



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:	2016.017aP	(to be completed by ICTV officers)			
Short title: Create <i>Pigeonpea sterility mosaic emaravirus 2</i> as a new species in the genus <i>Emaravirus</i> (e.g. 6 new species in the genus <i>Zetavirus</i>)					
Modules attached (modules 1 and 10 are required)	1 <input checked="" type="checkbox"/>	2 <input 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 type="checkbox"/>	10 <input checked="" type="checkbox"/>

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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)

Emaravirus study group

ICTV Study Group comments (if any) and response of the proposer:

Date first submitted to ICTV:

June 2016

Date of this revision (if different to above):

ICTV-EC comments and response of the proposer:

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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	2016.017aP	(assigned by ICTV officers)
To create 1 new species within:		
Genus:	<i>Emaravirus</i>	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.
Subfamily:		
Family:		
Order:		
Name of new species:	Representative isolate: (only 1 per species please)	GenBank sequence accession number(s)
<i>Pigeonpea sterility mosaic emaravirus 2</i>	Pigeonpea sterility mosaic virus isolate ICP8863	HF912243 to HF912246 HG939489, HG939490

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

Species demarcation criteria for the genus *Emaravirus* are:

1. Differences in relevant gene product sequences of more than 25%
2. Differences in host ranges
3. Differences in vector specificities

In addition to the pigeonpea sterility mosaic virus (PPSMV), pigeonpea sterility mosaic virus 2 (PPSMV-2) is the second emaravirus found to infect pigeonpea plant (*Cajanus cajan* L.) in nature (Elbeaino *et al.*, 2015). Similarly to its precedent, PPSMV-2 possesses all molecular, morphological and biological features to be considered as a new member of the genus *Emaravirus*, which currently comprises the following species: *Fig mosaic virus* (Elbeaino *et al.*, 2009a, 2009b, 2012); *Rose rosette virus* (Laney *et al.*, 2011); *Raspberry leaf blotch virus* (McGavin *et al.*, 2012); *Pigeonpea sterility mosaic virus* (Elbeaino *et al.*, 2014; Kumar *et al.*, 2003), *High Plains wheat mosaic virus* (Tatineni *et al.*, 2014; Skare *et al.*, 2006) and *European mountain ash ringspot-associated virus* as the type species of the genus (Mielke-Ehret & Mühlbach, 2007). However, redbud yellow ringspot-associated virus (RYRSaV) (Laney *et al.*, 2010; Di Bello *et al.*, 2016), actinidia chlorotic ringspot-associated virus (AcCRaV) (Zheng *et al.*, 2016) and woolly burdock yellow vein virus (WBV) (Bi *et al.*, 2012) are yet putative emaraviruses awaiting their final classification. Proposals for recognition of RYRSaV and AcCRaV as representatives of new species in the genus are submitted separately.

PPSMV-2 is transmitted by *Aceria cajani* mites to *Phaseolus vulgaris* cv. Topcrop plants and develops chlorotic mottling and mild mosaic symptoms 15-20 days post-inoculation. The RNA-dependent RNA polymerase (RdRp), glycoprotein precursor (GP),

nucleocapsid (NC) and P4 proteins exhibited different levels of sequence identity with ortholog proteins of other emaraviruses (Table 1).

Virus properties

- (i) Virus particles: no data. Assumed to be similar to characterized emaraviruses.
- (ii) Genome: The genome structure resembles that of members of the genus *Emaravirus*. It is composed of six segments of negative sense ssRNA. RNA-1: 7009 nt, RNA-2: 2229 nt, RNA-3: 1335 nt, RNA-4: 1491 nt, RNA-5: 1833 nt, RNA-6: 1194 nt (Fig.1) (in order from RNA-1 to RNA-6, GenBank accession numbers are: HF912243 to HF912246, HG939489 and HG939490) (Elbeaino *et al.*, 2015). Each segment is monocistronic, encoding a single protein translated from the complementary strand (Fig. 1). Untranslated regions (UTRs) at the 5' and 3' termini of all RNA segments extending from 46 to 106 nt and from 79 to 409 nt, respectively, and exhibiting a conserved stretch of 13 nt at their termini.
- (iii) Virus-encoded proteins: RNA-dependent RNA-polymerase (p1): 266.0 kDa; putative Glycoprotein precursor (p2): 74.3 kDa (Gc 22.1 kDa and Gc 52.2 kDa); putative Nucleocapsid protein (p3): 34.9 kDa; putative movement protein (p4): 40.7 kDa; p5 (function unknown): 55.0 kDa; p6 (function unknown): 27.0 kDa (Fig. 1).
- (iv) Phylogenetic relationships: Phylogenetic trees with RdRp, GP and NC proteins resulted in similar topologies, with all emaraviruses clustered into three distinct clades (Figure 2). PPSMV-2 groups together with FMV. However, these two viruses are distinct from each other, showing 70.5%, 62.2%, and 56.7% nucleotide identity between the RdRp, GP, and NC proteins, respectively (Table 1).
- (v) Experimental transmission: The virus is transmissible by *Aceria cajani* mites onto *Phaseolus vulgaris* cv. Topcrop plants.
- (vi) Natural host range: Pigeonpea plants.

MODULE 10: **APPENDIX**: supporting material

additional material in support of this proposal

References:

- Bi Y., Tugume A.K., Valkonen, J.P., 2012. Small-RNA deep sequencing reveals *Arctium tomentosum* as a natural host of Alstroemeria virus X and a new putative Emaravirus. *Plos One*, 7: e42758.
- Di Bello P.L., Laney A.G., Druciarek T., Ho T., Gergerich R.C., Keller K.E., Martin R.R., Tzanetakis I.E., 2016. A novel emaravirus is associated with redbud yellow ringspot disease. *Virus Research* 222: 41-47.
- Elbeaino T., Digiario M., Alabdullah A.K., De Stradis A., Minafra A., Mielke N., Castellano M.A., Martelli G.P., 2009a. A multipartite negative-sense single-stranded RNA virus is the putative agent of fig mosaic disease. *Journal of General Virology*, 90 (5): 1281-1288.
- Elbeaino T., Digiario M., Martelli G.P., 2009b. Complete nucleotides sequence of four viral RNAs segments of fig mosaic virus. *Archives of Virology*, 154 (11). 1719-1727.
- Elbeaino T., Digiario M., Martelli G.P., 2012. RNA-5 and -6, two additional negative-sense RNA segments associated with Fig mosaic virus. *Journal of Plant Pathology*, 94 (2): 421-425.
- Elbeaino T., Digiario M., Uppala M., Sudini H., 2014. Deep sequencing of Pigeonpea sterility mosaic virus discloses five RNA segments related to emaraviruses. *Virus Research*, 188: 27–31.
- Elbeaino T., Digiario M., Uppala M., Sudini H., 2015. Deep-sequencing of dsRNAs recovered from mosaic-diseased pigeonpea (*Cajanus cajan* L.) revealed the presence of a novel emaravirus: Pigeonpea sterility mosaic virus 2 (PPSMV2). *Archives of Virology*, 160:2019–2029.
- Kumar P.L., Jones A.T., Reddy D., 2003. A novel mite-transmitted virus with a divided RNA genome closely associated with pigeonpea sterility mosaic disease. *Phytopathology*, 93: 71-81.

additional material in support of this proposal

References:

- Laney A.G., Keller K.E., Martin R.R., Tzanetakis I.E., 2011. A discovery 70 years in the making: characterization of the Rose rosette virus. *Journal of General Virology*, 92: 1727–1732.
- McGavin W.J., Mitchell C., Cock P.J.A., Wright K.M., MacFarlane S.A., 2012. Raspberry leaf blotch virus, a putative new member of the genus *Emaravirus*, encodes a novel genomic RNA. *Journal of General Virology*, 93: 430–437.
- Mielke N., Muehlbach, H.P., 2007. A novel, multipartite, negative-strand RNA virus is associated with the ringspot disease of European mountain ash (*Sorbus aucuparia* L.). *Journal of General Virology*, 88: 1337–1346.
- Mühlbach H.P., Mielke-Ehret N., 2012. Emaravirus, p. 767–769. In King AMQ, Adams MJ, Carstens EB, Lefkowitz EJ (eds.). *Virus taxonomy: ninth report of the International Committee on Taxonomy of Viruses*, Elsevier-Academic Press, London UK.
- Olmedo-Velarde A., Ochoa-Corona F.M., Elbeaino T., 2016. Toward a broad detection of Emaraviruses: Endpoint RT-PCR. *Abstracts of the Annual Meeting of the American Phytopathological Society (APS)*. Tampa, Florida. U.S.A. July 30-August 3. 2016.
- Skare J.M, Wijkamp I., Denham I., Rezende J.A.M., Kitajima E.W., Park J.W., Desvoyes B., Rush C.M., Michels G., Scholthof K.B.G., Scholthof H.B., 2006. A new eriophyid mite-borne membrane-enveloped virus-like complex isolated from plants. *Virology*, 347: 343–353.
- Tatini S, McMechan AJ, Wosula EN, Wegulo SN, Graybosch RA, French R, Hein GL. (2014). An eriophyid mite-transmitted plant virus contains eight genomic RNA segments with unusual heterogeneity in the nucleocapsid protein. *Journal of Virology*, 88: 11834-11845.
- Zheng Y., Navarro B., Wang G., Wang Y., Yang Z., Xu W., Zhu C., Wang L., Di Serio F., Hong N., 2016. Actinidia chlorotic ringspot-associated virus: a novel emaravirus infecting kiwifruit plants. *Molecular Plant Pathology*: DOI: 10.1111/mpp.12421.

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.

Figure 1 Genomic organization of PPSMV-2. The terminal 13 nucleotides conserved at the 5' and 3' termini are indicated as black boxes on each segment. Letters (A–F) represent the conserved motifs in the RdRp (RNA1) gene. Expression products of each RNA (p1 to p6) are represented as dark gray boxes. The protein function and estimated molecular weight of each segment are indicated.

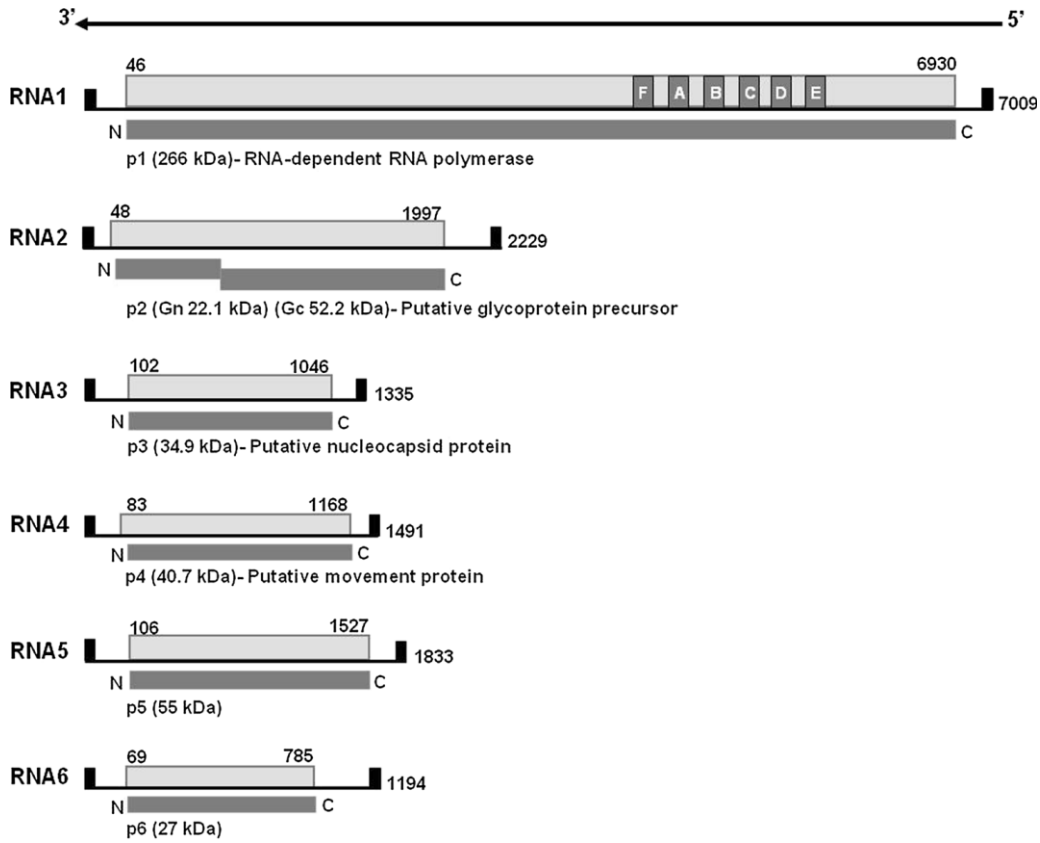


Figure 2 Phylogenetic trees constructed based on aligned amino acid sequences of P1, P2 and P3 of PPSMV-2 and other emaraviruses, using MEGA 6.0'. Trees were generated by the maximum-likelihood method, applying the JTT matrix and pairwise gap deletion. Branches with less than 70 % bootstrap support were collapsed, Sequences from the following viruses were used: European mountain ash ringspot-associated virus (EMARaV), fig mosaic virus (FMV), rose rosette virus (RRV), redbud yellow ringspot-associated virus (RYRSaV), raspberry leaf blotch virus (RLBV), wheat mosaic virus (WMoV), pigeonpea sterility mosaic virus 1 (PPSMV-1). The tospoviruses impatiens necrotic spot virus (INSV) and tomato spotted wilt virus (TSWV) were used as outgroups. Note that PPSMV-2 formed a separate clade with FMV, RRV and PPSMV-1 from other members of the genus *Emaravirus*.

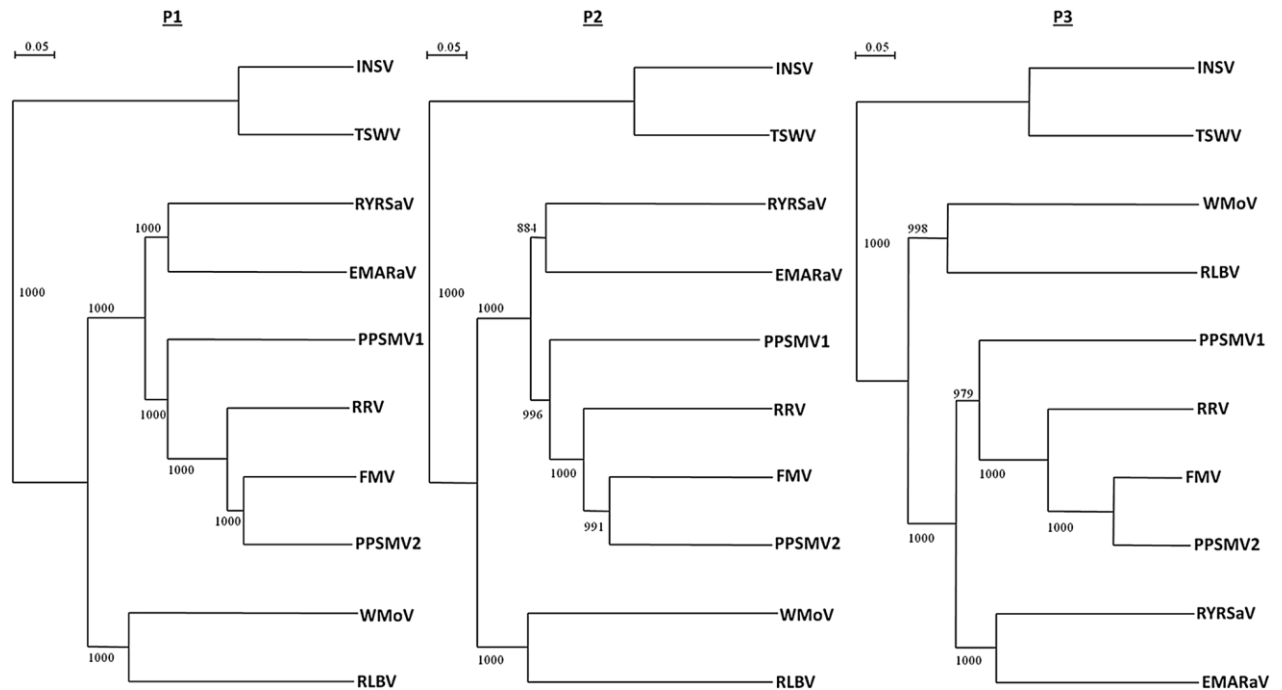


Table 1 Sequence identity matrix determined from nucleotide and deduced amino acid (in bold) sequences of multiple RNA segments of emaraviruses

Emaraviruses	RNA	Accession number	EMARaV	RYRSaV	PPSMV2	FMV	PPSMV1	RRV	RLBV	WMOv
EMARaV	1	AY563040		59.6	55.5	54.7	54.9	55.4	48.5	45.5
	2	AY563041	ID	48.9	48.1	48.5	46.8	47.9	38.8	35.3
	3	DQ831831		49.0	37.9	44.2	41.2	42.2	33.6	34.1
	4	DQ831828		35.7	33.4	33.8	34.1	32.8	28.6	29.2
1	JF795479	53.8			54.3	55.2	54.9	54.6	48.4	45.3
RYRSaV	2	JF795480	41.4	ID	50.5	50.6	50.0	51.3	38.9	35.6
	3	JF795481	44.6		42.2	42.0	43.9	41.6	36.0	35.6
	4	JF795482	5.2		45.0	44.7	42.6	45.6	36.7	33.4
	1	HF912243	48.9		46.4		70.5	62.5	67.6	48.7
PPSMV-2	2	HF912244	34.0	38.0		62.2	51.1	59.8	39.5	35.0
	3	HF912245	35.6	33.2	ID	56.7	48.5	50.7	34.0	34.0
	4	HF912246	6.0	30.8		68.7	49.4	60.8	37.3	34.4
	5	HG939489				43.1	61.1		37.6	35.1
	6	HG939490				50.6				29.1
	1	AM941711	48.2	46.7		72.1		59.0	67.7	48.4
FMV	2	FM864225	35.8	38.1		57.1		53.5	59.7	39.6
	3	FM991954	36.7	35.8	80.0	ID	46.7	57.5	34.8	34.7
	4	FM992851	6.0	28.7	75.0		46.3	60.7	35.2	32.9
	5	HE803826			30.6		43.2		39.8	38.6
	6	HE803827			23.9					29.9
	1	HF568801	47.3	46.8	52.9		51.5		60.4	48.0
PPSMV-1	2	HF568802	37.0	39.4	42.7		43.3		51.4	41.5
	3	HF568803	32.4	33.2	39.8	39.6	ID	47.3	38.1	36.6
	4	HF568804	5.2	25.9	39.3	38.2		48.8	37.9	35.6
	5	HF945448			70.4	29.2			37.2	35.7
	1	NC_015298	48.3	46.4	67.5	67.9		52.9		48.4
RRV	2	NC_015299	35.7	38.5	51.7	49.0		42.3	ID	38.8
	3	NC_015300	31.1	33.1	60.6	59.5	39.9	35.1		36.7
	4	NC_015301	6.6	30.3	61.2	59.0	38.2	38.4		35.0
	1	FR823299	32.9	33.2	32.8	31.8	32.0	31.9		
RLBV	2	FR823300	22.0	21.8	21.0	22.9	25.2	22.3		51.6
	3	FR823301	20.2	18.2	18.8	20.3	23.0	19.3	ID	48.4
	4	FR823302	5.8	19.6	18.0	16.7	19.6	19.8		54.8
	5	FR823303			12.2	11.0	11.8			36.8
	1	KJ939623	30.8	31.4	30.9	30.6	30.1	31.0		41.8
WMOv	2	FR823300	20.1	22.0	20.3	20.8	21.7	22.1		33.9
	3	KJ939625	15.1	17.8	16.0	17.9	18.4	16.1	26.6	
	4	KJ939627	5.4	14.8	17.9	17.7	18.5	19.5	44.0	ID
	5	KJ939628			12.1	10.0	11.7		18.1	
	6	KJ939629			7.5	5.4				