

# Template for Taxonomic Proposal to the ICTV Executive Committee To create a new Genus in an existing Family

Code<sup>†</sup>  To create a new genus in the family\*

Code<sup>†</sup>  To name the new genus\*

Code<sup>†</sup>  To create as type species in the new genus the species named\*

Code<sup>†</sup>  To designate the following as species of the new genus\*:

Code<sup>†</sup>  To designate the following as tentative species in the new genus\*:

<sup>†</sup> Assigned by ICTV officers

\* repeat these lines and the corresponding arguments for each genus created in the family

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- Order
- Family
- Genus
- Type Species
- Species in the Genus
- Tentative Species in the Genus
- Unassigned Species in the family

## New Taxonomic Order

- Order
- Family
- Genus
- Type Species
- Species in the Genus
- Tentative Species in the Genus

## Unassigned Species in the family

### ICTV-EC comments and response of the SG

Accepted. Move to 04

### Argumentation to choose the type species in the genus

Phage phiZ is the best-known species of the genus and the first sequenced.

### Species demarcation criteria in the genus

Species differ in DNA homology, restriction endonuclease patterns, and N-terminal sequences of the major capsid protein. The DNA of phiKZ is over 280 kb in size and has 306 ORFs, most of which are unrelated to any known proteins, while EL contains 201 ORFs within its 211 kb genome. These two phages and *Pseudomonas* phage Lin68 have recently been proposed as part of a genus “phiKZ viruses”. We now consider that the differences (number of ORFs, mol%G+C, protein homologs) between phiKZ and EL exclude EL from membership in the same genus, although a relationship at a possible future subfamily level can be envisioned. Indeed, the recent analysis of novel *Pseudomonas* phage 201phi2-1 showed this phage to have a strong correlation to phiKZ (167 similar proteins), showing that it is a true member of the “phiKZ-like viruses” (Figures 2 and 3).

### List of Species in the created genus

*Pseudomonas* phage phiKZ  
*Pseudomonas* phage Lin68  
*Pseudomonas* phage 201phi2-1

**List of Tentative Species in the created genus**

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## Argumentation to create a new genus:

Virions are very large (heads 120 nm in diameter, tails 184 nm long) and have tails surrounded by fibers. Heads contain a proteinic inner body as shown for 201phi2-1 in Figure 1.

## Origin of the proposed genus name

Name of type species

## References

- Ackermann H-W, Cartier C, Slopek S, Vieu J-F. 1988. Morphology of *Pseudomonas aeruginosa* typing phages of the Lindberg set. *Ann Inst Pasteur/Virol* 139:389-404
- Bourkaltseva MV, Krylov VN, Pleteneva EA, Shaburova OV, Krylov SV, Volkart G, Sykilinda NN, Kurochkina LP, Mesyanzhinov VV. 2002. Phenogenetic characterization of a group of giant  $\phi$ KZ-like bacteriophages of *Pseudomonas aeruginosa*. *Genetika* 38:1470-1479; *Russ J Genet* 1242-1250
- Campbell JIA, Albrechtsen M, Sørensen J. 1995. Large *Pseudomonas* phages isolated from barley rhizosphere. *FEMS Microbiol Ecol* 18:63-74
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- Krylov VN, Dela Cruz DM, Hertveldt K, Ackermann H-W: "phiKZ-like viruses", a proposed new genus of myovirus bacteriophages. *Archives of Virology* 2007, 152: 1955-1959.
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- Serwer P, Hayes SJ, Zaman S, Lieman K, Rolando M, Hardies SC. 2004. Improved isolation of undersampled bacteriophages: finding of distant terminase genes. *Virology* 239:412-424
- Thomas JA, Rolando MR, Carroll CA, Shen PS, Belnap DM, Weintraub ST et al.: Characterization of *Pseudomonas chlororaphis* myovirus 201varphi2-1 via genomic sequencing, mass spectrometry, and electron microscopy. *Virology* 2008, 376: 330-338.

## Annexes:

# PhiKZ (original name $\phi$ KZ)

## Background

The *Myoviridae* family of tailed phages has 6 genera named after their type species: P1, P2, Mu, T4 (enterics), SPO1 (*Bacillus*), phiH1 (*Halobacterium*). These phages comprise no more than a small part of the known myoviruses and differ in every respect from phiKZ. In turn, phiKZ and its relatives do not resemble any other tailed phages (except very superficially in size).

## Proposals

1. To establish a new genus within the family *Myoviridae*.
2. To name this genus 'phiKZ-like viruses.'
3. To designate *Pseudomonas* phage phiK as the type species of this genus.
4. To designate *Pseudomonas* phage phiKZ as the type virus of this species.
5. To establish two more species, named Lin68 and 201phi2-1, within the genus.
6. To designate *Pseudomonas* phages Lin68 and 201phi2-1 as type viruses of these species.

# **GENUS**        "**phiKZ-LIKE VIRUSES**"

*Type Species*        *Pseudomonas phage phiKZ*

## **Distinguishing Features**

Virions are very large and have tails surrounded by fibers. Heads contain a proteinic inner body and a genome of 280 kb that differs from any myovirus genome known.

## **Virion Properties**

### **Morphology**

Heads are icosahedra of 120 nm in diameter. Extended tails measure 184 x 20 nm and have a large collar, a base plate, and a fibrous network extending from the base plate and wrapped around the tail sheath. Heads contain a spring-like protein cylinder in addition to DNA.

### **Physicochemical and Physical Properties**

Infectivity is chloroform-resistant.

### **Nucleic Acid**

Genomes are ~280,000 bp in size, have a G+C content of 37%, and consist of 306 ORFs and 6 tRNAs. Unusual bases are absent. The genome of phage phiKZ has been fully sequenced. It contains many elements from other bacteriophages, eukaryote viruses, bacteria, and eukaryotic organisms.

### **Proteins**

Virions contain at least 40 proteins, five of which are strongly homologous collagen.

Lipids                      None known.

Carbohydrates None known.

## **Genome Organization and Replication**

Genomes are circularly permuted and terminally redundant and have a mosaic structure.

**Antigenic Properties        No group antigens are reported.**

## **Biological Properties**

Phages are virulent general transductants and specific for *Pseudomonas* bacteria. Most were isolated from soil. Plaques are extremely small (0.1 mm in diameter). Representatives have been found in many countries (USA, Russia, Georgia, Iran, Philippines).

## **List of Species Demarkation Criteria in the Genus**

Species differ in DNA homology, restriction endonuclease patterns, and N-terminal sequences of the major capsid protein. Species LU11 has spiral fibers laying along the tail shaft and particles sometimes present a double collar.

### **List of Species in the Genus**

Official virus species names are in italics. Tentative virus species names, alternative names (), strains, or serotypes are not italicized. Virus names, genome sequence accession numbers [], and assigned abbreviations are:

#### SPECIES IN THE GENUS

<i>Pseudomonas phage phiKZ</i>		
Pseudomonas phage phiKZ	[AF399001]	(phiKZ)
Pseudomonas phage Lin21		
Pseudomonas phage NN		
Pseudomonas phage PTB80		
<i>Pseudomonas phage Lin68</i>		
Pseudomonas phage Lin68		(Lin68)
Pseudomonas phage LBG22		
<i>Pseudomonas phage 201phi2-1</i>		
Pseudomonas phage 201phi2-1	[AJ697969]	(EL)

Similarity to Other Taxa            **Not reported.**

#### References

- Ackermann H-W, Cartier C, Slopek S, Vieu J-F. 1988. Morphology of *Pseudomonas aeruginosa* typing phages of the Lindberg set. *Ann Inst Pasteur/Virol* 139:389-404
- Bourkaltseva MV, Krylov VN, Pleteneva EA, Shaburova OV, Krylov SV, Volkart G, Sykilinda NN, Kurochkina LP, Mesyanzhinov VV. 2002. Phenogenetic characterization of a group of giant  $\phi$ KZ-like bacteriophages of *Pseudomonas aeruginosa*. *Genetika* 38:1470-1479; *Russ J Genet* 1242–1250
- Campbell JIA, Albrechtsen M, Sørensen J. 1995. Large *Pseudomonas* phages isolated from barley rhizosphere. *FEMS Microbial Ecol* 18:63-74
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- Krylov VN, Smirnova TA, Minenkova IB, Plotnikova TG, Zhazikov IZ, Khrenova EA. 1984. *Pseudomonas* bacteriophage  $\phi$ KZ contains an inner body in its capsid. *Can J Microbiol* 30:758-762
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- Serwer P, Hayes SJ, Zaman S, Lieman K, Rolando M, Hardies SC. 2004. Improved isolation of undersampled bacteriophages: finding of distant terminase genes. *Virology* 239:412-424

## COMMENTS

1. A G+C content of 48% that was reported in early work (1978 and 1980). This has been superseded by sequence data and is not cited.
2. I do not mention  $\phi$ KZ-like phages that were mentioned somewhere in the literature and then disappeared ( $\phi$ NZ, PMN17, see refs. Krylov et al., 1993; Sharibjanova, 1992; Sharibjanova et al., 1992) or which are as yet unpublished (phage 1615 or Ph-1615; Shaburova et al.).
3. The group was already called a genus (Bourkaltseva et al., 2002; Krylov et al., 2003)

## SUPPORTING MATERIAL

### Dimensions

References	Phage	Head, nm	Tail, nm
Ackermann 1988	21, 68	124	184 x 19
Krylov 1984	$\phi$ iKZ	120	180 x 20
Krylov 1978	$\phi$ iKZ	120	180 x 20
Adamia 1999	$\phi$ iST-1	140	240 x 30
Campbell 1995	Psp3	130	197
Serwer	201 $\phi$ i2-1	140	200 (Figure 1)



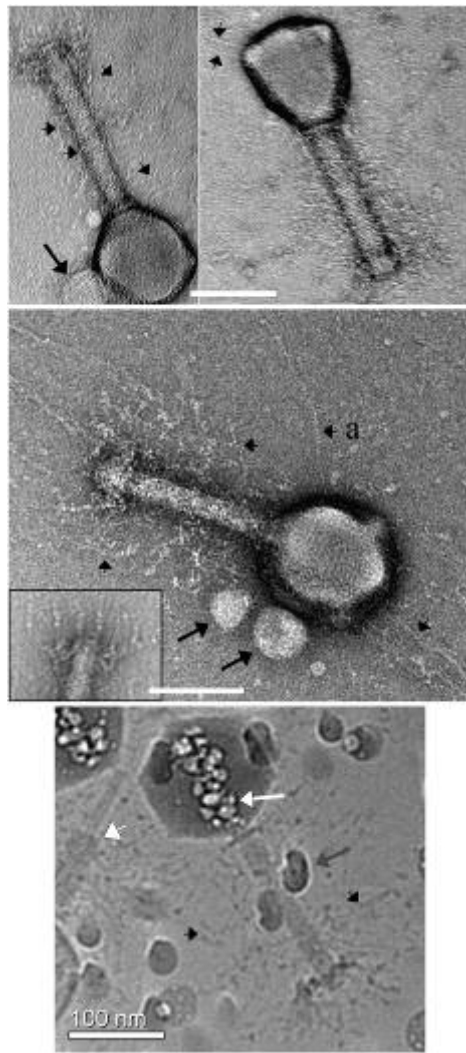


Figure 1: Electron micrographs of 201phi2-1. Black short tailed arrows in all panels highlight several fine fibers projecting from the tail sheaths or heads of 2012-1 virions. (Top panel) Two particles of purified 2012-1 that were treated with DNAase immediately prior to being negatively stained with uranyl acetate. (Middle panel) Negatively stained 2012-1 without prior DNAase treatment. The arrow marked “a” indicates a possible strand of DNA. The inset image highlights a well stained baseplate from a virion. (Bottom panel) Cryo-EM image of 2012-1, without DNAase treatment. The white short tailed arrow highlights a particle (at the image edge) that has a contracted tail sheath and an empty head. The large white arrow highlights gaseous bubbles which formed in the head despite the use of low-dose techniques (see text). In the top and middle rows, large black arrows highlight particles that co-purified with 2012-1 virions. In the bottom row, the large gray arrow highlights contamination on the cryo-EM specimen. All three scale bars = 100 nm.



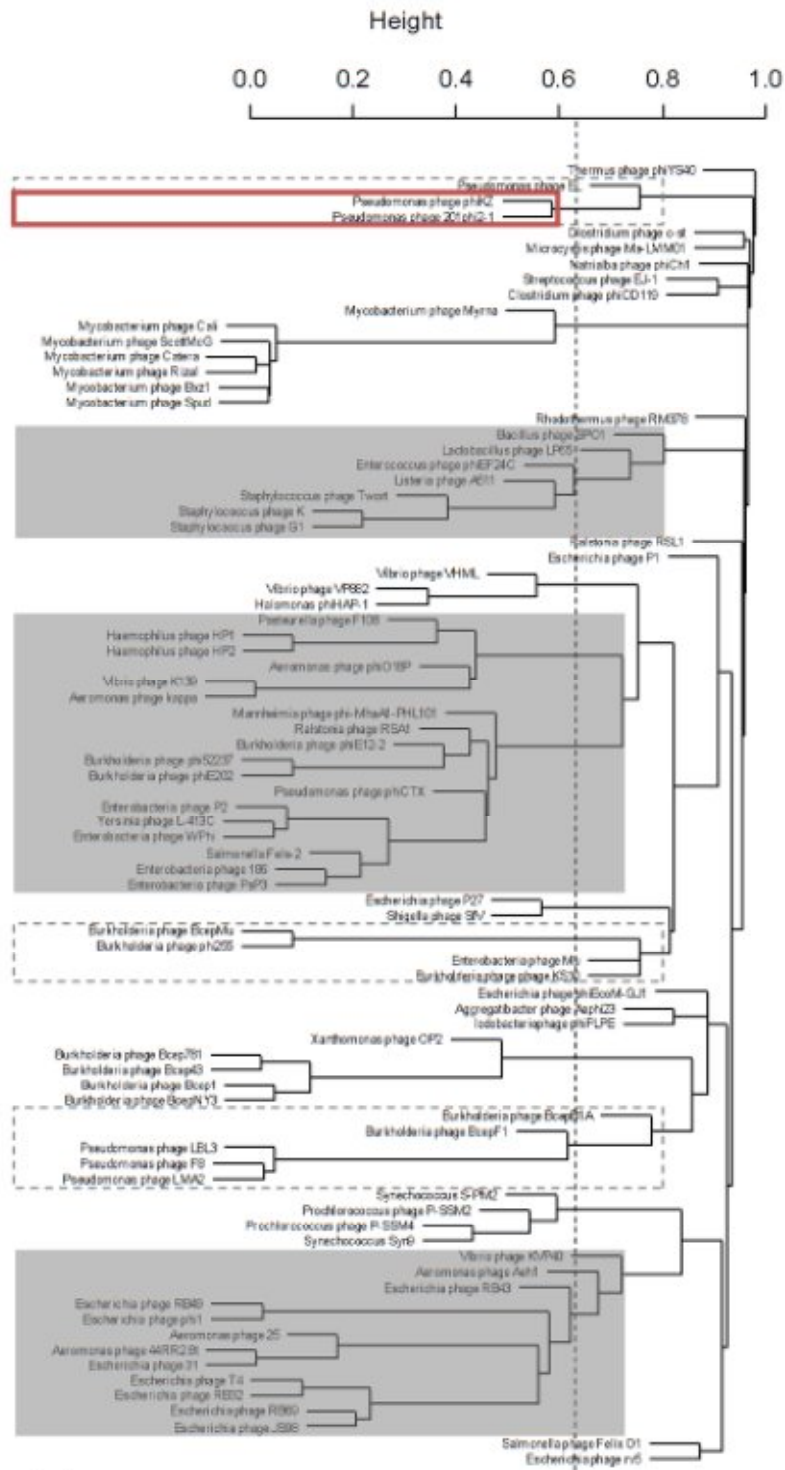


Figure 3: The relative dissimilarity between the phage proteomes (between 0.0 and 1.0) forms the basis for the proposed groupings. The dotted lines reflects the cut-off value used for the establishment of genera, used consistently for all Myoviridae and the previously defined Podoviridae (Lavigne et al., 2008). Subfamily and tentative subfamily groupings are indicated in the grey and dotted boxes, respectively. The “phiKZ-like viruses” are marked in red. Note the ‘proteomic divergence’ which excludes phage EL from this genus.

## References not to be cited in the Report:

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*Myoviridae*. In: Classification and Nomenclature of Viruses. Seventh Report of the International Committee on Taxonomy of Viruses, 69-84. Eds. MHV van Regenmortel, CM Fauquet, DHL Bishop, E Carstens, MK Estes, S Lemon, J Maniloff, MA Mayo, DJ McGeoch, CR Pringle, R Wickner. Academic Press, San Diego, CA

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Krylov VN, Bourkaltseva MV, Sykilinda NN, Pleteneva EA, Shaburova OV, Kadykov VA, Miller S, Biebl M. 2004.

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Investigation of *Pseudomonas aeruginosa* bacteriophages in view of their classification and the establishment of evolutionary relationships. Thesis, Moscow, Institute of Genetics and Selection of Industrial Microorganisms, Moscow

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A comparative study of DNA homology and morphology of *Pseudomonas aeruginosa* bacteriophages to reveal phylogenetic relationships. Genetika 28:24-32

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Some physicochemical properties of bacteriophage  $\phi$ KZ. Mol Biol (SSSR) 14:802-805 (transl.)