

First report of *Tamarillo fruit ring virus* in *Solanum* spp. in Uganda, Tanzania and Rwanda

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Between July and November 2021, diseased fruit of *Solanum melongena* from Rwanda, *S. aethiopicum* from Tanzania, and *S. macrocarpon* and *S. aethiopicum* from Uganda were intercepted by the Animal and Plant Health Agency at Heathrow Airport and submitted to Fera Science Ltd. for diagnosis. The fruit exhibited putative symptoms of virus infection including distortion, arcs and pale mottling.

The *S. melongena* sample was tested by ELISA for *Cucumber mosaic virus* (Agdia, USA), *Pepino mosaic virus* (Bioreba, Switzerland), *Tomato spotted wilt virus* (DSMZ, Germany), and potyvirus (DSMZ, Germany). It was positive only for potyviruses. RNA was extracted from the sample and tested by RT-PCR using universal potyvirus primers (van der Vlugt et al., 1999). The PCR product was sent to Eurofins (Germany) for Sanger sequencing. Sequence comparison by BLASTn confirmed the presence of a potyvirus, with 99% nucleic acid identity to *Tamarillo fruit ring virus* (TaFRV; GenBank Accession No. MZ031983) detected in *S. betacum* in Rwanda. The sequence was deposited in GenBank (OR963261).

Solanum aethiopicum fruits from Tanzania and Uganda were tested as above by ELISA with the addition of *Chilli vein mottle virus* (DSMZ,

Germany). They were positive only for potyvirus. Extracted RNA was tested by RT-PCR and the products were sent for Sanger sequencing as described previously. BLASTn comparison identified them to be TaFRV (Tanzania, OR963263; Uganda, OR963262) with 95–96% nucleic acid identity to GenBank Accession No. MZ031983.

Solanum macrocarpon was tested as above, with additional ELISA tests for *Eggplant mosaic virus* (Nano Diagnostics, USA), *Eggplant mottled dwarf virus* (Loewe, Germany), *Tomato mosaic virus* (DSMZ, Germany), *Tomato ringspot virus* (Agdia, USA) and tospovirus (Bioreba, Switzerland). Again only potyvirus was detected. The result was confirmed using RT-PCR as before, and the amplicon sequence was identified using BLASTn as TaFRV (OR963262) with 96% nucleic acid identity to GenBank Accession No. MZ031983.

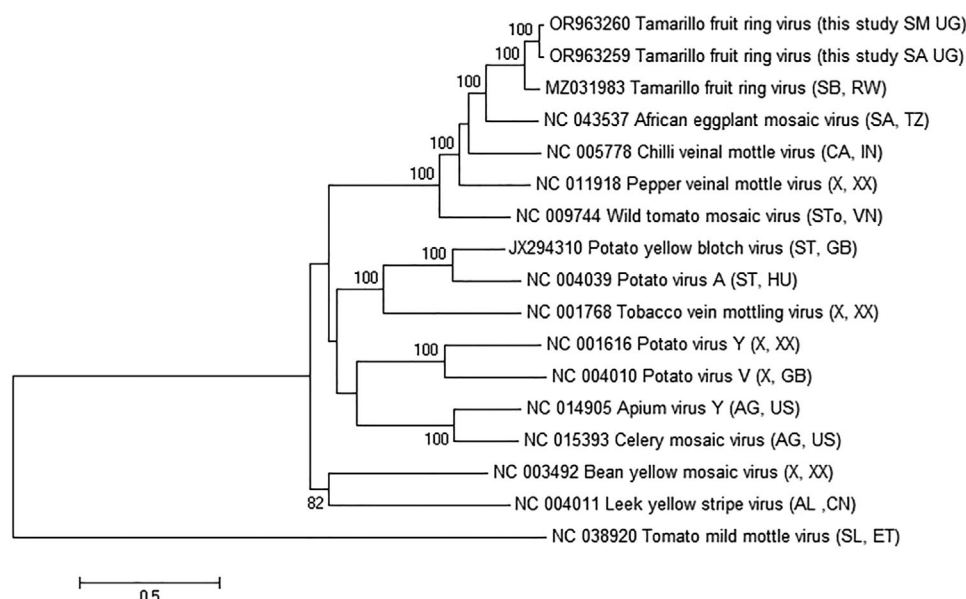
For confirmation, a specific real-time RT-PCR test for TaFRV was developed using Primer Express v.2 (Life Technologies Co., USA) (Table 1). All four samples tested positive. To obtain the genome of the virus, both *S. macrocarpon* and *S. aethiopicum* from Uganda were tested by high throughput sequencing on a MiSeq Sequencer (Illumina, UK) (Fowkes et al., 2021). The complete coding sequence was obtained

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**TABLE 1** Primer and probe sequences for specific real-time RT-PCR detection of Tamarillo fruit ring virus (TaFRV).

Primer/probe name	Sequence (5'-3')
TaFRV F	TGT CAA AGA TGC TAA CTG AGA AAC ACT
TaFRV R	GWT TTT GTC CCA TAT GTT TTG ATC TCT
TaFRV Pe	FAM-TAA GGT GAC AAA GGT TGA TGG TCG AAC AAT G-TAMRA

**FIGURE 1** Maximum-likelihood tree generated for the complete coding sequence for two samples from this study as well as other potyviruses publicly available on GenBank. Notation for sequences for this study as follows accession number, virus name, host name, country. Host viruses were isolated from: *Allium spp.*, AL; *Apium graveolens*, AS; *Capsicum annuum*, CA; *Solanum aethiopicum*, SA; *Solanum betaceum*, SB; *Solanum melongena*, SM; *Solanum torvum*, STo; *Solanum tuberosum*, ST; *Solanum lycopersicum*, SL; *Unknown host*, X. Original country that the virus was isolated in: China, CN; Ethiopia, ET; Great Britain, GB; Hungary, HU; India, IN; Rwanda, RW; Tanzania, TZ; Uganda, UG; United States of America, US; Vietnam, VN; Unknown origin, XX.

for each sample and the sequences were deposited in GenBank (*S. aethiopicum*-Uganda, OR963259; *S. macrocarpon*-Uganda, OR963260). A phylogenetic analysis for both sequences with other potyviruses was inferred by using the maximum-likelihood method (Jukes & Cantor, 1969) (Figure 1). This showed that the sequence isolates obtained in this study are closely related to MZ031983 and align alongside other solanaceous potyviruses.

Information on TaFRV is limited with a single unpublished Genbank accession from 2021 in *S. betaceum* in Rwanda. The additional reports here from *S. melongena* (Rwanda), *S. aethiopicum* (Uganda and Tanzania), and *S. macrocarpon* (Uganda) indicate that this virus has a wider host range and is more widely distributed than reported previously. In 2022, Rwanda produced 89,265 t of aubergine (FAOSTAT, 2024), and although production figures are not available for Uganda and Tanzania, the produce from crops in these three countries represent major export commodities. It is difficult to gauge the potential impact on production on the region and this should be monitored. The real-time RT-PCR primers used in this study can be used for easier and reliable virus detection.

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