

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:	2015.019		(to be completed by ICTV officers)				
Short title: To create a new species in the genus <i>Endornavirus</i>							
Modules attached (modules 1 and 10 are required)		1 ⊠ 6 □	2	3	4	5 ☐ 10 ⊠	
Author(s):							
Valverde, R. A. Okada, R. Kiyota, E. Moriyama, H. Fukuhara, T.							
Corresponding author with e-mail address:							
Valverde, R. A. (rvalverde@agcenter.lsu.edu)							
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> doubt, contact the appropriate schair (fungal, invertebrate, plant, pvertebrate viruses)	mittees.asp . If subcommittee	Two me	mbers of	the Endor	rnavirus st	udy group	
ICTV Study Group comments (if any) and response of the proposer:							
None							
Date first submitted to ICTV: Date of this revision (if different	nt to above):		June	15, 2015			
ICTV-EC comments and response of the proposer:							

## 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 $2015.019aP$ (assigned by IC			CTV officers)			
To create	e a ne	w species within:				
					all that apply.	
Ger	nus:	us: <i>Endornavirus</i>		If the higher taxon has yet to be		
Subfam	nily:				ated (in a later module, below) write ew)" after its proposed name.	
Family: Endornaviridae		If no genus is specified, enter				
Ore	der:				assigned" in the genus box.	
Name of new species:		Representative isolate: (only 1 per species please)		GenBank sequence accession number(s)		
Basella alba endornavirus 1		Rubra		AB844265		

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

Endornaviruses have been reported to infect economically important crops, such as, avocado, barley, broad bean, common bean, pepper, some plant pathogenic fungi, and the oomycete *Phytophthora* sp. (Fukuhara & Gibbs, 2012; Fukuhara et al., 2006; Okada et al., 2011, 2013; Villanueva et al., 2012). With the exception of *Phaseolus vulgaris endornavirus 1* and *Phaseolus endornavirus 2*, all of the other endornavirus species and tentative species have been isolated from a different host species (Okada et al., 2013). Currently endornavirus species are distinguished on the basis of their size, host-range, genome organization, and nucleotide sequence variations. The nucleotide sequences of different endornavirus species ranges from 30% to 75% identity (Fukuhara & Gibbs, 2012).

Recently, we found a 14 kbp dsRNA in plants of two cultivars (Eclipse and Rubra) of Malabar spinach (Basella alba). Sequencing the dsRNA from the two cultivars yielded a virus with properties similar to those of members of the *Endornaviridae* (Okada et al., 2014). Two isolates of the putative endornavirus (one from each cultivar) from, tentatively named Basella alba endornavirus (BaEV), were sequenced and their sequence analyzed and compared with sequences of accepted endornavirus species and tentative species. The results indicated that BaEV-Eclipse and BaEV-Rubra are strains of the same virus. Both BaEV-Elipse and BaEV-Rubra were transmitted to the progeny plants at rates close to 100% and viral dsRNA was detected in leaves, shoots, fruits, and seed tissues. These are biological properties typical of plant endornaviruses. The size of both BaEV strains was determined to be 14,027 nt. A BLAST search using amino acid sequences of BaEV revealed conserved domains of a putative RNA helicase-1, capsular polysaccharide synthase, UDP-glucose-glycosyltransferase, and an RNA-dependent RNA polymerase (Fig. 1). A unique molecular feature of many members of the family Endornaviridae is the presence of a site-specific nick in the 5' region of the coding strand RNA molecule. Sequence analyses of multiple RACE-generated clones of the BaEV strains indicated the presence of a nick in the coding strand at nt 1,063 for BaEV-Eclipse and nt 1,097 for BaEV-Rubra. Phylogenetic analyses of the putative Hel-1, UGT and RdRp showed that the two BaEV strains clustered with members of the family *Endornaviridae* (a phylogenetic tree using the viral RdRp is shown in (Fig. 2). When the complete nucleotide sequence of BaEV was compared with complete sequences of endornaviruses, proposed new species, and unclassified viruses, the closest endornaviruses to BaEV were Oryza sativa endornavirus, Oryza rufipogon endornavirus, Persea americana endornavirus 1 (this proposed new species is presented in a separate proposal), and Phaseolus vulgaris endornavirus 1 with an average of 48 % nucleotide sequence identity (**Table 1**). The biological and molecular properties of the virus isolated from *Basella alba* support creating a new species of the genus endornavirus and the name Basella alba endornavirus 1 (BaEV1) is proposed.

### MODULE 10: APPENDIX: supporting material

additional material in support of this proposal

#### **References:**

Fukuhara, T. & Gibbs, M. J. 2012. Family Endornaviridae. In Virus Taxonomy: Classification and Nomenclature of Viruses Ninth Report of the International Committee on Taxonomy of Viruses. Edited by Andrew M.Q. King, Michael J. Adams, Eric B. Carstens, and Elliot J. Lefkowitz. pp 519-521. Elsevier/Academic Press.

Fukuhara, T., Koga, R., Aoki, N., Yuki, C., Yamamoto, N., Oyama, N., Udagawa, T., Horiuchi, H., Miyazaki, S. & other authors (2006). The wide distribution of endornaviruses, large double-stranded RNA replicons with plasmid-like properties. Archives of Virology 151: 995-1002.

Okada, R., Kiyota, K., Moriyama, H. Fukuhara, T., and Valverde, R. A. 2014. A new endornavirus species infecting Malabar spinach (Basella alba L.). Archives of Virology 159:807-809.

Okada, R., Kiyota, E., Sabanadzovic, S., Moriyama, H., Fukuhara, T., Saha, P., Roossinck. M. J., Severin, A., and Valverde, R. A. 2011. Bell pepper endornavirus: molecular and biological properties and occurrence in the genus Capsicum. Journal of General Virology 92:2664-2673.

Okada, R., Young, C. K., Valverde, R. A., Sabanadzovic, S., Aoki, N., Hotate, S., Kiyota, E., Moriyama, H., and Fukuhara, T. 2013. Molecular characterization of two evolutionally distinct endornaviruses co-infecting common bean (*Phaseolus vulgaris*). Journal of General Virology 93:220-229.

Villanueva, F., Sabanadzovic, S., Valverde, R. A., and Navas-Castillo, J. 2012. Complete genome sequence of a double-stranded RNA virus from avocado. Journal of Virology 86:1282-1283.

#### Annex

**Table 1.** Percent as sequence identity of two Basella alba endornavirus 1strains domains compared with those of other endornavirus species (**black font**), proposed new species (**red font**, this or accompanying proposals), and unclassified viruses (**blue font**). The percentage of the complete nucleotide sequence identity is also presented

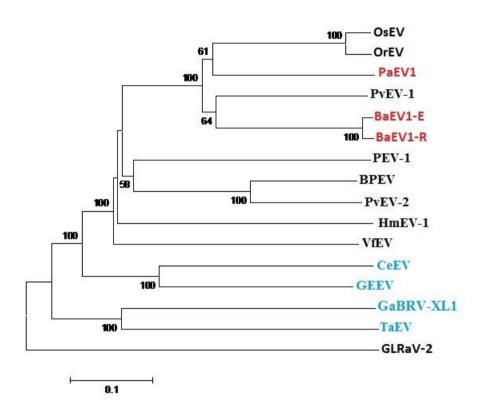
Virus	Hel-1	CPS	UGT	RdRP	Complete nucleotide Sequence	ACCESSION No.
Basella alba	100	100	100	100	100	AB844264
endornavirus 1-Rubra						
Basella alba	98.4	97.1	97.2	98.1	92.0	AB844265
endornavirus 1-Eclipse						
Oryza sativa	48.8	30.5	28.4	56.1	48.0	YP_438200.1
endornavirus						
Oryza rufipogon	48.6	32.3	29.1	55.4	48.1	YP_438202.1
endornavirus						
Persea americana	53.0	38.0	36.0	60.5	47.9	YP_005086952.1
endornavirus 1						
Phaseolus vulgaris	51.0	31.3	35.5	64.6	48.4	BAM68539.1
endornavirus 1						
Bell pepper	32.3	N.D.	29.7	42.2	46.6	BAK52155.1
endornavirus						
Phaseolus vulgaris	31.4	N.D.	32.4	42.7	45.4	BAM68540.1
endornavirus 2						
Vicia fava endornavirus	28.5	N.D.	N.D.	42.8	44.6	YP_438201.1
Phytophthora	30.6	N.D.	29.5	40.6	45.6	YP_241110.1
endornavirus 1						
Helicobasidium mompa	32.2	N.D.	29.2	41.0	44.6	YP_003280846.1
endornavirus 1						
Gremmeniella abietina	20.1	N.D.	N.D.	31.3	44.8	YP_529670.1
type B RNA virus-XL1						
Tuber aestivum	N.D.	N.D.	N.D.	35.1	45.5	YP_004123950.1
endornavirus						
Chalara elegans	28.5	N.D.	28.6	34.9	N.D.	ADN43901.1
endornavirus						
Grapevine endophyte	24.8	N.D.	N.D.	33.4	44.9	YP_007003829.1
endornavirus						

MTR: Viral methyltransferase; Hel-1: viral helicase 1; CPS: capsular polysaccharide synthase; UGT: UDP-glycosyltransferase; RdRp: viral RNA dependent RNA polymerase; N.D.: not determined.



**Fig. 1.** Schematic representation of the genome organization of Basella alba endornavirus 1. The size of the viral dsRNA consists of 14,027 bp. The box represents the large ORF coding for a 4,508aa. HEL: viral helicase 1; CPS: Capsular polysaccharide synthesis protein; UGT: UDP-glycosyltransferase; RDRP: viral RNA-dependent RNA polymerase.

V= Nick at nucleotide 1,063nt (Eclipse isolate).



**Fig. 2.** Neighbour-joining phylogenic tree using the deduced amino acid sequence of the RdRp region of endornaviruses (accepted species in **black**,-proposed species in **red**, and unclassified viruses in **blue**). Values at the nodes indicate bootstrap support values obtained for 1,000 replicates (only values above 50% are shown). OsEV: *Oryza sativa endornavirus*; OrEV: *Oryza rufipogon endornavirus*; PaEV1: Persea americana endornavirus 1; PvEV-1: *Phaseolus vulgaris endornavirus 1*; PvEV-2: *Phaseolus vulgaris endornavirus 2*; BaEV1-E: Basella alba Endornavirus 1-Eclipse; BaEV1-R: Basella alba endoravirus 1-Rubra; PEV-1: *Phytophthora endornavirus 1*; BPEV: *Bell pepper endornavirus*; HmEV-1: *Helicobasidium mompa endornavirus 1*; VfEV: *Vicia fava endornavirus*; CeEV: Chalara elegans endornavirus; GEEV: Grapevine endophyte endornavirus; GaBRV-XL1: Gremmeniella abietina type B RNA virus -XL1; TaEV: Tuber aestivum endornavirus. GLRaV-2: *Grapevine leafroll-associated virus 2* was used as outgroup. The scale bar indicates the number of substitutions per amino acid position.