First report of chayote yellow mosaic virus in Côte d'Ivoire: a divergent strain infecting bitter melon

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Abstract

In this report, we present the first description of nine complete genome sequences of a new strain of chayote yellow mosaic virus (ChaYMV, genus *Begomovirus*, family *Geminiviridae*), provisionally named Côte d'Ivoire (ChaYMV-CI), isolated from bitter melon (*Momordica charantia*) leaf samples showing yellow mosaic symptoms collected in the region of Abidjan in Côte d'Ivoire in 2013 and 2019. The nucleotide sequences of ChaYMV-CI DNA-A-like genome share the highest nucleotide sequence identities (91.8–92.5%) with those of ChaYMV isolates from Benin and Cameroon. Phylogenetic analysis confirmed the segregation of ChaYMV isolates into two distinct clades and their affiliation to Old World monopartite begomoviruses. The discovery of a new strain of ChaYMV in West Africa, at the limit of the taxonomic demarcation threshold for begomoviruses (91%), call for further investigations into the molecular diversity, geographical distribution and host range of ChaYMV in Western Africa.

Keywords Begomovirus · Geminiviridae · Bitter melon · Momordica charantia · Côte d'Ivoire · West Africa

Begomoviruses (genus *Begomovirus*, family *Geminiviridae*) are a group of plant viruses responsible for serious diseases on a wide range of cultivated plants (Navas-Castillo et al 2011). Chayote yellow mosaic virus (ChaYMV, genus *Begomovirus*, family *Geminiviridae*) was described for the first time on a chayote (*Sechium edule*) diseased plant in Nigeria. ChaYMV is a begomovirus with a monopartite genome and a unique DNA-A-like component (AJ223191). Leke et al. (2020) described ChaYMV on bitter melon plant samples from Benin, Nigeria and Togo exhibiting leaf yellow mosaic

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symptoms in 2014. Bitter melon or "margose" (*Momordica charantia*; *Cucurbitaceae*) is widely used in Africa as a traditional medicine for the treatment of various diseases (Johnson et al. 2016).

Leaf samples from three bitter melon plants showing leaf yellow mosaic symptoms were collected in open fields in 2013 (Isolate CI111) at Port-Bouet (5.260387° N; 3.934848° W) and in 2019 (Isolates CI404 and CI405) at Abobo (5.40475° N, -4.03016° W) in the region of Abidjan in the south of Côte d'Ivoire (CI). These samples were dehydrated for long-term conservation. The presence of begomoviruses in these samples was later investigated by PCR using a set of degenerate primers designed to amplify the coat protein gene of Old World begomoviruses' DNA-A-like component (Clust4CP-F342/Clust4CP-R1032, Séka et al. 2016). PCR products of the expected size (≈ 690 bp) were obtained from the three analyzed samples and sequenced by direct sequencing in both directions. When analyzed using BLASTn, all three sequences shared the highest nucleotide sequence identities (93.7 to 94.4%) with ChaYMV isolates from Nigeria (AJ223191, KT454826-7), Cameroon (KT454821- KT454824), Benin (KT454819) and Togo (KT454820), suggesting the presence of ChaYMV in Côte d'Ivoire (Brown et al. 2015). No PCR product was obtained



when using degenerate primers designed to amplify begomovirus DNA-B sequences (PBL1V2040-PCRC1; Rojas et al., 1993) or DNA betasatellite sequences (Beta1/Beta2; Briddon et al. 2002).

To confirm the molecular identification of this begomovirus, the full-length viral genome sequences were amplified by rolling-circle amplification (RCA) using Phi29 DNA polymerase and sequenced. Amplification products were digested with BamHI and PstI endonucleases. Unique digestion products (~2,8 kb) corresponding to the putative monomeric full-length viral DNA genome were obtained, cloned into the pJET1.2 vector and sequenced. Nine complete DNA-A-like sequences of 2,782 to 2,786 nucleotides in length were obtained from samples CI111 (Acc. numbers OL744264-OL74427), CI404 (OL744268-OL744269) and CI405 (OL744270-OL744272). The DNA-A-like components presented features typical of Old World monopartite begomoviruses, with six open reading frames (ORFs) corresponding to V1 and V2 on the viral strand and C1, C2, C3 and C4 on the complementary strand (Rojas et al. 2005). Intergenic region (IR) sequences were 315 nucleotides long and contained (i) a characteristic inverted repeat potentially capable of forming a stem-loop structure including the conserved nonanucleotide sequence TAATATTAC that is present in almost all geminivirus virion-strand replication origins, (ii) the TATA box sequences involved in the transcription of complementary and viron-sense genes, and (iii) the putative Rep protein high-affinity core binding site GGGT (Arguello-Astorga and Ruiz-Medrano 2001). The corresponding iteron-related domain (IRD; Rep N-terminal domain interaction with *ori*-associated iteron) was identified as MSPPKRFLIN (Arguello-Astorga and Ruiz-Medrano 2001).

Pairwise identity comparison of all nucleotide sequences was performed using SDT v1.2 with pairwise deletion of gaps (Muhire et al. 2014). The nine DNA-A-like sequences of ChaYMV shared 94 to 100% nucleotide identity to each other. They shared the highest nucleotide sequence identities (91.8 to 92.5%) with the Beninese (KT454819) and Cameroonian (KT454821 and KT454822) isolates of the monopartite begomovirus ChaYMV. In accordance with the proposed begomovirus strain demarcation criteria (94% nucleotide identity in DNA-A and DNA-A-like nucleotide sequences; Brown et al. 2015), these Ivorian isolates should be assigned to a new strain of ChaYMV tentatively named ChaYMV-CI. Alignment of the nine newly described sequences along with the nine complete DNA-A-like sequences of ChaYMV available from GenBank and selected Old World monopartite begomoviruses originating from Africa were performed using the MUSCLE alignment tool available in MEGA6 (Tamura et al. 2013; Table 1). The 18 complete nucleotide sequences of ChaYMV share between 90.4% and 100% pairwise nucleotide identity (Fig. 1). While the complete nucleotide sequences of ChaYMV isolates CI404 and CI405 collected in 2019 are genetically similar (between 99.2% and 99.9% identity), those of isolate CI111 collected in 2013 were more diverse (between 95.3% and



Fig. 1 Maximum-likelihood phylogenetic tree and color-coded matrix of pairwise nucleotide identity inferred from alignments of complete DNA-A-like sequences of chayote yellow mosaic virus and selected Old World monopartite begomoviruses originating from Africa. Only bootstrap values above 70% are shown (1 000 replicates). The matrix

uses a discontinuous range of three shades of color (red, green, and blue) differentiating two cut-off values representing the strain (94%, green–red) and the species (91%, blue-green) demarcation thresholds of begomoviruses. For begomovirus acronyms, see Table 1

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Table 1

D	Begomovirus isolates	Abbreviated names	Accessions numbers
_	Chayote yellow mosaic virus - Cote d'Ivoire [Cote d'Ivoire-Momordica charantia-CI405- BamA-2019]	ChYMV-CI [CI-Mch-Cl405-BamA-19]	OL744264
5	Chayote yellow mosaic virus - Cote d'Ivoire [Cote d'Ivoire-Momordica charantia-CI405- PstB-2019]	ChYMV-CI [CI-Mch-CI405-PstB-19]	OL744265
ũ	Chayote yellow mosaic virus - Cote d'Ivoire [Cote d'Ivoire-Momordica charantia-CI404- PstC-2019]	ChYMV-CI [CI-Mch-CI404-PstC-19]	OL744266
4	Chayote yellow mosaic virus - Cote d'Ivoire [Cote d'Ivoire-Momordica charantia-CI404- PstD-2019]	ChYMV-CI [CI-Mch-CI404-PstD-19]	OL744267
5	Chayote yellow mosaic virus - Cote d'Ivoire [Cote d'Ivoire-Momordica charantia-CI405- BamC-2019]	ChYMV-CI [CI-Mch-CI405-BamC-19]	OL744268
6	Chayote yellow mosaic virus - Cote d'Ivoire [Cote d'Ivoire-Momordica charantia- CI111-H1-2013]	ChYMV-CI [CI-Mch-CIIII-HI-13]	OL744269
L	Chayote yellow mosaic virus - Cote d'Ivoire [Cote d'Ivoire-Momordica charantia- C1111-H12-2013]	ChYMV-CI [CI-Mch-CI111-H12-13]	OL744270
8	Chayote yellow mosaic virus - Cote d'Ivoire [Cote d'Ivoire-Momordica charantia- C1111-H3-2013]	ChYMV-CI [CI-Mch-CI111-H3-13]	OL744271
6	Chayote yellow mosaic virus - Cote d'Ivoire [Cote d'Ivoire-Momordica charantia- C1111-H5-2013]	ChYMV-CI [CI-Mch-CI111-H5-13]	OL744272
10	Chayote yellow mosaic virus - [Nigeria-2005]	ChYMV-[NG-05]	AJ223191
11	Chayote yellow mosaic virus - [Cameroon-2014]	ChYMV-[CA-14]	KT454823
12	Chayote yellow mosaic virus - [Nigeria-2014]	ChYMV-[NG-14]	KT454827
13	Chayote yellow mosaic virus - [Cameroon-2014]	ChYMV-[CA-14]	KT454824
14	Chayote yellow mosaic virus - [Togo-2014]	ChYMV-[TG-14]	KT454820
15	Chayote yellow mosaic virus - [Benin-2014]	ChYMV-[BN-14]	KT454819
16	Chayote yellow mosaic virus - [Nigeria-2014]	ChYMV-[NG-14]	KT454826
17	Chayote yellow mosaic virus - [Cameroon-2014]	ChYMV-[CA-14]	KT454822
18	Chayote yellow mosaic virus - [Cameroon-2014]	ChYMV-[CA-14]	KT454821
19	Tomato interveinal yellowing virus - [Cote d'Ivoire-2019]	ToIYV-[CI-19]	MW057361

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100% identity) and divergent (between 94.0% and 94.9% identity). A maximum likelihood (ML) phylogenetic tree was constructed using MEGA6 (Tamura et al. 2013). The ML tree confirms the segregation of ChaYMV isolates into two distinct clades (bootstrap value 100%, Fig. 1).

In conclusion, for the first time we report complete genome sequences of ChaYMV isolates infecting bitter melon in Côte d'Ivoire. These isolates represent a new strain, at the limit of the taxonomic demarcation threshold for begomoviruses (91%) and tentatively named ChaYMV-CI, and form a distinct phylogenetic clade among ChaYMV isolates. Our results call for further investigations into the molecular diversity, geographical distribution and host range of ChaYMV in Western Africa.

GenBank accession numbers OL744264—OL744272.

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Declarations

Ethical approval This article does not contain any studies with human participants or animals performed by any of the authors.

Conflict of interest The authors declare that they have no conflict of interest.

References

- Arguello-Astorga GR, Ruiz-Medrano R (2001) An iteron-related domain is associated to Motif 1 in the replication proteins of geminiviruses: identification of potential interacting amino acidbase pairs by a comparative approach. Arch Virol 146:1465–1485. https://doi.org/10.1007/s007050170072
- Briddon RW, Bull SE, Mansoor S, Amin I, Markham PG (2002) Universal primers for the PCR-mediated amplification of DNA beta - A molecule associated with some monopartite begomoviruses. Mol Biotechnol 20:315–318. https://doi.org/10.1385/MB:20:3:315
- Brown JK, Zerbini FM, Navas-Castillo J, Moriones E, Ramos-Sobrinho R, Silva JCF, Fiallo-Olive E, Briddon RW, Hernandez-Zepeda C, Idris A, Malathi VG, Martin DP, Rivera-Bustamante R, Ueda S, Varsani A (2015) Revision of Begomovirus taxonomy based on pairwise sequence comparisons. Arch Virol 160:1593–1619. https://doi.org/10.1007/s00705-015-2398-y
- Johnson RC, Houéto EE, Boni G, Kpètèhoto WH, Dougnon V, Pognon E, Assogba F, Loko F, Boko M, Gbénou J (2016) Étude ethnobotanique et phytochimique de *Momordica charantia* Linn (Cucurbitaceae) à Cotonou au Bénin. J Appl Biosci 106:10249–10257. https://doi.org/10.4314/jab.v106i1.4

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D	Begomovirus isolates	Abbreviated names	Accessio
20	Tomato leaf curl Kunene virus - [Namibia-2019]	ToLCKunV-[NA-19]	MT0459
21	Tobacco leaf curl Comoros virus - [Comoros-2007]	TbLCKMV-[KM-07]	AM7017
22	Tobacco leaf curl Zimbabwe virus - [Zimbabwe]	TbLCZWV-[ZW]	AF3503
23	Tomato curly stunt virus - [South Africa]	ToCSV-[SA]	AF2618
24	Tomato leaf curl Mali virus - [Mali]	ToLCMLV-[ML]	AY5029
25	Tomato leaf curl Cameroon virus - [Cameroon-2008]	ToLCCMV-[CA-08]	FM2102

- Leke WN, Brown JK, Fondong VN (2020) First Report of Chayote Yellow Mosaic Virus (ChaYMV) and Its Associated Betasatellite Infecting Papaya (Carica papaya) in Cameroon. Plant Dis 104:1566. https://doi.org/10.1094/PDIS-11-19-2293-PDN
- Muhire BM, Varsani A, Martin DP (2014) SDT: A Virus Classification Tool Based on Pairwise Sequence Alignment and Identity Calculation. PLoS One 9:9
- Navas-Castillo J, Fiallo-Olivé E, Sánchez-Campos S (2011) Emerging virus diseases transmitted by whiteflies. Annu Rev Phytopathol 49:219–248
- Rojas MR, Gilbertson RL, Russel DR, Maxwell DP (1993) Use of degenerate primers in the polymerase chain reaction to detect whitefly-transmitted geminivirus. Plant Dis 77:340–347
- Rojas MR, Hagen C, Lucas WJ, Gilbertson RL (2005) Exploiting chinks in the plant's armor: evolution and emergence of geminiviruses. Annu Rev Phytopathol 43:361–394
- Séka K, Ouattara A, Assiri KP, Kra KD, Hoareau M, Lefeuvre P, Diallo HA, Lett JM (2016) First reports of Cotton leaf curl Gezira virus and Okra yellow crinkle virus associated with okra leaf curl disease in Côte d'Ivoire. New Dis Rep 34:8–8. https://doi.org/10. 5197/j.2044-0588.2016.034.008
- Tamura K, Stecher G, Peterson D, Filipski A, Kumar S (2013) MEGA6: Molecular Evolutionary Genetics Analysis Version 6.0. Mol Biol Evol 30:2725–2729. https://doi.org/10.1093/molbev/mst197