



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:	2013.005a-dB	(to be completed by ICTV officers)			
Short title: To create a new genus, <i>Phifllikevirus</i> , within the family <i>Siphoviridae</i> (e.g. 6 new species in the genus <i>Zetavirus</i>)					
Modules attached (modules 1 and 9 are required)	1 <input checked="" type="checkbox"/> 6 <input type="checkbox"/>	2 <input checked="" type="checkbox"/> 7 <input type="checkbox"/>	3 <input checked="" type="checkbox"/> 8 <input type="checkbox"/>	4 <input type="checkbox"/> 9 <input checked="" type="checkbox"/>	5 <input type="checkbox"/>

Author(s) with e-mail address(es) of the proposer:

Evelien Adriaenssens Evelien.Adriaenssens@gmail.com
Andrew M. Kropinski akropins@uoguelph.ca
Rob Lavigne rob.lavigne@biw.kuleuven.be
Mal Horsburgh m.j.horsburgh@liverpool.ac.uk

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-EC or Study Group comments and response of the proposer:

Date first submitted to ICTV: June 2013
Date of this revision (if different to above): July 2014

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	2013.005aB	(assigned by ICTV officers)
To create 3 new species within:		
Genus:	<i>Phifflikevirus (new)</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:	<i>Siphoviridae</i>	
Order:	<i>Caudovirales</i>	
		GenBank sequence accession number(s) of reference isolate:
<i>Enterococcus phage phifl1</i>		GQ478081
<i>Enterococcus phage phifl2</i>		GQ478084
<i>Enterococcus phage phifl3</i>		GQ478086

<p>Reasons to justify the creation and assignment of the new species:</p> <ul style="list-style-type: none"> • Explain how the proposed species differ(s) from all existing species. <ul style="list-style-type: none"> ○ 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 <p>BLASTN analyses reveal that these <i>Enterococcus</i> phages are related and distinct from any other phage. We have chosen 95% DNA sequence identity as the criterion for demarcation of species.</p>
--

MODULE 3: **NEW GENUS**

creating a new genus

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

Code	2013.005bB	(assigned by ICTV officers)
To create a new genus within:		
Subfamily:		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>Siphoviridae</i>	
Order:	<i>Caudovirales</i>	

naming a new genus

Code	2013.005cB	(assigned by ICTV officers)
To name the new genus: <i>Phiflikevirus</i>		

Assigning the type species and other species to a new genus

Code	2013.005dB	(assigned by ICTV officers)
To designate the following as the type species of the new genus		
<i>Enterococcus phage phifl1</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:		
3		

Reasons to justify the creation of a new genus:

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

This genus represents a group of temperate siphoviruses active against *Enterococcus faecalis*, possessing 36.3 to 40.3 kb genomes which encode 59 - 74 proteins and a mol% G+C content of 34.0-36.7 (1). Morphologically, their isometric heads measure 48 - 53nm in diameter; with tails 205-229 nm in length. Based on a shared DNA identity of over 95 %, the phages described by Yasmin and colleagues were classified in three separate species, *Enterococcus phage phifl1* comprising the isolates PhiFL1A, PhiFL1B and PhiFL1C, *Enterococcus phage phifl2* with the isolates PhiFL2A and PhiFL2B, and *Enterococcus phage phifl3* with PhiFL3A and PhiFL3B.

Origin of the new genus name:

Enterococcus phage PhiFL1A

Reasons to justify the choice of type species:

The original isolate of this group.

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 EMBOSS Stretcher algorithm.

MODULE 9: **APPENDIX**: supporting material

additional material in support of this proposal

References:

- 1: Yasmin A, Kenny JG, Shankar J, Darby AC, Hall N, Edwards C, Horsburgh MJ. 2010. Comparative genomics and transduction potential of *Enterococcus faecalis* temperate bacteriophages. J Bacteriol. 192(4):1122-30.
- 2: Darling AE, Mau B, Perna NT (2010) progressiveMauve: multiple genome alignment with gene gain, loss and rearrangement. PLoS One 5: e111147

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.

These represent a group of temperate siphoviruses active against *Enterococcus faecalis* which possess 36.3-40.3 kb genomes which encode 59-74 proteins; and a mol%G+C content of 34.0-36.7. Morphologically their isometric heads measure 48-53nm in diameter; with tails 205-229nm in length (1) (Figure 2).

Table 1. Phage genomes representing the proposed new species in this genus and comparison

Phage	GenBank Accession No.	Genome size (bp)	Mol%G+C	% DNA sequence identity (a)	% Shared proteins (b)
<i>Enterococcus phage phiFL1A</i>	GQ478081	38,764	36.7	100%	100%
<i>Enterococcus phage phiFL2A</i>	GQ478084	36,270	34.6	88.5	80.3
<i>Enterococcus phage phiFL3A</i>	GQ478086	39,576	34.5	64.1	55.7

(a) Calculated with EMBOSS Stretcher (relative to the type species phiFL1A)

(b) Calculated with CoreGenes 2.0

Table 2. Related phages

Species	Similar phage
<i>Enterococcus phage phiFL1</i>	<i>Enterococcus phage phiFL1B</i> , <i>Enterococcus phage phiFL1C</i>
<i>Enterococcus phage phiFL2</i>	<i>Enterococcus phage phiFL2B</i>
<i>Enterococcus phage phiFL3</i>	<i>Enterococcus phage phiFL3B</i>

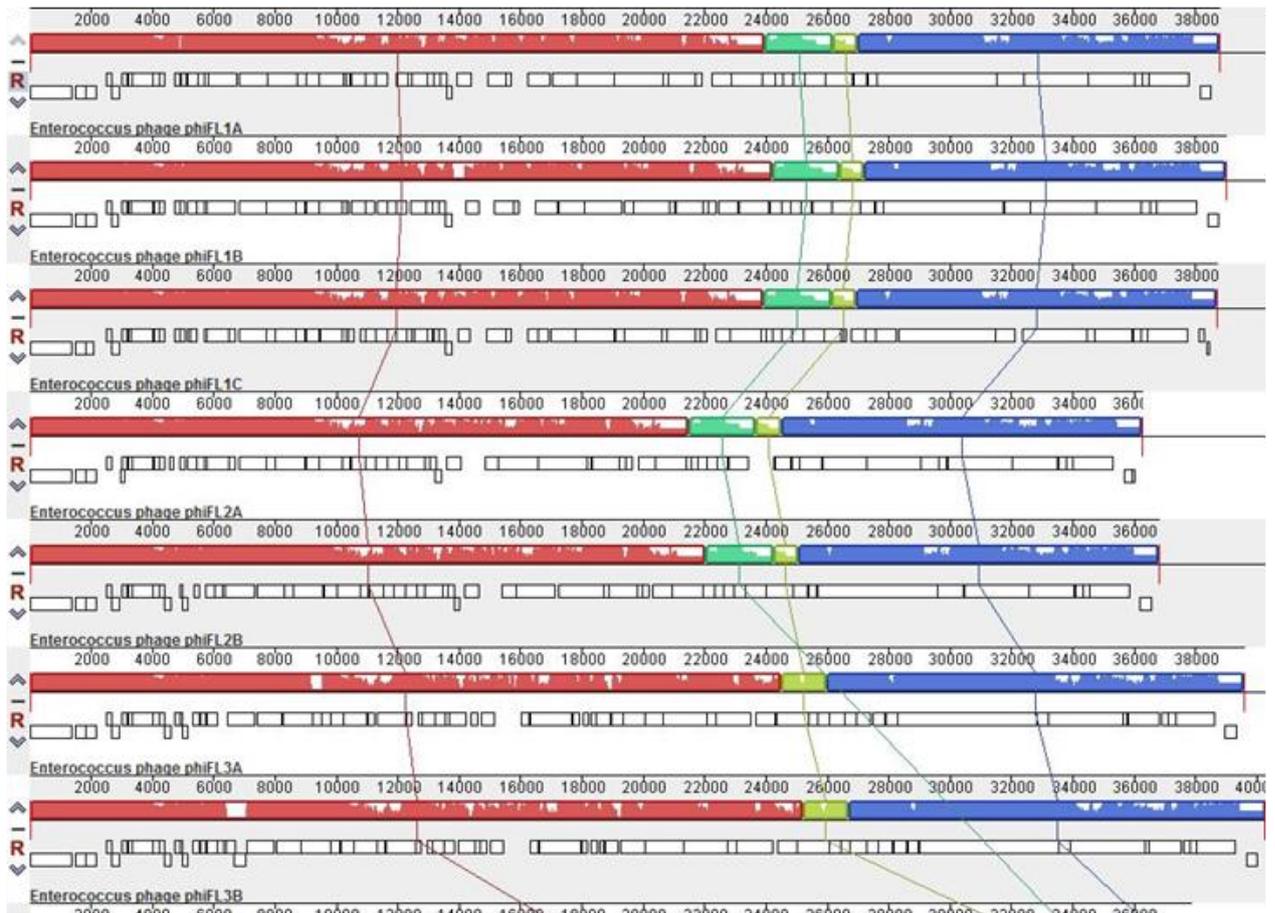


Figure 1. progressiveMauve alignment of the phage genomes (full genome represented by its annotated ORFs in white blocks) (2). Colored blocks indicate the regions of 1 to 1 best alignment with rearrangement breakpoints in a different random color. The degree of sequence similarity between regions is given by a similarity plot within the colored blocks with the height of the plot proportional to the average nucleotide identity.

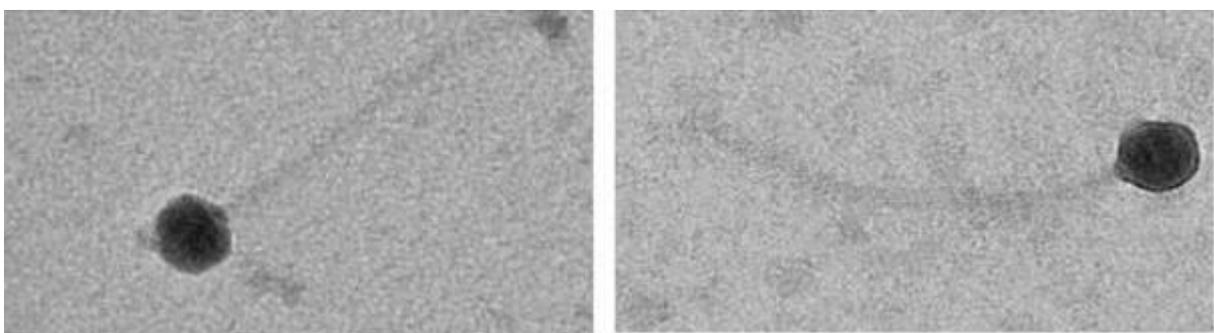


Figure 2. TEM images of phiFL2 (left) and phiFL3 (right), marking them as *Siphoviridae* members.

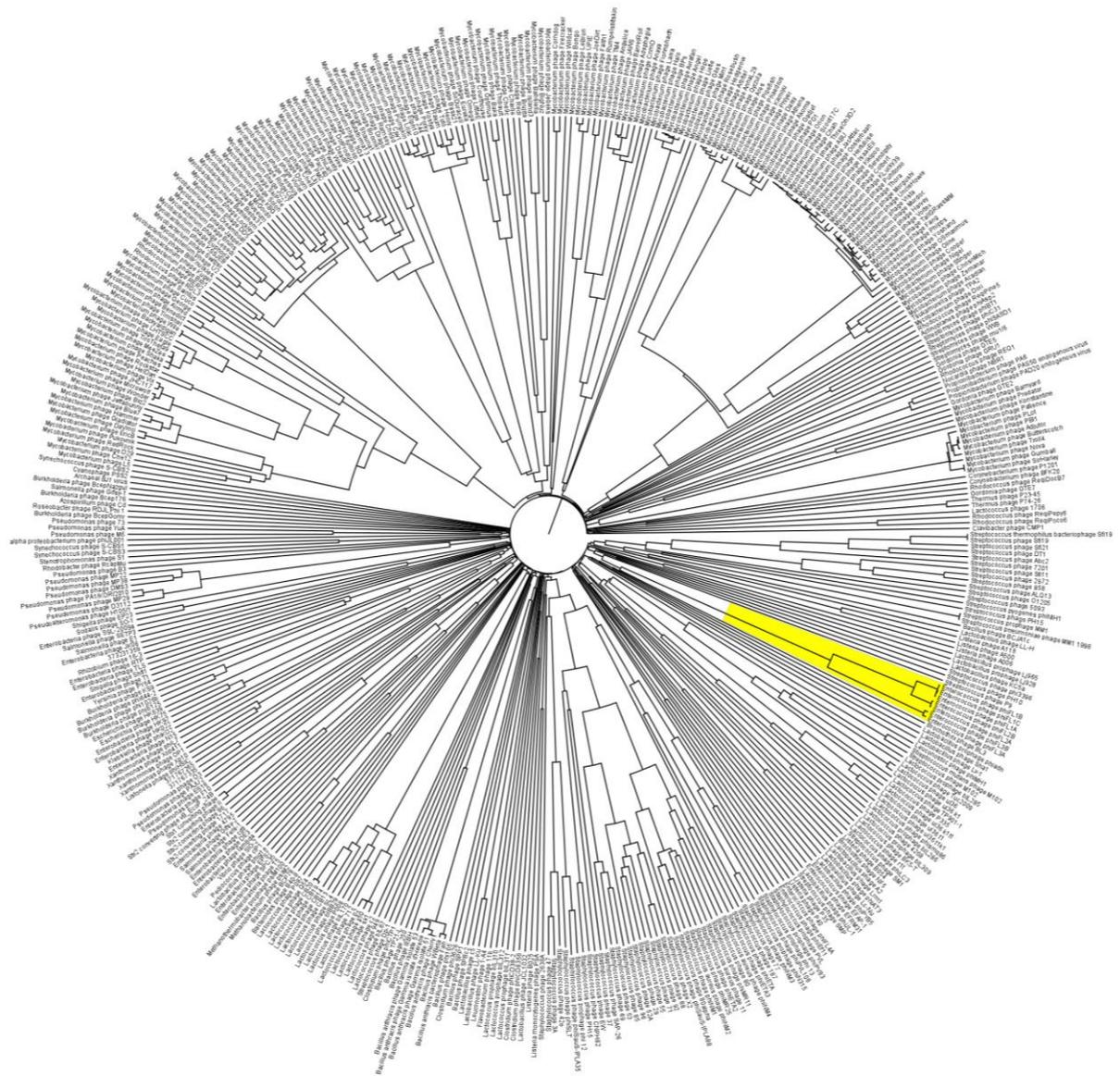


Figure 3: Whole genome phylogenetic tree of all *Siphoviridae* phages in the NCBI database in November 2012. Genome sequences were aligned and the tree (NJ) was created with ClustalW 2.0 and visualized with FigTree. The scale bar represents 0.06 substitutions per site. The newly proposed genus is highlighted in yellow.

