



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**

<b>Code assigned:</b>	<b>2010.003aP</b>	(to be completed by ICTV officers)
<b>Short title:</b> 2 new species in the genus Closterovirus (e.g. 6 new species in the genus <i>Zetavirus</i> )		
<b>Modules attached</b> (modules 1 and 9 are required)	1 <input checked="" type="checkbox"/> 2 <input checked="" type="checkbox"/> 3 <input type="checkbox"/> 4 <input type="checkbox"/> 5 <input type="checkbox"/> 6 <input type="checkbox"/> 7 <input type="checkbox"/> 8 <input type="checkbox"/> 9 <input checked="" type="checkbox"/>	

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

G.P. Martelli on behalf of the Study Group Closteroviridae martelli@agr.uniba.it

**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)

Closteroviridae

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

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Date first submitted to ICTV:

Date of this revision (if different to above):

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## 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	<b>2010.003aP</b>	(assigned by ICTV officers)
<b>To create 2 new species within:</b>		
Genus:	<i>Closterovirus</i>	Fill in all that apply. <ul style="list-style-type: none"> <li>• If the higher taxon has yet to be created (in a later module, below) write “<b>(new)</b>” after its proposed name.</li> <li>• If no genus is specified, enter “<b>unassigned</b>” in the genus box.</li> </ul>
Subfamily:		
Family:	<i>Closteroviridae</i>	
Order:		
<b>And name the new species:</b>		<b>GenBank sequence accession number(s) of reference isolate:</b>
<i>Strawberry chlorotic fleck-associated virus</i>		DQ860839
<i>Raspberry leaf mottle virus</i>		DQ357218

### 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

Strawberry chlorotic fleck-associated virus (SCFaV) is one of four closteroviruses discovered in strawberry plants affected by chlorotic fleck, an aphid-transmitted disease (Fulton, 1987). The virus was first reported from Oregon by Tzanetakis and Martin (2007), who have sequenced its genome and discussed the phylogenetic relationships with other members of the family *Closteroviridae*. The virus isolate in question came from a strawberry clone (CFRA 9018) kept in collection for more than 40 years and was propagated vegetatively with it.

Raspberry leaf mottle virus (RLMV) is one of the four or more viruses involved in raspberry mosaic disease (Converse, 1987). This virus was first isolated by Tzanetakis *et al.* (2007) in Oregon in the raspberry accession Glen Clova #8 (GC-8) of Scottish origin, then in a red raspberry plantation in Washington. These authors have sequenced the viral genome (which they termed raspberry mottle virus) and discussed phylogenetic relationships with other members of the family *Closteroviridae*. The majority of the study group (11/14) agrees with the use of the older name raspberry leaf mottle virus for the species as suggested by McGavin and MacFarlane (2010).

The criteria demarcating species in the genus *Closterovirus* are:

- Particle size,
- Size of CP, as determined by deduced amino acid sequence data,
- Serological specificity using discriminatory monoclonal or polyclonal antibodies,
- Genome structure and organization (number and relative location of the ORFs),
- Amino acid sequence of relevant gene products (CP, CPm, HSP70h) differing

by more than 10%,

- Vector species and specificity,
- Magnitude and specificity of natural and experimental host range,
- Cytopathological features (i.e., aspect of inclusion bodies and origin of cytoplasmic vesicles),

Both SCFaV and RLMV have filamentous, closterovirus-like virions of undetermined size and their monopartite ssRNA genomes have been completely sequenced. There are no serological or cytopathological data. Other properties are:

	<b>SCFaV</b>	<b>RLMV</b>
dsRNA	<i>ca.</i> 17 kb in size (estimated from sequence data)	a major band <i>ca.</i> 17 kb in size thought to be the replicative form of the viral genome and several bands with lower mol. wt, including a prominent band 1.2 kb in size, probably corresponding to a subgenomic or defective RNA
Coat protein size (deduced from sequence)	25 kDa	22 kDa
Nucleic acid	17,039 nt (DQ860839)	17,481 nt (DQ357218)
Genome	Monopartite, completely sequenced, containing 10 ORFs, including the quintuple gene block. Genome structure largely conforming to that of species in the genus <i>Closterovirus</i> , somewhat intermediate between the simplest ( <i>Beet yellows virus</i> , BYV) and the most complex ( <i>Citrus tristeza virus</i> CTV) genome type. See annex Figure 1.	
Phylogenetic relationships	SCFaV and RLMV are most closely related to one another and to CTV, with which they cluster in phylogenetic trees. See Annex Figure 2. None of the <b>proteins encoded by relevant genes (polymerase, HSP70, CP) has an identity at the amino acid level higher than 80% with comparable proteins of other closterovirus species.</b>	
Vector transmission	Transmission attempts from CFRA 918 with <i>Aphis gossypii</i> did not succeed with SCFaV. No data for RLMV.	
Mechanical transmission	Transmission to herbaceous hosts unsuccessful	

Although some of the biological properties expected of closteroviruses have not been confirmed (aphid and mechanical transmission attempts were unsuccessful or have not been made), phylogenetic analyses place both viruses clearly within the genus *Closterovirus* while genome organization and sequence identities separate them from the existing species.

MODULE 9: **APPENDIX**: supporting material

additional material in support of this proposal

**References:**

Converse, R.H., 1987. Virus and virus-like diseases of Rubus (Blackberry and raspberry). In: Converse, R.H. (ed.). Virus Diseases of Small Fruits. USDA-ARS Agricultural Handbook 631: 168-253..

Fulton, J.P., 1987. Strawberry chlorotic fleck. In; Converse, R.H. (ed.). Virus Diseases of Small Fruits. USDA-ARS Agricultural Handbook 631: 60.

McGavin, W.J. and MacFarlane, S.A., 2010. Sequence similarities between Raspberry leaf mottle virus, Raspberry leaf spot virus and the closterovirus Raspberry mottle virus. Annals of Applied Biology 156: 439-448.

Tzanetakis, I.E. and Martin, R.R., 2007. Strawberry chlorotic fleck: identification and characterization of a novel Closterovirus associated with the disease. Virus Research 124: 88-94.

Tzanetakis, I.E., Halgren, A., Mosier, N., Martin, R.R., 2007. Identification and characterization of raspberry mottle virus, a novel member of the Closteroviridae. Virus Research 127: 26-33. .

**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.

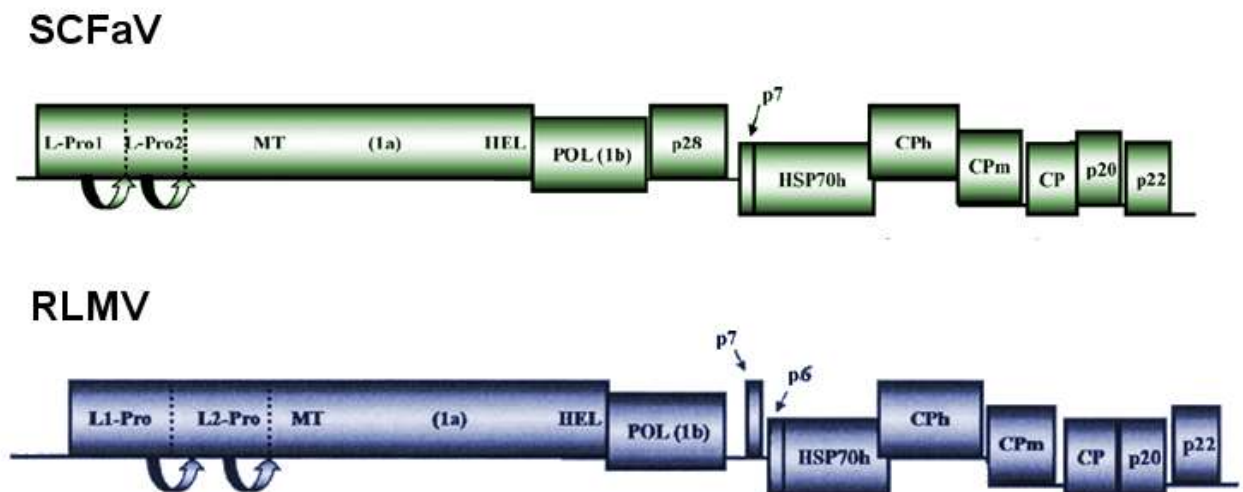


Fig. 1. Genomic organization of Strawberry chlorotic fleck associated virus (SCFaV) and Raspberry leaf mottle virus (RLMV). Abbreviations: L-Pro1 and L-Pro2, leader papain-like protease 1 and 2, respectively; MT, methyltransferase; Hel, helicase; Pol, RNA-dependent RNA polymerase; HSP70h, heat shock protein 70 homolog; CPh, coat protein homolog; CPm, minor coat protein; CP, major coat protein. Open reading frames are not to exact scale. Adapted from Tzanetakis and Martin (2007) and Tzanetakis *et al.* (2007).

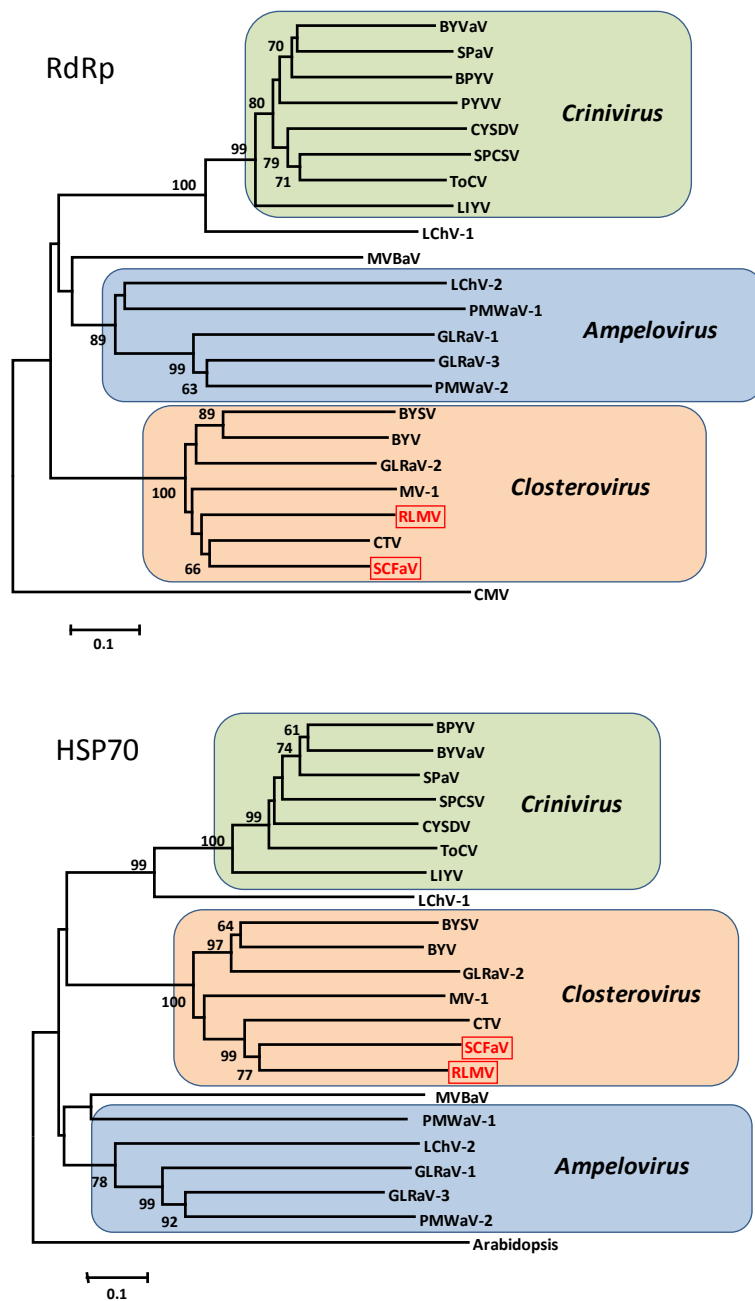


Fig. 2. Phylogenetic trees of the polymerase ORF1b (RdRp) and HSP70 homologue genes of viruses in the family *Closteroviridae* showing the positioning of the two proposed new species in the genus *Closterovirus*. The trees were generated from codon-aligned nucleotide sequences using MEGA4 (Neighbor-Joining, Maximum Composite Likelihood method with 10,000 bootstrap replicates. Bootstrap values shown when >60%). The RdRp trees uses Cucumber mosaic virus (CMV) as outgroup and the HSP70 tree is rooted with the *Arabidopsis thaliana* putative heat shock protein 70, AAN71949. The trees are based on those presented by Tzanetakis *et al.* (2007). Abbreviations are: BPYV, *Beet pseudoyellows virus*; BYSV, *Beet yellow stunt virus*; BYV, *Beet yellows virus*; BYVaV, *Blackberry yellow vein-associated virus*; CTV, *Citrus tristeza virus*; CYSDV, *Cucurbit yellow stunting disorder virus*; GLRaV-1, *Grapevine leafroll-associated virus 1*; GLRaV-2, *Grapevine leafroll-associated virus 2*; GLRaV-3, *Grapevine leafroll-associated virus 3*; LChV-1, *Little cherry virus 1*; LChV-2, *Little cherry virus 2*; LIYV, *Lettuce infectious yellows virus*; MV-1, *Mint virus 1*; MVBaV, *Mint vein banding-associated virus*; PMWaV-1, *Pineapple mealybug wilt-associated virus 1*; PMWaV-2, *Pineapple mealybug wilt-associated virus 2*; PYVV, *Potato yellow vein virus*; RLMV, *Raspberry mottle virus*; SCFaV, *Strawberry chlorotic fleck associated virus*; SPaV, *Strawberry pallidosis-associated virus*; SPCSV, *Sweet potato chlorotic stunt virus*; ToCV, *Tomato chlorosis virus*.