First report of apricot vein clearing-associated virus (AVCaV) infecting *Prunus domestica* L. revealed by high-throughput sequencing in Morocco

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Plum (*Prunus domestica* L., Rosaceae) trees, like many stone fruit trees, are known to be infected by numerous plant viruses, predominantly as consequence of their clonal mode of propagation and perennial cultivation (Jelkmann and Eastwell, 2011). Apricot vein clearing-associated virus (AVCaV) is a member of the genus *Prunevirus* in the family *Betaflexiviridae*. AVCaV was first reported in Italy infecting apricot (*P. armeniaca* L.) associated with foliar vein clearing symptoms (Elbeaino et al. 2014). It has also been detected in various *Prunus* species, like plum, Japanese plum (*P. salicina* L.), sour cherry (*P. cerasus* L.), and Japanese apricot (*P. mume* L.), apricot and peach (*P. persica* L.) sourced from Asian and European countries (Marais et al. 2015), as well as in the ornamental Myrobolan plum (*P. cerasifera* L.) in Australia (Kinoti et al. 2017). In 2018, during the vegetative season, a survey was carried out in two different apricot and plum orchards in the southern region of Agdez (Agadir, Morocco) where stone fruit trees are grown. Five branches with leaves were sampled from three apricot and three plum trees of unknown cultivars, all asymptomatic. Total RNA was extracted from 100 mg plant tissue (leaves and cambial scrapping) using RNeasy Plant Mini Kit (QIAGEN, Hilden, Germany) and separate samples (one per species) were used for library preparation (NEBNext Ultra RNA library kit; New England BioLabs, MA, USA), and sequencing (Illumina NextSeq v2, totRNA sequencing) at Admera Health (New Jersey, USA). All generated reads (6,756,881) from the plum sample were quality filtered and submitted to the VirusDetect pipeline (Zheng et al., 2017). The plum cDNA library, a total of 20 viral contigs (68-1928 bp) mapped to several AVCaV accessions in GenBank. A reference mapping (CLC Genomics Workbench 12, Qiagen, Denmark) was conducted against all four available AVCaV full genomes (KM507062-63, KY132099 and HG008921), revealing 100% coverage of the full sequence (8358 nt) with 97-98 % nucleotide (nt) identities (BLASTn). Analysis of the derived sequences allowed to identify the location of the four predicted ORFs i.e. (ORF1: 6066 nt/2,021 aa), (ORF2: 1383 nt/460 aa), (ORF3: 666 nt/221 aa) and (ORF4: 420 nt/139 aa), previously described for the AVCaV genome (Elbeaino et al. 2014). The amino acid sequences of the encoded proteins of AVCaV isolate from Morocco also shared 97-98% identities with the corresponding sequences of complete genome AVCaV isolates in GenBank. To confirm the detection of AVCaV in the three plum samples, specific RT-PCR primers (VC37657s: 5'-CCATAGCCACCCTTTTTCAA-3' / VC28239a: 5'-GTCGTCAAGGGTCCAGTGAT-3') (Elbeaino et al. 2014) were used and the expected 330 bp fragment from the replicase gene was amplified in all three samples and subsequently sequenced (MT980794-96). Sanger sequences were 100% identical to corresponding HTS derived sequence. This is the first report of AVCaV infecting plum in Africa. The incidence of AVCaV in Moroccan *Prunus* species is unknown. Plum trees from the surveyed orchards were also confirmed to be co-infected with little cherry virus 1 (LChV-1) using HTS. Further investigation is required to determine the impact of AVCaV on these asymptomatic plum trees and other stone fruits species.

References :

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