



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:	2013.006a-dB	(to be completed by ICTV officers)			
Short title: To create a new genus, the <i>Iebhlikevirus</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
Laurence Van Melderren lvmelder@ulb.ac.be

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	2013.006aB	(assigned by ICTV officers)
To create 2 new species within:		
Genus:	<i>Iebhlikevirus (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>Bacillus phage IEBH</i> <i>Bacillus phage 250</i>		EU874396 GU229986

<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 the phages IEBH and 250 are related and distinct from other <i>Bacillus</i> phages. 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	2013.006bB	(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	2013.006cB	(assigned by ICTV officers)
To name the new genus: <i>Iebhlikevirus</i>		

Assigning the type species and other species to a new genus

Code	2013.006dB	(assigned by ICTV officers)
To designate the following as the type species of the new genus		
<i>Bacillus phage IEBH</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:		
2		

Reasons to justify the creation of a new genus:

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

Two <i>Bacillus cereus</i> -infecting phages are currently classified in this genus, IEBH and 250 (1, 5). The type phage IEBH has an isometric head of 55 nm and a long non-contractile tail of 150 nm displaying transverse tail discs; for phage 250 no dimensions have been reported. Both phages share 81.7% DNA identity, but relative to phage IEBH they only have 54.7% shared proteins. This is due to a lower number of ORFs annotated in phage 250. BLASTN analyses reveal that they are related and distinct from other <i>Bacillus</i> phages. We propose 40% shared proteins with the type species <i>Bacillus phage Iebh</i> for new phages to belong to this genus.

Origin of the new genus name:

<i>Bacillus</i> phage IEBH

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.

MODULE 9: **APPENDIX**: supporting material

additional material in support of this proposal

References:

1. Lee Y-D & Park J-H (2010) Genomic sequence of temperate phage 250 isolated from emetic *B. cereus* and cloning of putative endolysin. *Food Science and Biotechnology* 19: 1643–1648
2. Mahadevan P, King JF, Seto D (2009) CGUG: *in silico* proteome and genome parsing tool for the determination of “core” and unique genes in the analysis of genomes up to ca. 1.9 Mb. *BMC research notes* 2: 168. doi:10.1186/1756-0500-2-168.
3. Mahadevan P, King JF, Seto (2009) Data mining pathogen genomes using GeneOrder and CoreGenes and CGUG: gene order, synteny and in silico proteomes. *International Journal of Computational Biology and Drug Design* 2: 100–114.
4. Zafar N, Mazumder R, Seto D (2002) CoreGenes: A computational tool for identifying and cataloging “core” genes in a set of small genomes. *BMC Bioinformatics* 3: 12. doi:10.1186/1471-2105-3-12.
5. Smeesters PR, Drèze P-A, Bousbata S, Parikka KJ, Timmery S, Hu X, Perez-Morga D, Deghorain M, Toussaint A, Mahillon J & Van Melderen L (2011) Characterization of a novel temperate phage originating from a cereulide-producing *Bacillus cereus* strain. *Research in Microbiology* 162: 446–459
6. Rohwer F & Edwards R (2002) The Phage Proteomic Tree: a genome-based taxonomy for phage. *Journal of Bacteriology* 184: 4529–4535
7. Darling AE, Mau B, Perna NT (2010) progressiveMauve: multiple genome alignment with gene gain, loss and rearrangement. *PLoS One* 5: e11147

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.

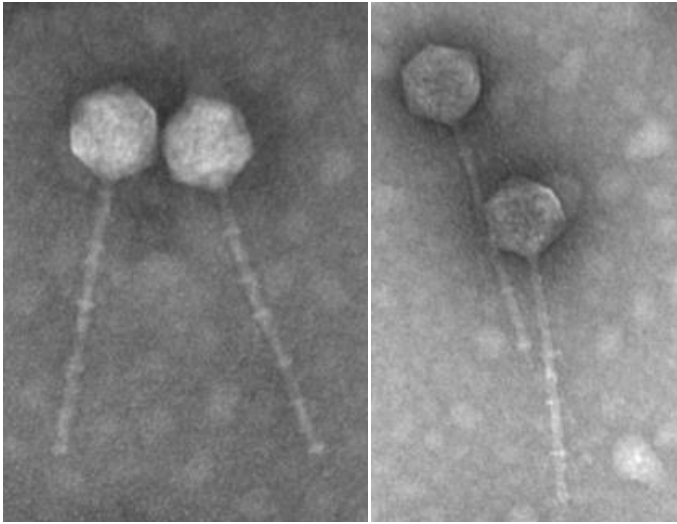


Figure 1. Electron micrograph of *Bacillus* phage IEBH (kindly provided by L. Van Melderen; Université Libre de Bruxelles, Gosselies, Belgium)

Table 1. Phage genomes

Phage	GenBank Accession No.	Genome size (bp)	Mol%G+C	% DNA sequence identity (a)	% Shared proteins (b)
IEBH	EU874396	53104	36.42	100%	100%
250	GU229986	56505	36.44	81.7%	54.7%

(a) EMBOSS Stretcher – reoriented genome to compensate for different origins

(b) CoreGenes 2.0 (2-4)

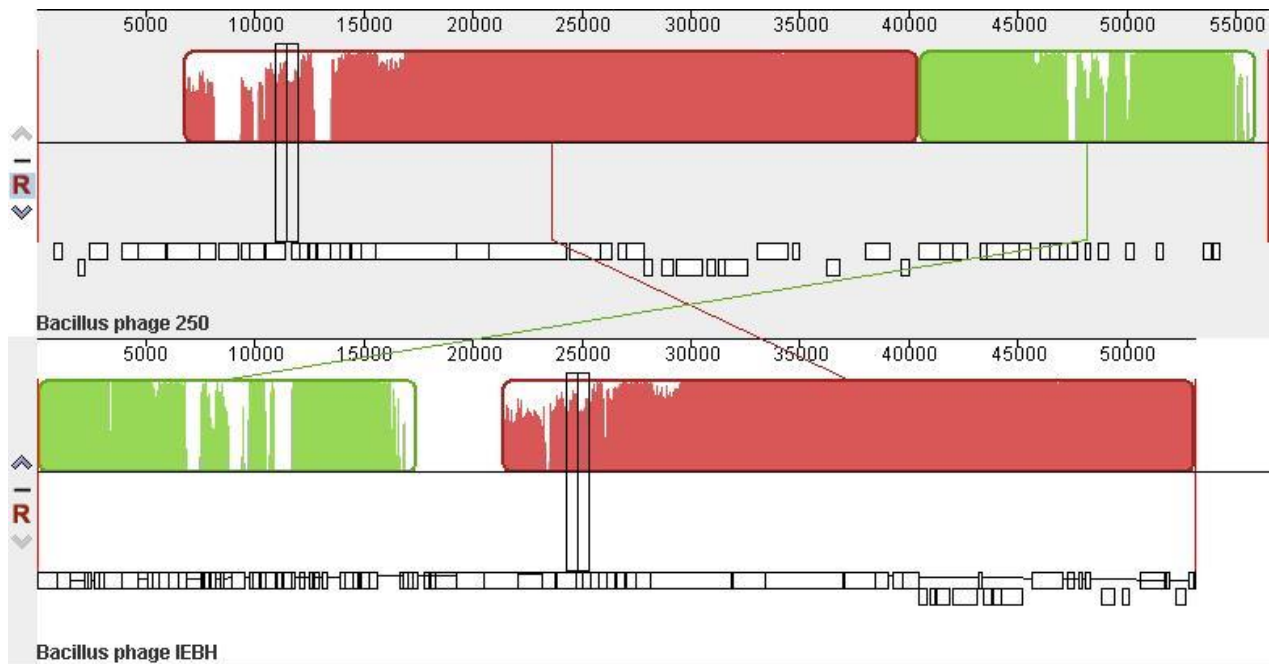


Figure 2. progressiveMauve alignment of the phage genomes of the proposed genus (full genome represented by its annotated ORFs in white blocks) (7). Colored blocks indicate the regions of 1 to 1 best alignment with rearrangement breakpoints in a different random color. The degree of sequence similarity 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 3: Phage Proteomic Tree (6) of all the *Siphoviridae* phages in the NCBI database November 2012. Briefly, all predicted proteins sequences are compared with all others

and a length-corrected protein distance matrix was calculated based on CLUSTALW alignment of sequences with a BLASTP e value < 0.001, with missing protein penalties of 10 and gap extension penalties of 0.20 (6). The tree was generated using FITCH. The proposed genus is highlighted. The scale bar represents protein distances of 2.0.

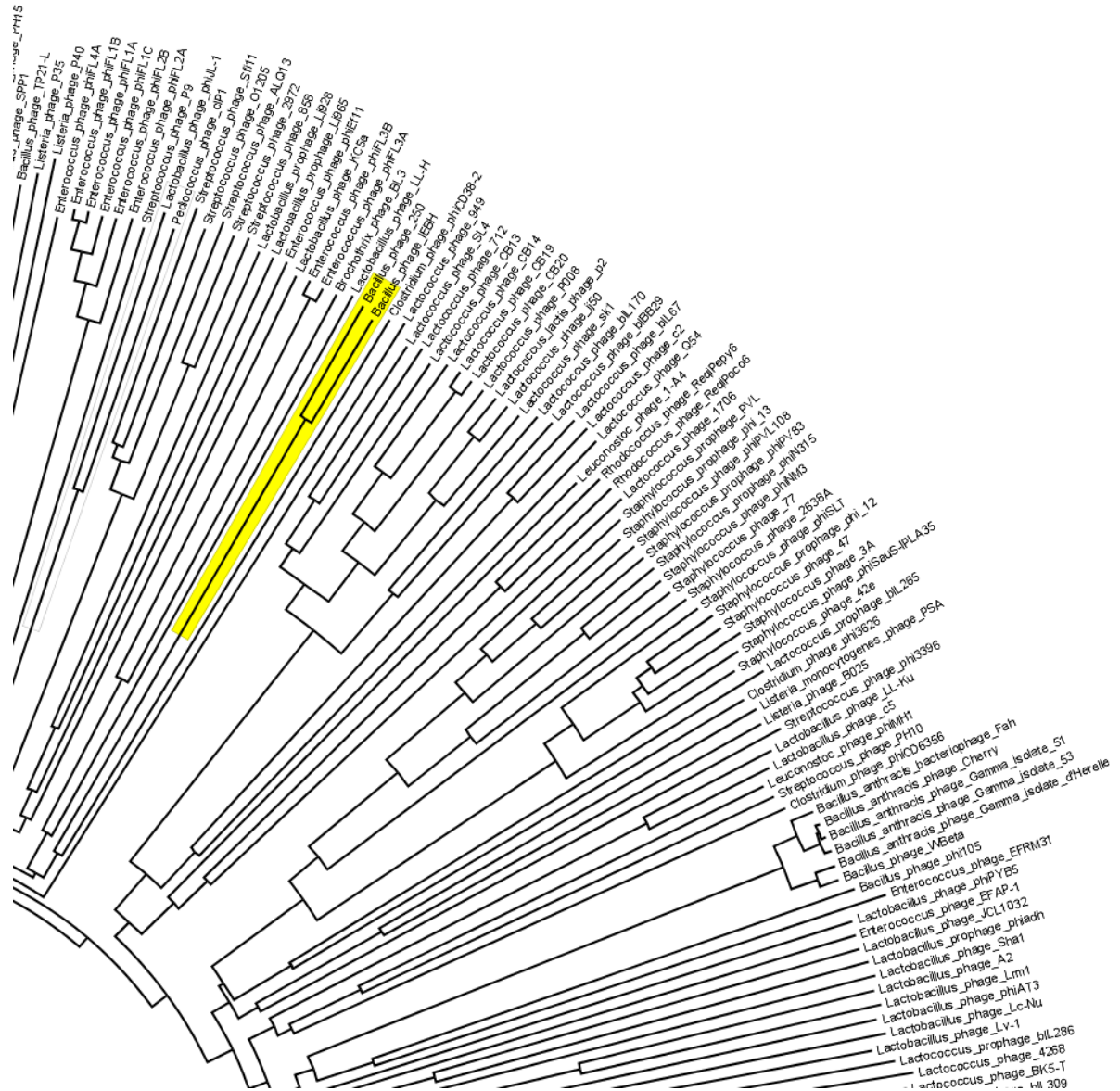


Figure 4: Fragment of the phylogenetic tree of Figure 3, zoomed in on the proposed genus.