pick up this free material; if they are not convinced, they will get planting material from their neighbours.

Therefore, the distribution programmes in Togo aim to make healthy material available in different areas, to give a minimum of material to individual farmers, and to encourage them to use sanitation. From these farmers, others will obtain planting material in due course. In the gari making areas, the farmers who multiply healthy cassava are given a free cassava grater. We ask them to produce, during two or three years, a planting material which the other farmers will come to collect. Nowadays, many farmers ask for cassava cuttings, but though this cassava is free, some farmers will not come to collect it, precisely because it is not being sold.

ARODOKOUN, D.Y.:

To control the disease of cassava each situation must be considered individually. In Benin, the outbreak of cassava bacterial blight led to the need to import food such as rice and wheat. This gave the State additional expense and this made a solution to this problem urgent. Our approach was to rely on IITA, which supplied us with its available varieties. This was very important and we could not have managed without it.

There is not yet any real sanitation programme in Benin, and it would be rather difficult to operate one because there are many locally cultivated varieties and 90 to 100% of the plants in the field are infected. That is why I am convinced that the sanitation programme can be useful only after the introduction of resistant varieties.

I distinguish two issues in the research programme:

(a) A complex issue: the thorough study of the disease and of epidemiology of the virus, something that can be done only in specialized institutes such as IITA or ORSTOM.

(b) Work by the national programmes which use the results and products of the specialized institutes, especially resistant varieties, and can make tests of adaptability, set up multilocation trials, and assess palatability characteristics and acceptability of farmers. When possible, they should also organize extension programmes. Thus, in my opinion, the control of the diseases of cassava must have a variety of combinations of elements. This means that, according to the region, the elements of control will be different. The aim should be to decrease the impact of cassava diseases by changing to resistant varieties, other agricultural methods, establish sanitation schemes, etc.

In my own case, the options are to rely entirely on resistant varieties, or to use them together with sanitation, or combine all the elements mentioned. Numerous varieties have been made available to Benin by IITA: of these, 30572, 30001 and 30395 behave comparatively well. Thus our main problem, which we have



pointed out to IITA, is to introduce new varieties with a lower cyanide content and which are also resistant. Nevertheless, the varieties I have mentioned are accepted by farmers, especially those farmers who grow cassava for processing. The farmers who usually eat unprocessed cassava roots do not accept these varieties and consequently do not grow them.

HARRISON, B.D.:

Could you tell us briefly on what scale you are growing the IITA varieties, what the result has been in terms of yield and how rapidly this material becomes infected with mosaic virus?

ARODOKOUN, D.Y.:

Unfortunately, we do not have precise data on the impact of IITA varieties in Benin. In 1987, we initiated a programme to re-establish cassava in Benin and we should soon be able to see how the improved varieties compare with the local varieties.

HARRISON, B.D.:

What is the reinfection rate in Togo, where you are using the propagation scheme?

TETEVI, K.:

The variety 321524 has been available to us for a long time; it was the result of an IRAT work to improve varieties and it worked well. This variety is tolerant of cassava mosaic because the plants become infected, but the effect on yield is insignificant. In fields planted with this variety, we do not find any plants which are stunted because their leaves have been affected early. The fields are generally healthy and cassava mosaic is not really a problem as such. When the problem of mealy-bug and green spider mite appeared, we noticed that this variety was very dear to us not only because it was tolerant of mosaic, but also because it was resistant to mealy-bug and green spider mite. We did not systematically assess the infection rate of mosaic although we did assess infestation by mealy-bug.

LYONGA, N.S.:

In Cameroon we depend on IITA for the supply of plant families, which are used in the national programme of clone selection, preliminary trials, multilocational trials, etc. Our programme began about 4 years ago, and this year cassava material is being provided to farmers for the first time. In this selection programme, we do some sanitation of the type which has been mentioned and we have produced material that is free from mosaic. But the problem is that although the IITA material is fairly resistant to mosaic, it is susceptible to attack by mealy-bug and cassava green mite. Secondly, the material tends to branch low

down so that farmers cannot use it for mixed cropping. Also it is bitter, which is not so important in my country because most of the cassava is processed. I think that for African countries where resources are limited we should be considering the range of constraints rather than simply seeking solutions to individual problems.

HARRISON, B.D.:

Dr. Rossel made this point earlier when he said that IITA was not breeding primarily for resistance to mosaic but trying to improve cassava in all kinds of ways. However, some of the varieties produced have not been acceptable because they are bitter. Can Dr. Rossel say how much attention is given to breeding sweet varieties?

ROSSEL, H.:

IITA is very much aware of low cyanide content as a useful characteristic to breed for, although this may not be so much of a problem for areas such as those where gari production is the major use. But it has always been part of our programme and is a major breeding objective. However, I do not believe that sweet cassava plays a very important role in Africa. I think the major areas grow mostly bitter types for a variety of reasons, and that bitterness is not considered a major constraint in these areas. I am surprised to hear this kind of remark.

HARRISON, B.D.:

Although my experience of cassava growing is very limited, I have heard several comments about bitter cassava not being acceptable. People may feel this very strongly, even if it applies to only a small area.

SAUTI, R.F.N.:

One of the problems we have in national programmes is that we feel that international institutes should have all the answers to our national problems. If the objective of the national programme is to improve cassava production in their own area, the eating habits of the people must be considered. For example, why import IITA cassava varieties which are bitter if farmers are not interested in bitter types. As long as we fail to import what the farmers require, there is no way we can make progress with IITA varieties. In my own country we have got one main rainy season, but I think environmental conditions in Nigeria are not very different from those in Cameroon, where they are experiencing the same problems as in Malawi with some of the IITA material.

I think it is important for national programmes to evaluate their own local varieties and only then to identify what the farmers need and what materials can be obtained to improve their own

programmes. No one has said "we are not giving you resistant varieties, we are only giving you material which you can use in your national breeding programmes".

N'DA, I.:

As one who is engaged in development, I must emphasize that research work must be based upon the needs of the population, of the consumers. In Ivory Coast, the population likes to eat sweet cassava. I am often being asked for planting material of cassava, always sweet cassava. In our country we eat cassava "attiéké" and also mixed with banana, as "foufou". In general, cassava is eaten in such a way that one really can taste its bitterness. The farmers will not accept bitter cassava. All the farmers to whom I offered bitter varieties refused them.

I therefore emphasize that if researchers breed varieties that are resistant to mosaic or to other diseases, they should also take into account the needs and eating habits of the regions, in order to place at their disposal the kind of variety that is preferred. Otherwise we will be compelled to process cassava differently, to try to get the new varieties accepted.

MAHUNGU, N.M.:

The problem of bitterness or sweetness of cassava is almost a false problem. Each national researcher should be able to evaluate his local material and decide what material he should ask international institutes or neighbouring countries to supply. This material should be assessed, not only as far as yield and diseases are concerned, but especially for its eating qualities.

Zaire has imported much material from IITA. A large part of this material has been kept in collections, so that its resistance can be made use of, and some varieties were multiplied, though they were bitter. Over 70% of the farmers in Zaire like bitter cassava, because they process its tubers very efficiently. However in some regions, the farmers like only sweet cassava, so there we try to introduce sweet varieties. I do not speak for IITA, but I believe that their breeding programme produces such varieties. Of the varieties used at the moment, the sweetest contains 400 ppm of cyanide by dry weight. I do not see where the problem is if countries know exactly what they need to import.

As for sanitation in Zaire, although cassava mosaic occurs in some regions, it is not such a major problem as bacterial blight. The local varieties are susceptible to bacterial blight which can severely affect whole fields, as in 1974 when production of cassava was much decreased. This was why the PRONAM was created. If sanitation is adopted it must be based on knowledge of the whole complex of pathogens, of resistance to them and on other agronomic qualities. For example varieties may be resistant to mosaic or to bacterial blight. Both are of concern to farmers, but bacterial blight can cause more damage than mosaic.

HARRISON, B.D.:

I think that we have considered several aspects of the situation in some western and central african countries, which occupy an enormous area, and include wide variations in conditions.

I now want to introduce the epidemiological element into the discussion, linking this perhaps, with needs for future research.

FARGETTE, D.:

I think that one lesson to be drawn from this seminar is that a connection is necessary between programmes of research and programmes of development. We noticed, for instance, that we may fail if we try to breed and maintain healthy material without having prior information on the rapidity of reinfection in field conditions. Similarly, I think that we could make better use of resistant varieties, if we had a better knowledge of the mechanisms of resistance. It might also be worth investigating the basis of what we have called reversion, i.e. the ability of an infected cassava plant to produce healthy cuttings. This could require relatively basic research on the physiology of the diseased plant, especially the diffusion and translocation of the virus within the plant. One might then be able, with the aid of this basic knowledge to maximise reversion by applying the optimum treatments and so produce healthy material in large quantities.

As to controlling mosaic disease, I do not think that one should regard the production of healthy stocks and the use of resistant varieties as alternatives, but that on the contrary they should be combined. Moreover, I am convinced that the best complementation between these two control strategies will be obtained, if we can understand the mechanisms of resistance of the varieties used.

Several approaches seem to be possible: one consists in distinguishing the components of resistance, and in studying the relationships that exist between them, i.e. between symptom expression, virus content, resistance to the vector and resistance to field infection. Dr. Rossel suggested another approach, which involves a study of the interaction between the plant and the virus, particularly in regard to limitation of virus spread within plants of certain varieties. This study would presuppose a thorough knowledge of the physiology of the diseased plant, which is still lacking at the moment.

As far as breeding resistant varieties is concerned, good progress was made by classical methods in East Africa, as well as in Nigeria. Nevertheless, further progress will be possible when a better knowledge of cassava genetic variability exists. Such studies are in progress in Ivory Coast. Modern methods of genetic engineering should also be tried as an aid to the control of cassava mosaic, particularly by the insertion of resistance genes or of virus-related nucleotide sequences into the nuclear genome of cassava.

I think that the papers presented at this meeting have clearly shown that the ecology of the disease is better understood now, and that a still more thorough knowledge of the disease will be obtained only when we understand better their biology of Bemisia tabaci on cassava. But I doubt whether it is necessary to make such a detailed analysis of the disease ecology in every situation. On the contrary, it would be worth ensuring that small multilocational trials are an integral part of development programmes. Indeed, wherever a method of control is proposed, a research programme should be added in which the reinfection rate is recorded and effects on other diseases and pests are noted. This has been emphasized already as well as the importance of socio-economic features. It is especially necessary to take notice of the reaction of farmers when new varieties are introduced or new farming methods proposed.

My final comment about research programmes is that we should study all whitefly-transmitted geminivirus diseases as a whole. These diseases cause many problems, not only in cassava but also in major food crops such as tomato, cowpea and okra, and in crops like tobacco. It seems desirable to study these diseases simultaneously, so that the research on one disease might benefit and be applied to another. I draw your attention to the fact that in Ivory Coast, for instance, we took advantage of knowledge about the epidemiology of African Cassava Mosaic in our studies of the epidemiology of okra leaf curl, another whitefly-transmitted geminivirus disease.

TRESH, M.:

I would like to make just one or two points. I believe that the biggest need is for multilocation trials. We have heard a good deal about the situations in Ivory Coast and in Kenya, but there are some awfully big gaps in all the other countries, between those two extremes. I think it is extremely important to know how quickly reinfection occurs in these countries, so that you have some idea of the degree of resistance they require.

I would also like to reiterate a point made by Mr. Robertson who pointed out that if you can use existing varieties, then at a stroke you will overcome the problem of acceptance. You do not have to worry about whether your growers are concerned about the bitter quality or not, and you do not have to go through consumers' preference trials or quality tests, and you go straight ahead and use the existing varieties.

There is a whole range of techniques available for producing healthy planting material,. We have heard about tip therapy and heat therapy, and about the reversion phenomenon, the ability to take individual nodes and to establish healthy material by quite simple techniques, as demonstrated in the Ivory Coast. We have also heard that with a good pair of eyes, you can often find a healthy mother plant, as they have done in Kenya, from which to produce healthy stocks that can be used in propagation schemes.

The other point I would like to emphasize is that in Kenya, at the outset, virtually all the planting material was infected, although

experiments later demonstrated a very slow rate of reinfection. That result was totally unexpected. When we look at the huge part of Africa where almost every cassava plant is infected we may be jumping to a false conclusion that the infection pressure in all these areas is very high. Undoubtedly, it is high at Adiopodoumé in the Ivory Coast, and apparently in parts of Ghana and Nigeria, but it is not necessarily high in other areas. Each country should therefore establish adequate stocks of healthy planting material to put in the field and assess rates of reinfection. Then we could begin to draw up the rules of the game, to ascertain the precise nature of the problem we are trying to deal with, and what constraints to progress exist. Until we get that sort of information, we can use and rely on resistance, if there is a range of suitable varieties available, but I think that we can deploy resistance more effectively than in the past.

HARRISON, B.D.:

I now propose to throw the discussion on these aspects open to the floor, in particular to consider the research needs there might be, either applied research or basic research.

LYONGA, N.S.:

I think one of the first issues is not so much what research you can do, but what the farmer can do. If what the farmer can do is expensive, it is out of question, and even if it is not expensive it must increase yield substantially. If this does not apply, you can forget about it.

An unrelated issue, which is overlooked now, is the matter of strains of the virus. This issue is very important, and should be examined.

HARRISON, B.D.:

What particular aspect of the strain question do you think is important?

LYONGA, N.S.:

At the moment there is a report of differential reactions of cultivars in certain places, and I would spend some substantial time determining exactly what is happening. For most diseases, you do not in general find any differential interaction unless you start growing you resistant material on a large scale. The existing observations may be completely unimportant, but I think the situation should be examined carefully.

HARRISON, B.D.:

As you say, this is a question I raised myself, largely in ignorance of what the situation was, except that I know there are other kinds of strain variation, from my own studies. The

indications seem to be, although experimental evidence is not very hard, that the kind of resistance that works in West Africa also works in East Africa and vice versa.

LYONGA, N.S.:

I agree with that, but in general I think the resistant varieties have not been grown over large areas for a long time. So if resistance-breaking strains are rare, it will take time to pick them up.

HARRISON, B.D.:

Dr. Malathi, is there any evidence whether the kind of resistance produced at IITA is effective against Indian form of the virus?

MALATHI, V.G.:

We did have certain well known lines from Nigeria, including line No. 30001 and some others. When we put these under Indian conditions, all of them developed serious disease symptoms, except for 3 particularly well known lines.

HARRISON, B.D.:

Do you know if they were supposed to be resistant in Nigeria, or not?

MALATHI, V.G.:

The reports of IITA indicate that most of the well known lines are resistant. But there is some ambiguity about this resistance.

I would like to comment on two things about the research programmes. IITA is an international organization with many contacts in South America and India, and so I was wondering why they do not conduct intensive breeding programmes using cassava lines taken from India and South America in a concerted effort to bring in sources of resistance both to mosaic and also to pests. Is it possible to develop such programmes?

The second point is about the multilocation trials. We have a system like this in India. Every cultivar that is to be introduced receives an extensive array of trials in different farmers fields, in what we call adaptive research. So, we do not run the risk of unacceptability of a cultivar. Perhaps some similar system should be implemented in all African countries.

HARRISON, B.D.:

I think IITA does use material from South America in their breeding programmes. What there may be is a lack of transfer of

material to Africa that is suitable for use at high altitudes.

NOLT, B.:

There has been a tradition of use of South American material in the IITA cassava breeding programmes. It is also true to say that most of the South American varieties tried in Africa have been susceptible to cassava mosaic. Perhaps only a limited number of lines were tested, so there may be scope for screening more lines from the centre of origin.

Regarding the highland material, that flow of germplasm has been less successful in the past but there have been some organisational changes in the work. It may now become possible to screen more highland material, which is available in large amount from Colombia in South America.

HARRISON, B.D.:

But perhaps mosaic is not such a problem in areas where you might wish to grow the highland material.

MABANZA, J.:

I would like to describe the situation concerning the sanitation of cassava in Congo. Congo is near Zaire, and those two countries have had the same problems, diseases and pests. Nevertheless, they did not choose the same solutions to their problems, though Congo and Zaire imported IITA material at about the same time.

I think that in Central Africa the problem of cassava mosaic is not similar to that in East or West Africa. It is less severe and, consequently, it is thought of differently. As we have just heard, Central African people prefer diseased to healthy leaves for eating. When bacterial blight appeared in Congo, the epidemic had a great impact and, in some areas, cassava crops almost disappeared. But we realized, some years later, that to replace the cultivars that had been destroyed, the farmers had managed to find some other cultivars among the local plant material, and to multiply them. These varieties have enabled them, up to now, to maintain a certain amount of production.

Though we have initiated programmes to import improved plant material from IITA, we have not yet succeeded in getting this improved foreign material to the farmers. However, we realized that the local varieties available to farmers had value and that we should pay attention to these.

Our experiments proved that we could breed varieties resistant to bacterial blight, from the clones and seeds imported from IITA.

However, in the beginning, those varieties were not always accepted by the farmers, either because they had low branches or



because their leaves were not good to eat. We nevertheless maintained this material in our research programmes, for breeding purposes. We also found that, in our varietal trials, some local cultivars could be almost as productive as imported cultivars. This induced us to use this local material in our sanitation schemes, because we thought that it might yield still better if it was freed from virus. We thought it was necessary to initiate sanitation schemes, as the planting material available to the farmers was 90-95% infected. We intend to reintroduce healthy material of these local varieties. At the moment, the healthy clones are only available from in vitro tissue culture.

We think it is necessary to consider the improvement problem as a whole: bacterial blight, mealy-bug, spider mite and mosaic. We must not try to solve the problem of cassava mosaic separately, but we must initiate the selection of varieties that are resistant at once to bacterial blight, to mosaic, to mealy-bug and to spider mite. In the meantime, the small farmer must be recommended to grow healthy material of his stock of local cultivars. We have already started this programme and it is going well. So, a sanitation scheme has already been decided upon in Congo, and must now only be organized and assessed.

HARRISON, B.D.:

Could you tell us on what scale you are producing this material, and also the reinfection rate in different areas in the country?

MABANZA, J.:

We have introduced this system experimentally, first in our research centres, then in State production centres which are used to grow cassava and also in developmental projects, which means through a State structure which is educating local farmers. However, at the moment, our unit can produce only 1500 micro-plants per month. As for the recontamination rate, we are involved only in the experimental side.

Actually, to study the rate of recontamination a certain space of time must elapse. We think that we shall win, because we started from the knowledge that the genetic stock of cassava in Central Africa is rather old. We want both to renew it and to free it from infection.

HARRISON, B.D.:

My question is, how rapidly do the stocks become reinfected when you put them in the field? Is it 10% a year, or 50% a year, for example?

MABANZA, J.:

Unfortunately I have no figures, though I think they could be obtained. Traditional sanitation schemes tend to get healthy

material. Once you have this healthy material, you can record what percentage of infection occurs.

HARRISON, B.D.:

There is one other topic I would like to have comments on. Obviously, whatever scheme of control is adopted by a particular country, a great deal of organization is required to implement it, whether it is based mainly on sanitation or mainly on resistance breeding.

VAN DER GRAAFF:

I think that the first issue with the cassava crop, which has not a high monetary value, is that schemes which are sustainable by the farmers themselves should need as little monetary input as possible. Also, I believe it is important that there should be as little legislation as possible involved in such schemes. Legislation and phytosanitary regulations may work when there is money involved. For example, Kenya exports flowers to Europe and it is important to keep flowers free of diseases, so this is done. If you want to use phytosanitary regulations, I think you need large plantings producing healthy material.

In my opinion, if you want to use sanitation, the best hope is to adopt a method that farmers can operate themselves. They might be able to select healthy material, plant it themselves and the result must be an increase in yield. I think we have already noted that you would need a high level of resistance to do it this way. You also need an extension service.

To return to the distribution of resistant material, this of course needs quite a long time and considerable effort, because the planting material has to be multiplied. In fact, as we have heard this week, all too often the resistant material has not left the research station. Obviously, government and research institutions should have effective links with an extension organization. Money to establish such links would be well spent. However, once you have a new and effective policy involving resistant material, it will soon spread. Farmers will always pick up good ideas from their neighbours. It is difficult to pinpoint any logistic issue. I think one of the most interesting experiences we have heard about is that in India, where there is a scheme which could be used elsewhere. If you can get locations where it is possible to use this scheme, I think it could work very well.

HARRISON, B.D.:

Is there any scope for an organization that promotes collaboration between different countries or arranges division of efforts between countries?

LYONGA, N.S.:

On the whole, national programmes and governments have little money to play with. So, I do not think that individual countries will undertake a whole spectrum of research. I think we should see what they can do and what can be undertaken by international institutes and organizations like ORSTOM and IITA. The more basic things like tissue culture, genetic engineering and so on, are best handled by those who have the ability and are accustomed to do this kind of work. The national programmes, of course, will be responsible for the simpler and more applied work of multiplying stocks and so on. We, in the national programmes, certainly know what we are doing, what each other is doing, and this has a collaborative element. We should encourage and increase the transfer of material, and seize this opportunity, as the President of the International Society for Tropical Root Crops, Africa branch, to say that the Africa branch exists for exactly this type of things. Our branch is very strong, and it holds a symposium every 3 years or so, which helps to stimulate interactions between those working on root crops in different institutes. The next meeting will be held in Zaire in 1988 or 1989. In this forum, the various national programmes are discussed.

ARODOKOUN, D.Y.:

I come back to the programme which has been proposed to ask whether this programme has taken account of actions leading to short-term as well as long-term results. We hear of reversion, of multicomponent resistance, and these studies must increase knowledge and give rise to more balanced measures. But at the moment we have problems, and short-term actions are needed. We should make this point to our various partners: international institute such as IITA and ORSTOM, organizations like CTA, FAO or CPI, and also national programmes.

KOUASSI, B.A.:

Since last Monday, we have learnt about cassava, its importance in Africa, and the economic impact of African Cassava Mosaic. Various possible solutions have been discussed, such as sanitation, or breeding varieties that are resistant to the virus or to the vector. Researchers are trying to find solutions that might be useful to the farmer. But I have heard no mention of Bemisia in any research programme, and I see no researcher who studies it. If you consider the problem of endemic human diseases, people are protected by controlling the vector. That is the case for sleeping sickness. Consequently, why is Bemisia not controlled? It may be difficult but it must not be neglected. Bemisia is not easily controlled by insecticides, but there is a danger that it will multiply more and more with the increase of cassava culture, because it is not controlled. From now on, entomologist researchers should therefore study Bemisia.

HARRISON, B.D.:

I think we have had some mention of Bemisia in earlier sessions, and of susceptibility and resistance to Bemisia. I do not think we can embark on a discussion of this question now. I would like to end this session by thanking the people who have led the different parts of the discussion this afternoon, and also all the others who have contributed to it. I have found it a valuable discussion and I hope you have too.

# **THEMATIC REPORTS**

Chairman  
Dr. DELLERE

## THE CURRENT SITUATION

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During this session, were successively presented the national situations, the statement of the research on African Cassava Mosaic, particularly in the Republic of Congo, the integration of methods of control of the disease and finally its impact upon cassava production. A summary of the specific case of India was also presented.

### GENERAL SYNTHESIS OF NATIONAL CIRCUMSTANCES

This general synthesis raised a major question: are the peasant farmers really aware of the existence of African Cassava Mosaic? Though the situation is quite different from one country to another and, within a given country, from one region to another, farmers seem to recognize the disease quite well and, if they were offered enough healthy material, they would certainly use it. The best evidence of this is that in countries where sanitation schemes exist, (i.e. Burundi, Rwanda, Malawi, Zaire, Benin, Uganda), the farmers quite willingly use such healthy material.

The acceptability of resistant varieties, which are mostly imported from IITA, depends on the eating habits of local people. Those varieties are easily accepted in East and Central Africa, but the populations of West Africa are reluctant because they prefer sweet varieties. The breeding programmes should consequently take such regional specificities into account. The populations who usually eat cassava leaves prefer those which are infected. An account should also be taken of this sociological data in the breeding and sanitation programmes.

As a matter of fact, the programmes of control of African Cassava Mosaic Disease should, as a priority, be devoted to sanitation schemes and to the breeding of resistant varieties. In addition, the farmers should be made aware of the problem and educated. The researchers would thus have an opportunity to meet the farmers and approach their everyday life, which would be the best way to evaluate the research needs.

## STATEMENT OF THE RESEARCH ON AFRICAN CASSAVA MOSAIC

In spite of numerous gaps, the data accumulated in East Africa as well as in Central and West Africa are considerable. Nevertheless, there still remains a lack of transfer to small farmers of the results acquired in laboratories and research stations. In other words, little information is available in the field and a closer relationship should be established between farmers and researchers. A greater effort should be made in this direction.

Some gaps to be filled are, for instance:

- On the one hand, the showing up of a real quality difference between cassava produced from healthy material and cassava produced from infected material. Work done in India tends to indicate that there is no difference in the roots but that there is a difference in the leaves;
- On the other hand, the explanation of the fact that the reinfection rate of healthy material is smaller in East Africa than in West Africa.

## STATEMENT OF THE RESEARCH ON AFRICAN CASSAVA MOSAIC IN CONGO

In developing countries, the research in the field of virology raises a problem. This kind of research implies the use of an expensive and sophisticated material, as well as the employment of highly skilled researchers. This need for researchers might be solved by a training policy which might be supported by the Interafrican Phytosanitary Council (CPI).

The existence of a programme of research on African Cassava Mosaic in Congo is accounted for by a certain number of factors such as:

- the fact that African Cassava Mosaic is present almost everywhere, in peasant farmers' fields as well as in State farms,
- the fact that the symptoms decrease with the age of the plant, which does not induce the local farmers to breed healthy planting material,
- the existence of a large genetic variability of cassava,
- the constraint on renewing the genetic stock,
- finally, the fact that some cultivars are genuine virus containers.

The programme consists of the creation of tissue cultures which are separated during 2 to 4 weeks, before they are controlled by means of a biological test.

The progress of research so far enables the assessment of the reinfection rate of the healthy material produced from tissue cultures. However, it seems necessary to make tissue cultures of

numerous clones at the same time, in order to constitute a basic culture stock, from the plants which are distributed to farmers.

It is necessary to add constant sanitation to this diffusion, in order to maintain the lowest possible reinfection rate.

#### IMPACT OF AFRICAN CASSAVA MOSAIC ON YIELD

The symptoms of the disease were evaluated by Cour's method (0 to 5) and their importance quantified thanks to the Index Severity of Symptoms (IGS). This index changes in the course of time and is specific to a given cultivar.

The impact of the disease on cassava production is analysed through four kinds of relations:

- the relation symptom/production shows that mosaic is a limiting factor of production. This should nevertheless be considered carefully, as a decrease in production is likely to happen, without any previous production of symptoms,
- the relation mode of contamination/decrease of production shows that the decrease, in terms of yield, caused by the use of infected material is higher than the decrease caused by whitefly contamination after plantation. The contamination seems to have no effect when it happens 100 days after plantation,
- the relation yield/environment shows that competition exists between all plants in a given field, so that the most infected plant will become weaker than the healthiest plant,
- finally, the relation yield/growth illustrates the relations that exist between some growth parameters (for instance the basal diameter of stems and the yield). However, those interrelations between growth parameters are not always clear.

As a matter of fact, if the assumption "symptoms-yield" does not always come true, nevertheless an infected plant will always show a greater yield decrease than a less infected plant or an uninfected one. The impact of the disease in terms of yield of cassava in Africa is quite real and it implies the setting up of programmes of constant sanitation and breeding of either resistant or tolerant varieties.

#### INTEGRATION OF METHODS OF CONTROL OF AFRICAN CASSAVA MOSAIC

The control of a viral disease presuppose knowledge of:

- the rate of severity of the disease
- the importance of infection, and
- the beginning of infection.

Later on, means are adopted to reduce the number of infected plants in order to delay or limit the infection.



In a further stage, the concept of tolerance and of resistance should be made clear within a breeding programme to which a policy of constant sanitation should always be added.

#### INDIA: A SPECIFIC CASE

Cassava in India is grown over an area of about 370,000 hectares and annual production reaches 5,5 million tonnes. The yield per hectare: 17 tonnes, is the highest among all cassava producing countries. The main region producing cassava is the State of Kerala.

As in Africa, cassava mosaic is one of the most important diseases infected cassava. The insect vector is Bemisia tabaci, but the disease spread is more related to farming methods than to whitefly contamination.

The geminivirus which is responsible for the disease in India presents an antigenic difference from the geminivirus of African Cassava Mosaic. In other respects, the yield losses caused by the virus are less important, as its hosts are less numerous. On the grounds of those differences, a distinction should be made between African Cassava Mosaic Disease and Indian Cassava Mosaic Disease.

The means of control is to be found in the identification of resistant cultivars. Those cultivars are cleansed by the culture of meristems. This material is then used both for the improvement of cultivars and for the culture of cassava. However, the very close screening and the systematic roguing of infected plants are two efficient methods of control.

## THE FACTORS

BOCK, K.R.  
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Five papers were included in this session, two dealing with the virus, one by Dr. Harrison, and one by Dr. Rossel, and 3 dealing with one vector, one by Mr. Robertson, one by Dr. Fishpool and co-authors, and one by Dr. Yao.

I would just comment that I am amazed at the amount of knowledge which has accumulated on ACMV: we isolated it in Kenya, not more than 10 years ago, and ACMV must now be among the best known of all plant viruses.

### FACTOR 1: THE VIRUS

In regard to the virus, two areas appeared to be of major interest. The first involved detection. We heard from Dr. Harrison of one sensitivity of Elisa (10ng/ml), and from Dr. Rossel that N.benthamiana was also sensitive.

In this regard, it is a pity that we did not have a paper on host virus interactions. We deduce, from what has been said at this meeting, that when there are no symptoms, there is no virus.

There were questions indirectly associated with this crucial point, and these of course dealt with detection. Dr. Harrison indicated that, using Elisa, sensitivity was to be done of 10 nanograms virus/ml. Dr. Rossel indicated that ACMV could be detected in young symptom bearing leaves, with more difficulty in older leaves, and not in symptomless leaves. Rossel also told us that, in the equilibrium of resistance, resistant plants outgrew infection, but that, nonetheless, some 20% of plants of the variety propagated exhibited symptoms. This, itself raises two questions, one of which was asked: the need for information from IITA on:

1. the concentration of virus in these plant tips,
2. whether whiteflies can acquire virus readily or not, from them. It was suggested that IITA should check this important point.

## FACTOR 2: WHITEFLIES

One very important point was field control of the vector. We were warned of the possible, indeed likely, disastrous chain of events that invariably ensues after insecticidal control of whiteflies, and to me the dangers are very clear.

It seems that at least two major parasites of whiteflies are known, but they have not been tested in Africa. Chances of success were not rated too highly.

Whitefly resistance: differences noted in Kenya and Malawi, in support of the results obtained by ORSTOM in Ivory Coast.

I personally find one difference in whitefly numbers and diseases incidence between East and West Africa interesting, but puzzling: Mr. Robertson's data on flies and our disease incidence indicates clearly that there is little, if any, correlation between peak populations and disease. In West Africa, ORSTOM and IITA results indicate a very close correlation. This suggests that more work needs doing in East Africa.

In conclusion, I would like to congratulate the ORSTOM team on their preliminary work and on their proposed program of work, on Bemisia tabaci in cassava, in West Africa.

One result of their work will be an essential prerequisite to a more comprehensive understanding of what is perhaps the most widespread and important disease in Africa.

## EPIDEMIOLOGY

THRESH, J.M.

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The point that I would make is that this is almost the first meeting on cassava mosaic where the deliberations have been underpinned by a very firm knowledge of the epidemiology of the disease, in at least some parts of Africa. In the early days, although the disease has been known for very many years, and it was appreciated that the disease spread more rapidly in the coastal areas of East Africa than it did in the high areas, there was very little information on the pattern of spread, on the sequence of spread, on the time of spread, and so on. So, I think that we did hear, on Wednesday, a very remarkable series of papers from Kenya and from the group here in ORSTOM, which do give us some very important facts, on which to base our further discussions.

If I could just remind you that when viruses, in general, spread into crops, the so-called primary spread is by very mobile vectors, which initiate the first infection. There is then a secondary spread within the crop.

In most virus diseases, the primary spread is of very great importance, in initiating new outbreaks, but the most of the spread that occurs is within the crop. Now we have quite a lot of evidence from the studies in Kenya, and in the Ivory Coast, that African Cassava Mosaic is really different in that the primary spread is to be all important and the secondary spread is of relative little importance.

If this is the pattern in all the cassava growing areas, and we know about this from Kenya, we also know about this from the Ivory Coast, it has a number of very important implications. One of them is that roguing out infected material, within the crop, is not going to have a big impact on the spread in that season's crop. I think this is a rather important point. Secondly, there is going to be a very limited opportunity, that even if we overcame the problem of using insecticides, and all the side effects that could occur, that approach would then likely be effective, because we know no insecticide that is able to halt the primary spread, even though it is effective in restricting the secondary spread. The third implication of this sort of finding is that, instead of looking into the crop we are concerned with, I suggest that we should look away from the crop, and look at the sources of

infection from which the whiteflies are coming in. So, I think that we have got to look at the sources of infection, in terms of number and of the extent of the primary infection from which spread is occurring into our crops. We also need to be concerned with the proximity effect, the distance.

Are the sources of infection near by, or are they far away? We also have to be concerned with the potency of the sources: Are they a good source of virus, or are they a poor source of virus? We have heard a number of points about this, and it appears that the wild cassava grown here is a relatively poor source of virus. We hear that these resistant varieties appear to be a poor source of virus although, as Dr. Bock pointed out, we do need some more information on this.

We should also be concerned with the orientation of the sources, in terms of wind direction, because all those factors are important.

If we look also at the plantings with which we are concerned, the size effect, because of the perimeter area ratio, is likely to be important in determining infection; the shape of the plots again influences the perimeter area ratio, the effects of windbreaks and of various other sorts of boundary around the plot.

We should also be concerned with the regional factors: how much cassava is being grown in the area around the planting that we are concerned with, what are the number of farms, their size and how they are distributed within the region?

I would just like to go through a number of points, that I think have arisen in the course of our discussion, in relation to epidemiology and, just in a particular order, a point raised by Dr. Harrison, that is: "Is the epidemiology of the different strains, the same?".

We know that there are quite big differences in the biochemistry and in some properties of the A, B and C strains. It appears that the strains, in this part of the world, are more difficult to control, than those in the coastal areas of East Africa. Is that an environmental effect, or is it something connected with the strains? We do not have any information on this point.

We heard that there was an urgent need for information on the Bemisia hancocki from Kenya. Is it, or is it not a vector? Mr. Robertson pointed out that Bemisia hancocki came from a large proportion of the whiteflies taken, caught in samples, in cassava fields. Dr. Bock has already covered my point about the potency of the so-called resistant varieties, just what sort of hazard do they pose to the other varieties near by?

Another important issue that I think arose is: what is the required frequency of roguing? Is it just sufficient to go through a planting in the early growth stages, to remove the obviously infected plants, and to remove the infected plant at the end of the growing season, to avoid carrying over to the next crop. Or is

something more frequent than that required? I do think that we need a lot more evidence, from a lot more different regions, on the requirements of roguing.

What are the basic requirements, what is the message we should give to the farmer, and to the extension service? Until we have that sort of information, our message, I suspect, is going to be a weak one, and we are not at all clear about exactly what we are going to do. Another important issue is the dispersal of whiteflies. If we are correct in saying that the primary spread is from outside sources, and not within the crop, then what are the determining factors that influence the numbers of whiteflies that disperse over a distance between plantings? What is the seasonal factor, what is the host factor that makes them get up and go?

We know from experience with a lot of other vectors, that there are some extremely important physiological cues that come from the host plant, or from the environment. They give the vector the impetus to leave the crop on which it has been developing and to suddenly move off over considerable distances. We know that whiteflies can be dispersed over big distances, but we are not at all sure whether this is a phenomenon that takes place at all times of the year, we are not at all sure of what is the stimulus that makes them get up and go. I suspect that this is a very important factor, that does need attention. I think that is one of the reasons why the ORSTOM group looked for some further entomological input into their programme here, to look at their problem.

Just one other point about the ORSTOM work, as well, is the surprising effects of temperature. Those results coming through the tropics tend to make us think it is all the year round hot and humid in the Ivory Coast. What the French ORSTOM workers have shown here is that, quite surprisingly, small differences in temperatures can have a very big effect on whitefly populations, on virus spread and on rates of disease development. This is not unknown with other physiological effects in the tropics and it is a general phenomena, although perhaps to us surprising, that quite small differences in temperature can exert very big differences on biological processes.

Just by comparison, there is the completely different seasonality of spread as shown by Mr. Robertson's work in Kenya and I think that we do need more information from other regions on the seasonality effect.

One other point is that although cassava, in general, throughout the continent, is growing in mixed stems with other crop species, we have heard nothing whatsoever about the impact of other crops on cassava and on the spread of cassava mosaic virus. I think this is a difficult area for experimentation, but I do suspect it is a very important one, because the mixture of crops that are grown may well have a very important effect on virus spread.

There are at least possibilities of looking at likely barrier crops to intercept some of these whiteflies, or to affect their deposition within the crop. This could have an important part to play in our control strategy.

I think we had some very exciting discussions, which are for the first time based on good scientific evidence on the behaviour of the virus and of the disease. I think we have got the basis for a lot more exciting work, which I am sure is going to have an impact, to make the control message, the extension message that we can put across more effective in the future, as the information accumulates.

AFRICAN CASSAVA MOSAIC  
FROM A GENETIC POINT OF VIEW

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The papers presented during this seminar concerning the breeding of cultivars resistant to African Cassava Mosaic enabled us to gather general information about the work done in different African countries and India by regional institutes such as IITA and ORSTOM and by national programmes (India, Uganda, Zaire, East Africa and national reports). Jennings and Silvester, moreover, presented an historical survey of the work done in East Africa and Madagascar. Several presentations made up for Dr. Hahn's absence and informed us about the evolution and applications of IITA's cassava breeding programme.

A basic question was discussed, i.e. which strategy should be adopted to develop slightly infected cassava crops.

SANITATION OR RESISTANT VARIETIES

A clear answer was given: the most efficient method consists of the combination of sanitation and breeding. Sanitation schemes allow a rather rapid multiplication of healthy plants, but in an infected environment their efficacy is limited. By contrast, the culture of resistant varieties following a long period of research and extension, enables a reduction of pressure of contamination in the field. This parallel evolution of both approaches was successfully achieved in East Africa, Madagascar and India.

African Cassava Mosaic was of course given the priority during this seminar, but several speakers insisted on the fact the other selection criteria should be taken into account, i.e. all parasitic diseases (bacteriosis, mealy bugs, mechanisms of level of production and of adaptation of cultivars (drought, altitude), of the plant morphology and of the product quality (sweet cassava, quality of leaves, starch, protein).

The selection schemes used in the different countries are arranged around breeding and sexual selection.

- In the course of sanitation, the regeneration of a variety must be carried out according to the cultivar breeding model.



Apart from the choice of healthy families, it allows the restoration of a clone.

- The virus resistance having a recessive, polygenic determinism, the IITA polycross method is recurrent selection. It allows the accumulation of favourable genes at each selection cycle; it causes a regular and cumulative increase of favourable genes. This procedure is moreover efficient for all other quantitative characters if they are positively correlated (mosaic and bacteriosis).
- This method as well as the genetic progress are limited by the parental clones used in the polycrosses. We noticed the important role played by hybrids descendants with M. glaziovii in the creation of varieties resistant to African Cassava Mosaic (Tanzania, IITA, Madagascar).
- National programmes develop the creation of cultivars based on breeding the descendants of the IITA material and the free descendants of local cultivars (example of PRONAM in Zaire), In Cameroon and in other African countries. This strategy makes the most of the variability of traditional varieties and of the sources of foreign genes, thanks to the allogamous reproductive mode of the species and to the vegetative multiplication. Thus, the choice individuals which have the characters researched for in a given country are multiplied in order to be distributed.

#### SEXUAL SELECTION AND VARIETAL SELECTION

These are two complementary strategies.

The work of cassava improvement enhances the role played by the outset material, traditional cultivars on the one hand and wild species on the other hand. Considering the importance of introgressions and of naturel genetic exchanges in the species Manihot, the use of germplasm came up against the risks associated with the transport of this material. At the moment, those barriers are technically overcome thanks to:

- the in vitro meristem culture associated with thermotherapy,
- the methods of indexing of mosaic,
- the in vitro conservation of germplasm.

Surveys and exchanges of both wild and cultivated material must be continued in order to increase the possibilities of breeding more efficient cultivars.

#### THEMES OF RESEARCH

From the discussions and propositions made, three themes of research have emerged to further progress in the control strategies of cassava, related to the varietal improvement:

- Study of the components of resistance to mosaic and of their relationships, arising out of their genetic determinism, so as to direct the selection towards different levels, the final aim remaining resistance in the field.
  
- Development of the knowledge about the genetic variability. This research is linked to the surveys and collections. It allows better identification of cultivars and appreciation of the genetic distance which exists between groups of cultivars having the same genomic combinations (choice of polycrosses). The relationship between wild, cultivated or adventitious species are stated, which allow for the determination of intermediate species for genetic exchanges. A regional coordination of such evaluations might be established.
  
- Review of biotechnologies suitable for cassava, particularly of genetic engineering. In cassava, the applications of genetic engineering are limited to regeneration and to molecular biology levels. The progress in such fields of research is rapid in specialized laboratories and we can be confident for the future.

## CONTROL STRATEGIES

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Control strategies were discussed for different geographical areas. For Kenya, sanitation procedures had been successful for 3 years and would probably be effective for much longer periods, providing that cultivars with moderate resistance to infection, were used. A range of cultivars with good agronomic characteristics is already available but further information is needed to assess reinfection rates of different local cultivars.

Similar approaches seem likely to be effective in several other parts of East Africa, and sanitation schemes are already operating in Malawi and Uganda, and could prove effective in Tanzania. It was important to site propagation plots at some distance, and certainly upwind of any infected material. For the future, breeding could play an important role by providing new cultivars with enhanced resistance and improved yield and other agronomic qualities. For the present, strong emphasis should be given to sanitation programmes and this would require education, sustained support from advisers, extension officers and farmers.

In India, sanitation schemes are also successful where they have had adequate and continued support from extension personnel.

In West and Central Africa, conditions are much more varied than in East Africa. While there is some evidence that sanitation procedures may be effective in some areas, precise information is lacking on rates of reinfection for large parts in most countries. However, sanitation alone will not be effective in areas with high inoculum pressure, and in such areas resistant varieties are essential. The IITA Cassava Improvement Programme is providing resistant genotypes for National Programmes. The need to suit these genotypes to local preferences was emphasized, e.g. tuber-sweetness, leaf palatability and branching habit. IITA is now able to distribute not only seed, but clonal material in tissue culture form, which has the advantage of maintaining favourable combinations of genes. In propagation schemes, the Congo tissue culture is already being used to build up stocks of pathogen-free clones and this approach may be appropriate in some other areas. It was also thought desirable that national programmes should produce healthy stocks of local cultivars and evaluate these, along side imported IITA material.

The discussions identified several further research objectives:

- i) multi-location trials are needed to assess infection rates in different areas,
- ii) the response of the resistant cultivars to different virus isolates, i.e. those occurring in West Africa, East Africa and India respectively, should be tested in detail. In addition, the possible occurrence of resistance-breaking strains should be recognized and, if observed, this should be reported promptly and follow-up studies initiated,
- iii) the extent and value of resistance to whiteflies needs to be examined further,
- iv) ways of accentuating the natural loss of virus from infected material should be explored,
- v) the prospect of improving cassava by genetic engineering was recognized, but this would require the application of expertise from developed countries.

Control strategies are most likely to be successful if they are simple, cheap, seen to be of value by farmers, do not require legislation and are supported by a strong extension service.

The need for greater exchange of information between African countries was recognised. Research needing sophisticated facilities is best done, for the present, through international organisations within Africa, or in collaboration with developed countries. Research of this type on other geminiviruses may well produce information applicable to cassava mosaic.

National programmes should give most emphasis to applied research and development.

## AFRICAN MOSAIC DISEASE AND ITS CONTROL

### CONCLUSIONS

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We have reached the end of the International Seminar on:

### AFRICAN CASSAVA MOSAIC AND ITS CONTROL

and, as Seminar Coordinator, it is my duty and my pleasure to weigh up our discussions and draw the conclusions of the Seminar.

The first objective of any Seminar is the dissemination of information; we may safely say that, in this case, the objective has been easily attained. Participation was considerable and the only regret is that there was not more time to exhaust all topics of discussion. People from three different domains were brought together: scientists, national representatives from 21 African countries and representatives of international organizations. As such, one might have expected a certain lack of inter-communication, but in the event this was not the case and I think everyone was satisfied with the exchange of ideas and concepts that occurred during the week of the Seminar.

The Seminar had three main aims:

The first was to describe the situation of ACMV within the African continent: and we were once more made aware, if it were still necessary, of the considerable importance of ACMV, amidst all the other diseases that affect cassava and which justify the development of large scale control methods. This assessment also showed that, within certain African countries, research and extension programmes were in progress and needed only to be encouraged. Research in 21 African countries showed that resistant varieties, originating from IITA, had been sent to many countries, but had not been widely distributed in most of these.

The second aim was to present an assessment of the state of our scientific knowledge of ACMV, and we were able to bring together all the scientists, except for one, either previously or currently

working in this field, from both Africa and Europe. In this way, it was possible to compare and contrast results obtained in regions as different as East and West Africa. It also underlined where information was still lacking: in certain geographical areas such as Central Africa and in certain scientific disciplines like the genetic variability of cassavas and in the cellular mechanisms of disease resistance. This survey had above all the effect of bringing new concepts into the study of the development of this disease and into the biological mechanisms of cassava resistance.

The third aim of the seminar was to identify methods of disease control relevant to Africa and one can say that a large measure of consensus was achieved throughout discussions. This consensus stresses:

#### THE COMPLEMENTARY AND SIMULTANEOUS USE OF RESISTANT VARIETIES AND SANITATION TECHNIQUES,

taking into account variation in local conditions. The realization of control programmes against this disease will require the support of international funding.

During the seminar, numerous participants stressed the necessity for international cooperation and coordination between states within Africa, to more effectively control ACMD. The national representatives were very pleased at the coming together of the European and Franco-African teams, so that their research and development programmes could be coordinated. As the ACMD does not respect national boundaries, a joint effort is required to control the disease.

All discussions highlighted the lack of development facilities in all the African countries concerned, and also the lack of training of the average small farmer. Sanitation techniques are extremely simple: however, they need to be explained and shown to local people. Similarly, resistant varieties exist, but the small farmer is reluctant to use them, since he does not understand neither their advantages nor their limitations. They need to be informed. Cassava, as everyone knows, is propagated by cuttings, which means that, compared with plants propagated by seed, distribution is relatively difficult. As a result, countries and extension workers need to take this into account and devise the means necessary to ensure the successful propagation and distribution of cassava.

What will be the consequences of the Seminar? These can be divided into three parts: Cooperation, publications and action.

With regard to cooperation, the contacts made between the scientists present were very important, helping to overcome language barriers or differences between research institutes. Equally, reinforcement of the liaison between African countries, in the form of their national representatives, many of whom will remain in mutual contact, can only help ease the problem.

As far as publications are concerned, a whole series ranging from the practical to the scientific, may be cited:

- technical information sheets in several vernacular languages, for use in explaining sanitation techniques and describing the new varieties to the small farmer.
- the seminar proceedings in French and English, to be distributed as quickly as possible to all participants.
- A synopsis of the seminar will be distributed in French and English, in a run of over 1000.
- the publication in the FAO bulletin of a synthesis of the work of the seminar and situation in Africa regarding ACMD.
- several articles on extension topics in various journals, widely available in Africa and the third world.
- the production of a scientific text on ACMD in French and English, aimed at scientists, universities and development agencies.
- broadcasting on Radio France International in French and English, on 19 African radio stations, and programmes on Ivorian Radio and Television.

Concerning research programmes, a number of propositions were made, and included:

- Evaluation of the variability of the resistance of local clones.
- Multilocal trials under field conditions of the improved varieties.
- Epidemiological studies in Central Africa,
- Studies of the variability of the different strains of the virus.
- Establishment of biotechnology programmes on cassava.

Regarding action programmes, nothing is yet decided, but now the international organizations, through their representatives, know what the position and views of the scientists and extension workers are, and the national representatives know that these organizations are ready to begin research and development programmes. As a result, everything is now ready so that the months and years to come will see the development of such programmes.

I hope therefore that this work and these good resolutions will bear fruit and I will make a date with you at the next International Seminar of ACMD, which doubtless, will have as its title:

#### THE CONTROL OF ACMD IN AFRICA

# POSTER SESSION



SOME AGRONOMIC ASPECTS OF RESEARCH ON CASSAVA  
IN THE LOWER IVORY COAST

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In the south of the Ivory Coast, almost all cassava is produced by traditional methods, where the yields are of the order of 5 to 6 t/ha. To increase the production of this food in a region where the availability of arable land is more and more limited, it is necessary to improve agricultural techniques.

Trials at an experimental station, from which some results are presented here, were aimed at determining the amount and quality of production of cassava in a monoculture, and analysing how this yield was affected by certain characteristics of the growth, edaphic and climatic factors, and technical procedures.

**EFFECT OF PLANTING DENSITY ON YIELD**

The behaviour of cassava (variety CB) at five planting densities was compared over a 14-month cycle.

Table 1. Effect of planting density on yield

Density (plants/ha)	5917	6944	8264	10000	12345
Spacing (in metres)	1.3x1.3	1.2x1.2	1.1x1.1	1.0x1.0	0.9x0.9
Yield (t/ha)	31.1	30.8	31.9	30.8	32.9
Mean of weight of tubers per plant (g)	580	573	513	439	408
Total of tubers per plant	9.1	7.9	7.0	6.6	6.0
Harvest index	0.44	0.41	0.42	0.40	0.39
Starch (% of dry weight)	64.2	68.9	66.6	68.6	66.3

The yield per hectare was close to 30 tonnes at all five

densities, but the number and mean weight of tubers per plant changed to compensate for the number of plants. The vegetative part developed more lateral branches and therefore had more leaves per unit surface area at the lower planting densities.

#### EFFECTS OF TILLING THE SOIL AND OF FERTILIZATION ON YIELD

The aim of this trial which is still in progress, was to observe the changes in yield of cassava (Bonoua variety) and of the physicochemical characteristics of the soils being tilled. The plots differed in their cropping history (old Panicum maximum fallow or secondary forest) and in the technical programmes followed (fertilization, traditional agricultural practices, etc.).

Table 2. Fresh weight of tubers (t/ha)

Tilling	Non-mechanized		Mechanized	
	A		B	C
Fertilisation	1st year	1st year	2nd year	1st year
Without fertilisation	18.6	28.6	21.0	20.6
With fertilisation	24.1	28.5	27.0	27.2

A: clearing of secondary forest in non-mechanized cultures;

B: clearing of secondary forest in mechanized cultures;

C: Panicum maximum fallow.

A comparison of the yields between fertilized and non-fertilized systems shows that better use was made of fertilizer on traditional clearance and mechanized fallow. In addition, the yields were better on tilled sites than on non-tilled sites.

#### WATER CONSUMPTION OF A CASSAVA CROP

From neutron measurements of soil humidity we measured the water consumption of a canopy of cassava (variety CB) for 12 months.

Table 3. Water consumption of a cassava crop

Period after planting		Real evapo- transpiration ETR (mm/day)
Beginning of cycle	3rd to 4th week	3.6
	4th to 5th week	3.4
Middle of cycle	25th to 26th week	3.1
	26th to 27th week	2.7
End of cycle	50th to 51st week	2.8
	51th to 52nd week	2.8

The yield was 30 t/ha; the mean measured water consumption was 3 mm per day. Compared with the potential water loss through transpiration at the station (ETPBAC w. 5.0 mm/day), the cassava, which is a sun-loving plant, could be said to have a relatively low demand for water. In certain conditions other plants such as maize consume up to 1.2\*ETP.

#### CONCLUSION

The plant is seen to respond to differences in planting density; the quality of the crop as a function of its intended use determines the choice of density. Working the soil (i.e. labour), even without fertilizer, makes it possible to obtain good yields; one of the beneficial effects of tilling the soil before planting is a better control of the structure of the plants. Cassava is a hardy plant and resists drought well, but nevertheless a prolonged water deficit could rapidly induce senescence of leaves thus reducing the foliar index, which might in some cases induce a loss of yield.

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CONSIDERATIONS ON THE GENETIC VARIABILITY OF CASSAVA  
IN AFRICA  
BASED ON INITIAL RESULTS OF ENZYME ANALYSIS

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Investigations on the genetic variability of species of the genus Manihot have so far been based on morphological criteria. Enzyme electrophoresis has highlighted the differences between the conclusions from morphological and biochemical analyses in the case of cassava (Zoundjihekpon, 1986).

Used as criteria for classifying cultivars, enzymatic markers have the advantage of not having been directly subject to human selection. Also, their simple genetic determination allows them to be used as descriptors of the organization and function of the genome.

This investigation concerns the cultivated species M. esculenta and a species formerly grown for its latex, M. glazovii. The latter, which is widespread in Africa, soon proved to be one of the best sources of resistance to the two principal African diseases of cassava: ACMV (Jennings, 1976), and vascular bacteriosis (Hahn *et al.*, 1980). At present, 10 enzyme systems can be detected by starch-gel electrophoresis. In M. esculenta, 10 loci can be distinguished displaying a structural polymorphism and 2 with no alleles.

We analysed three collections:

- a collection of Ivory Coast cultivars (168 of them, which finally yielded 78 electrophoretic "genotypes"),
- a collection of 49 clones (38 "genotypes") tested by the plant virology laboratory, showing the whole known range of resistance to ACMV (mainly consisting of improved varieties, from Africa, America, and Asia, and representing a wider genetic base than the collection just mentioned,
- a collection of M. glazovii and of M. esculenta x M. glazovii hybrids, prospected in the Ivory Coast (respectively 23 and 10 "genotypes").

For the 12 loci, the various zymograms encountered in M. esculenta may be interpreted assuming diploid genetic determination. Investigation of descendant lines (still under way) has already confirmed 8 of these interpretations. These 12 loci involve 28 alleles, of which 9 are shared with M. glazovii. We note that the allelic frequencies in the Ivory Coast cultivars of M. esculenta

are either less than 15% (which we call "infrequent" alleles) or more than 30% (which we call "common" alleles).

The analyses show that these cassava cultivars can be classified into seven groups. These are characterized as much by the particular combinations of common alleles as by the presence of certain infrequent ones. "Basal" and intermediate groups can be discerned.

The intraspecific variability available in the Ivory Coast therefore appears to be built up in a limited number of allelic combinations. The intermediate groups reflect an evolution involving recombinations and gene exchanges. At the same time, the analysis of the collection of varieties of cassava tested virologically did not reveal any new allele as compared to those in the Ivory Coast cultivars, except for two alleles of M. glazovii introduced into M. esculenta. Five of the seven infrequent alleles in the Ivory Coast were also at a low frequency in this collection. Groups of relationships defined from the same alleles were observed here also: some varieties showed up new allelic combinations, and others corresponded to the combinations found in the Ivory Coast.

In addition, the investigations made in 1985 in the Ivory Coast turned up hybrids between M. esculenta and M. glazovii. The observation of morphological characters (shape and size of the fruits, seeds, leaves, tuberization of the roots) suggests the existence of several levels of backcrossing with the cultivated parent. Thus different stages of introgression may be observed in the cultivated species.

This hypothesis was confirmed at the enzyme level: the spontaneous hybrids displayed at each locus an allele of both species; just one of these hybrids possessed a locus without any alleles of the M. glazovii type, and this was the one which seemed closest to M. esculenta morphologically.

Most of the traces of introgression found in the cultivated species (infrequent alleles in M. esculenta corresponding to common or even fixed alleles in M. glazovii), must nevertheless result from older exchanges. One may therefore observe gene exchanges, both interspecific (with a species which was probably introduced into Africa at the end of the last century), and intraspecific between groups. It may be assumed that two factors favour this evolution:

- allogamous sexual reproduction, accompanied by the absence of an absolute interspecific barrier (the use of seeds or of cuttings derived from natural germinations is not rare) (Beck, 1982).
- A vegetative multiplication, which has made possible not only the transfer of material (including the material derived from selection centers) but also the preservation of various stages in the process of introgression, thus increasing its chances of success.

Despite this evolution, the genetic variability of cassava remains structured, at least locally. For twelve loci observed, the two samples of M. esculenta differed more in the number of recombinations than in their allelic "richness". One wonders whether this situation is due merely to the recent history of the evolution of cassava on the African continent, or if it reflects some genetic disequilibrium.

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SEARCH FOR AND DISSEMINATION OF A CASSAVA CLONE  
THAT IS RELATIVELY TOLERANT TO  
AFRICAN CASSAVA MOSAIC

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#### INTRODUCTION

At the beginning of 1973, as part of a programme of agronomical support research for an agricultural development project in the maritime region (PRODERMA), the authorities in Togo asked IRAT to find a variety of cassava that offered a significant improvement in regional cassava production, which was then decreasing, and to disseminate it among the small scale farmers.

The main reasons for the low yields that were being obtained appeared to be:

- shortage of rain,
- exhaustion of the soils fertility,
- African Cassava Mosaic.

IRAT judged that a search for a variety with a significant resistance to the virus disease could provide the most rapid improvement in yields, so long as the proposed clone was accepted by the producers and consumers. To be accepted it would need, in particular, to be suitable for making gari, the speciality of the producers of the region, who earn part of their income from it.

#### THE WORK DONE

A programme of applied research was immediately set up, and this resulted five years later in the distribution among the farmers of a mosaic-resistant, high-yielding, well-received variety. The stages involved are described here.

##### Varietal collection

A bibliographic inventory of many plant materials was drawn up and the varieties reported for their tolerance to mosaic were chosen and then introduced into Togo at the Tsévié research station. In addition through searches in plantations in Togo and Benin, plant material notable for its vigour and its satisfactory resistance to the virus disease was collected.

Once they were planted, the varieties were observed and investigated particularly with respect to:

- their susceptibility to virus disease,
- their starch content,

- their production of roots,
- the attractiveness of the plant for the grower (colour of the petioles, habit of growth, easy roguing, etc.).

The results obtained with the best varieties in 1973 and 1974 are shown in Table 1, with the starch content calculated from the density and the table of G. Cours; the productivity was rated as 1=moderate, 2=acceptable, 3=average, 4=good; the attractiveness to the peasant farmers was rated as 1=acceptable, 2=average, 3=good; and mosaic was rated on Cours's scale, from 0 to 5, where 5=very susceptible.

Table 1. The range of cassava varieties investigated in one year at the Tsévié research station, 1973 and 1974.

No.	Variety name	% Starch content	Productivity	Attractiveness for peasant farmer	Mosaic on Cours's scale
1	312-524	19,10	4	3	1
2	NI	23,20	4	3	2
3	KONYEVE	16,60	4	2	4
4	H53	11,80	4	1	3
5	KATAOLI	20,30	3	3	3
6	C7	17,20	3	2	4
7	YOVOVI	21,60	3	2	3
8	H49	22,30	3	3	2
9	C3	13,10	3	1	4
10	HOUEDENOU	20,70	3	2	2
11	386 METE	24,40	3	3	2
12	N9	18,30	3	2	4
13	304 B.7	11,70	3	1	1
14	C2	20,20	3	1	4
15	H43	22,60	3	1	4
16	N11	21,90	3	1	4
17	HOUNLA	16,80	3	1	4
18	H35	16,60	3	1	2
19	HOMBETE	15,20	3	2	2
20	H51	16,40	3	1	2
21	DAZANEFETO	10,00	3	3	4
22	N4 C7	19,20	3	1	2
23	N3 C3	16,80	3	1	4
24	H56	21,20	3	2	4
25	H54	20,30	3	2	4
26	C1	14,00	3	1	4
27	AFAGNAN	13,20	3	3	4
28	C5	16,20	3	2	4
29	C6	13,60	3	2	4
30	ABIDJAN KOUTE	18,00	2	3	4
31	C9	17,70	2	1	2
32	N2	16,70	2	1	4
33	310-318	25,00	2	2	2
34	N5 C5	19,40	1	2	4
35	H34	20,40	1	1	4
36	N6 C9	25,30	1	1	4
37	ANKRAH	18,60	1	1	4



38	426-6	22,90	1	1	3
39	CRIOLINA	26,40	1	1	4
40	N7	19,40	1	1	4
41	N10	20,00	1	1	4
42	N8	16,20	1	1	4
43	GOULA	20,20	1	3	4
44	H44	11,80	1	1	4
45	C10	19,80	1	1	4
46	C4	15,90	1	1	4

For an initial selection of varieties from the collection, the clones with more than 19% starch and a susceptibility of 3 or less were chosen.

Initial classification of the varieties according to their susceptibility to the virus disease and starch content left us with these varieties:

No	Variety name	No	Variety name
1	312-524	10	HOUEDENOU
2	NI	11	386 METE
5	KATAOLI	22	N4 C7
7	YOVOVI	33	310-318
8	H49	38	425-6

Classification of the varieties according to their attractiveness to the peasant farmers and ability to produce high yields narrowed the choice of varieties down to six clones:

No	Variety name	No	Variety name
1	312-524	7	YOVOVI
2	N1	8	H49
5	KATAOLI	11	386 METE

These were satisfactory in respect of their susceptibility to the virus disease, starch content, productivity, and acceptability to the growers.

Finally, the roots harvested from these varieties and from the local varieties Goula and Kataoli were sold to gari makers in the Tsévié region. The processing procedures were followed by investigators from the research station, who gathered the impression and judgements of the personnel in charge of gari manufacture. The Goula variety gave complete satisfaction and was top of the list of preferences, immediately followed by 312-524 and Kataoli, which were judged as very acceptable. The other varieties were significantly desirable.

#### Comparison of production potentials

Even before knowing the outcome of the investigation of the collection, we made a preliminary comparison of the potentials of the varieties which, judging from the observations made during the investigations (312-524, 386 Mete) or from the gathered information (C3, C5, C7, C9), had a chance of being better accepted than the local varieties (Goula, Kataoli).

This first comparison was done in 1973 in the form of a block trial repeated eight-fold, with a planting density of 15,600 stalks per hectare, no fertilizer, on ground previously cultivated for three years with Centrosema pubescens and plough before the cassava planting (Table 2). The dates of planting and harvesting were 9 May 1973 and 10 May 1974, so that the growth cycle lasted 12 months. The rainfall recorded during this cycle was 1160 mm.

Table 2. Productivity and Disease Resistance of 8 Varieties of Cassava, 1973-1974.

Variety	Yield of fresh tubers (t/ha)	Starch content (%)	Susceptibility to mosaic (Cours'scale)
312-524	33,00-a	18,0	tolerant 1
C9	24,90-b	12,6	tolerant 2
C3	24,44-b	10,2	susceptible 3
C5	17,20---c	16,3	susceptible 4
C7	14,70---c	16,5	susceptible 4
386 METE	13,50---c	21,1	tolerant 2
KATAOLI	11,30---c	19,2	susceptible 3
GOULA	10,00---c	22,2	suceptible 4

- Coefficient of variation: 6,90%.

- Mean yield in the trial: 18,63 t/ha.

The variety 312-524 confirmed its high productivity and its low susceptibility to mosaic. On the other hand, the local varieties (Goula and Kataoli) had the best starch content.

The comparison of yields was then continued in 1974-1975, taking account of the data from the collection study: six-fold Fischer block trials were performed, using a planting density of 15,600 stalks/ha, and no fertilizer (Table 3). The previous crop was maize; the dates of planting and harvesting were 15 May 1974 and 11 June 1975, so that the growth cycle was 13 months long.

The rainfall recorded during trhe cycle was 1421 mm. Results obtained (Duncan's 0.05% test).

Table 3. Productivity and Disease Resistance of 6 Varieties of Cassava, 1974-1975.

Variety	Yield of fresh tubers (t/ha)	Susceptibility to mosaic (Cours'scale)
312-524	34,04 a	tolerant 1
H49	26,03-b	tolerant 2
386 METE	25,87-b	tolerant 2
KATAOLI	23,15-bc	susceptible 3
GOULA	21,08--c	susceptible 3
DAZANEFET	20,78--c	sensible 3

- Coefficient of variation: 13.65%

- Mean yield in the trial: 25.15 t/ha.

On the whole, yields were high, as the amount of rain recorded during the growing cycle was favourable. The variety 312-524 fulfilled its promise in respect of resistance to virus disease and productivity.

The experiments in 1976-1977 used 6-fold Fisher block trials, a planting density of 15,600 stalks/ha, and no fertilizer (Table 4). The previous crop was maize; the dates of planting and harvesting were 6 June 1976 and 6 May 1977, so that the growth cycle was 11 months long.

The rainfall recorded during the cycle was 547 mm. Results obtained (Duncan's 0.05% test).

Table 4. Productivity and Disease Resistance of 5 varieties of Cassava, 1976-1977.

Variety	Yield of fresh tubers (t/ha)	Susceptibility to mosaic (Cours'scale)	
N1	13,36 a	tolerant	2
312-524	13,24 a	tolerant	1
H49	11,40 a	susceptible	3
KATAOLI	8,72-b	susceptible	3
GOULA	7,17-b	susceptible	4

- Coefficient of variation: 18,70%.

- Average yield of the trial: 10,78 t/ha.

Note the general low level of the yields, mainly caused by the shortage of rainfall. Despite the difficult growing conditions, the variety 312-524 confirmed its resistance to the virus disease and its greater productivity compared with local varieties.

The experiments in 1977-1978 were 6-fold Fisher block trials, a planting density of 10,000 stalks, and fertilizer of N30-P205 30-K20 60 u/ha (Table 5). The previous crop was maize; the dates of planting and harvesting were 7 April 1977 and 1 May 1978, so the growth cycle was 13 months long.

The rainfall cycle recorded during the cycle was 1023 mm. Results obtained (Duncan's 0.05% test).

Table 5. Productivity and Disease Resistance of 5 Varieties of Cassava, 1977-1978.

Variety	Yield of fresh tubers (t/ha)	Susceptibility to mosaic (Cours'scale)	
N1	41,26 a	tolerant	2
312-524	39,00 a b	tolerant	1
H49	35,35-b	tolerant	3
KATAOLI	27,72--c	susceptible	3
GOULA	21,93---d	susceptible	3

- Coefficient of variation: 12,80%.

- Mean yield of trial: 33,05 t/ha.

The satisfactory rainfall allowed high yields. In addition, the provision of mineral fertilizer, mainly potassium, led to an increase in production, mainly for the new varieties. During these experiments over four years, with differing rainfalls, variety 312-524 was confirmed as a definite candidate for popularization, able to produce an improvement in production.

#### Offering the stocks to peasant farmers

From 1976 on, agents of the development project (PRODERMA) offered cuttings of 312-524 to growers who agreed to test the new variety on their plots. 312-524 soon drew attention in the countryside because of the vigor of the plant which was free of any sign of the virus disease infecting all vegetation around it. The harvest used for producing gari gave complete satisfaction to the growers. A constantly increasing demand for cuttings from the peasants ensured the success of 312-524.

#### Multiplication of the variety

To meet the demand, stocks of cuttings were set up at the Tsévié research station and in three other places in the region, Agbessi, Agbomedji, and Glope, and were made available to the development project. At present the National Institute for Tuberous Plants (INPT) of Togo is carrying on the propagation and dissemination of this clone from its base in Tabligbo.

It should be noted that from the beginning of this operation the cuttings planted were, as far as possible, systematically taken from healthy stalks. Though this could be done for 312-524, N1, and H49, which always had some virus-free stalks in the plots, this was not the case for Goula and Kataoli, all of whose stalks displayed some level of symptoms of the disease.

#### CONCLUSION

In a relatively short time, it has been possible to identify, test, and make available to the farmers a clone that is likely to improve the yield of cassava grown in the region. This improvement was possible because the growers concerned agreed to use the variety proposed, which did not alter their traditional mode of cultivation and lent itself to the traditional preparation of gari, while providing extra vigour and productivity. The success of this operation was due largely to the fact that from the beginning the factors favourable to unreserved adoption were identified and taken into account in the achievement of the work. So 312-524 has indeed "taken off" in Togo and at present it is also spreading into the neighbouring regions of Benin. However, though it represents a step up in increasing yields, improvement is still needed, especially in the starch content.

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## SOME EFFECTS OF AFRICAN CASSAVA MOSAIC DISEASE ON THE FIRST STAGES OF PLANT GROWTH

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### INTRODUCTION

African Cassava Mosaic is likely to infect cassava in two different ways:

- if the planting material is already infected and consequently the cuttings it produces develop plantlets that are infected from the beginning,
- if a plant that is healthy at the beginning becomes infected by Bemisia tabaci during cultural growth. Periods and pressures of infection in southern Ivory Coast are very unsteady (Fargette, 1985).

This paper aims to determine some effects of the virus at different plant growth levels: cutting, leaves, aerial parts, roots and tuberized roots, and their possible consequences likely to appear both at cycle end and during further cycles.

The results of several agronomical trials made between 1983 and 1986 at the ORSTOM experimental station at Adiopodoumé, are presented here. The CB variety was used in most of them, because the sanitation programme at ORSTOM virology laboratory enabled us to use a well defined planting material and virus-free cuttings. However, an infection was noticed in some plants from the early growth of young leaves, which enabled a comparison with other plants infected at different stages during the first weeks.

Though ACMV was not really studied, it was necessarily taken into account whenever the planting material was checked and all during young plants growth, because of the changes it caused in leaf surface.

The effects of ACMV on cuttings recovery and on planting material quality are first studied, then the evolution of the disease in time and as regards fertilization are presented.

### EFFECTS OF ACMV ON THE FIRST STAGES OF PLANT GROWTH

In two trials using the CB variety, it was possible among all plants available to gather similar cuttings as regards their date of virus infection. Thanks to a thorough weekly control, it was

possible to record for each plant the date of symptom emergence on 20 cm tall plantlets whose height as well as weight and number of internodes had been controlled from the beginning.

The chosen groups were composed of a minimum of 10 plants:

- plants that remained healthy for the whole period studied (S),
- plants on which virus symptoms were noticed from the early growth of young leaves (V),
- plants on which infection was noticed at 3rd, 4th and 5th controls - 2 to 4 weeks - (S-V, 2-4),
- plants on which infection was noticed at 5th, 6th and 7th controls - 6 to 8 weeks - (S-V, 6-8).

Only the first weeks of the growth cycle were studied, when competition between plant is considered unimportant: no competition either for light (incomplete soil cover), or for water (rain recovery or/and irrigation).

Growth measures made at the beginning of the cycle were weight of the different plant parts, number of stems, leaves, roots enabling a comparison between groups as regards their reaction to virus infection.

#### RESULTS AND DISCUSSION

Two types of results were gathered, on the one hand concerning the number of roots at 33 days (Table 1), and on the other hand those concerning the growth levels of aerial parts and tubers at 76 days (Table 2).

Table 1. Comparison between the number of nodal and basal roots of healthy cuttings (S) and of infected cuttings (V) for two different weights of cassava at 5 weeks.

Group	Cutting Fresh weight (g)	Number of Nodes	Number of roots	
			Nodal roots	Basal roots
S	40-65	6.9a	4.1a	16.7a
V	40-65	7.2a	3.4a	15.2a
S	90-120	10.8b	8.9b	21.4b
V	90-120	11.0b	7.6b	23.1b

a,b: along the columns, the followed values of a given letter are not significantly different for p=0.95.

Table 2. Action of virus infection at the beginning and during the cycle on the growth level of aerial parts (A) and on the initiation of tuberization (B) at 2,5 months.

A	Number of stems	Number of branches*	Leaves Dry weight (g)
V(n=18)	2.1a	2.1a	69.4a
SV.2-4 (n=11)	2.3a	2.6a	77.9ab
SV.6-8 (n=14)	1.9a	3.7b	92.0b
S (n=25)	2.2a	4.7c	127.2c

\* = branches that are not caused by flowering.

n = number of repetitions.

B	Roots % $\varnothing > 5$ mm	Total of $\varnothing > 5$ mm	Maximum M (mm)
V	23a	74.1a	8.7a
S-V.2-4	22a	91.3ab	8.2a
S-V.6-8	49b	126.4bc	13.1b
S	56b	163.6c	17.8c

a,b = The values that are followed by a same letter are not significantly different for  $p=0.95$ .

When considering the action of ACMV on rooting (Table 1), only the roots that were complete were counted. The number of roots developed by cuttings of equal weight was the same at the beginning, with the virus or without it. As all roots issued are similar anatomically and are all potentially likely to tuberize, it is the provision level in the cutting that will be able to act first upon the component of the yield number of tubers.

Results in Table 2 were obtained from the observation of plants whose original cutting weighed between 60 and 130 grammes for each group. No effect was noticed upon the number of stems produced per plant. Leaves' growth level was affected most as the virus was present early on the plant. The dry weight of leaves, grossly comparable to the photosynthesis surface, decreased by 45% as compared to the sample, in the extreme case of a plant infected from the beginning. This decrease was partly caused by a reduction in the number of branches that normally develop from side buds off the principal stems. This foliage limitation entails a limitation of the occupied space and the intraspecies competition for light, and/or with weeds and plants grown in association will occur at the expense of a given plant.

Taking into account the two yield component "number and weight" (represented here by diameter) of tuberized roots, infection is effective very early on tuberization, delaying the growing of primary axis both in number and on each root (Table 2B). For a tuberized root whose diameter is over 5 millimeters, the percentage of this type of root compared with the total number of roots is significantly decreased in plants infected in the first 5 weeks after planting. In other respects, the maximum diameter of



the biggest tuberized root on the plant decreases from 26 to 51% as compared with control. Whatever the phenomenon likely to arise after 2.5 months, the early disease occurrence causes a decrease in the accumulation of weight and in the number of tuberized roots on the individual plant.

#### ACTION OF ACMV UPON CUTTING QUALITY

When a 17 months old trial was harvested, a study was made to compare the stems of plant (CB variety) whose virus infection during the growth cycle had been recorded. In order to gather an adequate number of repetitions, the dates when the disease appeared on the last leaves were recorded during periods of 21 days for all plots with no significant difference. Only plants with two stems were included.

The diameter of the lowest part of each plant's biggest stem was measured about 5 cm above its insertion on the cutting. A single 20 cm long cutting was made from the stem's lowest part and its dry weight determined.

#### RESULTS AND DISCUSSION

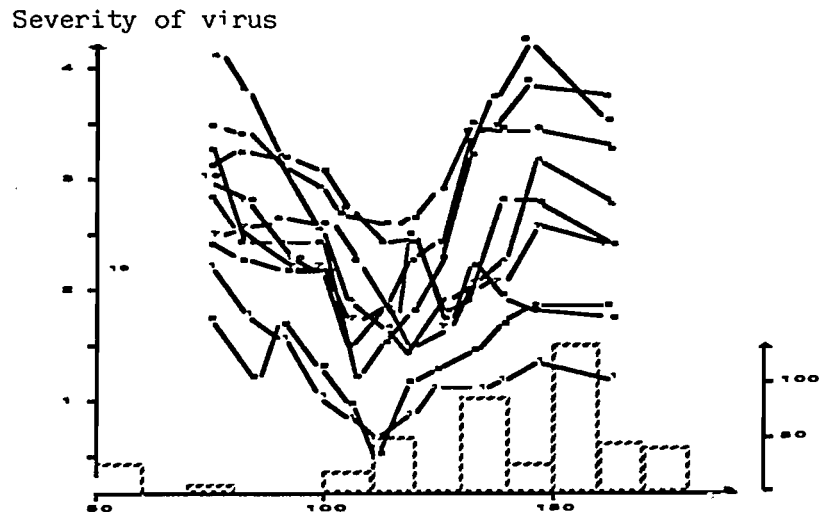
Table 3. Action of ACMV infection on a cassava stem at harvest and on a basic cutting provided by this stem.

Date of symptoms	Number of observations	Stem basal $\phi$	Basic cutting weight
2-4 weeks	17	25.9a	67.4a
6-8 weeks	25	26.3a	79.8a
10-12 weeks	27	31.4bc	109.8b
14-16 weeks	19	35.3c	117.4b

a,b = The values that are followed by a same letter are not significantly different for  $p=0.95$

The early infection of a cassava plant causes a decrease in stem diameter, which is in part the expression of an intraspecific competition in plant structure: healthy plants situated around an infected plant very early in the cycle bring shade for the latter. The reduction of limb surface of the plant itself may have caused a decrease in dry matter production. This has consequences on cuttings they will provide. In this manner the planting material quality is not as good and the number of roots decreases (cf. first part). A plant infected early in the cultivation cycle will, at harvest, provide cuttings which are likely to be more susceptible to climate changes (water stress for instance). The root number decreases, which is likely to change the first yield component, i.e. the number of tuberized roots.

Fig. 1: Evolution with time of the average intensity of the viral disease on the youngest completely developed leaf of 10 cassava clones infected at planting.

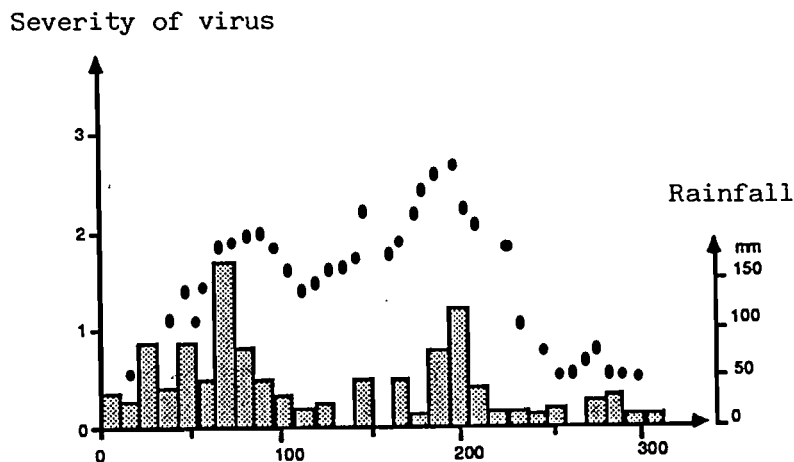


#### EVOLUTION OF ACMV DURING THE CULTIVATION CYCLE

Two trials enabled us to monitor the fluctuations of disease symptoms over time: one trial involved the comparison between 10 clones whose 20cm long cuttings had all been infected from planting, the other involved field preparation with the virus-free CB variety.

In both cases, a weekly control of leaf production rhythm was carried out by marking the youngest completely developed leaf from 0 (no symptom) to 5 (most severe symptoms), following the symptom scale of Cours (1951).

Fig. 2: Evolution of the average intensity of the viral disease on the youngest completely developed leaf in a plot of healthy cassava at planting (CB variety).



## RESULTS AND DISCUSSION

Figures 1 and 2 show the evolution of symptoms on the youngest completely developed leaf during 3.5 months and the whole cultural cycle, and present the amount of rainfall during the same period.

For the 10 clones considered, a general tendency of a decreasing severity of symptoms was recorded, coinciding with the end of the rainy season. Symptoms are minimum when the first rain recover and become more severe later on.

Figure 2 represents a different situation as plants were healthy from the beginning, and a first increase of symptoms can be seen on the trial, caused by the infection of a greater number of plants over time. At the end of the rainy season when contamination reached 100%, symptoms decreased then recovered with the arrival of the second rainy season. From the 7th month, after planting, the average mark heavily decreased.

The seasonal changes globally registered during the two trials are difficult to explain as the age of the plant must be considered. Nevertheless, it is interesting to note the effect of recovery of rains on symptom increase. Further experiments and a plant physiological control (nutritional state), coupled with the results of Fargette (1985) on whitefly population fluctuation, might give some explanation.

### EFFECTS OF ACMV ON CASSAVA LEAVES

To assess the nutritional levels of cassava plants by Foliar Diagnostic, a distinction was made between healthy and infected leaves. The reference leaf is the youngest completely developed leaf on each stem. Apart from the analysis of principal elements, area measures by planimetry and dry weight made it possible to record some effects of the disease on this type of leaf of the CB variety.

In a fertilization trial (Van Hanja, 1984, Raffaillac, 1984-b) with a local Bonoua-BR1 clone, the infection dynamics at cycle beginning could be analysed for the whole trial. For this purpose, three treatments were distinguished: N 0 = non fertilized sample, N 100 = 100 N units (urea) and N 200 = 200 N units (urea) applied at planting. Weekly recordings for infected plants were taken from the youngest completely developed leaf, whose median lobe length was measured, as an indication of limb surface.

## RESULTS AND DISCUSSION

Table 4: Effect of nitrogen fertilization upon ACMV cassava infection between 42-70 days of cycle and on a healthy leaf at 42 days.

Treatment	% of infected plants	Median lobe length of a healthy leaf (cm)	% N	% K
N 0	54.3a	16.6a	5.23	1.81
N 100	69.0b	17.5b	6.29	1.82
N 200	75.1c	17.6b	6.22	1.87

a,b,c = The values that are followed by a same letter are not significantly different for p=0.95

Nitrogen fertilization increased the contamination of the plants in this trial. This viral disease increase on fertilized plots must be compared to the healthy leaf's limb surface increase and to a tendency to concentrate, related to the total amount of nitrogen.

Table 5: Comparison at 72 days of the youngest completely developed leaf of healthy and infected cassava plants.

Group of plants	S (Healthy)	SV. 6-8 (Infected)	
		V3*	V5
Surface (cm <sup>2</sup> )	386a	208b	119c
Dry weight/ Surface (mg/cm <sup>2</sup> )	3.5a	3.9b	4.4c
% N	5.19a		5.85b
% P	0.42a		0.50b
% K	1.74a		2.07b
% Ca	0.66a		0.68a
% Mg	0.26a		0.31a

a,b,c = The values that are followed by a same letter are not significantly different for p=0.95.

\* = V3, V5 = Gravity index of Cours (1951).

This decrease in individual leaf area is nevertheless not constant in time and leaves infected differently may be observed on the same stem. This will not necessarily cause a photosynthesis decrease. The absorption of light is made easier by the canopy area decrease: an increased quantity of light was thus measured at mid-length of stems on a 3 month old plot of infected plants. This increase was between 20 and 65% depending the situation: one or two plants with 1 or 2 stems showed greater viral intensity than youngest leaves. In such situations, more leaves can work more efficiently; measurement of the efficiency of different types of leaves for the production of dry matter and its migration towards roots might bring some complementary information in this field.

The amounts of N, P and K elements in an infected leaf are significantly increased: the distinction between healthy and contaminated leaf must be made to estimate the nutritional level of cassava plants by "Foliar Diagnostic".

Intensity of infection depends upon how early infection takes place in the cultivation cycle, leaf level, ratio of healthy to infected plants in the plot and environmental factors. The prediction of yield loss from measurements of the decrease in limb area is likely to be inaccurate. Compensation takes place (Raffaillac, 1984-a), therefore individual yield loss must be distinguished from plot yield loss.

Loss of above ground vegetation caused by the disease does not necessarily imply yield loss in tuberized roots. As a matter of fact, an optimal level of Leaf Area Index (LAI) is known for each variety above which competition between aerial parts will limit tuberization (Cock et al, 1979). A small infection on vegetation where such a competition exists between aerial parts of plants will decrease competition for light and change the aerial/roots growth rate to the benefit of roots. Cours (1951) records such a phenomenon on a rich soil with an important foliar growth where increases reaching 20% from healthy plants to plants infected up to an intensity of 2 are recorded.

#### GENERAL CONCLUSION

The level of yields loss through ACMV varies for the different parts of the plant, and depending on whether the individual plant or total plant population is being considered.

The presence of the virus in the cutting is not the only factor which limits root tuberization. A decrease in stem growth leads to a decline in bulk (wet and dry weights). Propagation using infected plant material may lead to a cumulative decline in quality over a few generations.

Infection during a cultural cycle becomes less and less prominent on tuberization, as regards number and weight, as it occurs late in the season. The subsequent decrease in limb area will not necessarily imply a yield loss: the interaction between disease and competition must be taken into account.

Cassava's ability to compensate may conceal the effects of the disease on the individual plant, and therefore plant density on the trial plot becomes an important factor. The role played by environmental factors which encourage vegetation growth (e.g. nitrogen) must also be taken into account in the analysis of yield loss through ACMV. In the case of nitrogen fertilization, on the one hand plants are more rapidly infected but on the other hand an increased growth in aerial parts is likely to take place at the expense of the tubers (Raffaillac, 1984-b). In such a case leaf fall is likely to benefit tuberization if the Foliar Area Index already exceeds optimum value. Numerous experiments are still needed to confirm this.

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## USE OF ENZYME VARIABILITY FOR DESCRIBING AND SELECTING CASSAVA CULTIVARS

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Electrophoresis to reveal enzyme variability has been used in the biological study of cassava since 1982 (Zoundjihekpon, 1986). This technique has provided results used in the description and selection of cultivars resistant to African Cassava Mosaic.

### DESCRIPTION OF THE CULTIVARS

The description of cassava cultivars has until now been based on classic morphological and physiological characters such as the colour and shape of the various parts of the plant. The limitations of these characters have prompted a search for other, more reliable ones. Several authors (Cours, 1951; Caulliez and Fillias, 1979; Pouzet, 1980) have suggested that biochemical characters could be used; these have the advantage of being less affected by the environment and by human selection. The biochemical approach has therefore been considered for an improved description of cassava cultivars.

To describe the diversity of cassava cultivars and go on to specify the intraspecific systematics, the classic morphological and physiological characters have been used along with enzymatic ones. In this way 291 traditional cultivars, originating mainly from the Ivory Coast but also from the Central African Republic, and the Congo, have been analysed for 23 morphological variables: the shape and colour of the lobes of their leaves, the structure of the skin of the tubers, etc., and 9 enzymatic variables; this enables a final distribution of the cultivars of the collection into 8 classes. These have been described and defined by means of factorial analysis of correspondences and of hierarchical ascending classification (Zoundjihekpon, 1986). The discriminant analysis enabled us to show that this classification is satisfactory, since it leads to 91% of cultivars being well classified. Elsewhere, analysis of the enzyme variability showed that natural intracultivar hybridations occur. This idea had already been advanced by Cours (1951). In addition, it enabled us to show genotypic structures which were probably from the earliest ones introduced into Africa.

### SELECTION OF CULTIVARS RESISTANT TO CASSAVA MOSAIC

Twenty-eight clones tolerant of the viral disease, gathered and studied by plant virologists (Colon, 1984; Fargette, 1985) were also investigated electrophoretically. These were cultivars

originating from Kenya, India, and South America. The intraspecific hybrids were obtained in Kenya in programmes of improvement by Storey *et al.* (1938) and Idess (Bouakré). African Cassava Mosaic is one of the major phytosanitary problems, and several components of the resistance of cassava to this disease were investigated by Fargette (1985), who found that within the 28 clones investigated, there are 5 groups defined by their resistance to the vector (whitefly) and their resistance to the virus.

The electrophoretic analysis of each clone was performed. Their genetic structure were compared with the 5 classes previously defined. It seems that the clones that are resistant to the vector and those that are resistant to the virus belong to two quite different genetic groups. These results indicate that there is some genetic resistance to the virosis. In addition, most representatives of all the other genotypes are susceptible to the disease or to the vector. As this investigation was performed on a small sample (28 clones), more thorough and extensive study of a larger number of clones and enzyme systems is needed.

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**SEMINAR PROGRAMME**

**AFRICAN CASSAVA MOSAIC DISEASE AND ITS CONTROL**

**4 - 8 May 1987, Yamoussoukro, Côte d'Ivoire**

Sunday 3 May                   Arrival of participants at the IBIS Hotel  
                                  in Abidjan.  
                                  Registration in the Reception Hall

Monday 4 May  
morning                   9H00   Bus transport to Yamoussoukro

afternoon               15H00   Opening  
                                  **Chairman:**                   Dr. GODO

                                  Welcome address by the Mayor of Yamoussoukro

                                  Address by Mr. ASSOUMOU MBA, Director of CTA

                                  Address by Mr. DOUMENGE, President of ORSTOM

                                  Address by Mr. M'BIELE, Director of IAPC -  
                                  Official OAU Representative

                                  Opening address by the Minister of Scientific  
                                  Research of the Ivory Coast

Tuesday 5 May

morning

9H00 **SESSION A - CURRENT SITUATION**

**Chairman:** Mr. ASSOUMOU MBA (CTA)

**Rapporteurs:** Mr. HAINNAUX (ORSTOM)

Mr. VAN DER GRAAFF (FAO)

9H00 General introduction to the Seminar by  
Mr. FAUQUET, Seminar Coordinator

9H15 General presentation of African Cassava  
Mosaic Disease

Dr. GUTHRIE (SCOTLAND)

9H40 General summary of national  
questionnaires

Dr. GODO (ORSTOM)

10H30 Coffee break

11H00 African Cassava Mosaic Disease in Congo:  
importance, spreading and control  
methods

Dr. MASSALA (CONGO)

11H30 Impact of African Cassava Mosaic Disease  
on plant growth and yield

Dr. FAUQUET (ORSTOM)

12H00 Integration of control methods of the  
disease

Dr. THRESH (ODA)



Tuesday 5 May

afternoon

15H00 **SESSION B**

**Chairman:** Mr. FARGETTE (ORSTOM)

**Rapporteurs:** Dr. IGWEGBE (NIGERIA)

Prof. FOUABI (ENSA)

15H00 African Cassava Mosaic geminivirus:  
properties and geographical variations  
Dr. HARRISON (SCRI)

15H30 The etiology of Cassava Mosaic (CMD)  
in Nigeria  
Dr. ROSSEL (IITA)

16H00 The role of Bemisia tabaci Gennadius in  
the epidemiology of ACMV in East Africa.  
Biology, population dynamics and  
interaction with cassava varieties  
Mr. ROBERTSON

16H30 Coffee break

17H00 Monitoring Bemisia tabaci populations in  
cassava: field counts and trap catches  
Dr. FISHPOOL (TDRI)

17H30 Evaluation and exploitation of cassava  
genetic variability  
Dr. CHARRIER (ORSTOM)

18H00 Microclimate in a cassava field  
Dr. YAO (ORSTOM)

Wednesday 6 May

morning

9H00 **SESSION C - EPIDEMIOLOGY**

**Chairman:** Dr. ROSSEL (IITA)

**Rapporteurs:** Dr. FRISON (IPBGR)

Dr. NOLT (CIAT)

9H00 Virus/vector/plant relationships

Dr. FARGETTE (ORSTOM)

9H30 Distribution and propagation of African  
Cassava Mosaic Disease in a cassava field

Dr. FARGETTE (ORSTOM)

10H00 Automatic mapping of the spread of  
African Cassava Mosaic Disease

Dr. LECOUSTRE (CIRAD)

10H30 Coffee break

11H00 Development of the African Cassava  
Mosaic Virus

Dr. FARGETTE (ORSTOM)

11H30 Some aspects of the epidemiology of  
African Cassava Mosaic Disease in  
coastal districts of Kenya

Dr. BOCK (ICRISAT)

12H30 Epidemiology of African Cassava Mosaic  
Disease at regional level in the  
Ivory Coast

Dr. FAUQUET (ORSTOM)

Wednesday 6 May

afternoon

15H00 **SESSION D - SANITATION**

**Chairman:** Dr. DIOMANDE (AISA)

**Rapporteurs:** Dr. ATCHAM (CAMEROON)

Dr. SAUTI (MALAWI)

15H00 Tissue culture and cassava sanitation

Dr. FRISON (FAO)

15H30 Viral sanitation of potato crop  
in France: techniques, organisation  
and control

Dr. PERENNEC (INRA)

16H00 Selection of healthy cassava obtained  
by reversion in the cassava fields

Dr. FAUQUET (ORSTOM)

16H30 Coffee break

17H00 African Cassava Mosaic, cassava  
and farmers

Dr. MOLLARD (ORSTOM)

17H30 State of knowledge on cassava  
bacterial blight in Africa:  
epidemiology and vector control

Dr. DANIEL (ORSTOM)

Thursday 7 May

morning

9H00 **SESSION E - BREEDING - RESISTANCE**

**Chairman:** Dr. VAN DER GRAAFF (FAO)

**Rapporteurs:** Dr. KONATE (BURKINA FASO)

Dr. IGWEGBE (NIGERIA)

9H00 Host-virus relationships of resistant cassava and African Cassava Mosaic Disease, and some implications for breeding and disease control

Dr. JENNINGS (SCRI)

9H30 Statement of results at IRAT concerning the control of African Cassava Mosaic Disease in Madagascar and West-Africa

Dr. ARRAUDEAU (CIRAD)

10H00 Cassava resistance to African Cassava Mosaic Disease

Dr. FAUQUET (ORSTOM)

10H30 Coffee break

11H00 African Cassava Mosaic Disease in India: geographical and economic importance and control methods

Dr. MALATHI (INDIA)

11H30 The African Cassava Mosaic Disease in Zaire and its Control

Dr. MAHUNGU (ZAIRE)

12H00 Importance, production and utilization of cassava in Uganda

Dr. OTIM NAPE (UGANDA)

Thursday 7 May

afternoon 15H00 **SESSION F - CONTROL STRATEGIES/DISCUSSION SESSION**

**Chairman:** Dr. HARRISON (SCRI)

**Rapporteurs:** Dr. FISHPOOL (TDRI)

Dr. M'BAYE (SENEGAL)

**Discussants:** Dr. BOCK (ICRISAT)

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Dr. ROSSEL (IITA)

Dr. SYLVESTRE (CIRAD)

Dr. THRESH (ODA)

Dr. VAN DER GRAAFF (FAO)...

Friday 8 May

morning 9H00 **THEMATIC REPORTS ON THE SEMINAR**

**Chairman:** Mr. DELLERE (CTA)

9H00 Actual situation Dr. GODO (ORSTOM)

9H15 People involved Dr. BOCK (ICRISAT)

9H30 Epidemiology Dr. THRESH (ODA)

9H45 Genetic aspects Dr. CHARRIER (ORSTOM)

10H00 Control strategies Dr. HARRISON (SCRI)

10H15 Coffee break

Friday 8 May  
morning

**CLOSING**

- 10H45 Conclusions by the Coordinator  
Dr. FAUQUET (ORSTOM)
- 11H00 Closing address by the Minister for  
Rural Development of the Ivory Coast

Friday 8 May  
afternoon

- 13H00 Bus transport back to Abidjan
- 16H30 Visit to the ORSTOM Centre at Adiopodoumé
- 18H30 Bus transport back to the Hotel IBIS in  
Abidjan

## THE TECHNICAL CENTRE FOR AGRICULTURAL AND RURAL CO-OPERATION (CTA)

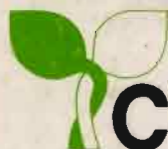
The Technical Centre for Agricultural and Rural Co-operation (CTA) was established in 1983 at Ede/Wageningen. It operates under the Lomé Convention between Member States of the European Community and the ACP States. CTA is at the disposal of the ACP States to provide them with better access to information, research, training and innovations in the field of agricultural and rural development and extension.

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