

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:	2009.013a,l		be comple	eted by IC	TV officers)		
Short title: create 2 (e.g. 6 new species in Modules attached (modules 1 and 9 are	new species in the the genus <i>Zetavirus</i> required)	e genus . s) I 🔀 5 🗌	Apscaviro 2 ⊠ 7 □	id, family 3 🗌 8 🗌	Pospiviro 4 9	idae 5 🗌	

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

Nuria Duran-Vila (nduran@ivia.es) and Ricardo Flores (rflores@ibmcp.upv.es) for CVd-V Takao Ito (itotaka@affrc.go.jp for CVd-VI

Has this proposal has been seen and agreed by the relevant study group(s)? Please select answer in the box on the right

Yes

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

It was the unanimous opinion of Study Group members that these two viroids meet all criteria for recognition as new viroid species.

Date first submitted to ICTV: Date of this revision (if different to above): May 2009

MODULE 2: NEW SPECIES

Part (a) to create and name one or more new species.

If more than one, they should be a group of related species belonging to the same genus (see Part b)

Code 2009.013aP (assigned by ICTV officers)

To create 2 new species with the name(s):

Citrus viroid V

Citrus viroid VI

Part (b) assigning new species to higher taxa

All new species must be assigned to a higher taxon. This is usually a genus although it is also permissible for species to be "unassigned" within a subfamily or family.

Code	2009.013bP	(assig
Couc	2009.01301	(000

To assign the species listed in section 2(a) as follows:

ned by ICTV officers)

Fill in all that apply. • If the higher taxon has yet to be Genus: Apscaviroid created (in a later module, below) write Subfamily: "(new)" after its proposed name. Family: *Pospiviroidae* If no genus is specified, enter Order:

"unassigned" in the genus box.

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 0 genus, explain how the new species meet these criteria.
 - If criteria for demarcating species need to be defined (because there will now be more 0 than one species in the genus), please state the proposed criteria.
 - Provide Genbank accession numbers (not RefSeg accessions) for genomic sequences
 - Further material in support of this proposal may be presented in the Appendix, Module 9

Sequence identity and biological properties (in particular, host range) are the primary criteria for viroid classification at the species level. Variation occurs within each species, and an arbitrary level of 90% sequence similarity is currently used to distinguish between viroid variants and new species.

Citrus viroid V (CVd-V) was recently isolated from citrus (Serra et al., 2008a) and has the typical features of members of the genus Apscaviroid:

- It contains the central conserved region (CCR) characteristic of members of this genus i) Apscaviroid, and a terminal conserved region (TCR);
- It has a GC-rich genome of 293-294 nucleotides with a predicted rod-like conformation ii) of minimum free energy with 68.7% paired nucleotides;
- iii) It has a restricted host range.

CVd-V (accession EF617306) exhibits less than 90% sequence identity with other members of the genus Apscaviroid: 73.5% (Apple scar skin viroid, ASSVd), 64.6% (Apple dimple fruit viroid, ADFVd, and Grapevine vellow speckle viroid 2, GYSVd-2), 62.6% (Citrus viroid III, CVd-III), 61.6% (Grapevine yellow speckle viroid 1, GYSVd-1), 59.9% (Citrus bent leaf viroid, CBLVd), 49.3% (Pear blister canker viroid, PBCVd) and 39.1% (Australian grapevine viroid, AGVd).

CVd-V induces <u>only</u> very small necrotic lesions and cracks in the stems of the indicator Etrog citron (*Citrus medica* L.), which sometimes are filled with gum. These symptoms differ from those induced by CBLVd ("variable syndrome" characterized by flushes of tissue showing mild leaf epinasty due to local midvein necrosis on the underside of the leaf, alternating with flushes of symptomless leaves; CBLVd also induces severe necrotic lesions and cracks releasing gum exudates, and the affected plants lose the apical dominance, stop growing, and undergo an unusual branching pattern) or by CVd-III (a "drooping leaf" pattern due to a moderate epinasty resulting from petiole and mid-vein necrosis). Co-infections with two previously identified members of the genus *Apscaviroid* (i.e., CBLVd and CVd-III) result in unusual synergistic interactions. This synergism supports that CVd-V is not a strain of CBLVd or CVd-III, because interactions between strains of the same viroid or between closely-related viroids result in interferece or cross-protection instead of synergism.

Recent work shows that CVd-V is more restricted in its distribution than other citrus viroids; nevertheless, distinct sequence variants have been identified in the USA, the Sultanate of Oman, and Nepal (Serra et al., 2008b).

Citrus viroid VI (CVd-VI) is the proposed species name for Citrus viroid Original Source (CVd-OS). The name in use for the viroid is not consistent with an ICTV rule prohibiting "newly designated serial numbers, letters or combinations thereof" as species epithets. Until a more descriptive name for CVd-OS becomes possible, the Study Group recommends that the species name should be *Citrus viroid-VI*.

Citrus viroid OS (CVd-OS) was isolated from citrus (Ito et al., 2001; 2002) and has the typical features of members of the genus *Apscaviroid*:

- i) It contains the central conserved region (CCR) characteristic of members of this genus *Apscaviroid*, and a terminal conserved region (TCR);
- ii) It has a GC-rich genome of 329-331 nucleotides with a predicted branched conformation of minimum free energy with 65.5% paired nucleotides.

CVd-OS (accession AB019508) exhibits less than 90% sequence identity lower with other members of the genus *Apscaviroid*: 66% (*Apple scar skin viroid*, ASSVd), 67% (*Apple dimple fruit viroid*, ADFVd), 68% (*Citrus viroid III*, CVd-III).

CVd-OS induces very mild leaf bending and petiole necrosis on the indicator Etrog citron (*Citrus medica* L.). It has recently been found in Japanese persimmon (Nakaune & Nakano, 2008).

MODULE 9: APPENDIX: supporting material

additional material in support of this proposal

References:

Ito, T., Ieki, H., Ozaki, K., and Ito, T. 2001. Characterization of a new citrus viroid species tentatively termed Citrus viroid OS. Arch. Virol. 146:975-982

Ito, T., Ieki, H., Ozaki, K., Iwanami, T., Nakahara, K., Hataya, T., Ito, T., Isaka, M., and Kano, T. 2002. Multiple citrus viroids in citrus from Japan and their ability to produce exocortis-like symptoms in citron. Phytopathology 92: 542-547

Nakaune, R., and Nakano, M. 2008. Identification of a new *Apscaviroid* from Japanese persimmon. Arch. Virol. 153:969-972

Serra, P., Barbosa, C.J., Daròs, J.A., Flores, R. and Duran-Vila, N. 2008a. Citrus viroid V: molecular characterization and synergistic interactions with other members of the genus Apscaviroid. Virology 370: 102-112.

Serra, P., Eiras, M., Bani-Hashemian, S.M., Murcia, N., Kitajima, E.W., Daròs, J.A., Flores, R., Duran-Vila, N. 2008b. Citrus viroid V: occurrence, host range, diagnosis, and identification of new variants. Phytopathology 98: 1199-1204.

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.

```
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           Viroids; Pospiviroidae; Apscaviroid.
REFERENCE 1 (bases 1 to 294)
  AUTHORS Serra, P., Barbosa, C.J., Daros, J.A., Flores, R. and Duran-Vila, N.
           Citrus viroid V: Molecular characterization and synergistic
  TITLE
           interactions with other members of the genus Apscaviroid
  JOURNAL
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REFERENCE
           2 (bases 1 to 294)
 AUTHORS Serra, P., Barbosa, C.J., Daros, J.A., Flores, R. and Duran-Vila, N.
           Direct Submission
  TTTLE
  JOURNAL
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           Instituto Valenciano De Investigaciones Agrarias, Apartado
Oficial,
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            Characterization of a new citrus viroid species tentatively termed
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  JOURNAL
Tree
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            859-2501, Japan (E-mail:itotaka@kcnt.affrc.go.jp,
            Tel:81-957-86-2306, Fax:81-957-86-4268)
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Figure. Unrooted phylogenetic tree of a representative of each member of the genus *Apscaviroid*. The tree was prepared in MEGA4 (Neighbor-Joining, Maximum Composite Likelihood method with 10,000 bootstrap replicates). The two proposed new species are highlighted in red. Representatives of existing species are in black and those in blue are viroids not currently recognised as species. Tree added to the proposal by the SC chair for information purposes.