Distribution and current status of cassava mosaic disease and begomoviruses in Guinea

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Abstract

Several begomovirus species and strains causing cassava mosaic disease (CMD) have been reported from cassava in Africa. A diagnostic survey was made in the four agroecological zones of Guinea to determine the status of CMD and cassava begomoviruses and to ascertain if the virulent Ugandan variant of the East African cassava mosaic virus (EACMV-Ug2) was present. In the southern Guinea savanna; humid forest, midaltitud savanna, and derived savanna, 88 farmers’ fields were visited. Each field was assessed for the incidence and severity of the disease. The CMD status was rated as mild, moderately severe, or severe. Cassava leaf samples were collected from plants showing the disease symptoms from farmers’ fields on which CMD severity was also rated on a 5-point scale. Whitefly populations were estimated in each field as low, moderate, or high. Samples of diseased leaves were sent to Germany, DSMZ Plant Virus Division, for the identification of associated pathogens. The CMD status in most farmers’ fields was moderately severe or severe. The lowland humid savanna had the highest severity (2.67) and ranged from 1.9 to 4.07. Only one field with an improved variety in the humid forest did not have any CMD symptoms. Across the country, the CMD symptoms were found in 73.61% of the farms. The highest incidence of the disease was in the derived savanna ecozone (81.9%). The mid altitude savanna had the lowest incidence (64.9%). Whitefly was found in all the fields surveyed. The highest populations were found in the midaltitude and derived savanna, while the lowest were obtained in the humid forest and southern Guinea savanna agroecologies. Differential PCR for ACMV, EACMV, EACMCV, and all other African and non-African begomoviruses in cassava conducted at DSMZ with leaf samples provided on FTA cards showed that only 22 PCR positive samples were obtained: five were infected with ACMV only; seven were infected with EACMCV only; and 10 were infected with mixtures of EACMCV and ACMV. There was no indication of other virus strains present in the country. This is the first report of EACMCV and mixtures with EACMCV and ACMV in Guinea.

Key word: cassava, mosaic virus, Guinea, whitefly

Introduction

Cassava (Euphorbiaceae: Manihot esculenta Crantz) is Africa’s second most important food crop; accounting for approximately one-third of the total staple food production. It was introduced into West and East Africa from Brazil at two independent events by Portuguese slave ships: western coast in the seventeenth century and eastern coast in the eighteenth century (Jones 1959). Both socioeconomic and biological constraints limit the average yield to about 10 t/ha, a value that is far below the potential yield of between 30.8 and 51 t/ha as estimated at research stations (Hahn et al 1989).

The most important disease and principal constraint affecting cassava production in sub-Saharan Africa is the cassava mosaic virus disease (CMD) (Hahn et al 1980; Legg and Fauquet 2004). The disease has been reported in all cassava-growing countries in Africa and.
in the Indian subcontinent. Yield losses are enormous, especially if susceptible cultivars are cultivated (Faragette et al 1988; Otim-Nape et al 1994). On such cultivars, 90 to 100% yield losses were recorded in Uganda during a recent CMD pandemic (Otim-Nape et al 2000). These yield losses are due mainly to a reduction in photosynthetic leaf area (Theberge 1985; Allem and Hahn 1991).

The disease is associated with several whiteflies (Bemisia tabaci)–transmitted geminiviruses belonging to the genus Begomovirus, family Geminiviridae, which are characterized by having ssDNA genomes with a twin-isometric particle morphology (Harrison 1985; Brown et al 1995). Several viral species of the begomoviruses occur in cassava (most common natural host) in sub-Saharan Africa. These include African cassava mosaic virus (ACMV) (Bock and Woods 1983; Guthrie 1987), East African cassava mosaic virus (EACMV), which has been reported from five East African countries, including Madagascar (Hong et al 1993; Swanson and Harrison 1993; Harrison et al 1997), as well as NIGERIA (Ogbe et al 1999) and Ghana (Offei et al 1999), East African cassava mosaic Cameroon virus (EACMCV) identified in Cameroon and first described by Fondong et al (1998); and Fondong te al (2000) has also been found in Nigeria (Ariyo et al 2003) and Côte d’Ivoire (Pita et al 2001). East African cassava mosaic Malawi virus (EACMMV), East African cassava mosaic Zanzibar virus (EACMZV) (Maruthi et al 2002), South African cassava mosaic virus (SACMV) is confined to the South African Republic (Berrie et al 1997, 1998, 2001; Rey and Thompson 1998) and Madagascar (Ranomenjanahary et al 2002) while a further distinct begomovirus species from cassava in Zanzibar described by Maruthi et al (2002) as the East African cassava mosaic Zanzibar virus (EACMZV) is confined to coastal regions of East Africa (Were et al 2004).

With the re-emergence of a severe epidemic form of CMD in Uganda in the 1980s, a new begomovirus type was described, referred to as either the Uganda variant virus (Zhou et al 1997) or a distinctive strain of EACMV (EACMVUG) (Deng et al 1997). This virus evolved as a recombination event between genomic segments of DNA-A of EACMV and a coat protein fragment of ACMV. Infections with this distinct strain of EACMV (EACMV-UG) (Deng et al 1997) resulted in severe disease symptoms, especially in mixed infections with ACMV (Harrison et al 1997). This virus is highly aggressive and was reported to be moving at a rate of 20 km/year (Gibson 1996; Otim-Nape et al 1997), devastating cassava fields. The rapid spread of this virus from Uganda to neighbouring countries (Legg and Ogwal 1998) and especially recent reports confirming the presence of EACMV-UG2 in the Democratic Republic of Congo (DRC) and Congo Republic (Neuenschwander et al 2002), as well as in Gabon (Legg, personal communication), indicated its movement towards the West African countries where ACMV predominates. This apparent westwards expansion poses a serious threat to cassava production in West Africa. In addition, mixed infection of ACMV and EACMV, which gave rise to the “Uganda variant” have already been reported from major cassava growing countries of West Africa. Nigeria, Ghana, Togo, Côte d’Ivoire, and Guinea (Fondong et al 1998; 2000; Offei et al 1999; Ogbe 2001; Ogbe et al 1999; 2003). Mixed infections are more severe than a single infection on cassava plants, and may provide further opportunities for the recombination into more virulent strains. In view of the current importance of cassava in the subregion, threats to production must be considered very seriously and preventive strategies should be vigorously pursued with urgent priority. This study was therefore undertaken to determine the status of CMD and cassava begomoviruses, and to ascertain if the virulent Ugandan variant of the East African cassava mosaic virus (EACMV-Ug2) was present in Guinea.

Materials and methods

Farmers’ fields were surveyed across the four agroecological zones of Guinea between 8 and 30 August 2005. The survey followed the method described by Ogbe et al (2003). The number of cassava farms examined in each ecozone varied, depending on availability. A total of 88 farmers’ fields were surveyed: derived savanna (26); humid forest (27); midaltitude savanna (17); southern Guinea savanna (18). The geographic position of the each field was recorded using the Global Positioning System (GPS). Cassava fields were selected at regular intervals of at least 20 km, and the assessment of disease severity was made on 30 randomly selected plants along two diagonals in each field. Each plant was rated on a scale of 1–5 in which 1 represented no symptoms and 5, the most severe symptoms including severe chlorosis, leaf distortion, and plant stunting (Hahn et al 1980). The whitefly population was estimated by recording the number of adult whiteflies on the five topmost apical leaves on a scale of 1–3 where 1=...
low (1 – 50 whiteflies/plant), 2 = moderate (50 to 100 whiteflies/plant), and 3 = high (> 100 whiteflies/plant). On average, two leaf samples were collected in each field, depending on the degree of CMD severity, using the FTA Plant Virus Division in Germany for identification.

Detection and differentiation of begomoviruses in cassava. To provide a high resolution of the virus(es) present in a respective sample, a polymerase chain reaction (PCR) assay was followed using differential primers amplifying specific virus species or, a subset of strains. Sixteen primers with unique sequences to each begomovirus were used to discriminate among virus species and strains. Two primers, Begomo 146 and 672, from sequences that are common to all cassava begomoviruses were included for virus detection purposes (Ariyo et al 2005).

For PCR analysis, leaf samples were subjected to DNA isolation, essentially following a plant DNA minipreparation method (Dellaporta et al 1983). From each DNA preparation, 1 µl was subjected to PCR in a total volume of 50 µl. The reaction consisted of 2.5 µl MgCl₂ (50 mm), 1 µl of each primer (100 qmol), 5 µl of 10X Taq polymerase buffer, 1 µl dNTPs (10 mm) and 0.5 µl (2.5 units) of Taq DNA polymerase (Gibco BRL, Karlsruhe, Germany). After an initial denaturation step of 3 min at 95 °C, 35 PCR amplification cycles were conducted, consisting of 1 min denaturation at 95 °C, 1.5 min primer annealing (at temperatures specified for the respective primer combinations) and 1 min strand extension at 72 °C, PCR amplification was terminated by a final extension period of 10 min at 72 °C. After PCR, 10 µl of each reaction was subjected to gel electrophoresis in 1% agarose gels to evaluate PCR-amplified virus genome fragments.

Discussion

This study on the distribution of geminiviruses in cassava was intended to present an update on the status of CMD and the occurrence of cassava mosaic begomovirus species and strains in Guinea. The highest incidence of CMD was obtained in the derived savanna, and the Kindia region. This is in agreement with the results obtained by Okao-Okuja et al (2004) during their survey from November to December 2002 in Guinea and Senegal. Wydra and Msikita (1998) noted that low average CMD incidence was regionally observed on different ecozones (Cameroon, wet savanna (29%), Cameroon, mountain forest (38%), Bénin, transition forest (45%), whereas in most other ecozones, plant incidence was between 64 and 97%. The higher severity (2.93), found in the derived savanna, is in agreement with the observations of Wydra and Msikita (1998) who reported that the average severity of infected plants varied between score 2.2 (Bénin, transition forest) and 3.4 (Cameroon, wet savanna) on the scale of 1 to 5.

The highest populations of whiteflies were found in the mid-altitude and derived savanna. This showed an apparent relationship between CMD incidence, severity, and whitefly populations and was in accordance with the findings of Muimba-Kankolongo et al (1998). They found higher incidence and severity of CMD in southern Africa during warm periods when whitefly populations, and activity were highest. However, in Nigeria, vector population, CMD incidence, and severity were highest on plants established in the forest-savanna transition, followed by those in the humid rain forest and were lowest in the northern Guinea savanna (Akano et al 1998). This suggested that the vector has great influence on CMD occurrence. There is a need for more detailed studies over the course of the season in the derived savanna of Guinea and for breeders to disseminate resistant varieties quickly to farmers. The virus distribution map of West Africa can now be extended to Guinea.

The need to protect cassava against this virus is, therefore, a crucial aspect in enhancing the production of the crop. Of the various measures that have been employed to control CMD, the two main approaches are the use of virus-resistant varieties and phytosanitation (Thresh and Cooter 2005). The use of resistant varieties has remained the most economical and ecologically sustainable control measure (Jennings 1994). Breeding for resistance to CMD and the search for virus-resistant genotypes started in

Results

Geographical distribution of cassava mosaic diseases. In all the four major ecozones, CMD was present and was observed in 96% of the fields visited in the humid forest (Fig. 1). The mid-altitude and southern Guinea savanna generally had low disease severity scores, while the humid forest and derived Guinea savanna had higher scores. The highest mean severity score was recorded in Kindia (Mambia) (4.07); other regions with high severity scores were Kindia (Bokaria) (3.77), Mamou (Ourekaba) (3.73), Mamou (Kenzy) (3.70) Pita (Bendougou) (3.73), N’zérékoré (Borta) (3.70) (Fig. 1).
Fig 1. Guinea, showing the distribution of farms with different levels of CMD severity across the various agroecological zones.

Fig 2. Severity of CMD in four ecologies in Guinea.

Fig 3. Incidence of CMD in four agroecologies in Guinea

Fig 4. Distribution of whitefly populations in cassava fields surveyed in Guinea.

Fig 5. Whitefly populations on cassava fields in four agroecologies in Guinea.
the late 1920s (Otím-Nape et al 1998). Since then, there have been trials of cultivars and selections in several countries including Kenya, Tanzania, Uganda, Madagascar, Zaire (now Democratic Republic of Congo) and Nigeria (Jennings 1994). TME 4 and 96/1089A, selected recently at IITA (Ibadan, Nigeria), remain solid and resistant to EACMCV or ACMV or both infections, even with high virus pressure.

Higher severity scores of 2.93 were obtained for cassava farms in the derived savanna; farms in the other ecozones generally had severity scores ranging from 2.46 to 2.67 (Fig. 2).

The incidence of CMD was higher in the derived savanna (81.9%) but lower in the midaltitude savanna (64.9%) (Fig 3).

More fields were recorded in the category of 90-100% incidence than in any other and more than 60% had an incidence score > 80% (Fig 4).

Whitefly populations varied across fields in all the agroecological zones. The highest number of whiteflies / plant was found in the midaltitude savanna; the humid forest and southern Guinea savanna had the lowest populations (Fig 5).

Low (37.5%), moderate (19.3%) and high (43.18%) populations of whitefly were recorded in all fields visited (Fig. 6).

From the 22 samples that tested positive: 5 were infected with ACMV only 7 were infected with EACMCV only 10 were infected with EACMCV and ACMV. This means that EACMCV is the most prevalent virus in all fields visited and we have a 50/50 situation of EACMCV and ACMV.

**References**


