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:	***2015.014aB***	(to be completed by ICTV officers)

Short title: To include four (4) new species within the genus *Phieco32virus* in the family *Podoviridae*.
(e.g. 6 new species in the genus *Zetavirus*)

Modules attached (modules 1 and 10 are required)	**1** ☒	**2** ☒	**3** ☐	**4** ☐	**5** ☐
	6 ☐	**7** ☐	**8** ☐	**9** ☐	**10** ☒

Author(s):

Andrew M. Kropinski – University of Guelph (Canada)
Anders S. Nilsson – The Wenner-Gren Institute Stockholm University (Sweden)
Hans-Wolfgang Ackermann – Laval University (Quebec)
Rob Lavigne – KU Leuven (Belgium)
Jens Kuhn – NIH (U.S.A.)
Evelien M. Adriaenssens – University of Pretoria (South Africa)

Corresponding author with e-mail address:

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 genus as *Phieco32virus* rather than *Phieco32likevirus*, since the Bacterial and Archaeal Virus Subcommittee of ICTV has voted overwhelmingly in favour of eliminating "*like*" from phage genus names.

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	***2015.014aB***	(assigned by ICTV officers)

To create 4 new species within:

Genus:	***Phieco32likevirus*** (proposed name ***Phieco32virus***)
Subfamily:	
Family:	***Podoviridae***
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.

Name of new species:	**Representative isolate: (only 1 per species please)**	**GenBank sequence accession number(s)**
Escherichia virus SU10	Escherichia phage vB_EcoP_SU10	KM044272
Escherichia virus NJ01	Escherichia phage NJ01	JX867715
Escherichia virus ECB2	Escherichia phage ECBP2	JX415536
Escherichia virus Septima11	Escherichia phage KBNP1711	KF981730

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

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.

BLASTN, CoreGenes, progressiveMauve (Fig. 2) and phylogenetic analyses (Fig. 1) all indicate that the genus, *Phieco32virus*, is cohesive and distinct from the other viral genera within the *Podoviridae*. The next closest related virus is *Salmonella* phage 7-11 (HM997019) which shares 6% DNA sequence identity.

The phages of this genus possess C3 morphology i.e. elongated capsids and a genome of ca. 77.1 kb (42.2 mol%G+C), and encode 122 proteins and 0-1 tRNAs; they share >61% DNA sequence identity and >69% protein homology (Table 1, Fig. 3.).

Please note that we have chosen to refer to this new genus as *Phieco32virus* rather than *Phieco32likevirus* since the Bacterial and Archaeal Virus Subcommittee of ICTV has voted overwhelmingly in favour of eliminating "*like*" from phage genus names.

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. CoreGenes3.5: 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. Mirzaei MK, Eriksson H, Kasuga K, Haggård-Ljungquist E, Nilsson AS. Genomic, proteomic, morphological, and phylogenetic analyses of vB_EcoP_SU10, a podoviridae phage with C3 morphology. PLoS One. 2014;9(12):e116294.

5. Li Y, Chen M, Tang F, Yao H, Lu C, Zhang W. Complete genome sequence of the novel lytic avian pathogenic coliphage NJ01. J Virol. 2012;86(24):13874-5.

6. Nho SW, Ha MA, Kim KS, Kim TH, Jang HB, Cha IS, Park SB, Kim YK, Jung TS. Complete genome sequence of the bacteriophages ECBP1 and ECBP2 isolated from two different *Escherichia coli* strains. J Virol. 2012;86(22):12439-40.

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.

Fig 1. Phylogenetic analysis of (top) major capsid proteins and (bottom) large subunit terminase proteins of phieco32viruses and their relatives 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."

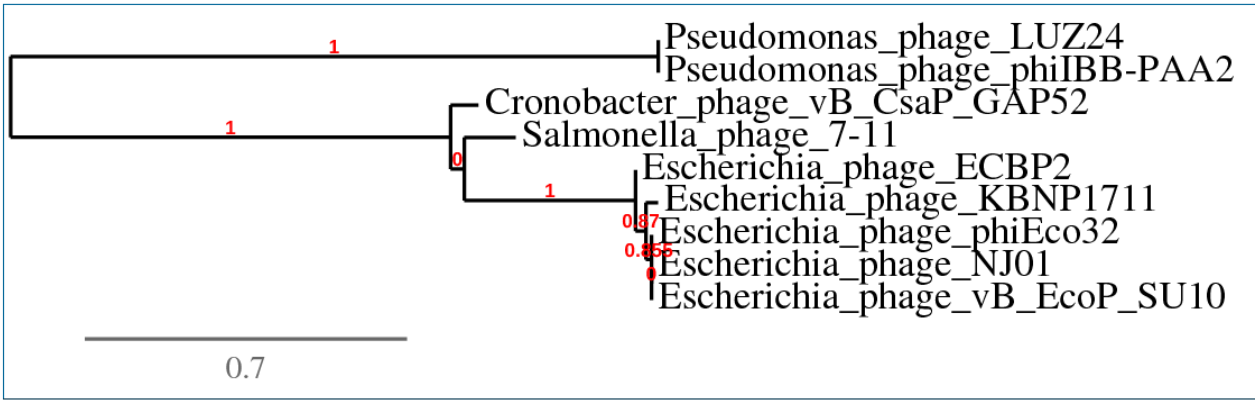

Figure 1: *Phylogenetic tree (the branch length is proportional to the number of substitutions per site).*

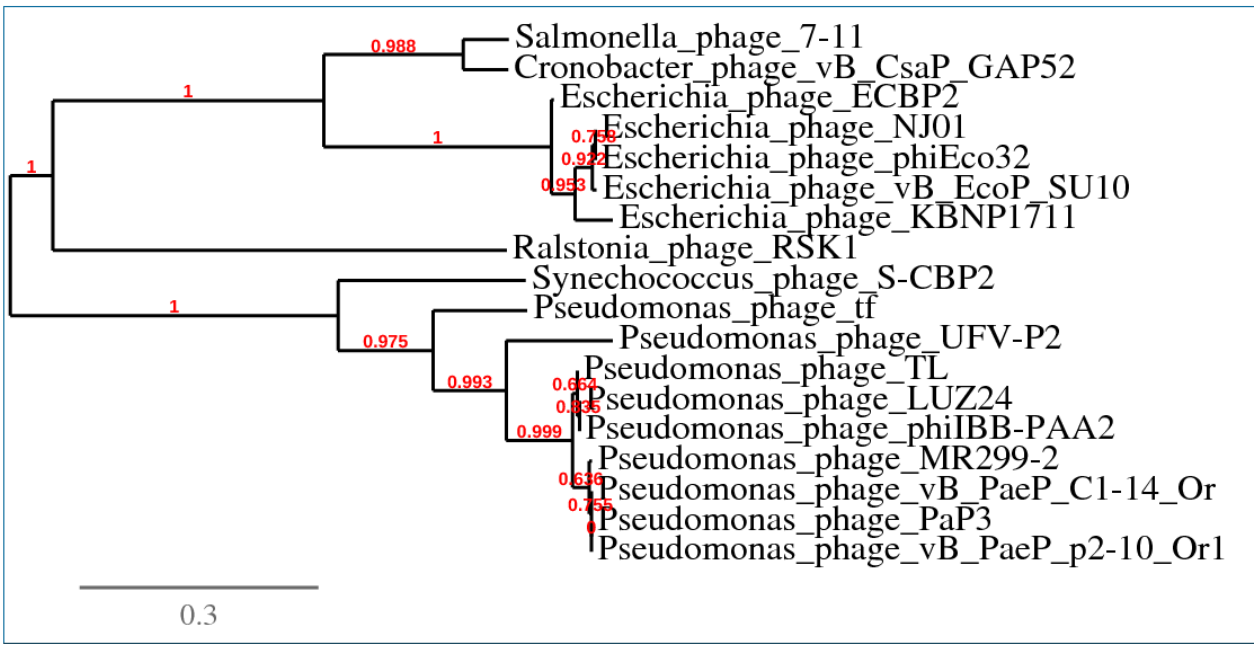

Figure 1: *Phylogenetic tree (the branch length is proportional to the number of substitutions per site).*

Table 1. Properties of the 5 phages belonging to the *Phieco32virus*, plus its closest relative *Salmonella* phage 7-11.

Phage	GenBank accession No.	Genome length (kb)	Genome (mol%G+C)	No. CDS	No. tRNAs	DNA (% sequence identity)*	Proteome (% homologous proteins)**
PhiEco32	EU330206	77.55	42.3	128	1	100	100
SU10	KM044272	77.33	42.1	125	0	91	89.1
NJ01	JX867715	77.45	42.0	109	0	86	68.8
ECBP2	JX415536	77.32	42.4	120	1	61	75.8
KBNP1711	KF981730	76.18	42.4	126	0	66	83.6
7-11	HM997019	89.92	44.1	151	6	6	32.0

* Determined using BLASTN; ** Determined using CoreGenes (2);

Fig. 2. progressiveMauve alignment of the annotated genomes of members of the *Phieco32virus* genus – from top to bottom: PhiEco32, HJ01, ECBP2, KBNP1711 and SU11 (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).

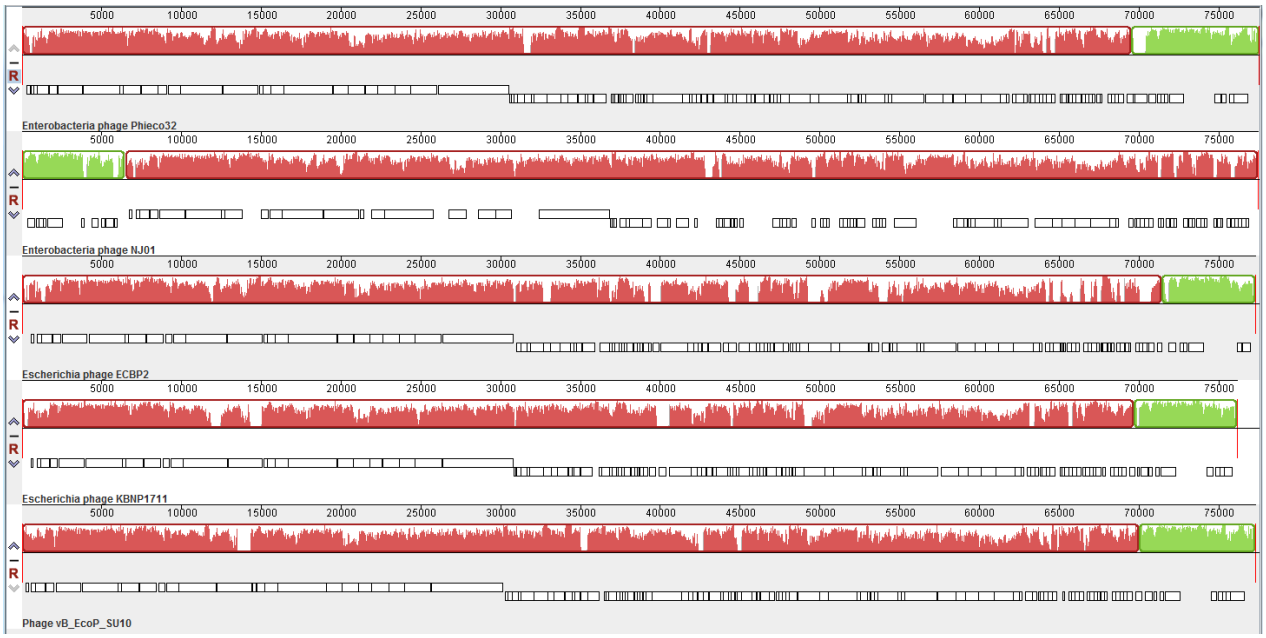

Fig. 3. Electron micrographs of SU10 (courtesy: Dr. Anders Nilsson). Left, a thin sectioning TEM and; right, a SEM.