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:	***2013.036a-dB***	(to be completed by ICTV officers)

Short title: To create a new genus *Sfi21dtunalikevirus* within the family *Siphoviridae*
(e.g. 6 new species in the genus *Zetavirus*)

Modules attached (modules 1 and 9 are required)	**1** ☒	**2** ☒	**3** ☒	**4** ☐	**5** ☐
	6 ☐	**7** ☐	**8** ☐	**9** ☒	

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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-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	***2013.036aB***	(assigned by ICTV officers)

To create 5 new species within:

Genus:	***Sfi21dtunalikevirus*** **(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.

	GenBank sequence accession number(s) of reference isolate:
Streptococcus phage DT1	AF085222
Streptococcus phage Sfi19	AF115102
Streptococcus phage Sfi21	AF115103
Streptococcus phage Abc2	FJ236310
Streptococcus phage 7201	AF145054

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 9

BLASTN analyses reveal that these five *Streptococcus* phages are related and distinct from any other phage. We have chosen 95% DNA sequence identity as the criterion for demarcation of species.

MODULE 3: **NEW GENUS**

creating a new genus

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

Code	***2013.036bB***	(assigned by ICTV officers)

To create a new genus within:

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 family is specified, enter "**unassigned**" in the family box

naming a new genus

Code	***2013.036cB***	(assigned by ICTV officers)

To name the new genus: *Sfi21dtunalikevirus*

Assigning the type species and other species to a new genus

Code	***2013.036dB***	(assigned by ICTV officers)

To designate the following as the type species of the new genus

Streptococcus phage DT1

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

Phages belonging to this genus have small, isometric capsids, and long non-contractile tails. Dimension for these phage are a capsid of 60 nm and a noncontractile tail of 260 x 8 nm [1]. Phages share a comparable genome size and GC content, and a common packaging mechanism with *cos* sites [2–5] (Table 1).
We propose 40% shared proteins with the type phage for new phages to be included in the new genus, as calculated with CoreGenes [6–8].

Origin of the new genus name:

The grouping Sfi21-like was coined first, but phage DT1 was isolated and described first, leading to the name combining both phages [1,4,9];

Reasons to justify the choice of type species:

Streptococcus phage DT1 was the original isolate of this group (Moineau, personal communication)

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. Tremblay DM, Moineau S (1999) Complete genomic sequence of the lytic bacteriophage DT1 of *Streptococcus thermophilus*. Virology 255: 63–76. doi:10.1006/viro.1998.9525.

2. Lamothe G, Lévesque C, Bissonnette F, Cochu A, Vadeboncoeur C, et al. (2005) Characterization of the cro-ori region of the Streptococcus thermophilus virulent bacteriophage DT1. Appl Environ Microbiol 71: 1237–1246. doi:10.1128/AEM.71.3.1237-1246.2005.

3. Desiere F, Lucchini S, Brüssow H (1998) Evolution of *Streptococcus thermophilus* bacteriophage genomes by modular exchanges followed by point mutations and small deletions and insertions. Virology 241: 345–356. doi:10.1006/viro.1997.8959.

4. Le Marrec C, van Sinderen D, Walsh L, Stanley E, Vlegels E, et al. (1997) Two groups of bacteriophages infecting *Streptococcus thermophilus* can be distinguished on the basis of mode of packaging and genetic determinants for major structural proteins. Appl Environ Microbiol 63: 3246–3253.

5. Guglielmotti DM, Deveau H, Binetti AG, Reinheimer JA, Moineau S, et al. (2009) Genome analysis of two virulent *Streptococcus thermophilus* phages isolated in Argentina. Int J Food Microbiol 136: 101–109. doi:10.1016/j.ijfoodmicro.2009.09.005.

6. 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.

7. 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.

8. 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.

9. Proux C, van Sinderen D, Suarez J, Garcia P, Ladero V, et al. (2002) The dilemma of phage taxonomy illustrated by comparative genomics of Sfi21-like *Siphoviridae* in lactic acid bacteria. J Bacteriol 184: 6026–6036. doi:10.1128/JB.184.21.6026-6036.2002.

10. Darling AE, Mau B, Perna NT (2010) progressiveMauve: multiple genome alignment with gene gain, loss and rearrangement. PLoS One 5: e11147.

References:

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.

Outline of additional studies performed on this viral clade:

- **Complete transcriptional map of phage DT1 using microarray (Duplessis et al., 2005)**
- **Multiplex PCR for the detection of the two groups of *S. thermophilus* phages (Quiberoni et al., 2006)**
- **Characterisation of the CRISPR-Cas system with phage DT1 (Deveau et al., 2008)**
- **Core genome of *S. thermophilus* bacteriophages, DT1 was used as a model phage for the cos-type group (Quiberoni et al., 2010)**
- **Identification of the receptor binding protein of phage DT1 (Duplessis & Moineau, 2001)**
- **Genomic comparison with other *S. thermophilus* phages (Levesque et al., 2005)**
- **Genomic comparison with other phages from different streptococcal species (Delisle et al., 2012)**

Table 1. Phage genomes belonging to the proposed genus

Phage	Accession No.	Size (bp)	Mol%G+C	Packaging – termini	% DNA sequence relatedness (a)	% Protein relatedness (b)
DT1	AF085222	34815	39	*cos* site	100	100
Sfi19	AF115102	37370	38	*cos* site	69.9	72.7
Sfi21	AF115103	40739	38	*cos* site	66.0	56.8
Abc2	FJ236310	34882	39	*cos* site	76.3	70.5
7201	AF145054	35466	39	*cos* site	Nd	68.2

(a) Calculated using EMBOSS Stretcher (relative to DT1)
(b) Calculated using CoreGenes 3.5 (relative to DT1)

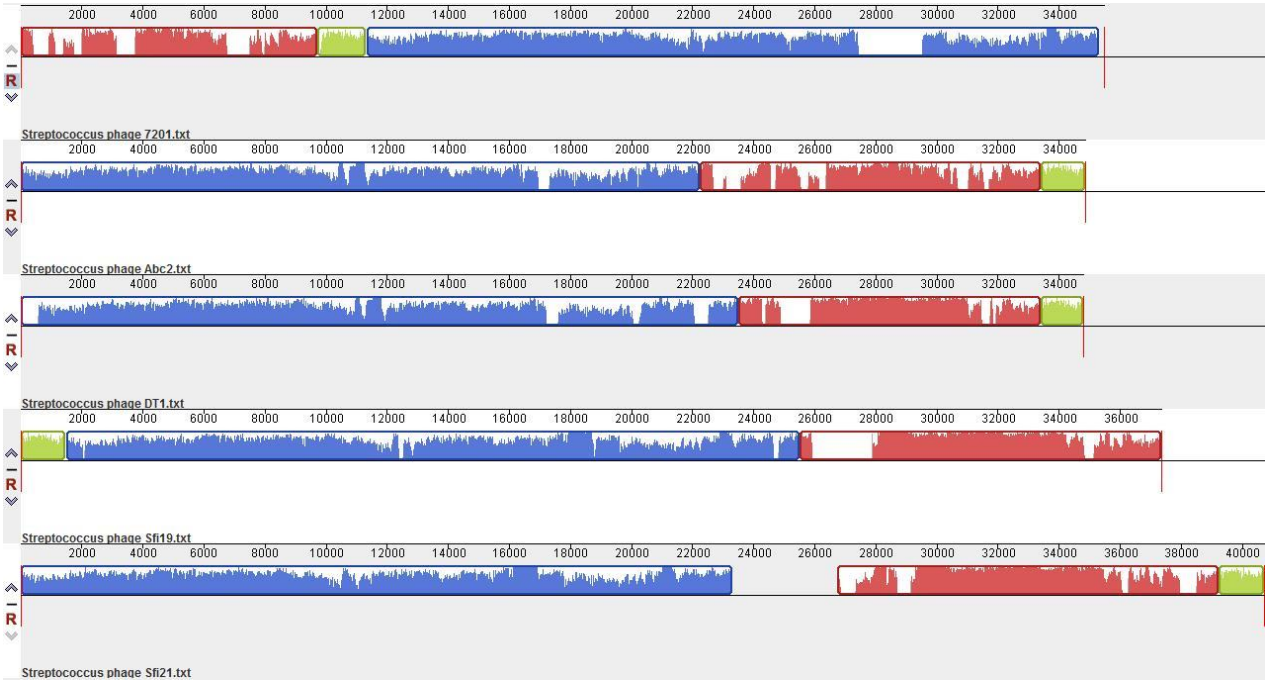

Figure 1. progressiveMauve alignment of the genomes of the these *Streptococcus* phages indicates that they are very similar [10]. The sequence of phage 7201 was deposited in GenBank with a different starting point from the others. Colored blocks indicate the regions of 1 to 1 best alignment with rearrangement 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.

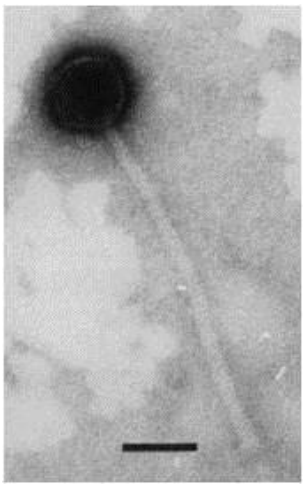

Figure 2. Electron microscopy photography of phage DT1 stain with uranyl acetate. Magnification 297,000x. Bar=50nm.

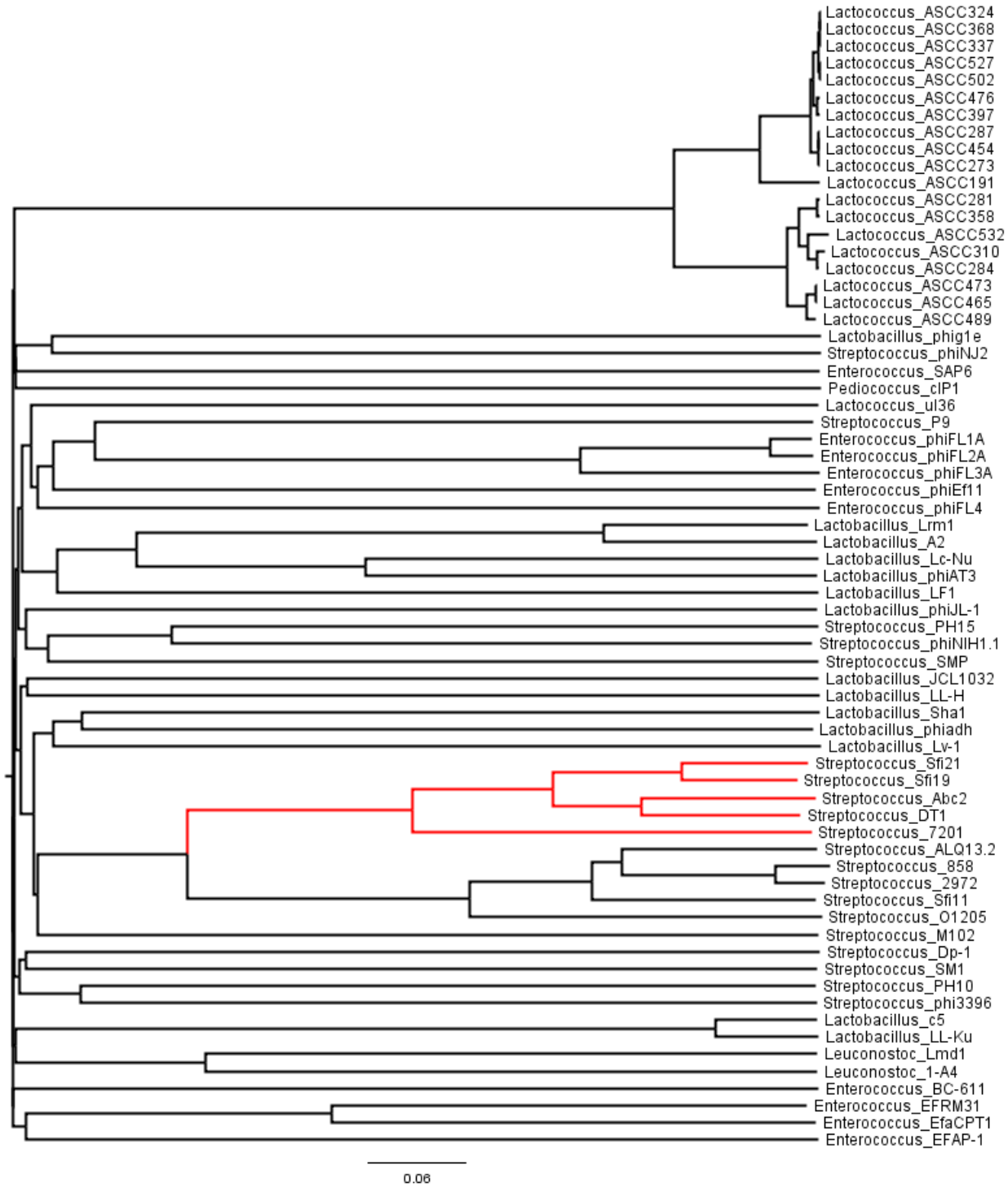

Figure 3: Whole genome phylogenetic tree of selected low GC content *Siphoviridae* phages in the NCBI database in November 2012. Genome sequences were aligned and the tree (NJ) was created with ClustalW 2.0 and visualized with FigTree. The newly proposed genus is colored in red.