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Evaluation of the seroprevalence of okra mosaic virus in Koulikoro, Mali

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Mosaic virus disease is transmitted by several types of viruses such as OMD, OYVMV, OLCV, CLCuGV, and CYCrV. Among these viruses, some of them are significant threats to okra production worldwide. To develop an integrated pest management strategy against this disease, this seroprevalence study was conducted at the Rural Polytechnic Institute (RPI) of Katibougou, Koulikoro region, Mali. Symptomatic leaves of okra plants were collected in Koulikoro. The symptoms were mosaic associated or not with other symptoms. For this purpose, 52 symptomatic samples and 3 asymptomatic samples collected in the villages of Niarébougou, RPI Katibougou, and Diakitébougou were analyzed. The DAS ELIZA Okra Mosaic Virus (OkMV) kit was used. Among the symptomatic samples, the analyses showed the presence of 13 positive samples. This indicates a prevalence of 25% in the study area. Okra is infected by the okra virus disease called Okra Mosaic Virus (OkMV), which is the first time in Mali to be detected by the ELIZA test. This study shows the prevalence of the okra mosaic virus in Mali and more precisely in Koulikoro. This study shows the importance of keeping OKMV in consideration among the threats to okra cultivation in this area.

Key words: Seroprevalence, okra mosaic virus, Mali.

INTRODUCTION

Okra, *Abelmochus esculentus*, is a tropical plant native to Africa. It is cultivated for its nutrient richness and importance in the diet of urban and rural populations (Khomsug et al., 2010). Okra does not require significant processing before consumption, the fruits are generally sold fresh, sometimes dried into a powder (Ouoba et al., 2010). These young leaves and fruits are edible as a sauce in most West African countries. The fruit contains many nutrients (calcium, iron, carbohydrates, proteins, vitamins, etc.) that are necessary supplements to the basic diet consisting mainly of starch (cereals and tubers) among African populations.

The economic importance of okra, its diverse uses, and

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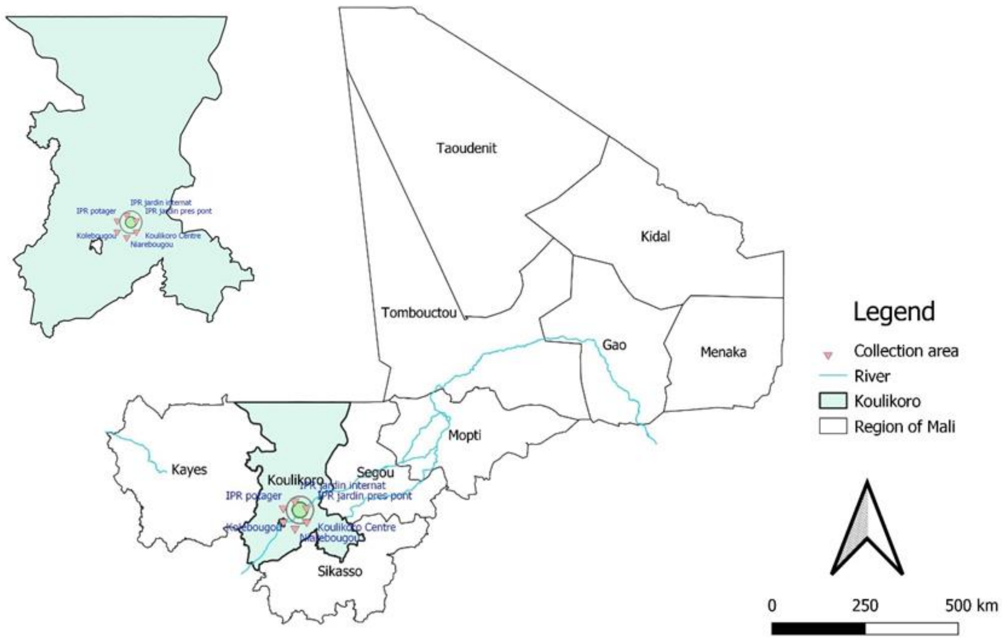

Figure 1. Okra sample collection site.

its nutritional value in the diet of the population make this crop a real tool for poverty alleviation in rural, urban, and peri-urban areas (Fondio et al., 2011). Unfortunately, this crop is confronted with the harmful action of several diseases and pests. These diseases can be of several types: viral (mosaic, leaf curl) and fungal (Cercosporiosis, fusariosis) (Ugwoke and Onyishi, 2009). In West Africa, okra mosaic virus is widespread and is usually transmitted by insect vectors (Givord and Hirth, 1973; Asare-Bediako et al., 2017). Vector control, therefore, seems to be an effective means of prevention. Chemical control of these insect vectors is widely used but the resulting intensive use of insecticides has a heavy environmental impact (Asare-Bediako et al., 2014a). Because of these undesirable effects, it is necessary to seek effective control methods that respect human and environmental health, such as farming techniques of crop associations (Kadri et al., 2013). Okra mosaic virus disease is manifested according to okra species, leaves may be covered with a coarse chlorotic mosaic or a band of veins and a pale green or yellow mosaic or irregular chlorotic areas (Givord and Hirth, 1973; Givord, 1977). Symptoms also vary according to growth stage, on the first diseased leaves a few days (7-8) after infection, they may show a green or yellow mosaic or chlorosis of the regular veins, and on the second and third diseased leaves, one to three of the central veins may be bordered by wide chlorotic bands of variable width. More rarely, symptoms are a mixture of dark green and light or whitish areas of irregular shape on the lamella (Givord and Hirth, 1973; Givord, 1977). This study aimed to search for these symptoms in Koulikoro, Mali, and to estimate the disease seroprevalence.

MATERIALS AND METHODS

Study location

This is a cross-sectional study. Okra leaf samples were collected in Koulikoro, Mali's second administrative region, during the year 2020. The study area has a Sudano-Sahelian climate characterized by a short rainy season of 4 to 5 months (late June to September) and a long dry season of 8 to 9 months (October to June). Annual rainfall is low at the beginning and reaches its maximum in August (600 to 900 mm).

Sampling involved young leaves with symptoms resembling those caused by okra viruses. Symptoms sought on the leaves were: mosaics, discoloration, waffling and rolling.

Samples were collected in the Koulikoro region, specifically in the vegetable garden, the boarding school garden, near the bridge and Diakitébougou (at the RPI), Niarébougou and the center of Koulikoro (Figure 1). A total of three non-symptomatic samples were collected. The collection of symptomatic samples was done during the pre-wintering and post-wintering period. Also, three young asymptomatic leaves samples were collected to serve as a control. One sample consisted of a diseased plant. The samples collected were labeled according to the place of collection and were then transported at 4°C to the Laboratory of Applied Molecular Biology (LBMA) in Bamako within the same week. Samples were stored in a freezer at -80°C until analysis.

Serological detection

Identification of Okra Mosaic Virus (OkMV) in the collected okra leaf samples was done by a Double Antibody Sandwich - Enzyme Linked Immuno Sorbent Assay (DAS-ELISA). The test was conducted according to the manufacturer's instructions (ACD, Inc.,

Table 1. Characteristics of okra leaves samples collected.

Site	Symptoms			Total number
	Leafroll + Mosaic	Leafroll + Mosaic + Yellow spots	Leafroll mosaic + Waffling	
RPI Diakitebougou	2	0	0	**2**
RPI boarding garden	0	1	0	1
RPI garden near bridge	2	0	0	2
RPI garden	6	0	0	6
Kolebougou	15	0	0	15
Koulikoro Downtown	5	0	0	5
Niarebougou	0	0	21	21
Total number	30	1	21	52

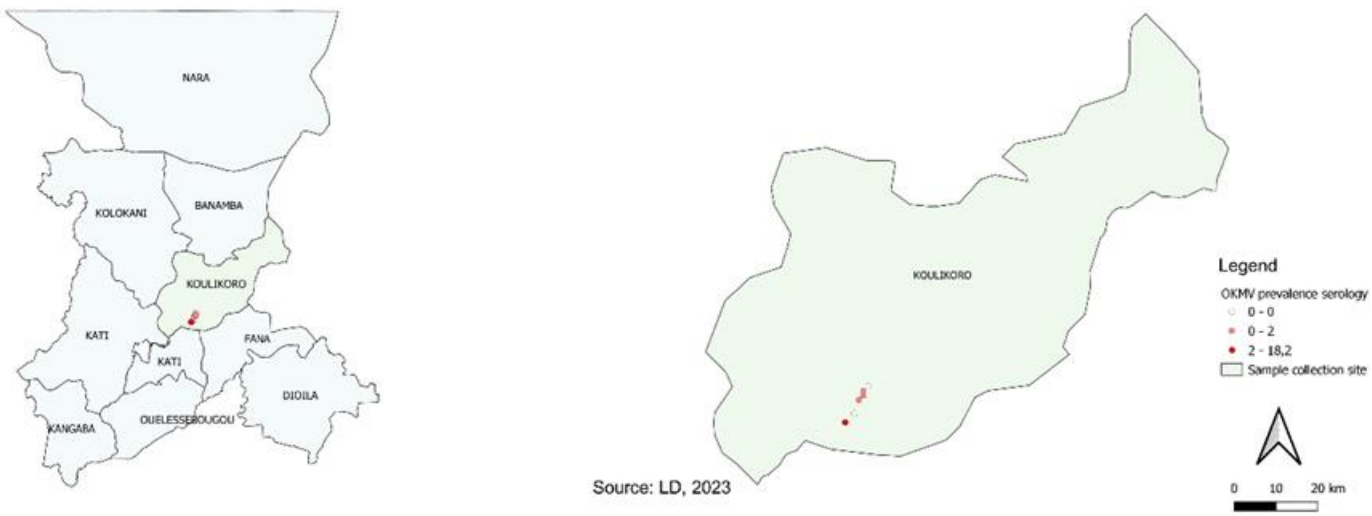

Figure 2. Distribution of Okra Mosaic Virus seroprevalence in the collection site.

Nano Diagnostics, LLC, Fayetteville, USA; https://www.nanodiaincs.com).

Reading of positive or negative reactions was done using an automated 405 nm ELISA plate reader (Biotek ELx808).

Test results were validated only if the positive control wells gave a positive result (yellow staining) and the negative control wells remained clear.

Data entry and analysis

The data were recorded in a logbook, the results obtained after the serological analysis were entered in an Excel file. Data analysis was done using R software. Mapping was done using QGIS software version 3.16.3-Hannover.

RESULTS AND DISCUSSION

Characteristics of collected samples

The study was carried out in the Koulikoro region, which is an agricultural and market gardening area per excellence. Table 1 presents the symptoms observed and the number of samples collected; a total of 52 symptomatic samples and 3 non-symptomatic samples. Symptoms encountered during this study were: mosaic-associated or not, leafroll plus yellow spot (RPI, boarding garden), leafroll plus waffling (Niarebougou), and leafroll alone in the other collection sites.

It was found that mosaic was most often associated with other symptoms in this study. This finding was reported by Konaté et al. (1995) in their study. Waffling and curling symptoms generally cause much more losses (N'Guessan et al., 1992; Kumar et al., 2010; Tiendrébéogo et al., 2010; Haruna and Jabil, 2017).

The study found an overall okra mosaic virus (OkMV) seroprevalence of 25% (13/52) (Figure 2 and Table 2). A study carried out by Zaharaddeen Samaila et al., 2021 in Nigeria found similar results to this study in their different sample collection areas: 20%, 22%, and 31% in Zamfara, Kaduna, and Lere states respectively (Zaharaddeen Samaila et al. 2021).

The highest prevalence was found in an area closer to the river (Kolebougou site) among the other collection sites (Figure 2 and Table 2). The results in Table 3 are in

Table 2. Distribution of samples according to collection sites and serological results.

Collection site	Serology OkMV	
	Negative (%)	**Positive (%)**
RPI Diakitebougou	2 (3.6)	0 (0)
RPI boarding garden	0 (0)	1 (1.8)
RPI garden near bridge	1 (1.8)	1 (1.8)
RPI garden	5 (9.1)	1 (1.8)
Kolebougou	5 (9.1)	10 (18.2)
Koulikoro Downtown	5 (9.1)	0 (0)
Niarebougou	24 (43.6)	0 (0)
Total	42 (76.3)	13 (23.6)

Table 3. Classification of symptoms according to optical density.

Symptoms	Number	Optical density mean
Mosaic	0	0
Leafroll	0	0
Waffling	0	0
Mosaic + Leafroll	12	2.865
Mosaic + Leafroll + Waffling	0	0
Mosaic + Waffling	0	0
Mosaic + Leafroll + Yellow spots	1	4.023

agreement with the research work of Kouamé (2016) in Ivory Coast who mentioned that the symptom groups (mosaic plus embossing plus upward curl and embossing plus upward curl) as well as the group (mosaic plus 14.52%. Several studies report variation in the incidence of okra virus disease by symptoms and geographical areas (Kouame, 2016). Appiah et al., 2020, reported more cases of okra mosaic virus (OkMV) than okra yellow vein mosaic virus (OYVMV) in symptomatic okra leaves in their study. However, they also reported that okra yellow vein mosaic virus was more abundant than OkMV (Appiah et al. 2020). A study in Ghana reported the incidence of okra mosaic disease varying from 78 to 83% and okra leaf curl disease varying from 63 to 70% (Asare-Bediako et al., 2014b). Moreover, Asare-Bediako in 2019 reported that okra mosaic diseases are commonly observed in okra crops in Ghana, with disease incidence up to 100% depending on the okra cultivar and growth stage (Asare-Bediako 2019). According to Konaté et al. (1995), the average incidence varies by geographical area, 51% for Sahelian zone, 36% for Savannah-Sudanese zone, and 39% for north Guinean zone (Konaté et al., 1995). Osundare et al., 2024, showed that okra mosaic virus (Okmv) susceptibility, viral incidence, and disease severity are cultivar-dependent.

Successful okra mosaic disease management is very important in order to improve crop yields. Different methods can therefore be used, such as preventing the insect vector contacting the early stage plant (Fajinmi and Fajinmi, 2010), treating the plant with plant extracts (Ali et al., 2005; Asare-Bediako et al., 2014a), induced plant resistance, and chemical control of insect vectors (pesticide) (Asare-Bediako et al., 2014b; Nagendran et al., 2017). According to Fajinmi and Fajinmi (2010), in order to prevent okra mosaic virus disease (OKMV), it is necessary to control the vectors by using a 2 m high net barrier around okra plots, until the plants become more than 21 days old after emergence. Hence, an effective control measure is essential during the early stages of okra growth (Fajinmi and Fajinmi, 2010). It has also been reported in Ghana that majority of the farmers (75%) managed with synthetic pesticides (Asare-Bediako et al., 2014b).

One of the strengths of the study is the choice of a vegetable cultivation area. However, there are few limitations. Some of these are the limited number of samples tested, which makes it difficult to generalize to other okra cultivation areas in Mali, the descriptive nature of the study, and the lack of recent studies in the literature on the prevalence of OKMV. Further research is necessary to consider the severity, environmental factors (such as temperature, humidity, and soil conditions) with OKMV, and molecular characterization; however, this study provides information on the possible circulation of this virus in Mali, which constitutes a valuable addition to future research work.

Conclusion

This study shows the presence of okra mosaic virus in Mali. This is the first time OkMV has been found in Mali. However, molecular characterization is necessary to identify the viruses involved and to evaluate the incidence of okra mosaic disease at the national level, especially according to the symptoms of the geographical location.

CONFLICT OF INTERESTS

The authors have declared any conflict of interests.

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