



## First reports of Cotton leaf curl Gezira virus and Okra yellow crinkle virus associated with okra leaf curl disease in Côte d'Ivoire

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Okra leaf curl disease (OLCD) is commonly observed in okra (*Abelmoschus esculentus*) crops in several African countries (N'Goussan *et al.*, 1992). Affected plants are severely stunted with apical leaf curl (upward or downward), distortion and thickening of the veins. In Africa, OLCD is associated with a complex of several strains of two begomovirus species: Cotton leaf curl Gezira virus (CLCuGV; Idris & Brown, 2002) and Okra yellow crinkle virus (OYCrV; Shih *et al.*, 2007).

In 2012 and 2013, severe symptoms of leaf curling, deformation, and vein thickening (Fig. 1), resembling those of okra leaf curl disease were observed on okra in four localities in south-eastern Côte d'Ivoire (Table 1). Fourteen leaf samples with symptoms were collected and tested for the presence of begomoviruses using a polymerase chain reaction (PCR) assay with a set of degenerate primers designed to amplify the coat protein gene of Old World begomoviruses (Clust4CP-F342, 5'-TATMATCATTTCCACBCCVG-3'; Clust4CP-R1032, 5'-GCATGAGTACATGCCATATAC-3'). PCR products of the expected sizes were obtained for nine samples suggesting the presence of Old World monopartite begomoviruses in all four localities (Table 1).

PCR positive samples were further processed and full-length viral genomes were amplified from four samples (Table 1) by rolling-circle amplification, cloned using the *Bam*HI restriction enzyme and sequenced. One complete genome sequence (GenBank Accession No. KX100570) showed the highest pairwise sequence identity of 99% (100% coverage) with isolates of the Niger strain of CLCuGeV from Niger (CLCuGeV-NE[NE:Sad:NG2FL:Ok:07], FJ469627) and Burkina Faso (CLCuGeV-NE[BF:Kap:Ok4:08], FN554524). The other three sequences (KX100571 to KX100573) showed the highest pairwise sequence identity of 98-99% (100% coverage) with isolates of the Mali strain of OYCrV from Mali (OYCrV-ML[ML:Mlo1:04], DQ902715; OYCrV-ML[ML:Bam4:06], EU024119). A maximum likelihood phylogenetic tree, produced from alignments of publicly available begomovirus sequences (MEGA6; Tamura *et al.*, 2013), confirmed the genetic relationship of Ivorian CLCuGeV and OYCrV isolates with isolates from three neighbouring countries, Burkina Faso, Mali and Niger (Fig. 2).

To our knowledge, this is the first report of CLCuGeV and OYCrV

implicated in OLCD in Côte d'Ivoire. Our results confirm that OLCD is associated with a complex of several strains of CLCuGeV and OYCrV in sub-Saharan Africa (Tiendrebeogo *et al.*, 2010) and show that Ivorian isolates are genetically closely related to strains described in West Africa.

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Figure 1

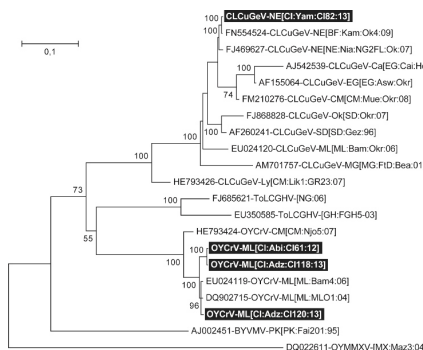


Figure 2

Table 1 | Geographical origin of okra samples (each corresponding to a distinct leaf from a different plant) collected in Côte d'Ivoire and description of Cotton leaf curl Gezira virus (CLCuGeV) and Okra yellow crinkle virus (OYCrV) isolates

Sample	Locality*	Sampling date (dd/mm/yyyy)	Location (Latitude, Longitude)	PCR diagnosis (CLCuGeV/OYCrV) (1032/3059)	Accession	GenBank Accession No.
01016	Toumodi	16/01/2012	06°51'07.30"N, 06°02'02.20"W	-	Not available	-
01032	Toumodi	16/01/2012	06°51'07.30"N, 06°02'02.20"W	+	Not available	-
01041	Toumodi	16/01/2012	06°51'07.30"N, 06°02'02.20"W	+	Not available	-
01042	Toumodi	16/01/2012	06°51'07.30"N, 06°02'02.20"W	+	Not available	-
01045	Abidjan	16/01/2012	06°16'30.20"N, 04°18'08.70"W	-	Not available	-
01061	Abidjan	16/01/2012	06°16'30.20"N, 04°18'08.70"W	+	OYCrV-Mali (OYCrV-ML) [ML: Mlo1:04]	KX100571
01062	Yamoussoukro	24/08/2013	06°52'00.00"N, 06°24'53.40"W	+	OYCrV-Mali (OYCrV-ML) [ML: Bam4:06]	KX100570
01065	Yamoussoukro	24/08/2013	06°52'00.00"N, 06°24'53.40"W	-	Not available	-
01119	Abidjan	26/08/2013	06°10'23.66"N, 03°54'18.89"W	+	OYCrV-Mali (OYCrV-ML) [ML: Mlo1:04]	KX100572
01119	Abidjan	26/08/2013	06°10'23.66"N, 03°54'18.89"W	-	Not available	-
01120	Abidjan	26/08/2013	06°10'23.66"N, 03°54'18.89"W	+	OYCrV-Mali (OYCrV-ML) [ML: Mlo1:04]	KX100573
01128	Yamoussoukro	22/07/2013	06°52'36.80"N, 06°28'50.83"W	-	Not available	-
01129	Yamoussoukro	22/07/2013	06°52'36.80"N, 06°28'50.83"W	+	Not available	-
01132	Yamoussoukro	22/07/2013	06°52'36.80"N, 06°28'50.83"W	+	Not available	-

Figure 3

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