



First report of *Schlumbergera virus X* infecting dragon fruit (*Selenicereus* spp.) in Portugal

Natália Tomás Marques¹ · Ana Rita Trindade¹ · Amílcar Duarte¹

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Dragon fruit (*Selenicereus* spp.) is a fruit crop with a high commercial value. In recent years, there has been an increased interest in this crop in the southern region of Portugal, with the introduction of new cultivars (Trindade et al. 2023). In June 2022, a survey was conducted to monitor the sanitary status of a plantation in Moncarapacho. Seven two- to three-year-old plants of *Selenicereus stenopterus* (F.A.C. Weber) D.R.Hunt × *S. undatus* (Haw.) D.R.Hunt cv. ‘Connie Mayer’ were sampled. The plants exhibited a range of symptoms on their cladodes, including systemic mottling, small irregular chlorotic spots which may also appear ring-shaped, and chlorotic patches. Total RNA was extracted from the symptomatic cladodes and one asymptomatic plant. The RNA was subjected to reverse transcription (RT)-PCR using Ribolock (Thermo Scientific, USA), M-MuLV Reverse Transcriptase (Roche, Sigma-Aldrich), and Dream Taq DNA polymerase (Thermo Scientific, USA). The degenerate primer pair Potex5Fw and Potex2RC (van der Vlugt and Berendsen 2002) were used for the amplification of a 584-bp sequence. No amplification was observed in the asymptomatic plant sample. The amplified sequences were cloned into the pGEM[®]-T Easy vector and sequenced in both directions. *Schlumbergera virus X* (SchVX) was identified in the seven plant samples.

Pairwise multiple sequence alignment of all consensus sequences identified three distinct sequences, E3(D1) (E3 is the name of the isolate, D1 is the identification of the clone), P2(F7) and P3(C9), which shared 98.2% nucleotide identity. The 584 bp cloned sequences were confirmed by BLASTn in NCBI (<https://blast.ncbi.nlm.nih.gov/>) to share nucleotide sequence homology of 99.41–99.59% with an RNA-dependent RNA polymerase (RdRp) sequence from SchVX from Spain (GenBank Accession No. MZ614940, Janssen et al. 2021). The nucleotide sequences E3(D1), P2(F7) and P3(C9) were deposited in the GenBank database under the Acc. Nos. PP993267, PP993268 and PP993269, respectively. As a secondary confirmation, a primer pair was designed targeting the RdRp gene in a region upstream of the previously amplified sequence. The primer pair SchVXF1 (5'-CCCGMARAACCTGAAATGCCA-3') and SchVXR1 (5'-GTTTTRTCCCAGAAGGCYTC-3') was designed by multiple alignment of available complete gRNA sequences (AY366207, KP090203, KU854929, NC_011659, and OL584345) and amplifies 768 bp. The RT-PCR amplicons were cloned and sequenced as previously described, yielding two distinct nucleotide sequences with 98.7% identity. BLASTn analysis showed that P2(G9) and P3(H9) had the highest identity (88.93% and 88.93%, respectively) with the SchVX isolates OQ559395 and OQ559395 from Ecuador, respectively. The two nucleotide sequences P2(G9) and P3(H9) were deposited in GenBank under the Acc. Nos. PP993270 and PP993271, respectively. The RdRp of isolates P2 and P3 was therefore amplified in two different regions. To the best of our knowledge, this is the first report of the virus in Portugal.

✉ Natália Tomás Marques
nmarques@ualg.pt

Ana Rita Trindade
artrindade@ualg.pt

Amílcar Duarte
aduarte@ualg.pt

¹ MED - Mediterranean Institute for Agriculture, Environment and Development, CHANGE- Global Change and Sustainability Institute, Faculdade de Ciências e Tecnologia, Universidade do Algarve, Campus de Gambelas, Edif. 8, Faro 8005-139, Portugal

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Data Availability All data analysed during this study are included in this published article.

Declarations

Conflict of interest The authors declare no conflict of interest.

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