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

MODULE 1: TITLE, AUTHORS, etc

MUDULE 1: TITLE, AUTHORS, Etc						
Code assigned:	2013.014a-	dB		(to be co officers)	mpleted by	ICTV
Short title: To create a new genus, Cjwunalikevirus, within the family Siphoviridae						
(e.g. 6 new species in the ge Modules attached (modules 1 and 9 are require	1		2 × 7 □	3 ⊠ 8 □	4 ☐ 9 ⊠	5 🗌
Author(s) with e-mail ad	dress(es) of the propo	ser:				
Evelien M Adriaenssens, a Andrew M Kropinski, akro Rob Lavigne, rob.lavigne John Nash, john.nash@ph	opins@uoguelph.ca @biw.kuleuven.be	gman.	<u>com</u>			
List the ICTV study grou	ıp(s) that have seen tl	nis pro	posal:			
A list of study groups and co http://www.ictvonline.org/sub in doubt, contact the appropr chair (fungal, invertebrate, pl vertebrate viruses)	committees.asp . If riate subcommittee					
ICTV-EC or Study Group comments and response of the proposer:						
Date first submitted to ICT	ΓV:		June	2013		
Date of this revision (if dit	fferent to above):		Julv	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 <b>2013.014aB</b>		(assigned by ICTV officers)	
To create 9 n	ew species within:		
Genus: Cjwunalikevirus (new) Subfamily: Family: Siphoviridae Order: Caudovirales		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.	
And name th	e new species:	GenBank sequence accession number(s) of reference isolate:	
Mycobacterium phage CJW1 Mycobacterium phage pumpkin Mycobacterium phage 244 Mycobacterium phage porky Mycobacterium phage Bask21 Mycobacterium phage sirduracell Mycobacterium phage eureka Mycobacterium phage kostya Mycobacterium phage toto		AY129331 GQ303265 DQ398041 EU816588 JF937091 JF937106 JN412590 EU816591 JN006061	

## 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**.
  - o 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

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 (Table 1).

## **MODULE 3: NEW GENUS**

creating a new genus

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

Code 2	201	3.014bB	(assigned by I	CTV officers)
To create a	new	genus within:		Fill in all that apply.
Subfam	ily:			If the higher taxon has yet to be created
Fami	ily:	Siphoviridae		(in a later module, below) write "(new)" after its proposed name.
Ord	der:	Caudovirales		<ul> <li>If no family is specified, enter "unassigned" in the family box</li> </ul>

naming a new genus

Code	2013.014cB	(assigned by ICTV officers)	
To name the new genus: Cjwunalikevirus			

Assigning the type species and other species to a new genus

Code	2013.014dB	3.014dB (assigned by ICTV officers)				
To design:	To designate the following as the type species of the new genus					
Mycobacterium phage CJW1		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:						
9						

### 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 was originally recognized by the Mycobacteriophage group (<a href="www.phagesdb.org">www.phagesdb.org</a>) as belonging to cluster E. Phages belonging to this genus share a comparable genome size (74 - 76 kb), a comparable GC content (~63.1%), the presence of about 2 tRNAs and a genome with defined physical end with a comparable 9 bp 3' overhang. Members of this genus also have a comparable morphology, with an isometric head and a long, non-contractile tail (Figure 1). A ClustalW analysis of the complete genomes of this genus with all other *Mycobacterium* phages belonging to the *Siphoviridae* reveals that this genus is a clearly separate group (Figures 2 and 3). We propose a shared protein content of at least 40% with the type phage, *Mycobacterium phage CJW1*. We performed a CoreGenes 3.5 analysis [1–3] with selected phages based on the results of the ClustalW analysis (Table 1). The CoreGenes analysis was also performed against the type species of other proposed genera of siphoviruses infecting *Mycobacterium* and the shared protein content was consistently below 40% (data not shown).

#### **Origin of the new genus name:**

Mycobacterium phage CJW1

### Reasons to justify the choice of type species:

The genus *Cjwunalikevirus* is named after the first isolated and sequenced phage of this group, *Mycobacterium* phage Cjw1 [4]. The number '1' is replaced with the word 'una' to avoid confusion with the adjacent letter 'l' in the genus name.

# 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. Mahadevan P, King JF, Seto (2009) Data mining pathogen genomes using GeneOrder and CoreGenes and CGUG: gene order, synteny and in silico proteomes. Int J Comput Biol Drug Des 2: 100–114.
- 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 Res Notes 2: 168. doi:10.1186/1756-0500-2-168.
- 3. 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.
- 4. Pedulla ML, Ford ME, Houtz JM, Karthikeyan T, Wadsworth C, et al. (2003) Origins of highly mosaic mycobacteriophage genomes. Cell 113: 171–182.
- 5. Darling AE, Mau B, Perna NT (2010) progressiveMauve: multiple genome alignment with gene gain, loss and rearrangement. PLoS One 5: e11147. doi:10.1371/journal.pone.0011147.

#### 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.

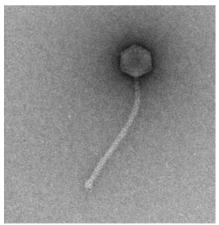


Figure 1: EM picture of phage Cjw1, a member of the genus Cjwunalikevirus (http://phagesdb.org/media/emPics/Cjw1.tif).

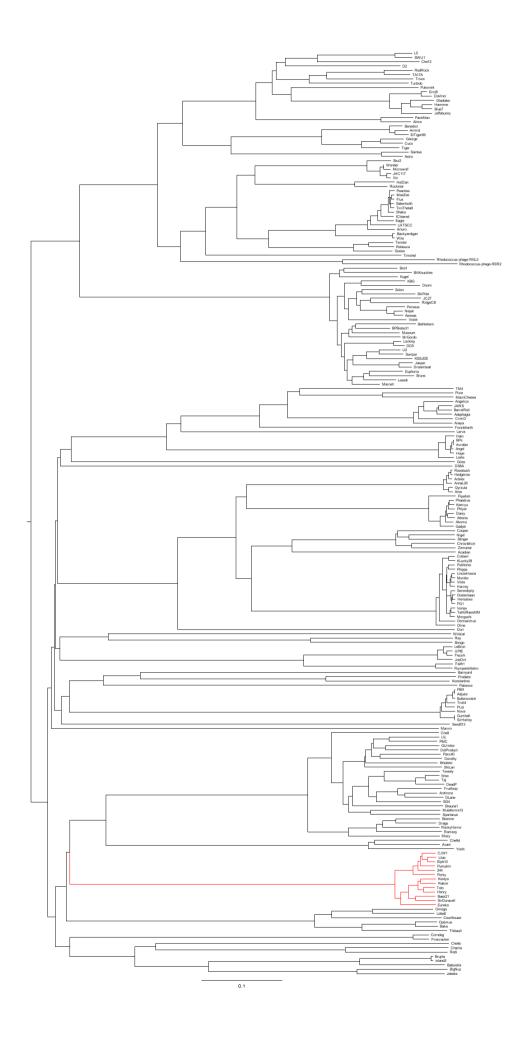


Figure 2: ClustalW phylogenetic tree (NJ) of complete genomes of all *Mycobacterium* siphoviruses in the NCBI database in November 2012. The proposed genus is colored in red.

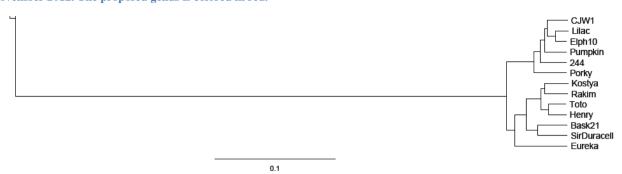


Figure 3: ClustalW phylogenetic tree of complete genomes of the isolates belonging to the genus *Cjwunalikevirus*, excerpt of Figure 2.



Figure 4: progressiveMauve alignment of the phage genomes belonging to the proposed [5]. 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.

Table 1: Selected EMBOSS Stretcher results in the genus Cjwunalikevirus.

Isolate Name	Isolate name	% DNA identity	Species
CJW1	Elph10	95.1	Mycobacterium phage Cjw1
Lilac	Elph10	95.8	Mycobacterium phage Cjw1

Kostya	Rakim	94.9	Mycobacterium phage Kostya
Toto	Henri	95.6	Mycobacterium phage Toto
CJW1	Pumpkin	93.1	Different species

Table 2: CoreGenes 3.5 and EMBOSS Stretcher analysis, showing the shared protein content and DNA similarity of selected phages of the genus *Cjwunalikevirus* with the type species of this genus.

Phage name	% proteins shared with Cjw1	% DNA similarity with Cjw1
Mycobacterium phage pumpkin	95.7	93.1
Mycobacterium phage 244	93.6	92.3
Mycobacterium phage porky	89.4	89.0
Mycobacterium phage Bask21	90.1	85.3
Mycobacterium phage sirduracell	87.9	84.2
Mycobacterium phage eureka	89.4	88.6
Mycobacterium phage kostya	87.9	87.8
Mycobacterium phage toto	90.1	90.0