



Molecular and Biological Features of Sweet Potato Leaf Curl Virus in Burkina Faso

Ezechiel Bionimian Tibiri^{1,2,3,4}, Fidèle Tiendrébéogo^{1,3*}, Justin Simon Pita⁵, Koussao Somé^{2,3}, Martine Bangratz^{3,6}, James Bouma Néya^{1,3}, Christophe Brugidou^{3,6} and Nicolas Barro⁴

¹Laboratoire de Virologie et de Biotechnologies Végétales, Institut de l'Environnement et de Recherches Agricoles (INERA), Burkina Faso

²Laboratoire de Génétique et de Biotechnologies Végétales, INERA, Burkina Faso

³Laboratoire Mixte International Patho-Bios, IRD-INERA, Burkina Faso

⁴Laboratoire d'Epidémiologie et de Surveillance des bactéries et virus Transmissibles par les Aliments et l'eau. LabESTA/UFR/SVT, Université Ouaga 1 Professeur Joseph Ki-Zerbo, Burkina Faso

⁵Université Félix Houphouët-Boigny (UFHB), Pôle scientifique et d'innovation de Bingerville, Côte d'Ivoire

⁶IRD, Cirad, Université Montpellier, Interactions Plants Microorganismes et Environnement (IPME), Avenue Agropolis Montpellier Cedex, France

*Corresponding Author: Fidèle Tiendrébéogo, Laboratoire de Virologie et de Biotechnologies Végétales (LVBV), INERA, Burkina Faso.

Received: September 16, 2019; Published: September 30, 2019

Abstract

Begomoviruses (family *Geminiviridae*, genus *Begomovirus*) that infect sweetpotato (*Ipomoea batatas*) are known as sweepoviruses and are a major constraint to sweetpotato production. To date, the known distribution of sweepoviruses in Africa has been limited to Kenya, Uganda, South Africa, Sudan and Tanzania. Weeds can act as sources or reservoirs for important crop pathogens, but their role in crop plant diseases in Africa has not been well documented. Begomoviruses infect a wide range of dicotyledonous plants, including cultivated crops and weeds.

Sweetpotato plants and weeds from Burkina Faso exhibiting severe leaf curling, suggesting the presence of begomoviruses, were tested and characterized using rolling circle amplification followed by cloning and sequencing. Six full genomes were successfully obtained, and molecular analysis revealed a monopartite genomic organization typical of sweepoviruses. Sequence analyses and phylogenetic data showed the virus was closely related to *Sweet potato leaf curl virus*-United States (SPLCV-US) with 98% nucleotide identity for sweetpotato plant samples. From *Ipomoea alba*, the sequenced genomes were also closely related to SPLCV-US with 96% as the highest nucleotide sequence identity.

When healthy *Ipomoea setosa* plants were grafted with infected scions, plants showed systemic upward leaf-curling and leaf-deformation symptoms.

This is the first published full-length genome of a sweepovirus reported and first time in West Africa that the common weed *I. alba* has been found to be sweepovirus-positive. This discovery raises concerns about its presence in Burkina Faso and its potential spread in the region.

Keywords: *Begomovirus*, *Ipomoea alba*, Sweepovirus, RCA, Sweetpotato, Weed

Introduction

Begomovirus (family *Geminiviridae*, genus *Begomovirus*) is the largest viral genus, with 388 species currently recognized by the International Committee on Taxonomy of Viruses [1]. These viruses have circular single-stranded DNA encapsulated in twinned icosahedral particles. They are either monopartite (only DNA-A) or bipartite (DNA-A and DNA-B components). In the bipartite begomoviruses, the two components share a common region (CR) of approximately 200 base pairs (bp) within the intergenic region (IR) that includes the replication origin. DNA-A encodes a coat protein (CP, AV1/V1), a putative movement protein (MP, AV2/V2), a replication-associated protein (Rep, AC1/C1), a transcriptional ac-

tivator (TrAP, AC2/C2), a replication enhancer (REn, AC3/C3) and C4 (AC4/C4). DNA-B encodes a nuclear shuttling protein (BV1) and an MP (BC1). The genomes of monopartite begomoviruses resemble the bipartite DNA-A component [2,3]. The 200 nucleotides of the CR are similar (usually 80 -100% sequence identity) [4,5]. The CR encompasses the fully conserved stem-loop structure containing the nonanucleotide sequence TAATATT↓AC and plays a role in initiation of the rolling-circle replication [6].

Begomoviruses infect a wide range of dicotyledonous plants, mostly in tropical and subtropical regions. In the Old World (OW), most of begomoviruses are monopartite and some have a bipartite genome. Begomoviruses native to the New World (NW) are almost

exclusively bipartite, with one exception [7]; however, a number of monopartite begomoviruses have been introduced from the OW [8]. All begomoviruses known to infect sweet potato (*Ipomoea batatas*, Convolvulaceae) are monopartite and are known as “sweepoviruses” [9]. Phylogenetically, the sweepoviruses have been grouped in a monophyletic cluster distinct from the OW and NW *Begomovirus* branches [5,10]. In Africa, the only sweepoviruses infecting sweetpotato have been reported in Kenya, Uganda, South Africa, Tanzania and more recently in Sudan [11-15].

In recent years, a number of sweepoviruses infecting *Ipomoea* species have been identified in various parts of the world [16-18]. In addition to sweetpotato (*Ipomoea batatas*), sweepoviruses can infect other congeneric hosts such as *I. nil*, *I. alba*, *I. cordatotriloba*, *I. hederacea*, *I. lacunosa*, *I. tiliacea*, *I. trifida* and *I. setosa* [19]. The symptoms caused by sweepoviruses depend on the specific host but usually consist of leaf curling and vein yellowing, although infections can also be asymptomatic [19]. In addition to the experimental host *Nicotiana benthamiana*, two species are hosts for Sweet potato leaf curl virus (SPLCV) in Japan (both in family Convolvulaceae): *Calystegia japonica* and *Convolvulus tricolor*. *Ipomoea aquatica*, *I. fistulosa* and *I. lobata* have also been shown to be hosts for SPLCV-United States (SPLCV-US) [16]; *I. indica* is a natural host for Ipomoea yellow vein virus [20], and *Merrimeia* spp. and *I. purpurea* are natural hosts for SPLCV [21].

In Burkina Faso, begomoviruses are major pathogens for common crops including cassava, okra, pepper and tomato [22,23], but begomoviruses have never been documented on sweetpotato.

Globally, sweetpotato is one of the most important subsistence crops and the third most important root crop after potato (*Solanum tuberosum*) and cassava (*Manihot esculenta* Crantz) [24]. Sweetpotato plants are propagated vegetatively by directly planting cuttings from a plant from the previous season or by using sprouted tubers. Sweetpotato is prone to virus infections and accumulation of viruses when thus propagated [17,25].

The aim of the current study was to characterize the viruses that were specifically inducing symptoms consistent with sweepoviruses. It also aimed to test whether the above-mentioned weeds commonly associated with sweetpotato fields are hosts to sweepoviruses and are thus potential sources of infection affecting sweetpotato plants.

Materials and Methods

Sampling

Three samples (BFA43, BFA270 and BFA271) of sweetpotato plants exhibiting leaf-curling symptoms typical of infection with sweet potato leaf curl virus (SPLCV) were collected around Bama (N11°22'45.3"; W004°23'31.3") and Tiébélé (N11°07'10.5"; W000°57'50.5") (Figure 1a and b). Then, weeds showing severe leaf curling and yellowing symptoms were sampled, as follows: ten *I. alba*, four *Physalis ixocarpa*, two *I. asarifolia*, two *Sida acuta* and

three *Sida* spp., (Figure 1c-f). We were particularly interested in *I. alba*, commonly called moon vine (Figure 1e), a night-blooming morning glory native to tropical and subtropical regions of the New World. In tropical regions of Africa, *I. alba* is a weed and in Burkina Faso it is widespread and could be a reservoir of sweepoviruses.

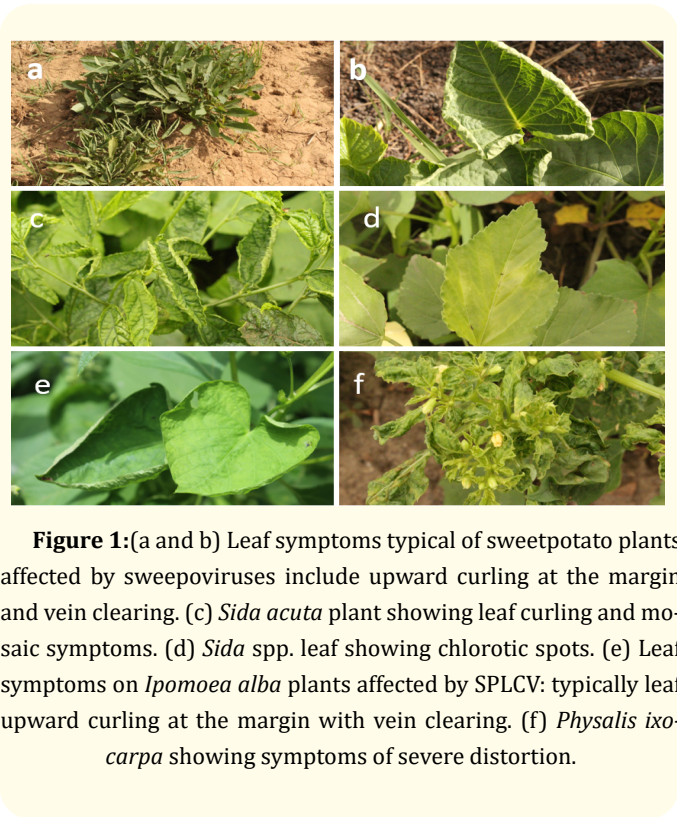


Figure 1:(a and b) Leaf symptoms typical of sweetpotato plants affected by sweepoviruses include upward curling at the margin and vein clearing. (c) *Sida acuta* plant showing leaf curling and mosaic symptoms. (d) *Sida* spp. leaf showing chlorotic spots. (e) Leaf symptoms on *Ipomoea alba* plants affected by SPLCV: typically leaf upward curling at the margin with vein clearing. (f) *Physalis ixocarpa* showing symptoms of severe distortion.

Checking for presence of sweepoviruses by PCR for amplification of CP gene of SPLCV

Total DNA was extracted from sweetpotato and weeds samples using the cetyltrimethylammonium bromide (CTAB) protocol [18]. PCR was performed using the degenerated primers SPG1 5'-CCCCK-GTGGWRAATCCAT-3' and SPG2 5'-ATCCVAAYWTYAGGGAGC-TAA-3' [26] to test for the presence of begomoviruses.

Cloning and sequencing of the full genomes

PCR-positive samples were amplified by the rolling-circle amplification (RCA) method using the φ29 DNA polymerase TempliPhi DNA amplification kit (Amersham Biosciences Corp., Sunnyvale, CA, USA) as described in Inoue-Nagata., *et al* [27]. The RCA products were digested with the *Bam*HI restriction enzyme to identify suitable sites for cloning the full-length genomes (~2.8 kb). The restricted fragments that corresponded to putative full-length monomer genomes were cloned into the vector pGEM-3Zf (+) (Promega Corp., Madison, WI, USA) and fully sequenced by the primer walking method by Macrogen Europe, Amsterdam, Netherlands.

Phylogenetic relationships and recombination analyses

Contigs obtained were mapped to a reference genome using Geneious v. 8.1.7. All sequences were analyzed using the BLAST search tools of NCBI and by pairwise sequence comparison (Geneious)

[28]. The homologous sequences were selected for phylogenetic analysis. Using ClustalW in MEGA 7.0.14, sequences were aligned with sequences from other parts of the world retrieved from GenBank [29]. Comparisons were made between 34 full-genome sequences obtained from GenBank, six new genome sequences from sweetpotato and weed samples obtained in this study. Our full genome sequences were compared with sequences from the

world and especially sequences from Africa (Table 1). Evolutionary history was inferred by the maximum-likelihood method using the Tamura-Nei model; phylogenetic reconstruction was performed using Fast Tree v. 2.1 [30], with bootstrap support values of 1,000. The tree was visualized and edited using Fig Tree v. 1.4.3. Then, pairwise identity comparisons of nucleotide sequences were performed using SDT v. 1.2 with pairwise deletion of gaps [31].

Table 1: Isolates from Burkina Faso used in this study (bold) and strains of sweepoviruses used as references.

Virus isolate	Strain	Host	Geographical origin	Accession no.
BFA43	Sweet potato leaf curl virus	<i>Ipomoea batatas</i>	Burkina Faso	LS991864
BFA270	Sweet potato leaf curl virus	<i>I. batatas</i>	Burkina Faso	LS990769
BFA271	Sweet potato leaf curl virus	<i>I. batatas</i>	Burkina Faso	LS990768
BFA181	Sweet potato leaf curl virus	<i>Ipomoea alba</i>	Burkina Faso	LS991865
BFA1598	Sweet potato leaf curl virus	<i>Ipomoea alba</i>	Burkina Faso	This study
BFA1599	Sweet potato leaf curl virus	<i>Ipomoea alba</i>	Burkina Faso	This study
[ES-CI-BG4-02]	Sweet potato leaf curl Canary virus	<i>I. batatas</i>	Spain	EF456742
[US-Geo-16]	Sweet potato leaf curl Georgia virus	<i>I. batatas</i>	USA	AF326775
[CN-Hn10-12]	Sweet potato leaf curl Henan virus	<i>I. batatas</i>	China	KC907406
[BR-SP-AlvM-09]	Sweet potato leaf curl Sao Paulo virus	<i>I. batatas</i>	Brazil	HQ393477
[CN-Sc15-12]	Sweet potato leaf curl Sichuan virus 1	<i>I. batatas</i>	China	KC488316
[CN-Sc14-12]	Sweet potato leaf curl Sichuan virus 2	<i>I. batatas</i>	China	KF156759
[US-SC-648-B9-06]	Sweet potato leaf curl South Carolina virus	<i>I. batatas</i>	USA	HQ333144
[UG-KAMP-08]	Sweet potato leaf curl Uganda virus	<i>Ipomoea setosa</i>	Uganda	FR751068
[CN-Yn-RL31-06]	Sweet potato leaf curl virus-China	<i>Ipomoea purpurea</i>	China	EU253456
[BR-PA-Bel1-08]	Sweet potato leaf curl virus-Brazil	<i>I. batatas</i>	Brazil	FJ969829
[CN-Fuj-Ip3-07]	Sweet potato leaf curl virus-Fujian	<i>Ipomoea purpurea</i>	China	FJ515898
[IT-Sic-02]	Sweet potato leaf curl virus-Italy	<i>Ipomoea indica</i>	Italy	AJ586885
[JR-Miy-96]	Sweet potato leaf curl virus-Japan	<i>I. batatas</i>	Japan	AB433786
[BR-PB-Sou1-08]	Sweet potato leaf curl virus-Paraiba	<i>I. batatas</i>	Brazil	FJ969830
[BR-RO-PV-08]	Sweet potato leaf curl virus-Pernambuco	<i>I. batatas</i>	Brazil	HQ393456
[PR-80-N2-06]	Sweet potato leaf curl virus-Puerto Rico		Puerto Rico	DQ644562
[BR-BA-Uti-08]	Sweet potato leaf curl virus-Rondonia	<i>I. batatas</i>	Brazil	HQ393447
[BR-SP-AlvM-09]	Sweet potato leaf curl virus-Sao Paulo	<i>I. batatas</i>	Brazil	HQ393473
[US-SC-646-B11-06]	Sweet potato leaf curl virus-South Carolina	<i>I. batatas</i>	USA	HQ333138
[ES-CI-BG6-02]	Sweet potato leaf curl virus-Spain	<i>I. batatas</i>	Spain	EF456744
[US-Lou-94]	Sweet potato leaf curl virus-United States	<i>I. batatas</i>	USA	AF104036
[BR-BSB1-08]	Sweet potato mosaic virus	<i>I. batatas</i>	Brazil	FJ969831
[SD: KTR: 402-16]	Sweet potato leaf curl virus	<i>I. batatas</i>	Sudan	KY270781
[ZA: WP:2011]	Sweet potato mosaic virus	<i>I. batatas</i>	South Africa	JQ621843
[ES: 98]	Sweet potato leaf curl virus	<i>Ipomoea indica</i>	Spain	AJ132548
[TZ: SNG7:12]	Sweet potato leaf curl Sao Paulo virus	<i>I. batatas</i>	Tanzania	KF836891
[CN: 05]	Sweet potato leaf curl China virus		China	DQ512731
Outgroup [ZA: ToCSV:04]	Tomato curly stunt virus	<i>Solanum lycopersicum</i>	South Africa	AF261885

Given that recombination is as been a major evolutionary mechanism for begomovirus [32,33], we performed a recombination analysis using our sequences and a set of sequences representing the whole begomoviral diversity representing an alignment of 444 sequences. Detection of potential recombinant sequences, identification of likely parental sequences, and localization of possible recombination breakpoints was carried out by using the RDP [34], GENECONV [33], BOOTSCAN [35], MAXIMUM CHI SQUARE [36], CHIMAERA [37], SISTER SCAN [38] and 3Seq [39] recombination detection methods as implemented in RDP5 [37,40]. The analysis was performed with default settings for the different detection methods and a Bonferroni corrected P-value cut off 0.05. Only events detected with 3 methods or more were accepted.

Virus transmission by grafting

Grafting was performed with sample clones, which came from samples BFA43, BFA270 and BFA271 from sweetpotato. Grafting was used for transmission tests and indexing of viral symptoms on *I. setosa*. Ten grafts from each of the sweetpotato samples (BFA43, BFA270 and BFA271) and ten grafts from healthy sweetpotato samples were taken and each of these scions were side-grafted to ten healthy *I. setosa* plants. Symptoms were recorded weekly for 8 weeks. The *I. setosa* leaf samples were harvested 15, 25, 45 and 60 days after grafting and were tested for virus presence using the method described by Li., *et al* [26].

Results and Discussion

Detection of CP gene of SPLCV by PCR

Three sweetpotato samples (BFA43, BFA270 and BFA271) plants were PCR-positive. Despite the sweepovirus-like symptoms (Figure 1), all *I. asarifolia*, *P. ixocarpa* and *S. acuta* samples were negative using SPG1 and SPG2. Only the *I. alba* samples (BFA181, BFA1598 and 1599) showed positive results from the PCR test.

Analyses of full genomes obtained

The full genome sequence of three cloned isolates (BFA43, BFA270 and BFA271) corresponding to putative full-length sweepovirus genomes were successfully obtained from sweetpotato samples and deposited in European Nucleotide Archive (ENA) (Table 1). All genomes (2,829 bp for BFA43 and BFA270, and 2,830 bp for BFA271) had the typical organization of monopartite begomoviruses with two open reading frames (ORFs) in the virion sense (V1/CP and V2/MP) and four ORFs in the complementary sense (C1/Rep, C2/TrAP, C3/REn and C4). The BFA181, BFA1598 and BFA1599 samples, from the weed, *I. alba*, was successfully cloned and sequenced. The BFA181 sequence (Table 1) consisted of 2830 nucleotides ([European Nucleotide Archive (ENA) LS991865], with six encoding open reading frames (ORFs). Of these, two ORFs, AV1 encoding a capsid protein (CP) (nucleotides 296 - 1060) and AV2 encoded a movement protein (MP) (nucleotides 73 - 471), were in the viral-sense strand. The remaining four ORFs were located in the complementary-sense strand, as follows: AC1 encoding a replication-associated protein (Rep) (nucleotides 1582

- 2676), AC2 encoding a transcriptional activator protein (nucleotides 1227 - 1673), AC3 encoding a replication enhancer protein (nucleotides 1076 - 1510) and AC4 (nucleotides 2262 - 2796). The genomes of BFA1598 and BFA1599 consisted of 2826 and 2795 nucleotides length, respectively, and had the same organization. All sequences contained the conserved nonanucleotide sequence 5'-TAATATT↓AC-3' and a non-coding intergenic region (IR) consisting of 110 nucleotides was located between AC4 and AV2. The iterative elements (with the consensus GGWGA sequence core [6]), three in the forward direction (I, II and III) and one in the reverse direction (IV), were located in the IR surrounding the C1 ORF TATA box (Table 2). These elements were identified for the three isolates as being similar to the two other sweepoviruses (SPLCV and SPLCV-US). The iteron-related domain (IRD) in the N-terminal region of the replication-associated protein (Rep) was also identified [6]. We found sequences that contained Rep IRDs (MAPPKRFRISS and MAPPKRFKIQA) that differed from those previously described by Albuquerque., *et al.* [25] for sweepoviruses (Table 2).

BLASTn searches revealed that isolate BFA43 from sweetpotato and BFA181 from *I. alba* showed the highest nucleotide identity (96%) with SPLCV-US, accession numbers AF104036 (USA) and KC253233 (Peru). Isolates BFA270 and BFA271 shared highest nucleotide identity (95% and 96%, respectively) with the SPLCV isolate from South Korea (GenBank accession number KT992061). According to the current demarcation threshold for *Begomovirus* species (< 89% identity for DNA-A) recommended by the *Geminiviridae* Study Group of the ICTV [41], this study showed that isolates BFA43, BFA270 and BFA271 (from sweetpotato sample) and BFA181, BFA1598 and BFA1599 (from *I. alba*) belonged to species SPLCV-US.

Phylogenetic relationships

Phylogenetic maximum-likelihood tree and a pairwise comparison matrix of full genome sequences was constructed using multiple alignment of 32 selected begomovirus sequences. Phylogenetic relationships of the nucleotide sequences of the Burkina Faso SPLCV isolates to those of closely related sweepoviruses were deduced using the available sweepovirus species from around the world (Table 1). The sequences of isolates BFA43 and BFA181 grouped together with the SPLCV-US (AF104036). The isolates BFA270 and BFA271 shared a common clade and grouped separately from SPLCV-US. It is also the case of BFA1598 and BFA1599 which shared a common clade. The six Burkina Faso isolates clustered separately and showed that the sequences are different and further analyses are needed to investigate more SPLCV in Burkina Faso.

Symptoms indexing on *Ipomoea setosa*

The *I. setosa* plants developed systemic infections within two weeks post-grafting. Early symptoms were relatively mild, consisting of leaf curling and vein clearing after three weeks (Figure 2a). After four weeks of growth, the symptoms were severe and consisted of upward leaf curling, vein clearing and leaf distortion;

Table 2: Iterative elements, consensus sequences and corresponding iteron-related domains in the N-terminal regions of the replication protein (Rep IRD) of sweepoviruses.

Virus	Accession number	Iterative elements					Consensus	Rep IRD
		I	II	III	TATA box	IV		
Sweet potato leaf curl virus	LS991864	ATTTGGAGA	ATTGGAGA	GGAGAC	TATATA	TCTCC	GGWGAC	MAPPKRFKIQA
Sweet potato leaf curl virus	LS991865	ATTTGGAGA	ATTGGAGA	GGAGAC	TATATA	TCTCC	GGWGAC	MAPPKRFKIQA
Sweet potato leaf curl virus-United States	AF104036	ATTTGGAGA	ATTGGAGA	GGAGAC	TATATA	TCTCC	GGWGAC	MAPPKRFKIQA
Sweet potato leaf curl virus-Fujian	FJ515898	ATTTGGAGA	ATTGGAGA	GGAGAC	TATATA	TCTCC	GGWGAC	MAPPKRFKIQA
Sweet potato leaf curl virus-South Carolina	HQ333138	ATTTGGAGA	ATTGGAGA	GGAGAC	TATATA	TCTCC	GGWGAC	MAPPQRFKIQA
Sweet potato leaf curl virus	LS990769	ATTTGGTGA	ATTGGTGA	GGTGAC	TATA	TCACC	GGWGAC	MAPPKRFRISS
Sweet potato leaf curl virus	LS990768	ATTTGGTGA	ATTGGTGA	GGTGAC	TATA	TCACC	GGWGAC	MAPPKRFRISS
Sweet potato leaf curl virus	KY270781	ATTTGGTGA	ATTGGTGA	GGTGAC	TATA	TCACC	GGWGAC	MAPPKRFRISS
Sweet potato leaf curl virus-Spain	EF456744	ATTTGGTGA	ATTGGTGA	GGTGAC	TATA	TCACC	GGWGAC	MAPPKRFRISS
Sweet potato leaf curl virus-China	EU253456	ATTTGGTGA	ATTGGTGA	GGTGAC	TATA	TCACC	GGWGAC	MAPPKRFRISS
Sweet potato leaf curl virus-Italy	AJ586885	ATTTGGTGA	ATTGGTGA	GGTGAC	TATA	TCACC	GGWGAC	MAPPKRFRISS
Sweet potato leaf curl virus-Pernambuco	HQ393456	ATTTGGTGA	ATTGGTGA	GGTGAC	TATA	TCACC	GGWGAC	MAPPKRFRISS
Sweet potato leaf curl virus-Puerto Rico	DQ644562	ATTTGGTGA	ATTGGTGA	GGTGAC	TATA	TCACC	GGWGAC	MAPPKRFRISS

within 45 - 60 days post-grafting, inoculated plants showed signs of stunting (Figure 2b and c). The infections were confirmed to be caused by SPLCV by using PCR and sequencing using the degenerate primers (SPG1 5'-CCCCKGTGCGWRAATCCAT-3' and SPG2 5'-ATCCVAAYWTYCAGGGAGCTAA-3') [26].

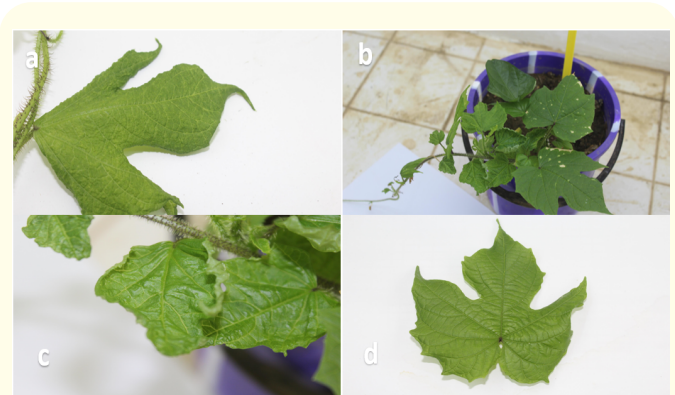


Figure 2: Sweet potato leaf curl virus-associated symptoms in *Ipomoea setosa* plants infected by grafting with *I. batatas*. (a–c) Leaf-curling and deformation symptoms on *Ipomoea setosa* after inoculation by grafting with infected sweetpotato (*I. batatas*). (d) A healthy *I. setosa* leaf.

Recombination analyses

Five distinct recombination events (a, b, c, d and e) were successfully detected within the full genome sequences of SPLCV isolates (Figure 4), using a large sequence alignment of begomoviruses. These recombination events have identified four genetic groups of Sweet potato leaf curl viruses (G1 to G4; Figures 3 and 4). Recombinant event (a) involves an Unknown major parent and a minor parent related to TYLCV [KSA:Has3:13] (KF435137). The event (a) is present in groups G1 and G3. For the event (b), it is an intra-strain recombination involving SPLCHnV-[CN:Hn10:12] (KJ476507) as a major parent and SPLCCNV-[CN:Gu:12] (KJ013576) as minor parent. Events (c) and (e) have involved SPLCV [BF:Leg:BFA181:16] (LS991865) and SPLCV [BF:Kam:BFA1599:17] as major parents, respectively. The event (e) is present in the groups G3 and G4 and concern the end of the gene C1 and the whole intergenic region. The event (d) is only present in the group G4 with an Unknown major parent and a minor parent related to BDMV [CO:87] (M88179). These recombinant events highlight the complex nature of sweepoviruses in Burkina Faso and the intra-strain recombination seems to prove the Chinese origin of sweet potato introduced in Vallée du Kou, Burkina Faso. The biological effects of the recombination events should be more investigated in the future to more understand the widespread of sweet potato leaf curl virus in Burkina Faso.

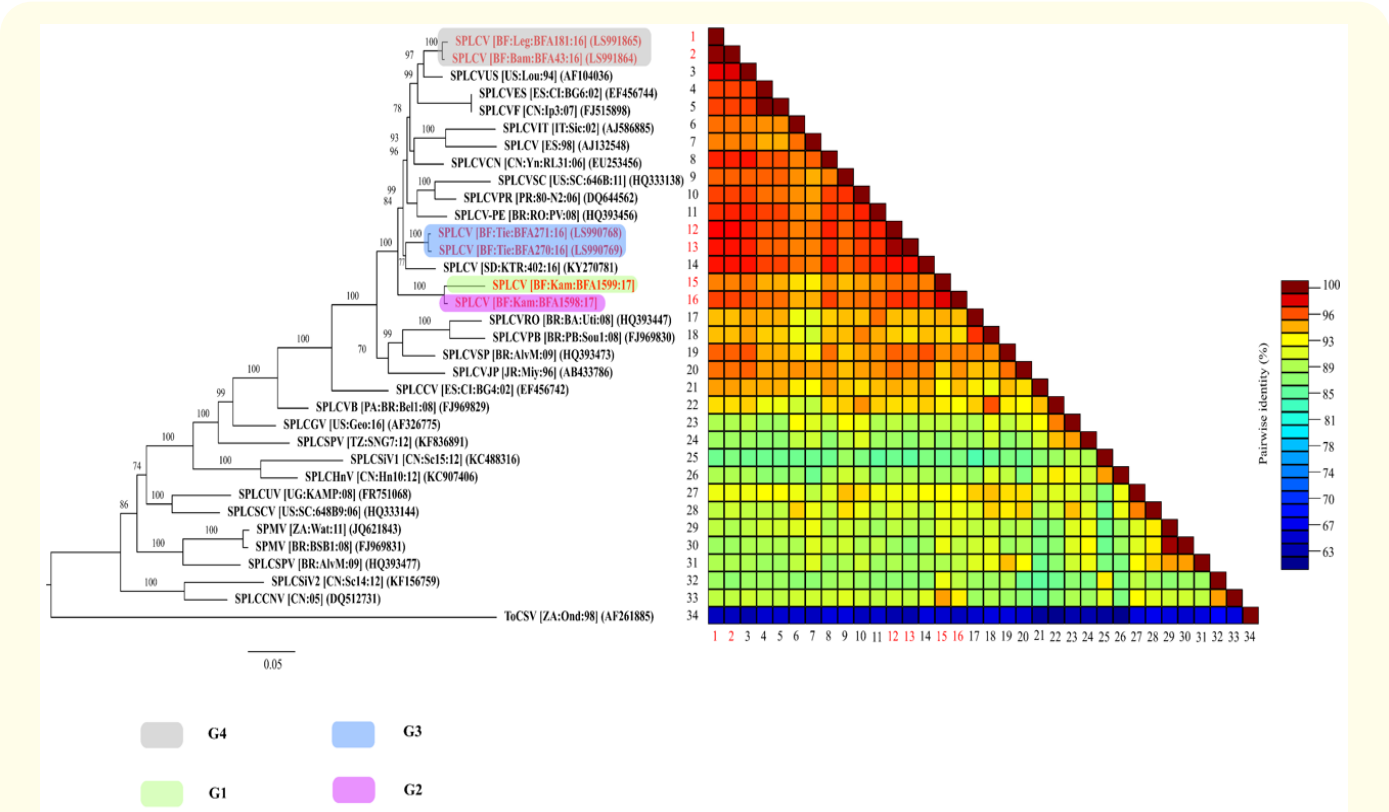


Figure 3: Maximum-likelihood phylogenetic tree obtained from alignment of 27 full genome nucleotide sequences of sweepoviruses and genome sequences for BFA43, BFA 181, BFA270, BFA271, BFA1598 and BFA1599 DNA-A. Bootstrap values are percentages with values shown at the nodes. The tree was rooted with a tomato curly stunt virus isolate (AF261885). Isolates from this study are red, GenBank sequences are black. The scale bar represents the number of nucleotide substitutions per site. A pairwise comparison matrix of complete genomes was performed using the same dataset and the identity values are presented in the lower triangular matrix, ordered according to the phylogenetic tree. The matrix uses a discontinuous range of color (red, yellow, green and blue) to differentiate two cut-off values representing the strain (93–96%, yellow–red) and the species (90–91%, blue–green) demarcation thresholds of begomoviruses.

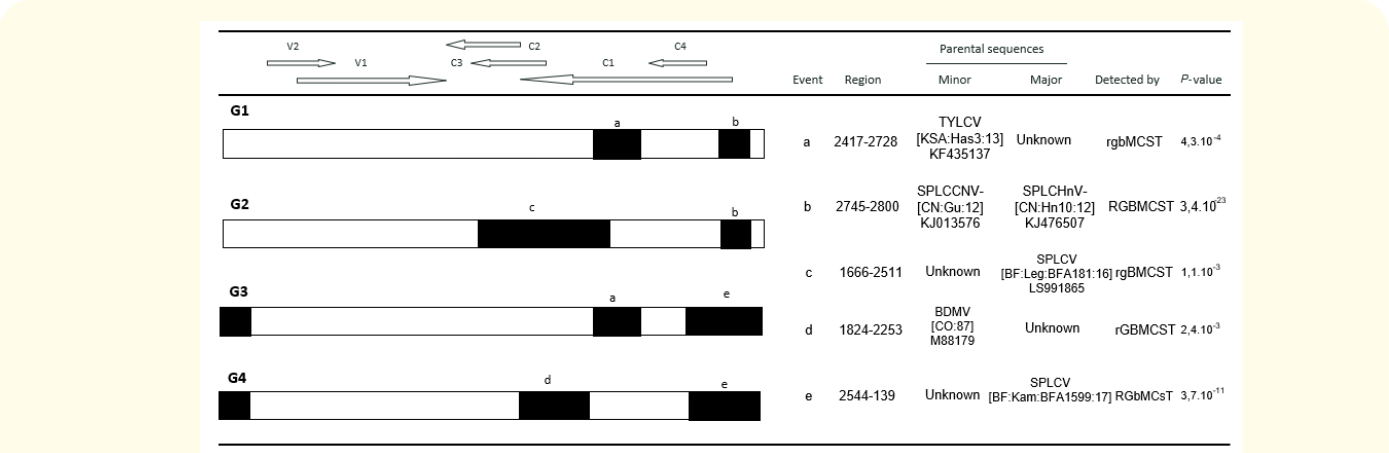


Figure 4: Recombinant regions detected within the sequences using RDP5. The genome at the top of the figure corresponds to the schematic representation of sequences below. Region coordinates are nucleotide positions of detected recombination breakpoints in the multiple sequence alignment used to detect recombination. Wherever possible, parental sequences are identified. “Major” and “Minor” parents are sequences that were used, along with the indicated recombinant sequence, to identify recombination. Whereas for each identified event the minor parent is apparently the contributor of the sequence within the indicated region, the major parent is the apparent contributor of the rest of the sequence. Note that the identified “parental sequences” are not the actual parents but are simply those sequences most similar to the actual parents in the analysed dataset. Recombinant regions and parental viruses were identified using the RDP (R), GENECONV (G), BOOTSCAN (B), MAXIMUM CHI SQUARE (M), CHIMAERA (C), SISTER SCAN (S) and 3Seq (T) methods. Whereas upper case letters imply a method detected recombination with a multiple comparison corrected P-value <0.01, lower case letters imply the method detected recombination with a multiple comparison corrected P-value <0.05 but >= 0.01.

In conclusion, this study confirms the presence of SPLCV on sweetpotato and *Ipomoea alba* in Burkina Faso and its recombinant nature. The six isolates form three distinct clades with known SPLCV isolates worldwide. This is the first report of the presence of sweepovirus on sweetpotato and *Ipomoea alba* plants in West Africa. This virus raises concerns for sweetpotato cultivation and food security in the region due to the weak of controls in Burkina Faso regarding the movement of plant material across borders.

Acknowledgments

The authors are grateful to the AgShare. Today team for their editorial support. This work was fully funded by the Bill and Melinda Gates Foundation and the Department for International Development of the UK [grant no. OPP1082413: “West African Virus Epidemiology (WAVE) for root and tuber crops”] through a subgrant from Université Félix Houphouët-Boigny (UFHB).

Bibliography

1. ICTV. International Committee on Taxonomy of Viruses (2018).

2. Rojas MR., et al. “Exploiting chinks in the plant’s armor: evolution and emergence of geminiviruses”. *Annual Review of Phytopathology* 43 (2005): 361-394.

3. Zerbini FM., et al. “ICTV virus taxonomy profile: Geminiviridae”. *Journal of General Virology* 98.2 (2017): 131-133.

4. Márquez-Martín B., et al. “Diverse population of a new bipartite begomovirus infecting tomato crops in Uruguay”. *Archives of Virology* 157.6 (2012): 1137-1142.

5. Briddon RW., et al. “Distinct evolutionary histories of the DNA-A and DNA-B components of bipartite begomoviruses”. *BMC Evolutionary Biology* 10 (2010): 97-114.

6. Argüello-Astorga GR and Ruiz-Medrano R. “An iteron-related domain is associated to Motif 1 in the replication proteins of geminiviruses: Identification of potential interacting amino acid-base pairs by a comparative approach”. *Archives of Virology* 146 (2001): 1465-1485.

7. Sánchez-Campos S., et al. “Fulfilling Koch’s postulates confirms the monopartite nature of tomato leaf deformation virus: a Begomovirus native to the New World”. *Virus Research* 173 (2013): 286-293.

8. Duffy S and Holmes EC. “Multiple introductions of the old world Begomovirus tomato yellow leaf curl virus into the new world”. *Applied and Environmental Microbiology* 73.21 (2007): 7114-7117.

9. Fauquet CM and Stanley J. “Revising the way we conceive and name viruses below the species level: a review of Geminivirus taxonomy calls for new standardized isolate descriptors”. *Archives of Virology* 150.10 (2005): 2151-2179.

10. Lozano G., et al. “Novel Begomovirus species of recombinant nature in sweet potato (*Ipomoea batatas*) and *Ipomoea indica*: taxonomic and phylogenetic implications”. *Journal of General Plant Pathology* 90 (2009): 2550-2562.

11. Miano DW., et al. “First report of a Begomovirus infecting sweetpotato in Kenya”. *Plant Disease* 90 (2006): 832.

12. Wasswa P., et al. “First identification of a sweet potato Begomovirus (sweepovirus) in Uganda: characterization, detection and distribution”. *Plant Pathology* 60 (2011): 1030-1039.

13. Esterhuizen LL., et al. “. Genetic identification of two sweet potato infecting begomoviruses in South Africa”. *Archives of Virology* 157 (2012): 2241-2245.

14. Mohammed HS., et al. “First report of sweet potato leaf curl virus infecting sweet potato in Sudan”. *Plant Disease* 101 (2017): 8-9.

15. Mbanzibwa DR., et al. “Small RNA deep sequencing-based detection and further evidence of DNA viruses infecting sweetpotato plants in Tanzania”. *Annals of Applied Biology* 165 (2014): 329-339.

16. Ling K-S., et al. “Experimental host range and natural reservoir of sweet potato leaf curl virus in the United States”. *Crop Protection* 30 (2011): 1055-1062.

17. Cuellar WJ., et al. “Synergistic interactions of begomoviruses with sweet potato chlorotic stunt virus (genus Crinivirus) in sweet potato (*Ipomoea batatas* L.). *Molecular Plant Pathology* 16.5 (2015): 459-471.

18. Doyle JJ and Doyle JL. “A rapid DNA isolation procedure for small quantities of fresh leaf tissue”. *Phytochemical Bulletin* 19 (1987): 11-15.

19. Trenado HP., et al. “Sweepoviruses cause disease in sweet potato and related *Ipomoea* spp.: fulfilling Koch’s postulates for a divergent group in the genus Begomovirus”. *PLoS One* 6 (2011): e27329.

20. Banks GK., et al. “A novel Geminivirus in *Ipomoea indica* (Convolvulaceae) from southern Spain”. *Plant Disease* 83.5 (1999): 486-495.

21. Yang CX., et al. “First Report of the occurrence of sweet potato leaf curl virus in tall morningglory (*Ipomoea purpurea*) in China”. *Plant Disease* 93.7 (2009): 764.

22. Tiendrébéogo F., *et al.* "Evolution of African cassava mosaic virus by recombination between bipartite and monopartite begomoviruses". *Virology Journal* 9 (2012): 67-73.

23. Ouattara A., *et al.* "Tomato leaf curl Burkina Faso virus: a novel tomato-infecting monopartite begomovirus from Burkina Faso". *Archives of Virology* 162.5 (2017): 1427-1429.

24. FAOSTAT. "FAO Statistical Databases" (2018).

25. Albuquerque LC., *et al.* "Genetic diversity and recombination analysis of sweepoviruses from Brazil". *Virology Journal* 9 (2012): 241-254.

26. Li R., *et al.* "Detection of geminiviruses in sweetpotato by polymerase chain reaction". *Plant Disease* 88 (2004): 1347-1351.

27. Inoue-Nagata AK., *et al.* "A simple method for cloning the complete Begomovirus genome using the bacteriophage ϕ 29 DNA polymerase". *Journal of Virological Methods* 116.2 (2004): 209-211.

28. Bao Y., *et al.* "Improvements to pairwise sequence comparison (PASC): a genome-based web tool for virus classification". *Archives of Virology* 159.12 (2014): 3293-3304.

29. Kumar S., *et al.* "MEGA7: Molecular evolutionary genetics analysis version 7.0 for bigger datasets". *Molecular Biology and Evolution* 33.7 (2016): 1870-1874.

30. Price MN., *et al.* "FastTree 2 - Approximately maximum-likelihood trees for large alignments". *PloS One* 5 (2010): e9490.

31. Muhire BM., *et al.* "SDT: A virus classification tool based on pairwise sequence alignment and identity calculation". *PLoS One* 9 (2014): e108277.

32. Lefeuvre P., *et al.* "Begomovirus "melting pot" in the south-west Indian Ocean islands: Molecular diversity and evolution through recombination". *Journal of General Virology* 88.12 (2007): 3458-3468.

33. Padidam M., *et al.* "Possible emergence of new geminiviruses by frequent recombination". *Virology* 265.2 (1999): 218-225.

34. Martin D and Rybicki E. "RDP: detection of recombination amongst aligned sequences". *Bioinformatics* 16.6 (2000): 562-563.

35. Martin DP., *et al.* "A modified bootscan algorithm for automated identification of recombinant sequences and recombination breakpoints". *AIDS Research and Human Retroviruses* 21.1 (2005): 98-102.

36. Maynard-Smith J. "Analyzing the mosaic structure of genes". *Journal of Molecular Evolution* 34 (1992): 126-129.

37. Martin DP., *et al.* "RDP2: Recombination detection and analysis from sequence alignments". *Bioinformatics* 21.2 (2005): 260-262.

38. Gibbs MJ., *et al.* "Sister-scanning: a Monte Carlo procedure for assessing signals in recombinant sequences". *Bioinformatics* 16 (2000): 573-582.

39. Boni ME., *et al.* "An exact nonparametric method for inferring mosaic structure in sequence triplets". *Genetics* 176 (2007): 1035-1047.

40. Martin DP., *et al.* "RDP4: Detection and analysis of recombination patterns in virus genomes". *Virus Evolution* 1 (2015): 1-5.

41. Brown JK., *et al.* "Revision of Begomovirus taxonomy based on pairwise sequence comparisons". *Archives of Virology* 160.6 (2015): 1593-1619.

Volume 2 Issue 10 October 2019

© All rights are reserved by Ezechiel Bionimian Tibiri, *et al.*