This form should be used for all taxonomic proposals. Please complete all those modules that are applicable (and then delete the unwanted sections). For guidance, see the notes written in blue and the separate document "Help with completing a taxonomic proposal"

Please try to keep related proposals within a single document; you can copy the modules to create more than one genus within a new family, for example.

MODULE 1: **TITLE, AUTHORS, etc**

Code assigned:	***2013.003aP***	(to be completed by ICTV officers)

Short title: One new species in the family *Alphaflexiviridae*
(e.g. 6 new species in the genus *Zetavirus*)

Modules attached (modules 1 and 9 are required)					
	1 ☒	**2** ☒	**3** ☐	**4** ☐	**5** ☐
	6 ☐	**7** ☐	**8** ☐	**9** ☒	

Author(s) with e-mail address(es) of the proposer:

Jan Kreuze (j.kreuze@cgiar.org) on behalf of the *Flexiviridae* SG

List the ICTV study group(s) that have seen this proposal:

A list of study groups and contacts is provided at http://www.ictvonline.org/subcommittees.asp . If in doubt, contact the appropriate subcommittee chair (fungal, invertebrate, plant, prokaryote or vertebrate viruses)	*Flexiviridae* SG

ICTV-EC or Study Group comments and response of the proposer:

Date first submitted to ICTV:
Date of this revision (if different to above):

MODULE 2: **NEW SPECIES**

creating and naming one or more new species.

If more than one, they should be a group of related species belonging to the same genus. All new species must be placed in a higher taxon. This is usually a genus although it is also permissible for species to be "unassigned" within a subfamily or family. Wherever possible, provide sequence accession number(s) for one isolate of each new species proposed.

Code	***2013.003aP***	(assigned by ICTV officers)

To create 1 new species within:

Genus:	***Mandarivirus***
Subfamily:	
Family:	***Alphaflexiviridae***
Order:	***Tymovirales***

Fill in all that apply.

- If the higher taxon has yet to be created (in a later module, below) write "**(new)**" after its proposed name.
- If no genus is specified, enter "**unassigned**" in the genus box.

And name the new species:	**GenBank sequence accession number(s) of reference isolate:**
Citrus yellow vein clearing virus	JX040635

Reasons to justify the creation and assignment of the new species:

- Explain how the proposed species differ(s) from all existing species.
  - If species demarcation criteria (see module 3) have previously been defined for the genus, **explain how the new species meet these criteria**.
  - If criteria for demarcating species need to be defined (because there will now be more than one species in the genus), please state the proposed criteria.
- Further material in support of this proposal may be presented in the Appendix, Module 9

The family *Alphaflexiviridae* contains viruses with flexuous filamentous virions that infect plants and a few viruses discovered in plant-infecting fungi. They share a distinct lineage of alphavirus-like replication proteins that is unusual in lacking any recognized protease domain. Currently throughout the family, isolates of different species should have less than about 72% nt identity (or 80% aa identity) between their respective CP or polymerase genes. Based on this proposal (see below) however we now recommend that biological and serological differences should also be taken into account in case of closely related viruses. Viruses from different genera usually have less than about 45% nt identity in their CP or polymerase genes.

Table 1: Distinguishing properties of genera in the family *Alphaflexiviridae*

Genus	Host	Virion length (nm)	ORFs	Rep[a] (kDa)	CP[b]
Allexivirus	plants	ca. 800	6	170–195	26–29
Botrexvirus	fungi	ca.720	5	158	43
Lolavirus	plants	640	6	196	32
Mandarivirus	plants	650	6	187	34
Potexvirus	plants	470–580	5	150–195	18–27
Sclerodarnavirus	fungi	n/a[c]	1	193	n/a[c]

[a]Rep, replication protein size (kDa).
[b]CP, coat protein size (kDa).
[c]No virions found.

Citrus yellow vein clearing virus CYVCV (Loconsole et al., 2013)

Yellow vein clearing disease (YVCD) was first observed in Pakistan in 1988 in lemon and sour orange trees and has since been reported by several authors associated with a filamentous virus. The sequence of CYVCV isolate Y1 from Adana, Turkey, was determined by deep sequencing small RNA fractions from a yellow vein clearing disease (YVCD) affected lemon (cultivar Kütdiken) and the virus transmitted by mechanical and graft inoculation of herbaceous and citrus indicator plants. A polyclonal antiserum was developed from CYVCV-Y1 and used in western blot assays to characterize the coat protein of CYVCV-Y1 and determine its serological relationship with related viruses. Contigs assembled from the Illumina sequenced short reads were used to construct the whole genome of Citrus yellow vein clearing virus (CYVCV), consisting of a positive-sense RNA of 7,529 nucleotides and containing six predicted open reading frames. CYVCV is closely related to *Indian citrus ringspot virus* (ICRSV) (Mandarivirus, Alphaflexiviridae) with an overall nucleotide sequence identity of ≈74%, just above the suggested species demarcation limit defined for the family. Although the two viruses were similar with regard to genome organization, viral particles, and herbaceous host range, CYVCV causes a different disease in citrus and is serologically distinct from ICRSV. Virus specific primer pairs were designed and used to detect the virus by conventional and quantitative reverse transcription-polymerase chain reaction on yellow vein clearing symptomatic field trees as well as graft- and mechanically inoculated host plants, confirming the association of the virus with the disease. Collectively, these data suggest that CYVCV is the causal agent of YVCD and represents a new species in the genus *Mandarivirus*.

This is the second mandarivirus species to be recognized and the criteria applied have relied heavily on biological and serological differences to justify the slightly higher nucleotide and amino acid sequence found than recommended for species demarcation in the family. Since we feel these attributes should always be taken into consideration throughout the family for species demarcation, we recommend that these two criteria be added as principal criteria for species demarcation within the entire family, besides the sequence similarity criteria which already exist.

MODULE 9: **APPENDIX**: supporting material

additional material in support of this proposal

References:

Loconsole, G., Önelge, N., Potere, O., Giampetruzzi, A., Bozan, O., Satar, S., De Stradis, A., Savino, V., Yokomi, R. K., and Saponari, M. 2012. Identification and characterization of Citrus yellow vein clearing virus, a putative new member of the genus Mandarivirus. Phytopathology 102:1168-1175.

Annex:

Include as much information as necessary to support the proposal, including diagrams comparing the old and new taxonomic orders. The use of Figures and Tables is strongly recommended but direct pasting of content from publications will require permission from the copyright holder together with appropriate acknowledgement as this proposal will be placed on a public web site. For phylogenetic analysis, try to provide a tree where branch length is related to genetic distance.

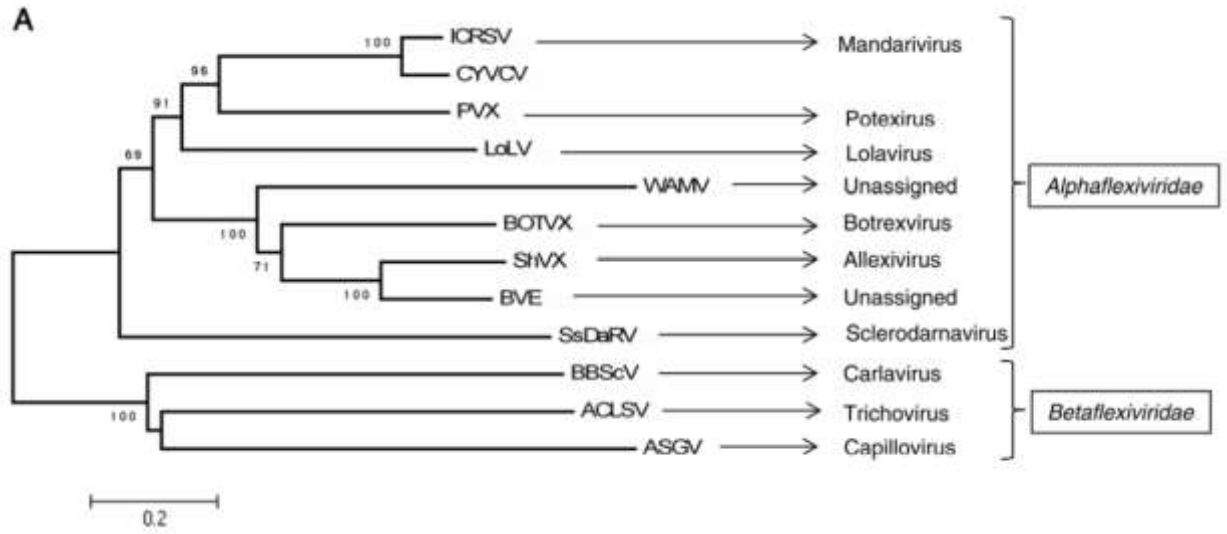

Fig 1 (from Loconsole et al., 2013). Phylogenetic trees generated by the neighbor-joining method from the alignment of the entire RNA-dependent RNA-polymerase proteins of Citrus yellow vein clearing virus (CYVCV) and members belonging to the families *Alpha-* and *Betaflexiviridae*, using MEGA (Version 5). Bootstrap values for 1,000 replicates are indicated at the main branches. Branch length is proportional to number of amino acid changes. Accession numbers of reference sequences are as follows: *Apple chlorotic leaf spot virus* (ACLSV, NC_001409); *Apple stem grooving virus* (ASGV, NC001749); *Blackberry virus E* (BVE, JN053266); *Blueberry scorch virus* (BBScV, NC 003499); *Botrytis virus X* (BOTVX, NC005132); *Indian citrus ringspot virus* (ICRSV, NC003093); *Lolium latent virus* (LoLV, NC010434); *Potato virus X* (PVX, NC011620); *Sclerotinia sclerotiorum debilitation-associated RNA virus* (SsDaRV, NC_007415); *Shallot virus X* (NC003795); *Sweet potato chlorotic fleck virus* (SPCFV, NC_006550); and *White ash mosaic virus* (WAMV, DQ412998).