This form should be used for all taxonomic proposals. Please complete all those modules that are applicable (and then delete the unwanted sections).

Code(s) assigned:	***2008.042-045BB***	(to be completed by ICTV officers)

Short title: create new genus named "LUZ24-like viruses" in the family Podoviridae
(e.g. 6 new species in the genus *Zetavirus*; re-classification of the family *Zetaviridae* etc.)

Modules attached (please check all that apply): **1** ☐ **2** ☐ **3** ☐ **4** ☒ **5** ☐ **6** ☐ **7** ☐

Author(s) with e-mail address(es) of the proposer:

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ICTV-EC or Study Group comments and response of the proposer:

MODULE 4: **NEW GENUS**

(if more than one genus is to be created, please complete additional copies of this section)

Code	***2008.042B***	(assigned by ICTV officers)

To create a new genus assigned as follows:

Subfamily:	
Family:	***Podoviridae***
Order:	***Caudovirales***

Fill in all that apply. Ideally, a genus should be placed within a higher taxon, but if not put "unassigned" here.

Code	***2008.043B***	(assigned by ICTV officers)

To name the new genus "LUZ24-like viruses"

Code	***2008.044B***	(assigned by ICTV officers)

To assign the following as species in the new genus:

- *Pseudomonas phage PaP3*
- *Pseudomonas phage LUZ24*

Code	***2008.045B***	(assigned by ICTV officers)

To designate the following as the type species in the new genus:
Pseudomonas phage *LUZ24*

Argument to justify the creation of a new genus:

The proposed taxonomic classification is based on available proteomic data. Using developed programs (CoreExtractor & CoreGenes) and careful review of available literature data, phages can be grouped. These programs parse-out/quantify the relationship between two phages into a single correlation score (= the relative number of homologous proteins between two sequenced phages).

Analysis and biological interpretation of the molecular correlations among all tailed phages (*Caudovirales*) with known genome sequence, shows this approach supports the current ICTV classification and proves that horizontal gene transfer does not mask the evolutionary relationship between phages.
Using a cut-off score of 40% homologous proteins between two phages, phages cluster correctly within existing genera. Hence, using this parameter, new genera may be defined.

The virulent *Pseudomonas aeruginosa* bacteriophage LUZ24 (45,625 bp) was isolated from hospital sewage. It belongs to the family of the Podoviridae, and carries a bidirectionally transcribed dsDNA genome delineated by two direct terminal repeats of 184 bp. Phage LUZ24 encodes 68 proteins, 47 of which are arranged in rightward orientation while 21 aim leftward.

Argument to justify the creation of a new genus:

Pseudomonas aeruginosa bacteriophage PaP3 contains contains 45 503 bp with GC content of 52.1% and is described as a temperate phage.

The genome of LUZ24 displays an overall nucleotide identity of 71% to phage PaP3. Eight insertions/deletions are present and 88% of the encoded gene products are related between both phages (Figure 1). However, the cos site identified in PaP3 is not conserved in LUZ24, and no difference in restriction patterns could be observed if samples were heated to 80 °C prior to separation on a 1% agarose gel (data not shown). Moreover, only two tRNA genes (tRNAAsn and tRNAPro) are present at the right end of the LUZ24 genome, compared to four in PaP3. This suggests these phages should be considered as separate species within this genus. Since LUZ24 (and PaP3) diverged strongly from other currently investigated phages, only a limited number of protein functions could be predicted by similarity searches. Especially proteins presumed to be involved early in the infection process can differ considerably, even between very closely related phages.

It seems unlikely that the newly isolated phage LUZ24 has a temperate nature, despite being very homologous to PaP3. LUZ24 shares its bidirectional genome organization with a vast number of virulent phages like cyanophages P60, Pf-WMP3 and Pf-WMP4, roseophage SIO1 and vibriophage VpV262. The closely related gene map and a strikingly conserved order of structural genes suggest (very ancient) divergence from phages like P60, Pf-WMP4, SIO1 and VpV262.

Origin of the new genus name:

After the type species *Pseudomonas phage LUZ24*

Argument to justify the choice of type species:

Although PaP3 was the first phage sequenced, the recent genome paper of LUZ24 casts doubt on a number of physiological characteristics of PaP3 (e.g. lysogenic state).

Species demarcation criteria in the 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.

The presence of several unique genes in both phages; proteomic correlation

References:

** Ceyssens PJ, Hertveldt K, Ackermann HW, Noben JP, Demeke M, Volckaert G, Lavigne R. (2008) The intron-containing genome of the lytic *Pseudomonas* phage LUZ24 resembles the temperate phage PaP3. Virology. 1;377(2):233-8.
** Tan Y, Zhang K, Rao X, Jin X, Huang J, Zhu J, Chen Z, Hu X, Shen X, Wang L, Hu F. (2007) Whole genome sequencing of a novel temperate bacteriophage of *P. aeruginosa*: evidence of tRNA gene mediating integration of the phage genome into the host bacterial chromosome. Cell Microbiol. 9(2):479-91.

Annexes:

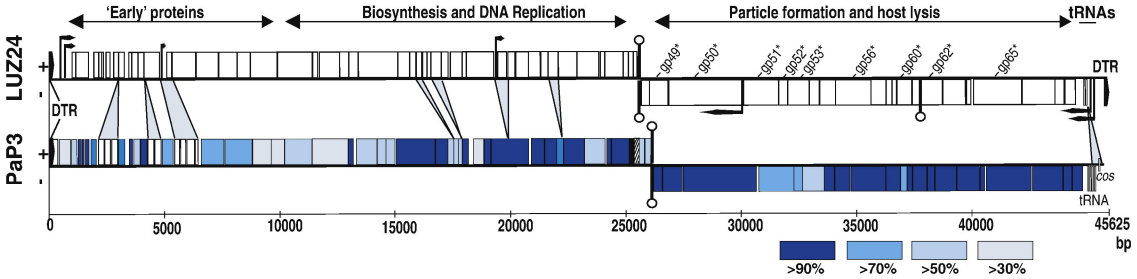

Figure 1: Side-by-side comparison of the LUZ24 and PaP3 genomes, marking the amino acid identities of the corresponding ORFs in different shades of blue. Insertions and deletions are indicated, as are the experimentally identified σ70 promoters (arrows with length according to the promoter strength) and rho-independent terminators (open circles). One copy of the LUZ24 DTR (black arrow) is also present in the genome of PaP3. The identified structural proteins are marked with asterisks, and the newly annotated PaP3 gene is hatched.