



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:	2016.015a-dM	(to be completed by ICTV officers)
Short title: One (1) new species in a new genus <i>Peropuvirus</i> , family <i>Nyamiviridae</i> (e.g. 6 new species in the genus <i>Zetavirus</i>)		
Modules attached (modules 1 and 11 are required)	6 <input type="checkbox"/> 7 <input type="checkbox"/> 8 <input type="checkbox"/> 9 <input type="checkbox"/> 10 <input 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)

ICTV *Nyamiviridae* Study Group

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

Date first submitted to ICTV:

July 18, 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	2016.015aM	(assigned by ICTV officers)
To create 1 new species within:		
Genus:	<i>Peropuvirus</i> (NEW)	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:	N/A	
Family:	<i>Nyamiviridae</i>	
Order:	<i>Mononegavirales</i>	
Name of new species:	Representative isolate: (only 1 per species please)	GenBank sequence accession number(s)
<i>Pteromalus puparum peropuvirus</i>	<i>Pteromalus puparum</i> negative-strand RNA virus 1 (PpNSRV-1) HZ	KX431032

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 11

Pteromalus puparum negative-strand RNA virus 1(PpNSRV-1) was originally isolated from the laboratory parasitoid strain of the pteromalid wasp *Pteromalus puparum*. PpNSRV-1 is present in various tissues and life stages of the parasitoid, and is transmitted vertically through females and males. The complete viral genome is 12, 230 nucleotide in length with a G+C content of 46.09%, lacking a poly(A) tail at the 3' terminus. The PpNSRV-1 genome contains five large ORFs located at sequence positions 153 to 2,015, 2,109 to 2,564, 2,608 to 3,816, 3,842 to 5,566, and 5,612 to 12,112 (Fig. 1A). The leader and trailer regions in the PpNSRV-1 genome are 152 and 118 bp in length, respectively (Fig. 1A) and their terminal nucleotides do not exhibit obvious complementarity. By comparing the 5' and 3' untranslated regions and intergenic regions of PpNSRV-1, we found the putative transcription initiation and termination signals for each of five ORFs. A conserved transcription initiation motif of 3'-(G/U)UUCUAUAUUU(C/U)UU-5' was identified upstream of each putative ORF at 4–44 nt before the start codon (Fig. 1B). Similarly, the transcription termination motif of 3'-CUAUAUUUCUUUUG-5' was detected downstream of every ORF at 1–104 nt behind the stop codon (Fig. 1C). To further confirm the transcriptional strategy of PpNSRV-1, primers were designed for RACE of each gene. RACE results indicated that all five genes could be transcribed independently as shown in the transcript map (Fig. 1D).

The amino acid sequence of PpNSRV-1 ORF V is similar to that of the Midway virus (MIDWV) RNA-dependent RNA polymerase (RdRp) (Table 1). Moreover, ORF IV protein contains seven potential O-linked and four potential N-linked glycosylation sites and 29 potential phosphorylation sites (Table 1), suggesting that ORF IV may encode the viral

glycoprotein. The ORF I protein was predicted to have 37 potential phosphorylation sites, whereas ORF II protein has nine (Table 1). The ORF III protein contains 37 potential *O*-linked glycosylation sites (Table 1). Based on the conserved genomic organization of mononegaviruses, ORF II was suggested to encode a phosphoprotein. ORF III may encode the glycosylated matrix protein and ORF I may encode the viral nucleoprotein.

Phylogenetic analysis indicated that PpNSRV-1 clusters with and is a novel member of the family *Nyamiviridae* (Fig. 1E). Multiple alignments of the core RdRp motifs of PpNSRV-1 with those of other mononegaviruses revealed four highly distinct and conserved motifs (A to D) (Fig. 1F) in the PpNSRV-1 ORF V (L) protein.

PASC analysis reveals that the PpNSRV-1 genome is 13.8% identical to that of soybean cyst nematode virus 1 (SbCNV-1; nyamiviral genus *Socyvirus*), 12.3 % identical to that of Nyamanini virus (NYMV; nyamiviral genus *Nyavirus*), 12.1% identical to that of Midway virus (MIDWV; nyamiviral genus *Nyavirus*), and 10.9% identical to that of Sierra Nevada virus (SNVV; nyamiviral genus *Nyavirus*) which is lower than the 17% identity measured between members of the two accepted nyamiviral genera (*Nyavirus* and *Socyvirus*).

MODULE 3: **NEW GENUS**

creating a new genus

Ideally, a genus should be placed within a higher taxon.

Code	2016.015bM	(assigned by ICTV officers)
To create a new genus within:		
Subfamily:	N/A	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 family is specified, enter “ unassigned ” in the family box
Family:	<i>Nyamiviridae</i>	
Order:	<i>Mononegavirales</i>	

naming a new genus

Code	2016.015cM	(assigned by ICTV officers)
To name the new genus: <i>Peropuvirus</i>		

Assigning the type species and other species to a new genus

Code	2016.015dM	(assigned by ICTV officers)
To designate the following as the type species of the new genus		
<i>Pteromalus puparum peropuvirus</i>		Every genus must have a type species. This should be a well characterized species although not necessarily the first to be discovered
The new genus will also contain any other new species created and assigned to it (Module 2) and any that are being moved from elsewhere (Module 7b). Please enter here the TOTAL number of species (including the type species) that the genus will contain:		
<i>1</i>		

Reasons to justify the creation of a new genus:

Additional material in support of this proposal may be presented in the Appendix, Module 11

See species justification.

Origin of the new genus name:

Sigil of <i>Pteromalus puparum</i> and <i>virus</i> .

Reasons to justify the choice of type species:

There is currently only one species in the genus.

Species demarcation criteria in the new genus:

If there will be more than one species in the new genus, list the criteria being used for species demarcation and explain how the proposed members meet these criteria.

N/A because there is currently only one species in the genus.

MODULE 11: **APPENDIX**: supporting material

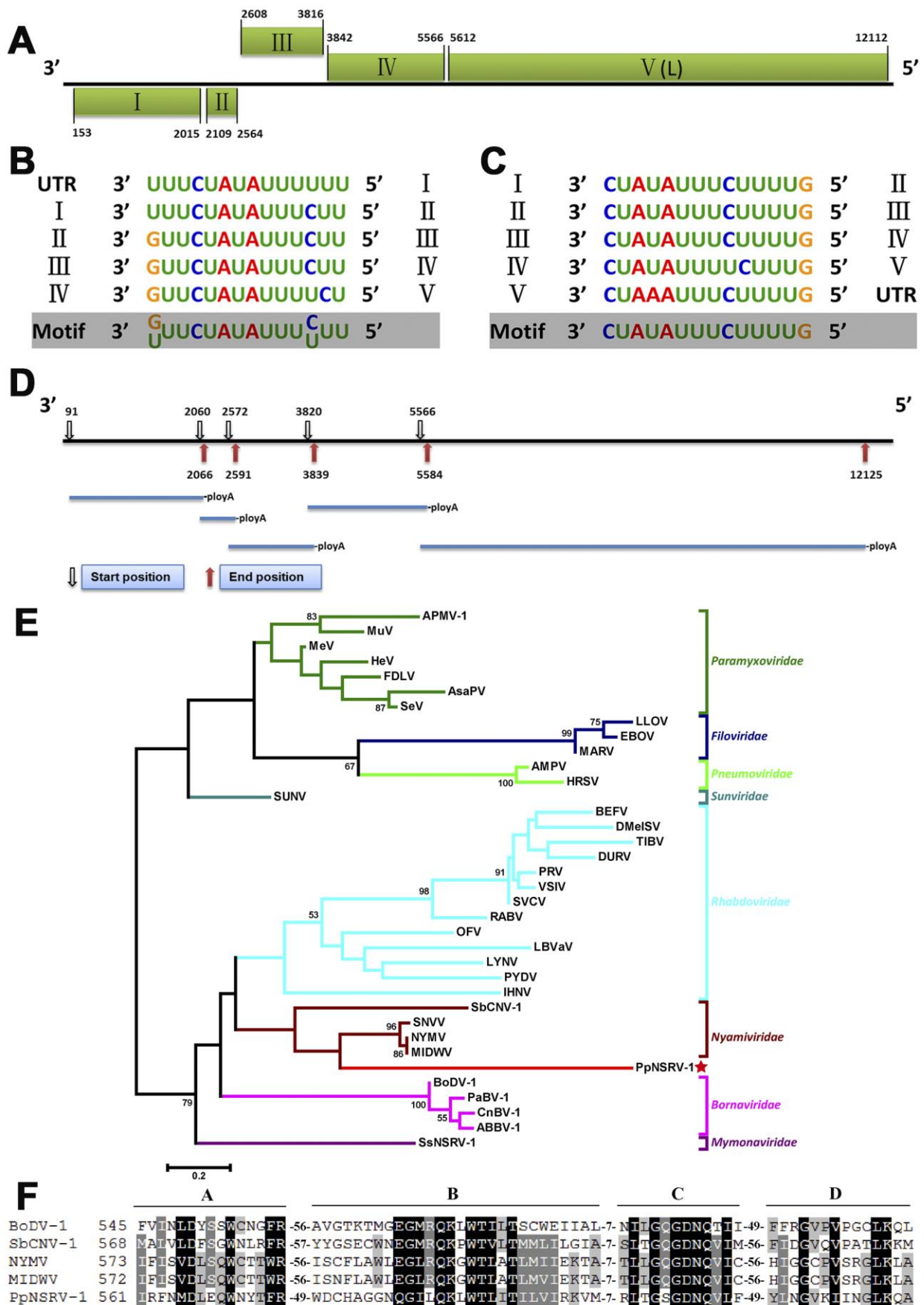


Fig 1. Genomic organization of PpNSRV-1. (A) Genome length and organization of PpNSRV-1. ORF I and ORF II are in the same frame, ORF IV and ORF V are in another frame, and ORF III is in third frame. Boxes indicate the position and length of each ORF, which are labeled with Roman numerals. ORF V encodes the RNA-dependent RNA polymerase (RdRp) protein (L). (B) Putative transcription initiation sequences. (C) Putative transcription termination sequences. The consensus motif is shown below the sequences for each viral ORF in 3' to 5' orientation. (D) The possible transcript map of PpNSRV-1 based on the 5' and 3' RACE. (E) Phylogenetic analysis of PpNSRV-1. A phylogram of the core RdRp motifs of PpNSRV-1 and selected mononegaviruses from the families *Paramyxoviridae*, *Filoviridae*, *Pneumoviridae*, *Sunviridae*, *Rhabdoviridae*, *Nyamiviridae*, *Bornaviridae*, and *Myonnaviridae* are shown. The position of PpNSRV-1 is indicated with a red star. Bootstrap values in 1,000 replications are shown. (F) Multiple sequence alignment of the core RdRp motifs of PpNSRV-1 and selected mononegaviruses. Identical residues are shaded in black, and similar residues are in grey. Conserved motifs are marked A to D. Numbers at the beginning of the sequences represent the amino acid positions from the N-terminus of the L protein.

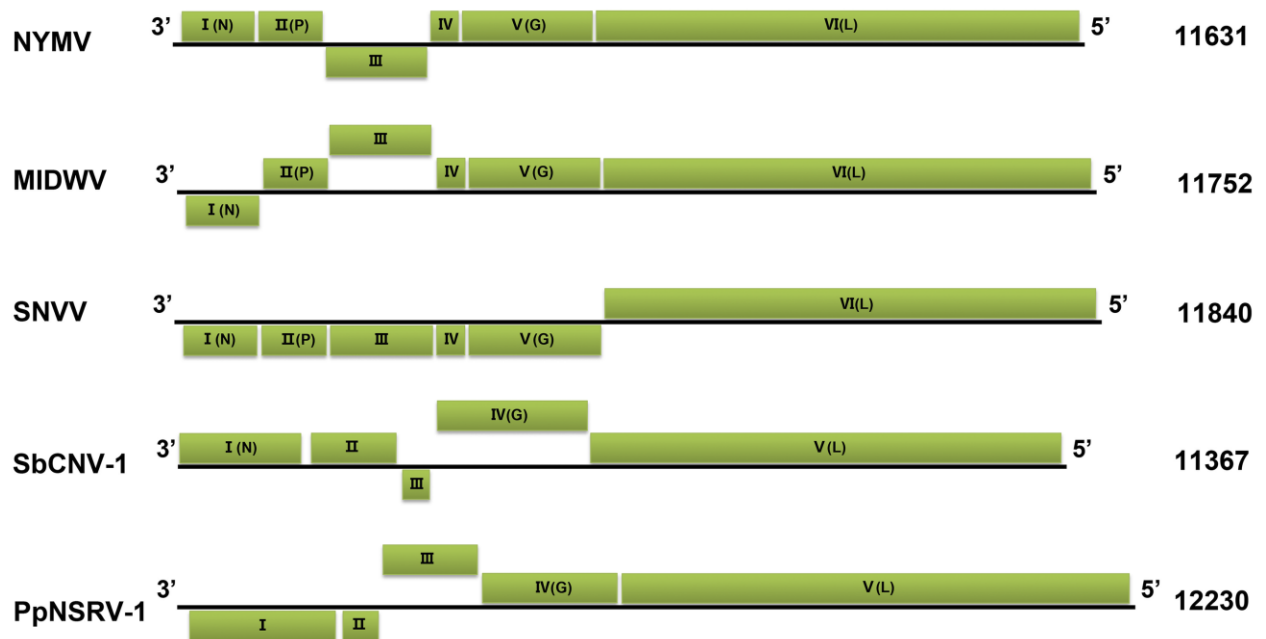


Fig 2. Comparison of the genome organization of PpNSRV-1 and other nyamiviruses. Boxes indicate the position and length of each ORF. The roman numeral in the right indicate the whole genome length of each virus. Boxes which are in the same horizontal position indicate the ORFs are in the same frame.

Table 1. Calculated and predicted properties of PpNSRV-1 ORFs

Viral ORF	ORF length (nt)	Size (aa)	Mass (kDa)	pI	Signal peptide	No. of glycosylation sites		No. of phosphorylation sites			Top BLASTX match	E value
						O-linked	N-linked	Ser	Thr	Tyr		
ORF I	1863	620	69.13	5.47	-	11	0	15	16	6	ND	-
ORF II	456	151	16.86	5.45	-	15	0	6	3	0	ND	-
ORF III	1209	402	43.5	8.73	-	37	0	24	15	2	ND	-
ORF IV (G)	1725	574	63.36	6.39	+	7	4	16	10	3	ND	-
ORF V (L)	6501	2166	245.9	7.24	-	15	0	43	24	16	RdRp (<i>Midway virus</i>)	2E-83

ND, No significant homology was detected.

References:

Wang F., Fang Q., Wang B.B., Yan Z.C., Hong J., Bao Y. M., Kuhn J. H., Werren J.H., Song Q.S., Ye G.Y., 2016. A novel negative-strand RNA virus mediates sex ratio in a parasitoid host. In revision.

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.