Journal of Experimental Biology and Agricultural Sciences

<http://www.jebas.org>

ISSN No. 2320 – 8694

PATHOGENIC APTITUDES OF THREE Yam Mosaic Virus (YMV) GENETIC GROUPS RESPONSIBLE FOR YAMS VIRAL DISEASES OF THE COMPLEX

Dioscorea cayenensis-rotundata IN TOGO

Kwasi Dzola Ayisah^{1*}, Gbenonchi Mawussi², Larounga Tchaniley¹

¹Department of Plant Breeding and Crop Sciences, Ecole Supérieure d'Agronomie, Université de Lomé, B. P. 1515, Lomé, Togo.

²Laboratory of Research on Aggroresources and Environmental Health, Ecole Supérieure d'Agronomie, Université de Lomé, B. P. 1515, Lomé, Togo.

Received – September 01, 2020; Revision – November 22, 2020; Accepted – December 15, 2020

Available Online – December 30, 2020

DOI: [http://dx.doi.org/10.18006/2020.8\(6\).810.817](http://dx.doi.org/10.18006/2020.8(6).810.817)

KEYWORDS

Yam mosaic virus,

D. cayenensis-rotundata

Pathogenicity

Genetic groups

Molecular variability

Inoculation

ABSTRACT

This work was aimed to study the pathogenicity of genetic groups of *yam mosaic virus* (YMV) infecting *Dioscorea cayenensis-rotundata* in Togo. The virulence and aggressiveness of six YMV isolates (P3-20, C9-25, S2-21, M9-30, K9-5, P8-15) belonging to three genetic groups, were evaluated at the seedlings stage of three accessions of *D. cayenensis-rotundata*. These isolates were collected from the five yam-producing regions of Togo. For testing virulence and aggressiveness, two isolates per genetic group were mechanically inoculated to ten yam seedlings (five seedlings in two repetitions) per accession. Positive accessions were used to transmit YMV with the help of insect vector *Aphis craccivora*. RT-PCR test was performed to detect the presence of YMV in yam seedlings using two primers viz., YMV1 & YMV2 (196 bp). The results of the study revealed that all six YMV isolates were able to infect the three yam accessions. A significant difference ($p = 0.05$) was observed in the aggressiveness of the selected six isolates which clustered into three aggressiveness levels: group 1 included isolate P3-20 which was reported as very aggressive, group 2 have three isolates viz., S2-21, M9-30, K9-5 which considered as a moderately aggressive while group 3 have rest two isolates C9-25, P8-15 with a low level of aggressiveness. However, apart from isolates S2-21 and M9-30 belonging to the same genetic and aggressiveness group, most of the isolates from the same genetic group had different levels of aggressiveness; suggesting that the variability in pathogenicity of these isolates was not, at first blush, related to the genetic variability of the virus. Also, it was reported that isolate S2-21 could not transfer through the vector *A. craccivora*. Results of the study suggested that among the tested YMV isolates, P3-20 was highly pathogenic while S2-21 and M9-30 were reported moderately pathogenic.

* Corresponding author

E-mail: dayisah@yahoo.fr (Kwasi Dzola Ayisah)

Peer review under responsibility of Journal of Experimental Biology and Agricultural Sciences.

Production and Hosting by Horizon Publisher India [HPI]
(<http://www.horizonpublisherindia.in/>).
All rights reserved.

All the articles published by [Journal of Experimental Biology and Agricultural Sciences](#) are licensed under a [Creative Commons Attribution-NonCommercial 4.0 International License](#) Based on a work at www.jebas.org.

1 Introduction

Yams (*Dioscorea spp.*) is considered an important food crops, not only in Togo but throughout West Africa. *Dioscorea cayenensis-rotundata* is one of the cultivated tropical species of *Dioscorea* in the country (Kpemoua & N'Kpenu, 2010). In Togo, the estimated annual production was 858,783 tones and this is the second most important tuber crops after cassava (FAOSTAT, 2020). Under climatic conditions of the country, yams cultivation is hampered by viral diseases caused by Potyviruses including *yam mosaic virus* (YMV). Under West African climatic conditions including Togo, YMV is the most prevalent virus on yams (Eni et al., 2010), and causing annually up to 89% yield loss (Lamptey et al., 2016). In Togo, the incidence rate of YMV in *Dioscorea cayenensis-rotundata* is 55.48% for the five major yam-producing regions of the country (Ayisah, 2012). Information regarding the genetic characterization of local isolates of YMV collected from the five major yam-production areas are in scarcity. As per the previous studies, genetic variability leads to changes in the expression of the concerned virus's pathogenicity (Granier et al., 1993; Weiland & Edwards, 1996). These changes in pathogenicity are often causing explosive epidemics. Therefore, knowledge of the pathogenicity of different genetic groups of YMV identified in Togo is necessary. This will make possible to better understand the different virus populations from pathogenicity point of view which will help in the selection of appropriate control methods.

The objective of this work was to study the pathogenic aptitudes of three genetic groups of YMV isolates identified from the yams complex *D. cayenensis rotundata* cultivated in Togo. Results of this work not only help in identifying highly pathogenic YMV strains but also help in the screening of virus-resistant yam genotypes under climatic conditions of Togo.

2 Materials and Methods

2.1 Viral isolates studied

To carry out this study, six isolates of YMV (P3-20, C9-25, S2-21, M9-30, K9-5, P8-15) were selected from three genetic groups of the virus isolates previously sequenced and characterized genetically in 2012 at CIRAD- AMIS at Montpellier, France (Ayisah & Gumedzoe, 2012). Two YMV isolates were selected per genetic group for the evaluations. All the collected viral isolates were stored at -20 ° C in the Laboratory of Plant Virology and Biotechnology (LVBV) at Higher School of Agronomy, University of Lomé – Togo. The list of the six YMV isolates selected and studied is established in Table 1.

2.2 Plant material inoculated

The pathogenic aptitudes of the six YMV isolates were evaluated by using artificial inoculations on yam seedlings produced from seed germination. The yam seeds were collected from three yam plants (or accessions) complex of *D. cayenensis-rotundata* from Ogou, Tchoudjo, Oti, Zio, and Kloto Districts (Table 2). The seeds were disinfected with the help of fungicides Mancozeb (6.6g/L of water) and germinated in the Petri dishes containing a moistened filter paper. All seeds were germinated in twenty-one days of inoculation on filter paper. The seedlings at the first leaf stage were transplanted into 5 cm x10 cm polyethylene pots containing sterilized soil. After transplanting, the seedlings were stored away from insects under greenhouse conditions at 25-34°C day temperature and 21-23 ° C at night temperature with 70-90 percent relative humidity. These yam seedlings are produced from seed germination that's why these are free from YMV because the virus is not transmitted by yam seeds (Therry, 1977).

Table 1 List of six YMV isolates evaluated on yam seedlings: genetic groups, localities of origin, identification numbers (Ayisah, 2012)

YMV isolates	Identification number	Localities	Districts	Genetic groups
P3-20	20-601/03	Plateaux region	Ogou	G1
C9-25	25-182/03	Centralregion	Tchaoudjo	G1
S2-21	21-11/05	Savannah region	Oti	G2
M9-30	30-27/03	Maritime region	Zio	G2
K9-5	05-232/03	Kara region	Bassar	G3
P8-15	15-781/03	Plateaux region	Kloto	G3

Table 2 List of the three yam accessions inoculated

Yam accessions	Yam species	Districts of collection
Cay-rtAg1	<i>D. cayenensis-rotundata</i>	Agou (Amouzoukopé)
Cay-rtAv1	<i>D. cayenensis-rotundata</i>	Avé (Mékoviade)
Cay-rtK1(I)	<i>D. cayenensis-rotundata</i>	Kloto (Agomé – Tomégbé)

2.3 Mechanical inoculation of YMV to yam seedlings

The yam seedlings were artificially inoculated by YMV isolates at the three leaf stage. To do this, yam leaf samples infected by YMV isolates were selected and were ground in sodium phosphate buffer (0.03M Sodium Phosphate, pH8.3) supplemented with 0.2% DIECA (Sodium Diethyl-dithiocarbamate). The ground materials were filtered through Hydrophilic cotton and then a pinch of activated charcoal was added to the collected filtrates. The prepared leaf extracts were rubbed on the leaves of the yam seedlings in the presence of carborundum. The inoculated leaves were rinsed with distilled water to remove the excess inoculum. Ten yam seedlings (five seedlings in two replicates) per YMV isolate were inoculated. Two plants were inoculated by only buffer solution which served as negative controls per yam accession and YMV isolate. The inoculated seedlings were kept in cages away from insects. The yam seedlings were reinoculated after 5 days to ensure infection.

2.4 Inoculation of yam seedlings by vector *A. craccivora*

The yam seedlings inoculated mechanically and showed symptoms of virus disease and are positive in RT-PCR tests, served as sources of inoculum for *A. craccivora*. This insect vector was used for the transmission of the six YMV isolates to yam seedlings. Indeed, in fields, YMV is transmitted to yam plants by *A. craccivora* in non-persistent mode. The use of this method of inoculation will give an idea of the behavior of the virus in a real environment. These aphids were previously multiplied in cages on healthy cowpea (*V. unguiculata*) plants. The cowpea plants were regularly replaced to allow the rapid multiplication of the aphids. Before inoculations, the aphids were first fasted for 2 hours and then placed on the leaves of infected yam seedlings for 5 minutes to acquire YMV particles. After the viral acquisition, ten aphids maintained in micro-cages were placed on each yam seedling for 10 minutes (Figure 1). A total of ten yam seedlings (five seedlings in two replicates) were inoculated per YMV isolate and per yam accession and this was followed by the subsequent elimination of

Figure 1 Inoculation of YMV isolates to yam seedlings using *A. craccivora* maintained in micro-cages.

aphids. Two yam seedlings inoculated with virus-free aphids served as negative controls for each of the treatments.

2.5 Molecular detection of YMV inoculated yam seedlings

After four weeks of inoculations, leaf samples were taken from the inoculated yam seedlings and analyzed by RT-PCR for YMV detection of the presence of virus by using QiAgen one-step RT-PCR kit. A pair of primers: YMV1: 5'-TGCGGAACCTCAAAGAAC-3' and YMV2: 5'-TGCCATCAAATCCAAACA-3' was used at the concentration of 10 μ M in the RT-PCR reaction mixture (Bousalem et al., 2000).

2.6 Observation and measurement techniques

The pathogenicity of the inoculated YMV isolates on inoculated Yam plants was evaluated for the presence of viral disease symptoms. The symptom severity on yam seedlings was evaluated using a rating scale from 1 to 5 which defined as 1 = symptom-free; 2 = moderate symptoms; 3 = severe symptoms; 4 = very severe symptoms; 5 = distortion, malformation of leaves or stems, stunting of plants (Mignouna et al., 2001). Observations began 15 days after inoculations and lasted 30 days.

2.7 Data analysis

The statistical analysis of the results was carried out using the software STATISTICA vers. 5.5 (StatSoft Inc., 1999). STATISTICA for Windows. 2300 East 14Th Street Tulsa, OK 74104 USA).

3 Results

3.1 Behavior of yam accessions to mechanical inoculations

The results of the study are presented in tables 3, 4, and 5. Before this study, the susceptibility levels of YMV to three inoculated yam accessions were not yet known. These yam accessions were chosen based on high incidence rates of YMV in the localities where they were cultivated. Results given in table 3 revealed that four weeks after inoculations, all six YMV isolates showed virus disease symptoms on the selected three yam accessions. Depending on the yam accessions and YMV isolates, the time to onset of viral symptoms varied. As shown in figure 2, the symptoms induced by YMV isolates were varied and the most common symptoms were yellow mosaic, chlorosis, and leaf deformity mosaic. However, these symptoms could not be identified to any of the YMV isolates or any of the yam accessions inoculated.

The results compiled in table 3 showed that the susceptibility of the selected three accessions to the six YMV isolates was highly variable. According to this, yam accessions Cay-rtAg1 and Cay-

Table 3 Reactions of yam seedlings to mechanical inoculations of six YMV isolates: Infected seedlings proportions

Yam accessions	YMV Genetic groups						Proportions of Yam plants Infected	Yam plants Infected rates
	Genetic group G1 P3-20	C9-25	Genetic group G2 S2-21	M9-30	Genetic group G3 K9-5	P8-15		
Cay-rtAg1	9/10	6/10	8/10	7/10	7/10	6/10	43/60	71.67
Cay-rtAv1	9/10	7/10	8/10	7/10	8/10	6/10	45/60	75.00
Cay-rtK1(I)	7/10	4/10	5/10	6/10	7/10	5/10	34/60	56.67
Proportions of Yam plants Infected	25/30	17/30	21/30	20/30	22/30	17/30		
Yam plants Infection rates	83.33	56.67	70	66.67	73.33	56.67		

Table 4 Reactions of yam seedlings to mechanical inoculations of six YMV: Symptom severity

Yam accessions	YMV Genetic groups					
	Genetic group G1		Genetic group G2		Genetic group G3	
	P3-20	C9-25	S2-21	M9-30	K9-5	P8-15
Cay-rtAg1	3.5±1.08	2.1±1.12	2.6±0.97	2.5±1.18	2.8±1.32	2.0±0.94
Cay-rtAv1	3.6±1.17	2.5±0.83	3.0±1.15	2.9±1.10	3.0±1.15	2.1±0.99
Cay-rtK1(I)	2.4±1.17	1.6±0.70	2.0±0.94	1.9±0.99	2.4±1.07	1.6±0.70
Symptom severity average	3.17±0.51	2.07±0.31	2.53±0.36	2.43±0.36	2.73±0.22	1.90±0.20

Table 5 Statistical analysis (variance) of averages severity of viral symptoms induced by six YMV isolates on the seedlings of all three yam accessions

Variables	ddl	Value of F	Probability
Cay-rtAg1	5*	2.41	0.048*
Cay-rtAv1	5	2.16	0.071
Cay-rtK1(I)	5	1.139	0.243

Figure 2 Symptoms induced by YMV isolates on inoculated yam seedlings: a) systemic mosaic; b) chlorosis; c) mosaic with deformation of leaves; d) healthy plant

rtAv1 were most susceptible to YMV isolates. Plants of these two yam accessions (Cay-rtAg1 and Cay-rtAv1) had the highest rates of YMV infections viz. 71.67% and 75%, respectively. While the yam accession Cay-rtK1 (I) was found to be less sensitive among the three yam accessions inoculated with YMV isolates (56.67%). All the seedlings showing viral symptoms were found to be positive for the RT-PCR test. No cases of asymptomatic plants were observed among the seedlings inoculated.

3.2 Aggressiveness of YMV isolates in mechanical inoculations

The results reported in table 3 indicated that the tested six YMV isolates had varying levels of pathogenicity against selected three yam accessions. Further, the results of study suggested that the isolate P3-20 has the highest infection percentage to the yam plant (83.33%). The proportions of yam seedlings infected by isolate P3-20 was 9/10, 9/10, and 7/10 for the three yam accessions Cay-rtAg1, Cay-rtAv1, Cay-rtK1(I) respectively. This isolate is followed by isolates K9-5 (73.33%), S2-21 (70%), and M9-30

(66.67%). Isolates C9-25 and P8-15 each infected the average 56.67% of the all yam seedlings (Cay-rtAg1, Cay-rtAv1, Cay-rtK1(I)) and this is the weakest contamination rate obtained. In the selected three yam accessions, YMV isolates C9-25 infected yam seedlings proportions of 6/10, 7/10, 4/10 respectively while this infection rate was 6/10, 6/10, 5/10 for all the selected yam accessions for isolate P8-15 respectively.

Regarding the severity of the viral symptoms induced on the inoculated yam seedlings, table 4 and table 5 indicated an important and significant variation in the aggressiveness of the six YMV isolates on the inoculated yam seedlings. The isolate P3-20 induced the most severe symptoms with an average severity of 3.17 ± 0.51 on the three yam accessions. In fact, on yam accessions Cay-rtAg1 and Cay-rtAv1, the isolate P3-20 caused average severities estimated at 3.5 ± 1.08 and 3.6 ± 1.17 respectively. This isolate was followed by the isolate K9-5 with an average severity of 2.73 ± 0.22 on three yam accessions. Isolates C9-25 and P8-15 induced the less severe symptoms on yam seedlings with the severity means estimated at 2.07 ± 0.31 and 1.90 ± 0.20 , respectively.

Statistical analysis of the average severity (Table 5) revealed a significant difference between all six YMV isolates in the case of yam accession Cay-rtAg1 ($p = 0.05$). In contrast, all six YMV isolates did not show a significant difference in case of rest two yam accessions i.e. Cay-rtAv1 and Cay-rtK1 (I). Besides, no significant interaction was observed between the six isolates of YMV and the inoculated yam accessions.

The Newman-Keuls test ($p = 0.05$) of average severity on the accession Cay-rtAg1, classified all six YMV isolates into three major groups of aggressiveness and these three were denoted as A, B, and AB (table 6). Group A contains the YMV isolate P3-20 which has the highest level of aggressiveness on the yam seedlings while group B consisted of isolates C9-25 and P8-15 which have the lowest level of aggressiveness on yam plants and group AB consists of isolates K9-5, S2-21, and M9-30 which were moderately aggressive.

However, according to the results mentioned in table 6, the levels of aggressiveness of the six YMV isolates were not related to the genetic groups to which they belong. Indeed, isolate P3-20 was the most aggressive isolates among the selected six YMV isolates while isolate C9-25 was reported as the least aggressive one but both these belong to the genetic group G1. Likewise, isolate K9-5 was moderately aggressive, and isolate P8-15 was the weakly aggressive isolate and both belonged to the genetic group G3. Further, isolates S2-21 and M9-30 were moderately aggressive and belonged to the genetic group G2.

3.3 Inoculation of YMV isolates with *A. craccivora*

The results of the YMV inoculations with *A. craccivora* are presented in table 7. Results mentioned in table 7 revealed that all six evaluated YMV isolates, apart from the isolate S2-21, were transmitted by *A. craccivora* to yam seedlings. Further, the infectivity of the isolate P3-20 was reported highest (73.33%) for the seedlings of all three yam accessions. This isolate infected the proportions of 8/10, 7/10, and 7/10 of seedlings in the three yam accessions, Cay-rtAg1, Cay-rtAv1, Cay-rtK1 (I) respectively. This isolate is followed by isolates K9-5, C9-25 and M9-30 which infected 63.33%, 60.00%, and 56.67% of yam seedlings respectively. Isolate P8-15 infected the lowest number of yam seedlings (46.67%). The proportions of yam seedlings infected in the accessions Cay-rtAg1, Cay-rtAv1, Cay-rtK1 (I) by isolate P8-15, were 6/10, 6/10, 2/10, respectively.

Regarding symptom severity, table 8 indicates that isolate P3-20 induced the most severe virus symptoms on yam seedlings with an average severity of 3 ± 0.4 . In the three inoculated yam accessions, Cay-rtAg1, Cay-rtAv1, Cay-rtK1 (I), isolate P3-20 caused average severities of 3.1 ± 1.52 , 3.5 ± 1.43 , 2.4 ± 1.17 , respectively. This isolate P3-20 was followed by isolates K9-5 with an average severity of 2.53 ± 0.42 ; M9-30 (2.17 ± 0.31) and C9-25 (2.03 ± 0.42) isolates. Isolate P8-15 was the least aggressive with a severity average of 1.93 ± 0.29 caused on the three inoculated yam accessions.

Table. 6 Classification of the six YMV isolates according to the Newman-Keuls test ($p = 0.05$): groups of aggressiveness.

YMV Genetic groups	YMV isolates	Symptom severity averages	Aggressiveness groups
G1	P3-20	$3.5 \pm 1.08a^*$	A
	C9-25	$2.1 \pm 1.12b$	B
G2	S2-21	$2.6 \pm 0.97ab$	AB
	M9-30	$2.5 \pm 1.18ab$	AB
G3	K9-5	$2.8 \pm 1.32ab$	AB
	P8-15	$2.0 \pm 0.94b$	B

*Numbers followed by the same lowercase letter are not significantly different for $p = 0.05$ (Newman and Keuls test).

Table 7 Reactions of yam seedlings to *A. craccivora* inoculations of six YMV isolates: Infected plants proportions

Yam accessions	YMV Genetic groups						Proportions of Yam plants Infected	Yam plants Infected rates
	Genetic group G1 P3-20	C9-25	Genetic group G2 S2-21	M9-30	Genetic group G3 K9-5	P8-15		
Cay-rtAg1	8/10	6/10	0/10	6/10	7/10	6/10	33/60	55.00
Cay-rtAv1	7/10	7/10	0/10	7/10	7/10	6/10	34/60	56.67
Cay-rtK1(I)	7/10	5/10	0/10	4/10	5/10	2/10	23/60	38.33
Proportions of Yam plants Infected	22/30	18/30	0	17/30	19/30	14/30		
Yam plants Infected rates	73.33	60.00	0.00	56.67	63.33	46.67		

Table 1 Reactions of yam seedlings to *A. craccivora* inoculations of six YMV isolates: Symptom severity

Yam accessions	YMV Genetic groups					
	Genetic group G1 P3-20	C9-25	Genetic group G2 S2-21	M9-30	Genetic group G3 K9-5	P8-15
Cay-rtAg1	3.1±1.52	2.1±1.12	-	2.4±1.26	3.0±1.49	2.2±1.14
Cay-rtAv1	3.5±1.43	2.6±1.11	-	2.4±1.07	2.7±1.25	2.1±1.10
Cay-rtK1(I)	2.4±1.17	1.4±0.34	-	1.7±0.95	1.9±0.88	1.5±0.85
Symptom severity average	3±0.4	2.03±0.42		2.17±0.31	2.53±0.42	1.93±0.29

However, statistical analysis of the severity averages revealed no significant difference between the tested isolates ($p = 0.05$). All five YMV isolates showed a statistically identical level of aggressiveness on the three yam accessions.

4 Discussion

All three yam accessions inoculated with six YMV isolates were infected but the infection rates were variable. These variations in the responses of the yam accessions infections suggest a genetic diversity within these accessions. This might be because of cross-pollination in yam crops. Thus, seeds formed on the same yam plant can be genetically different, and therefore plants produced from these seeds can have different levels of resistance to the same viral strain. This would be responsible for the variations in the viral symptoms induced on the inoculated yam seedlings and the non-specificity of these symptoms to any of the six YMV tested isolates. The variability and non-specificity in the symptoms induced by YMV isolates inoculated *D. cayenensis-rotundata* seedlings were also observed by Adjata (1991). According to Attiri et al. (2003), these variations in symptoms caused by YMV on yam plants might be due to great genetic variability in the virus or due to the exploited yams cultivars response. On the other hand, statistical analysis of the data from mechanical inoculations revealed a significant difference ($p = 0.05$) between the symptom severity means in the yam accession Cay-rtAg1. This suggests an important variability in the aggressiveness of YMV isolates; which could confirm the great genetic diversity within these isolates.

However, the differences observed in the pathogenicity of all six virus isolates are not at first blush, all related to the genetic groups to which they belong. Only the isolates S2-21 and M9-30 are moderately aggressive and belonged to the same genetic group. For the rest four isolates, the same levels of aggressiveness are often found in different genetic groups. This suggests that the variability's observed in the pathogenicity of six YMV isolates were mostly independent to the molecular diversity observed within populations of the pathogen in Togo. The genetic variabilities identified in YMV isolates of Togo in 2012, would not have much influence on the pathogenic aptitudes of the virus isolates (Ayisah & Gumedzoe, 2012). Indeed, according to Astier et al. (2001), the genetic variability of a virus does not always translate into variations in its pathogenicity. Mutations are the initial source of variation in viral populations (Drake et al., 1998), but in reality, only a tiny part of these results is noticeable phenotypic variations like symptoms and transmissibility etc. (Astier et al., 2001).

According to the data of mechanical inoculations, YMV isolate P3-20 was the most pathogenic strain while isolates S2-21, M9-30, and K9-5 were moderately aggressive. Isolate P3-20, because of its high aggressiveness, could be used for the search for resistant genotypes of yam. However, isolates S2-21, M9-30 are moderately aggressive and their genetic group G2, was widely represented in all yam-production areas of Togo (Ayisah & Gumedzoe, 2012). In this regard, they could also be used effectively in the screening of resistant yam genotypes.

Yam seedlings inoculations with the help of *A. craccivora* did not show any significant difference ($p = 0.05$) between the viral symptom severity means and level of aggressiveness in all six YMV isolates. On the other hand, during this inoculation, isolate S2-21 could not be transmitted to any of the yam accessions and the proportions of yam seedlings infected by YMV isolates, were somewhat lower than the results obtained using mechanical inoculations. Failed transmission of YMV isolates by *A. craccivora* in Togo, has already been reported by Fétéké(1997). The non-transmissibility of YMV strains by aphids and several cases of potyvirus strains having lost their ability to be transmitted by aphids have already been reported by Goudou-Sinha (1995) and Astier et al. (2001). According to Astier et al. (2001), the loss of transmissibility of potyviruses by aphids is often linked to natural mutations in a conserved amino acid triplet DAG present in the N-terminus of the capsid protein of most of these viruses. The low efficiency of transmission of the six YMV isolates by *A. craccivora*, including the non-transmissibility isolate S2-21, could be associated with many factors such as the YMV isolates genetic variability and geographic origin, and the aphid population. Indeed, Maruthi et al. (2002) demonstrated that the efficiency of virus transmission by *Bemisia tabaci* varied greatly and depending on the geographic origin of the isolates. In Potato Leaf Roll Virus (PLRV), Bourdin et al. (1998) and Guyader (2003) demonstrated that the efficiency of aphid virus transmission greatly differs with the variability of isolates and the aphid population. It would be, therefore, desirable to continue inoculations with other populations of aphids; this will provide more information about the isolate S2-21.

The absence of interactions between YMV isolates and inoculated yam accessions suggested that the isolate P3-20 and other moderately aggressive YMV isolates could be used effectively for the screening of yam resistant genotypes in all yam cultivar groups of the complex *D. cayenensis-rotundata*.

Conclusion

This study showed that six isolates of YMV evaluated were virulent against the three accessions of inoculated yams. The results of the mechanical inoculations revealed great variability in the aggressiveness of the six isolates which clustered into three levels of aggressiveness. The first group consisted of the isolate P3-20 with a very high level of aggressiveness, group 2 consisted of the isolates S2-21, M9-30, K9-5 moderately aggressive, and group 3 contained the isolates C9- 25, P8-15, which have a low level of aggressiveness. However, apart from the isolates S2-21, M9-30, moderately aggressive, which belonged to the same genetic group G2, mostly of the isolates, even belonging to the same genetic groups, had different levels of aggressiveness. This suggests that the variability in pathogenicity of the six isolates was not related to the molecular variability of the virus in Togo.

Inoculation of the isolate S2-21 by *A. craccivora* revealed failed transmission, suggesting either the non-transferability of the isolate by the vector or the inefficiency of the aphid population to transmit it. While the isolates P3-20 was the most aggressive and S2-21, M9-30 moderately aggressive but genetically represented in all yam-production regions of Togo, could be used for the screening of yam genotypes resistant to YMV. Maritime, Central, and Plateaux regions, which host the isolates of the three aggressiveness levels, could serve as search areas for yam resistant sources against YMV.

Conflict of interest

The authors declare that they have no conflict of interest.

References

- Adjata KD (1991) Application of Enzyme-Linked Immunosorbent Assay (ELISA) to the detection of yam potyviruses (*Dioscorea spp.*). End of agronomic studies thesis n°90/01/PV, E.S.A.- UL. (Lomé-Togo) Pp ;102-110(French).
- Astier S, Albouy J, Maury Y, Lecoq H (2001) Principles of plant virology: Genome, pathogenicity, virus ecology. INRA ed. Versailles (French)
- Attiri GI, Winter S, Alabi JO (2003) Yam. In: Loebenstein G, Thottappilly G 2003. Virus and Virus -like Diseases of Major Crops in Developing Countries. Kluwer Academic Publishers, Dordrecht, Netherlands.
- Ayisah KD (2012) Molecular variability of the yam mosaic virus (YMV) on the complex *Dioscorea cayenensis-rotundata* in Togo and search for sources of resistance in local yam populations (cultivated and wild). PhD thesis submitted to the Faculty of Sciences of University of Lomé, Togo, B.P.: 1515 Lomé-Togo N°d'ordre 418. French.
- Ayisah KD, Gumedzoe YMD (2012) Genetic diversity among yam mosaic virus (YMV) isolates infecting yams of the complex *Dioscorea cayenensis-rotundata* in Togo. International Journal of Biological and Chemical Sciences 6(3): 1090- 1101
- Bourdin D, Rouzé J, Tanguy S, Robert Y (1998) Variation among clones of *Myzus persicae* (Sulzer) *Myzus nicotianae* Blackman in the transmission of a poorly and a highly aphid-transmissible isolate of potato leafroll luteovirus (PLRV). Plant Pathology 47: 794-800
- Bousalem M, Dallot S, Guyader S (2000) The use of phylogenetic data to develop molecular tools for the detection and genotyping of Yam mosaic virus. Potential application in molecular epidemiology. Journal of Virological Methods 90: 25-36.

- Drake JW, Charlesworth B, Charlesworth D, Crow JF (1998) Rates of spontaneous mutation. *Genetics* 148:1667–1686
- Eni AO, Hughes Jd'A, Asiedu R, Rey MEC (2010) Survey of the Incidence and Distribution of Viruses Infecting Yams (*Dioscoreaspp*) in Ghana and Togo. *Annals of Applied Biology* 156(2): 243-151
- FAOSTAT- FAO (2020) Agriculture database. FAO statistical yearbook. Viale delle Terme di Caracalla 00153 Rome, Italy.
- Fétéké F (1997) Prevalence and incidence of *yam mosaic virus* (YMV) in Sotouboua District and its evolution in yam seed production plots (*Dioscoreacayenensis-rotundata*); End of agronomic studies thesis. University of Lomé, E.S.A., N° d'ordre 97/16/PV (French).
- Goudou-Sinha C (1995) Yam mosaic: Epidemiological aspects in Burkina Faso and variability of the virus. Doctoral thesis, University of Montpellier II, Sciences et Techniques du Languedoc.
- Granier F, Durand-tardif M, Casse-Delbard F, Lecocq F, Robaglia C (1993) Mutation in zucchini yellow mosaic virus helper component protein associated with loss of aphid transmissibility. *Journal of General Virology* 74:2737-2742.
- Guyader S (2003) Evaluation du potentiel de variabilité du *Potato Leafroll Virus* (*Luteoviridae*, *Polyerovirus*) et identification de quelques facteurs de sélection. Thèse de doctorat, Université de Rennes I.
- Lampitey JNL, Oppong A, Ofori FA (2016) Impact of yam mosaic virus (YMV) on tuber yield of white yam (*Dioscorea rotundata* Poir) in Ghana. *Agricultural and Food Science Journal of Ghana* 9 : 730-741
- Mignouna HD, Njjukenge P, Abang MM, Asiedu R (2001) Inheritance of resistance to *yam mosaic virus*, genus *Potyvirus*, in white yam (*Dioscorea rotundata*). *Theoretical and Applied Genetics* 103(8): 1196-2000.
- Maruthi MN, Colvin J, Seal S, Gibson G, Cooper J (2002) Co-adaptation between cassava mosaic geminiviruses and their local vector populations. *Virus Research* 86: 71-85;
- Kpemoua KE, N'Kpenu KE (2010) Etat des lieux de la recherche sur l'igname au Togo. Renforcement de capacité de la recherche pour le développement de l'igname en Afrique de l'Ouest et du Centre. ITRA-Togo, IITA-Ibandan (Nigeria).
- Therry ER (1977) Incidence symptomatology and transmission of yam virus in Nigeria. *Proceedings of the Fourth symposium of the International Society of Tropical Root Crops*, Cali, Columbia. Pp 170-173
- Weiland JJ, Edwards MC (1996) A single nucleotide substitution in the aa gene confers oat pathogenicity to barley stripe mosaic virus strain ND18. *Molecular Plant-Microbe Interactions* 9: 62-67.