

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:	2015.011a-dB			(to be completed by ICTV officers)				
Short title: To create one (1) refamily Siphoviridae. (e.g. 6 new species in the genus 2 Modules attached (modules 1 and 10 are required)	_	uding five 2 7	•	species w 4 9	5 ☐ 10 ⊠			
Author(s):								
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Andrew M. Kropinski Phage.Canada@gmail.com								
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) Bacterial & Archaeal Virus Subcommittee								
ICTV Study Group comments (if any) and response of the proposer:								
Please note that we have chosen to refer to this new genus as <i>P70virus</i> rather than <i>P70likevirus</i> since the Bacterial and Archaeal Virus Subcommittee of ICTV has voted overwhelmingly in favour of eliminating " <i>like</i> " and " <i>Phi</i> " from phage genus names.								
Date first submitted to ICTV: Date of this revision (if different	Date first submitted to ICTV: May 2015 Date of this revision (if different to above):							
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	2015.011aB (assigned by IC			ICTV office	CTV officers)			
To crea	To create 5 new species within:							
					Fill in all that apply.			
Genus: P70virus (new)			w)		 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 			
Subfamily:								
Family: Siphoviridae								
	Order: Caudovirales				"unassigned" in the genus box.			
		Representative isolat per species please)	e: (only 1	GenBank sequence accession number(s)				
Listeria	a virus l	P70	Listeria phage P70		JX442241			
		Listeria phage LP-026	•	KJ094020				
Listeria virus LP110 Listeria		Listeria phage LP-110)	JX126919				
Listeria virus LP114 Listeria		Listeria phage LP-114		KJ094021				
Listeria virus LP37			Listeria phage LP-037	,	JX126920.2			

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

- Explain how the proposed species differ(s) from all existing species.
 - o 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

Listeria phage P70 features an elongated head measuring 128 nm in length and 57nm in diameter and a tail length of 141 (B3 morphotype). P70 has a broad host range lysing isolates of serovars 1/2a, 1/2b, 1/2c, 4a, 4c, 4d, 4e, 5, 6a, and 6b (4). While being strictly lytic it encodes a number of proteins indicative of "a temperate lifestyle, such as ORF44 (Holliday junction resolvase), ORF49 (DNA replication inhibitor), or ORF53 (C1-like repressor protein) (4)" By comparison the US isolates LP-026, LP-114, LP-037, LP-110 measurements are: head 123 x 44 nm and tail, 162 x 7–8 nm (5). The latter manuscript recognized that these phages and P70 were related.

BLASTN, CoreGenes (Table 1; 2), progressiveMauve alignment (Fig. 2; 1) and phylogenetic analyses (Fig. 3; 3) all indicate that the proposed genus, *P70virus*, is cohesive and distinct from the other genera of viruses. The phages of this genus possess genome of approx. 66 kb (36.4 mol%G+C), and encode 115 proteins and 0 tRNAs. They share 90-93% DNA sequence identity and >86% homologous proteins (Table 1).

We have chosen 95% DNA sequence identity as the criterion for demarcation of species in this new genus. Each of the proposed species differs from the others with more than 5% at the DNA level as confirmed with the BLASTN algorithm.

Please note that we have chosen to refer to this new genus as P70virus rather than P70likevirus since the

Bacterial and Archaeal Virus Subcommittee of ICTV has voted overwhelmingly in favour of eliminating "*like*" and "*Phi*" from phage genus names.

MODULE 3: NEW GENUS

creating a new genus

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

Code	201	15.011bB	(assigned by	ned by ICTV officers)			
To create	a new	genus within:		Fill in all that apply.			
Subfa	mily:			If the higher taxon has yet to be created (in a later module, helper) write "(read)"			
Fa	mily:	Siphoviridae		(in a later module, below) write "(new)"after its proposed name.			
C	Order:	Caudovirales		If no family is specified, enter "unassigned" in the family box			

naming a new genus

Code	2015.011cB	(assigned by ICTV officers)				
To name the	he new genus: P70virus					

Assigning the type species and other species to a new genus

Assigning the type species and other species to a new genus							
Code	2015.011dB	(assigned by ICTV officers)					
To designa	To designate the following as the type species of the new genus						
Listeria vir	rus P70	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: 5							

Reasons to justify the creation of a new genus:

Additional material in support of this proposal may be presented in the Appendix, Module 9

BLASTN, CoreGenes (Table 1; 2), progressiveMauve alignment (Fig. 2; 1) and phylogenetic analyses (Fig. 3; 3) all indicate that the proposed genus, *P70virus*, is cohesive and distinct from the other genera of viruses.

Origin of the new genus name:

Named after the first phage of its type to be sequenced: *Listeria* phage P70

Reasons to justify the choice of type species:

First phage of its type to be sequenced

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.

We have chosen 95% DNA sequence identity as the criterion for demarcation of species in this new genus. Each of the proposed species differs from the others with more than 5% at the DNA level as confirmed with the BLASTN algorithm.

MODULE 10: APPENDIX: supporting material

additional material in support of this proposal

References:

- 1. Darling AE, Mau B, Perna NT. progressiveMauve: multiple genome alignment with gene gain, loss and rearrangement. PLoS One. 2010; 5(6):e11147.
- 2. Turner D, Reynolds D, Seto D, Mahadevan P. CoreGenes 3.5: a webserver for the determination of core genes from sets of viral and small bacterial genomes. BMC Res Notes. 2013; 6:140.
- 3. Dereeper A, Guignon V, Blanc G, Audic S, Buffet S, Chevenet F, Dufayard JF, Guindon S, Lefort V, Lescot M, Claverie JM, Gascuel O. Phylogeny.fr: robust phylogenetic analysis for the non-specialist. Nucleic Acids Res. 2008; 36(Web Server issue):W465-9.
- 4. Schmuki MM, Erne D, Loessner MJ, Klumpp J. Bacteriophage P70: unique morphology and unrelatedness to other *Listeria* bacteriophages. J Virol. 2012; 86(23):13099-102.
- 5. Denes T, Vongkamjan K, Ackermann HW, Moreno Switt AI, Wiedmann M, den Bakker HC. Comparative genomic and morphological analyses of *Listeria* phages isolated from farm environments. Appl Environ Microbiol. 2014; 80(15):4616-25. [LP-114, LP-110, LP-037 & LP-026]

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.

Table 1. Properties of the two phages belonging to the *P70virus* genus.

Phage	GenBank	Genome	Genome	No.	No.	DNA (%	Proteome
	accession No.	length (kb)	(mol%G+C)	CDS	tRNAs	sequence	(%
						identity)*	homologous
							proteins)**
Listeria virus	JX442241	67.17	36.5	119	0	100	100
P70							
Listeria virus	KJ094020	67.15	36.3	114	0	93	86.5
LP26							
Listeria virus	JX126919	65.13	36.3	113	0	90	88.2
LP110							
Listeria virus	KJ094021	66.68	36.4	116	0	91	89.1
LP114							
Listeria virus	JX126920.2	64.76	36.6	114	0	91	89.9
LP37							

^{*} Determined using BLASTN; ** Determined using CoreGenes (2)

Fig. 1. Electron micrographs of negatively stained *Listeria* phage P70 (left two panels) and *Listeria* phage LP-032 (right panel).

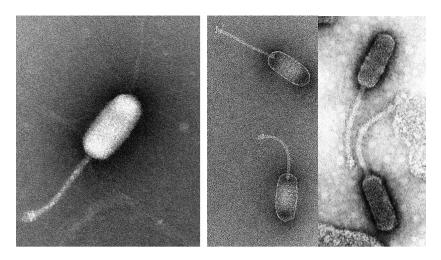


Fig. 2. progressiveMauve alignment of the annotated genomes of *Listeria* phages (from top to bottom: LP-026, LP-037, LP-110, LP-114, P70) (1). Colored blocks indicate the regions of 1 to 1 best alignment with rearrangement breakpoints in a different random color. The degree of sequence similarity between regions is given by a similarity plot within the colored blocks with the height of the plot proportional to the average nucleotide identity (Aaron Darling, personal communication). Please note that these genomes are not collinear.

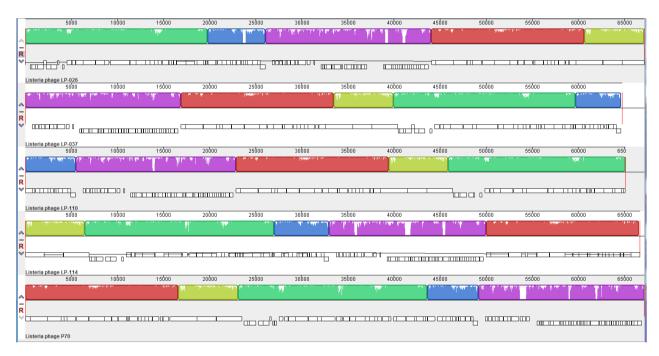


Fig. 3. Phylogenetic analysis of the large subunit terminase of p70viruses and some related phages constructed using "one click" at phylogeny.fr (3). "The "One Click mode" targets users that do not wish to deal with program and parameter selection. By default, the pipeline is already set up to run and connect programs recognized for their accuracy and speed (MUSCLE for multiple alignment and PhyML for phylogeny) to reconstruct a robust phylogenetic tree from a set of sequences." It also includes the use of Gblocks to eliminate poorly aligned positions and divergent regions. "The usual bootstrapping procedure is replaced by a new confidence index that is much faster to compute. See: Anisimova M., Gascuel O. Approximate likelihood ratio test for branches: A fast, accurate and powerful alternative (Syst Biol. 2006;55(4):539-52.) for details."

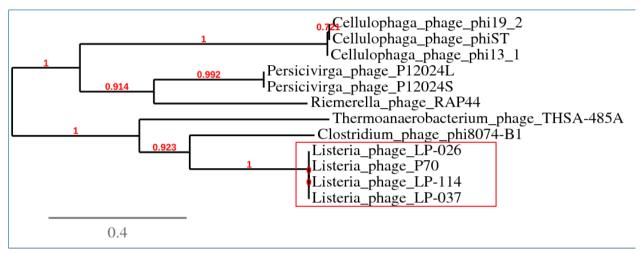


Figure 1: Phylogenetic tree (the branch length is proportional to the number of substitutions per site).