

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:	2012.004aP		(to be completed by ICTV officers)					
Short title: create 8 new species in the genus 7 (e.g. 6 new species in the genus Zetavirus)  Modules attached (modules 1 and 9 are required)		Говаточ 1 ⊠ 6 □	<i>virus</i> , fami 2 ⊠ 7 □	ly <i>Virgav</i> 3 □ 8 □	iridae 4 □ 9 ⊠	5 🗌		
Author(s) with e-mail address(es) of the proposer:								
Mike Adams ( <u>mike.adams@rothamsted.ac.uk</u> ) on behalf of the <i>Virgaviridae</i> SG								
List the ICTV study group(s) that have seen this proposal:								
A list of study groups and contact <a href="http://www.ictvonline.org/subcommin">http://www.ictvonline.org/subcommin</a> in doubt, contact the appropriate schair (fungal, invertebrate, plant, portebrate viruses)	mittees.asp . If subcommittee	Virgaviridae						
ICTV-EC or Study Group comments and response of the proposer:								
Date first submitted to ICTV:	nt to above):		1Q To	ine 2012				

### **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	201	2.004aP	(assigned by ICTV officers)				
To create 8 new species within:							
Subf F	Fill in all that apply.  Genus: Tobamovirus  ofamily: Family: Virgaviridae  Order:  Fill in all that apply.  If the higher taxon has yet to be created (in a later module, below "(new)" after its proposed name  If no genus is specified, enter "unassigned" in the genus box.						
And na	me the	e new species:	GenBa	ank sequence accession er(s) of reference isolate:			
	_	ottle virus	DQ355				
		ottle virus mottle virus	EU043 JN566				
Cucumber mottle virus		AB261	1167				
Maracuja mosaic virus		DQ356	DQ356949				
Passion fruit mosaic virus		HQ389	9540				
Rattail	cactus	necrosis-associated vi	rus JF7294	471			
Tropica	al soda	apple mosaic virus	AY956	6381/2 (MP and CP only)			

## 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.
  - o 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 genus *Tobamovirus* contains viruses with a monopartite ssRNA+ genome encapsidated in rigid rod-shaped particles. They have no known natural vectors but are readily transmitted by mechanical inoculation. Other genera in the family *Virgaviridae* have divided genomes with 2 or 3 components and have different modes of transmission (seed, nematodes, fungoid protists) but there are clear phylogenetic relationships between some of the major gene products. Genome organization is conserved within each of the genera. In the genus *Tobamovirus*; the first ORF is a replication protein of 126-130 kDa with a 'leaky' stop codon that, when suppressed, extends the protein into an RdRp domain and results in a product of about 185 kDa. Two other smaller ORFs are located downstream and are translated from separate subgenomic mRNAs. The first is a cell-to-cell movement protein of about 30 kDa and the second is the single coat protein of about 18 kDa.

Species discrimination criteria within the genus as listed in the 9<sup>th</sup> report are:

- Sequence similarity: less than 10% overall nt sequence difference is considered to characterize strains of the same species, although most of the sequenced species have considerably less than 90% sequence identity
- Host range: many of these viruses have wider and more overlapping host ranges in

experimental rather than natural situations

• Antigenic relationships between the CPs

All the proposed new species have particle morphology and genome organisation typical of a member of the genus *Tobamovirus* (but see comments below on Passionfruit mosaic virus). For most, a complete genome sequence is available, and in all cases BLAST searches show that the sequences are most closely related to known tobamoviruses. Some details of each proposed species are provided below and a phylogenetic tree based on the movement and coat protein amino acid sequences of these and representative isolates of all existing tobamovirus species is provided in Appendix Figure 1. Trees based on the entire genome or entire proteome have a similar topology but lack species (or proposed species) for which there is not a complete sequence (data not shown).

### Bell pepper mottle virus (BPMV)

The complete sequence has been determined of a Netherlands eggplant isolate (DQ355023; Rhie et al., 2007). It is 6375 nt long and is most closely related to isolates of *Tomato mosaic virus* (76-77% nt identity over the entire genome; 86-87% aa identity over the entire proteome). Local necrotic lesions developed in the inoculated leaves of *Chenopodium amaranticolor*, *C. quinoa*, *Capsicum annuum*, *Datura stramonium*, *Nicotiana benthamiana*, *N. clevelandii*, *N. debneyii*, *N. glutonosa*, and *N. tabacum*, cvs Samsun NN and Xanthi-nc. Systemic mosaic symptoms occurred in *N. benthamiana*, *N. clevelandii*, and *Physalis floridana*. No symptoms were shown and no virus multiplication was detected in *Cucurbita pepo* and *Cucumis sativus*. In Western blots, antiserum raised to the virus reacted weakly with the coat protein of tomato mosaic virus and more strongly with that of pepper mild mottle virus (PMMoV) but in the reciprocal reaction antiserum raised to PMMoV showed little reaction with BPMV. Phylogenetic analysis places BPMV with other tobamoviruses infecting solanaceous hosts (Appendix Figure 1).

#### **Cactus mild mottle virus (CMMV)**

The complete sequence has been determined of a Korean isolate from *Gymnocalycium mihanovichii* (EU043335; Min et al., 2009). It is 6449 nt long and is only distantly related to existing tobamoviruses. It is most closely related to rattail cactus necrosis-associated virus (see below) but the two sequences are only 62% identical. The two viruses form a distinct branch of the phylogenetic tree, representing viruses that infect members of the family *Cactaceae*. Local necrotic lesions developed in the inoculated leaves of *Chenopodium amaranticolor* and *Vigna sesquipedalis*, and local chlorotic local lesions in the inoculated leaves of *C. quinoa*, *C. murale* and *N. tabacum* cv. White Burley. Systemic necrotic lesions were shown by the inoculated and systemic leaves of *Gomphrena globosa*.

### Clitoria yellow mottle virus (ClYMV)

A suspected tobamovirus was detected in symptomatic leaves of the African forage legume *Clitoria ternata* growing in Australia. Local chlorotic and pale necrotic lesions developed in the inoculated leaves of *Chenopodium quinoa, Nicotiana clevelandii, N. glutinosa* and *Pisum sativum*, and chlorotic lesions with dark spreading haloes in the inoculated primary leaves of *Phaseolus vulgaris* (cv Purple King) and with systemic chlorotic flecking. When sap extracts from these plants were tested by inoculating the primary leaves of Purple King bean seedlings, all produced the same chlorotic lesions with spreading dark haloes. No symptoms were shown by inoculated seedlings of *Brassica chinensis, Cucumis sativa, Lactuca sativa, Lycopersicon esculentum, Ocimum basilicum* or *Zinnia elegans*, and when sap extracts from them were inoculated to the leaves of Purple King beans no symptoms appeared. The complete sequence

has been determined of an Australian isolate (JN566124; Wei et al., 2012). It is 6514 nt long and is most closely related to sunn-hemp mosaic virus although sequences are only about 64% identical. The two viruses both infect leguminous plants (family *Fabaceae*).

### **Cucumber mottle virus (CMoV)**

The complete sequence has been determined of a Japanese isolate (AB261167; Orita et al., 2006). It is 6485 nt long and is most closely related to isolates of *Cucumber green mottle mosaic virus*, with which it shares only about 62% identity. Phylogenetic analysis places it on the same branch as other tobamoviruses infecting cucurbit hosts. Local necrotic lesions developed in the inoculated leaves of *Chenopodium amaranticolor*. Systemic mild mottle mosaic symptoms were shown by the inoculated and systemic leaves of *Cucumis sativus*.

### Maracuja mosaic virus (MarMV)

The complete sequence has been determined of a Peruvian isolate from *Passiflora edulis* (DQ356949; Song et al., 2006). It is 6794 nt long and is only distantly related to existing tobamoviruses. It is most closely related to passionfruit mosaic virus (see below) with which it has about 73% nt identity. The two viruses form a distinct branch of the phylogenetic tree, representing viruses that infect *Passiflora*. Local necrotic lesions developed in the inoculated leaves of *Chenopodium amaranticolor*, *C. quinoa*, *Nicotiana clevelandii*, *N. glutinosa*, and *N. tabacum* cvs. Samsun NN and Xanthi-nc, and local chlorotic local lesions in the inoculated leaves of *Datura stramonium*. Systemic mosaic symptoms were shown by the inoculated and systemic leaves of *N. benthamiana* and *Passiflora edulis*.

### Passion fruit mosaic virus (PFMV)

The complete sequence has been determined of a US isolate (HQ389540; 6791nt; Song & Ryu, 2011). A sequence of a second (TGP) isolate that lacks only the extreme 5'-terminus has also been reported (JF807914; Stobbe et al. 2012; 6674nt). An additional ORF that appears to encode a cysteine-rich protein has been identified downstream of the RdRp and partially overlapping with the movement protein. The sequence is only distantly related to existing tobamoviruses. It is most closely related to MarMV (see above) with which it has about 73% nt identity. The two viruses form a distinct branch of the phylogenetic tree, representing viruses that infect *Passiflora*. Local necrotic lesions developed in the inoculated leaves of *Nicotiana tabacum* cvs Samsun NN and cv. Xanthi-nc. Unlike MarMV, symptomless systemic infection occurred after inoculation of *N. benthamiana*. Antiserum raised to PFMV recognized PFMV coat protein and (more weakly) that of MarMV in Western blots but MarMV antiserum did not recognize PFMV.

#### Rattail cactus necrosis-associated virus (RCNaV)

The complete sequence has been determined of a Korean isolate from symptomatic plants of the cactus *Aporocactus flagelliformis* (JF729471; Kim et al., 2012). It is 6506 nt long and is only distantly related to existing tobamoviruses. It is most closely related to cactus mild mottle virus (see above) but the two sequences are only 62% identical. The two viruses form a distinct branch of the phylogenetic tree, representing viruses that infect members of the family *Cactaceae*. Following mechanical inoculation of purified virions, systemic mosaic and leaf crinkling developed on *Nicotiana benthamiana* while necrotic local lesions were produced on *N. tabacum* cvs Samsun NN and *Xanthi*-nc. By contrast, *N. tabacum* cv. Samsun, *N. glutinosa*, *N. rustica*, *Cucumis sativus* cv. Baekdadaki, *Vigna unguiculata* and *Chenopodium amaranticolor* did not show symptoms.

### Tropical soda apple mosaic virus (TSAMV)

The MP and CP sequences have been determined of a Florida isolate from the invasive weed

Solanum viarum (AY956381/2; Adkins et al., 2007a, 2007b). It appears to be widespread in *S. viarum*, where it may interfere with a biological control program using the beetle *Gratiana boliviana* (Overholt et al., 2009). TSAMV often occurs together with tomato mosaic virus but the two viruses are serologically distinct. The virus can readily be transmitted to a range of solanaceous plants. In sequence and serological comparisons, and phylogenetic analysis, the virus is most closely related to isolates of *Pepper mild mottle virus*, with which it shares about 78% nt identity, placing it with other tobamoviruses infecting solanaceous hosts.

### MODULE 9: APPENDIX: supporting material

additional material in support of this proposal

#### **References:**

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- Song Y.S., Ryu K.H. (2011). The complete genome sequence and genome structure of passion fruit mosaic virus. Arch. Virol. 156(6):1093-1095.
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Figure 1. Phylogenetic (Distance) tree of the concatenated movement and coat protein sequences of tobamoviruses. A single sequence has been chosen to represent each of the existing species as the sequences of isolates within a species are all very closely related to one another. Tree prepared in MEGA4 with JTT amino acid differences. Bootstrap percentages (from 1000 replicates) are shown where >60%. Viruses proposed as new species are shown in red. Families of natural hosts are shown in green. BPMV, Bell pepper mottle virus; BrMMV, Brugmansia mild mottle virus; ClYMV, Clitoria yellow mottle virus; CMMV, Cactus mild mottle virus; CFMMV, Cucumber fruit mottle mosaic virus; CGMMV, Cucumber green mottle mosaic virus; CMOV, Cucumber mottle virus; FrMV, Frangipani mosaic virus; HLFPV, Hibiscus latent Fort Pierce virus; HLSV, Hibiscus latent Singapore virus; KGMMV, Kyuri green mottle mosaic virus; MarMV, Maracuja mosaic virus; ObPV, Obuda pepper virus; ORSV, Odontoglossum ringspot virus; PaMMV, Paprika mild mottle virus; PFMV, Passion fruit mosaic virus; PMMoV, Pepper mild mottle virus; RCNaV, Rattail cactus necrosis-associated virus; RheMV, Rehmannia mosaic virus; RMV, Ribgrass mosaic virus; SFBV, Streptocarpus flower break virus; SHMV, Sunn-hemp mosaic virus; TMGMV, Tobacco mild green mosaic virus; TMV, Tobacco mosaic virus; ToMV, Tomato mosaic virus; YMoV, Youcai mosaic virus; ZGMMV, Zucchini green mottle mosaic virus.

