

**Part 1:** **TITLE, AUTHORS, APPROVALS, etc**

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| **Code assigned:** | **2021.038B** |  |
| **Short title:** Create one new subfamily (*Kantovirinae*) including two new genera (*Caudoviricetes*) | | |
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**Author(s) and email address(es)**

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| Andrew M. Kropinski |

**List the ICTV Study Group(s) that have seen this proposal**

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| Caudoviricetes Study Group, Bacterial Viruses Subcommittee |

**ICTV study group comments and response of proposer**

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**Authority to use the name of a living person**

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| **Is any taxon name used here derived from that of a living person (Y/N)** | N |

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| **Taxon name** | **Person from whom the name is derived** | **Permission attached (Y/N)** |
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**Submission dates**

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| --- | --- |
| Date first submitted to SC Chair | May 2021 |
| Date of this revision (if different to above) |  |

**ICTV-EC comments and response of the proposer**

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| The first proposed subfamily name was *Ibarakivirinae* which may be confused with Ibaraki virus in veterinary science. The name was changed to *Kantovirinae* in response.  Acceptance of proposal 2021.001B.abolish\_Caudovirales by EC53 results in removal of the order *Caudovirales* and families *Myoviridae*, *Podoviridae* and *Siphoviridae*. All underlying taxa are to be assigned directly to the class *Caudoviricetes*. The Excel module of this proposal has been altered to reflect the future changes; however, the Word module has been unaltered while awaiting the ratification vote. |

**Part 2:** **NON-TAXONOMIC PROPOSAL**

**Text of proposal**

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**Part 3:** **TAXONOMIC PROPOSAL**

**Name of accompanying Excel module**

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| 2021.038B.R.Kantovirinae |

**Abstract**

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| The genus *Naesvirus* currently consists of five species: *Burkholderia virus Bcep1, Burkholderia virus Bcep43, Burkholderia virus Bcep781, Burkholderia virus BcepNY3* and *Xanthomonas virus OP2*. Our proteomic (ViPTree), genomic (VIRIDIC) and phylogenetic analyses (phylogeny.fr) indicate that the Burkholderia phages are readily distinguishable from the Xanthomonas viruses, and that the genus *Naesvirus* should be restricted to the former viruses, while two new genera *Tsukubavirus* and *Beograduvirus* (Subfamily: *Kantovirinae*) constituted for the Xanthomonas phages. These two genera all fit within a single subfamily the members of which possess, on average, genomes of 46.6 kb (61.8 mol%G+C) and encode for 69 proteins and 0 tRNA. They share ca. 45% DNA sequence similarity. Using CoreGenes 5.0 (https://coregenes.ngrok.io/) revealed 40 homologs shared by this group of phages (48%). These homologs included: large terminase subunit, DNA helicase and Holliday junction resolvase / crossover junction. At this time we have chosen not to classify Xanthomonas phage XPV2 which VIRIDIC analysis reveals is closely related to XPV1, but phylogenetic analysis reveals may be distinct. |

**Text of proposal**

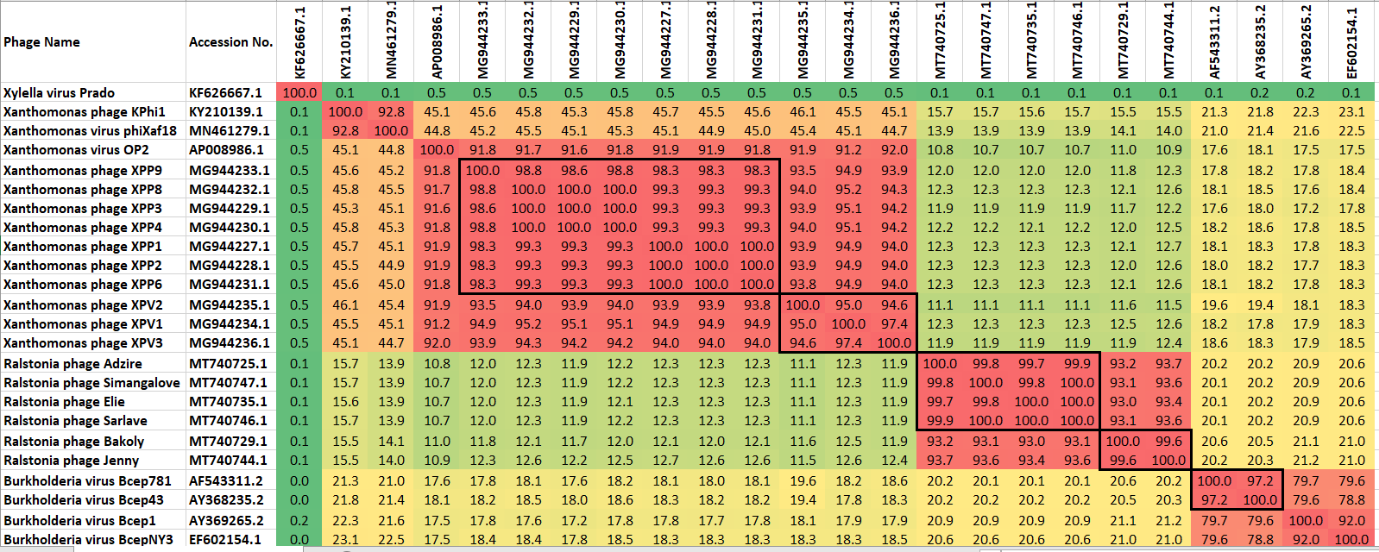
|  |  |
| --- | --- |
| |  | | --- | | **Species demarcation criteria:** Two phages are assigned to the same species if their genomes are more than 95% identical over their genome length for isolates.  These values can be calculated by a number of tools, such as BLASTn – usually calculated using intergenomic distance calculator VIRIDIC [3].  **Genus demarcation criteria:** In search for criteria that create cohesive and distinct genera that are reproducible and monophyletic, the Bacterial Viruses Subcommittee has established 70% nucleotide identity of the genome length as the cut-off for genera. Genus-level groupings should always be monophyletic in the signature genes, as tested with a phylogenetic tree.  **Subfamily demarcation criteria:** Subfamilies are to be created when two or more genera are related below the family level. In practical terms, this usually means that they share a low degree of sequence similarity (here minimum 45%) and that the genera form a clade in a marker tree phylogeny. [9] | |

**Supporting evidence**

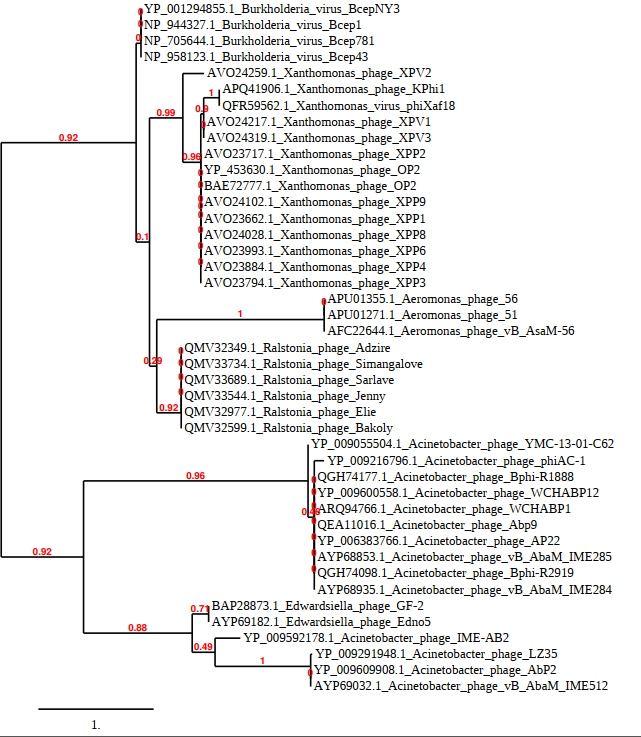
**ViPTree analysis:** ViPTree analysis ([https://www.genome.jp/viptree/](about:blank); [1]) is based upon Rohwer and Edwards (2002) famous Phage Proteomic Tree [2]. The phages of interest are indicated with a **blue bar**.

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**VIRIDIC heat map:** VIRIDIC (Virus Intergenomic Distance Calculator; [3]; [http://rhea.icbm.uni-oldenburg.de/VIRIDIC/](about:blank)) computes pairwise intergenomic distances/similarities amongst phage genomes. Values in black boxes correspond to strains.



**Phylogeny:** The phylogenetic tree was constructed using the terminase large subunit proteins of thesephages with phylogeny.fr in “one click” mode [5]. "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 [6] for details."

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**Proposals:**

1. **To create a new genus, *Tsukubavirus* with three species**
2. **To create a new genus, *Beograduvirus* with two species**
3. **To create a new subfamily *Kantovirinae***

**To create a new genus, *Tsukubavirus* with three species**

**Source of the name of this taxon:** Xanthomonas oryzae phage OP2 was isolated at the National Agricultural Research Center, Tsukuba, Ibaraki, Japan. Tsukuba is a city in Japan’s Kantō region, on Honshu island.

**History:** OP2 is a lytic Xanthomonas oryzae phage. Xanthomonas phage XPP1 and Xanthomonas phage XPV1 were isolated by Enviroinvest Corp., Pecs, Baranya 7632, Hungary. Though OP2 is reported to have 71 bp terminal repeats, these have not been found for XPP1 or XPV1 [Kovács T et al. 2019]

**Electron micrograph:** None available

**Specific Reference:** Inoue,Y., Matsuura,T., Ohara,T. and Azegami,K. Sequence analysis of the genome of OP2, a lytic bacteriophage of Xanthomonas oryzae pv. oryzae. J. Gen. Plant Pathol. 72, 104-110 (2006)

Kovács T, Molnár J, Varga I, Nagy IK, Valappil SK, Papp S, Vera Cruz CM, Oliva R, Vizi T, Schneider G, Rákhely G. Complete Genome Sequences of 10 Xanthomonas oryzae pv. oryzae Bacteriophages. Microbiol Resour Announc. 2019 Jul 3;8(27):e00334-19. doi: 10.1128/MRA.00334-19. PMID: 31270189; PMCID: PMC6606903.

**GenBank Summary:**

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| --- | --- | --- | --- | --- | --- | --- | --- |
| Phage name | INSDC | Size (Kb) | GC% | Protein | tRNA | Overall DNA sequence identity (\*) | % common proteins (\*\*) |
| Xanthomonas phage OP2 | [AP008986.1](about:blank) | 46.64 | 60.9 | [62](about:blank) | 0 | 100 | 100 |
| Xanthomonas phage XPP1 | [MG944227.1](about:blank) | 46.2 | 61.0 | [73](about:blank) | 0 | 91.9 | 96.8 |
| Xanthomonas phage XPV1 | [MG944234.1](about:blank) | 46.5 | 61.1 | [77](about:blank) | 0 | 91.2 | 96.8 |

**(\*) Determined using VIRIDIC [3]**

**(\*\*) Determined using CoreGenes 3.5 at** [**http://binf.gmu.edu:8080/CoreGenes3.5/**](about:blank) **[4]**

**To create a new genus, *Beograduvirus* with two species**

**Source of the name of this taxon:** This taxon is named in honour of Belgrade, Serbia where at the Institute for Plant Protection and Environment the first virus of its type, Xanthomonas phage KPhi1 was isolated in 2018.

**History:** Lytic Xanthomonas euvesicatoria phage KPhi1 was isolated from pepper rhizosphere soil in Serbia, while Xanthomonas vesicatoria phage phiXaf18 was isolated from Mexican soil.

**Electron micrograph:**. None available

**Specific Reference:** NA

**GenBank Summary:**

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| --- | --- | --- | --- | --- | --- | --- | --- |
| Phage name | INSDC | Size (Kb) | GC% | Protein | tRNA | Overall DNA sequence identity (\*) | % common proteins (\*\*) |
| Xanthomonas phage KPhi1 | [KY210139.1](about:blank) | 46.08 | 62.8 | [66](about:blank) | 0 | 100 | 100 |
| Xanthomonas virus phiXaf18 | [MN461279.1](about:blank) | 47.41 | 63.0 | [67](about:blank) | 0 | 92.8 | 89.4 |

**(\*) Determined using VIRIDIC [3]**

**(\*\*) Determined using CoreGenes 3.5 at** [**http://binf.gmu.edu:8080/CoreGenes3.5/**](about:blank) **[4]**

**To create a new subfamily *Kantovirinae***

**Source of the name of this taxon: “**The Kanto region is a geographical area of Honshu, the largest island of Japan. In a common definition, the region includes the Greater Tokyo Area and encompasses seven prefectures: Gunma, Tochigi, Ibaraki, Saitama, Tokyo, Chiba and Kanagawa.” (Wikipedia)

**Rationale:**

These two genera all fit within a single subfamily the members of which possess, on average, genomes of 46.6 kb (61.8 mol%G+C) and encode for 69 proteins and 0 tRNA. They share ca. 45% DNA sequence similarity. Using CoreGenes 5.0 ([https://coregenes.ngrok.io/](about:blank)) revealed 40 homologs shared by this group of phages (48%). These homologs included: large terminase subunit, DNA helicase and Holliday junction resolvase / crossover junction.

**References:**

1: Nishimura Y, Yoshida T, Kuronishi M, Uehara H, Ogata H, Goto S. ViPTree: the viral proteomic tree server. Bioinformatics. 2017; 33(15):2379-2380. doi:10.1093/bioinformatics/btx157. PubMed PMID: 28379287.

2: Rohwer F, Edwards R. The Phage Proteomic Tree: a genome-based taxonomy for phage. J Bacteriol. 2002 Aug;184(16):4529-35. PubMed PMID: 12142423

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7: Lowe, T.M. and Chan, P.P. (2016) tRNAscan-SE On-line: Search and Contextual Analysis of Transfer RNA Genes. Nucl. Acids Res. 44: W54-57.

8: Zimmermann L, Stephens A, Nam SZ, et al. A Completely Reimplemented MPI Bioinformatics Toolkit with a New HHpred Server at its Core. J Mol Biol. 2018;430(15):2237-2243. doi:10.1016/j.jmb.2017.12.007

9: Turner D, Kropinski AM, Adriaenssens EM. 2021. A Roadmap for Genome-Based Phage Taxonomy. Viruses 2021, 13, 506. https://doi.org/10.3390/v13030506