This Word module should be used for all taxonomic proposals.

Please complete **Part 1** and:

either **Part 3** for proposals to create new taxa or change existing taxa

or **Part 2** for proposals of a general nature.

Submit the completed Word module, together with the accompanying Excel module named in Part 3, to the appropriate ICTV Subcommittee Chair.

The Word module explains and justifies your proposal. The Excel module is a critical document that will be used to implement the proposed taxonomic changes once they are approved and ratified. If proposals presented in the Word module are not presented accurately in the Excel module, the taxonomic changes cannot proceed.

For guidance, see the notes written in blue, below, and the Help Notes in file Taxonomic\_Proposals\_Help\_2019.

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

|  |  |  |  |
| --- | --- | --- | --- |
| **Code assigned:** | ***2019.034B*** | |  |
| **Short title:** Create one new genus (*Squashvirus*) including two new species in the family *Siphoviridae* | | | |
|  | | | |
| **Author(s) and email address(es):** | | | |
| List authors in a single line *Archives of Virology* citation format (e.g. Smith AB, Huang C-L, Santos, F) | | Provide email address for each author in a single line separated by semi-colons | |
| Kropinski AM, Adriaenssens EM | | [Phage.Canada@gmail.com](mailto:Phage.Canada@gmail.com); [Evelien.Adriaenssens@](mailto:Evelien.Adriaenssens@)quadram.ac.uk | |
| **Author(s) institutional address(es) (optional):**   |  | | --- | | Provide institutional addresses, each on a single line followed by author(s) initials (e.g. University of Woolloomooloo [SAB, HCL]) | | University of Guelph, Canada [AMK]  Quadram Institute Bioscience, UK [EMA] | | | | |
| **Corresponding author** | | | |
| Andrew M. Kropinski | | | |
| **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 (there are six virus subcommittees: animal DNA and retroviruses, animal ssRNA-, animal ssRNA+, fungal and protist, plant, bacterial and archaeal) | | **Bacterial and Archaeal Viruses Subcommittee**  **Caudovirales Study Group** | |
| **ICTV Study Group comments (if any) and response of the proposer:** | | | |
|  | | | |
|  | | | |
| Date first submitted to ICTV: | | |  |
| Date of this revision (if different to above): | | |  |

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

**Part 2:** **NON-STANDARD**

Template for any proposal regarding ICTV procedures, rules or policy, not involving the creation of new taxonomy.

| **Text of proposal:** |
| --- |
|  |

**Part 3:** **PROPOSED TAXONOMY**

|  |
| --- |
| **Name of accompanying Excel module:** 2019.034B.A.v1.Squashvirus.xlsx |

The taxonomic changes you are proposing should be presented on an accompanying Excel module, 2019\_TP\_Template\_Excel\_module. Please enter the file name of the completed module in this box.

**Supporting material:**

| additional material in support of this proposal |
| --- |
| Please explain the reasons for the taxonomic changes you are proposing and provide evidence to support them. The following information should be provided, where relevant:   * **Species demarcation criteria**: Explain how new species differ from others in the genus and demonstrate that these differences meet the criteria previously established for demarcating between species. If no criteriahave previously been established, and if there will now be more than one species in the genus, please state the demarcation criteria you are proposing. * **Higher taxa**:   + There is no formal requirement to state demarcation criteria when proposing new genera or other higher taxa. However, a similar concept should apply in pursuit of a rational and consistent virus taxonomy.   + Please indicate the **origin of names** assigned to new taxa at genus level and above.   + For each new genus a **type species** must be designated to represent it. Please explain your choice. * **Supporting evidence**: The use of Figures and Tables is strongly recommended (note that copying from publications will require permission from the copyright holder). For phylogenetic analysis, please provide a tree where branch length is **proportional to genetic** distance, generated using an appropriate algorithm (Neighbour-Joining, Maximum Likelihood, or Bayesian) and provide evidence of the reliability of the branching (e.g., by bootstrapping).   Please refer to the Help Notes file (Taxonomic\_Proposals\_Help\_2019) for more information. |

| **References:** |
| --- |
| Brister, J. R., Ako-adjei, D., Bao, Y. & Blinkova, O. NCBI Viral Genomes Resource. Nucleic Acids Res. 43, D571–D577 (2015).  Sayers EW, Agarwala R, Bolton EE, Brister JR, Canese K, Clark K, et al. Database resources of the National Center for Biotechnology Information. Nucleic Acids Res. 2019;47(D1):D23-D28.  Tolstoy I, Kropinski AM, Brister JR. Bacteriophage Taxonomy: An Evolving Discipline. Methods Mol Biol. 2018;1693:57-71.  O'Leary NA, Wright MW, Brister JR, Ciufo S, Haddad D, McVeigh R, et al. Reference sequence (RefSeq) database at NCBI: current status, taxonomic expansion, and functional annotation. Nucleic Acids Res. 2016;44(D1):D733-45.  Agren J, Sundstrôm A, HÂfstrôm T, Segerman B. Gegenees: fragmented alignment of multiple genomes for determining phylogenomic distances and genetic signatures unique for specified target groups. PLoS One. 2012;7(6):e39107.  Chan PP, Lowe TM. tRNAscan-SE: Searching for tRNA Genes in Genomic Sequences. Methods Mol Biol. 2019;1962:1-14.  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.  Darling AE, Mau B, Perna NT. progressiveMauve: multiple genome alignment with gene gain, loss and rearrangement. PLoS One. 2010;5(6):e11147.  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.  Anisimova M, Gascuel O. Approximate likelihood-ratio test for branches: A fast, accurate, and powerful alternative. Syst Biol. 2006;55(4):539-52. |

**Species demarcation 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 BLASTN algorithm.

**Source of the name of this taxon:** The name of this genus is derived from the name of the second phage of its type, Microbacterium phage Squash.

**History:** Phage Squash was isolated in 2016 from using Microbacterium foliorum NRRL B-24224 SEA the host bacterium. It was isolated by Ann-Catherine Stanton (University of Pittsburgh, USA) as part of the Phage Hunters Integrating Research and Education Program. The genome of this lytic virus possesses 204 bp direct terminal repeats. It is classified in the Actinobacteriophage Database to Cluster EG (<https://phagesdb.org/phages/Squash/>).

**GenBank Summary:**

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Phage name | RefSeq No. | INSDC | Size (Kb) | GC% | Protein | tRNA | Overall DNA sequence identity (\*\*) | % common proteins (\*\*\*) |
| Microbacterium phage Squash |  | [MH153813.1](https://www.ncbi.nlm.nih.gov/nuccore/MH153813.1) | 62.31 | 66.8 | 108 | 0 | 100 | 100 |
| Microbacterium phage Hyperion |  | [MH153803.1](https://www.ncbi.nlm.nih.gov/nuccore/MH153803.1) | 61.77 | 67.0 | 104 | 0 | 81.5 | 84.3 |

**\*\* Determined using BLASTn at NCBI**

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

**BLASTN homologs:** The next most closely related phage is Microbacterium phage OneinaGillian which shares 38.4% DNA sequence relatedness. While this may constitute part of a subfamily we do not choose to propose one at this time.

**Electron micrograph:** Electron micrograph of negatively stained Microbacterium phage Squash. Limited permission was granted by The Actinobacteriophages Database, funded by the Howard Hughes Medical Institute, to use this electron micrograph for this taxonomy proposal; it cannot be reused without permission of The Actinobacteriophages Database.



**Phylogeny:** The phylogenetic tree was constructed using the large subunit terminase protein homologs of Squash and related phages with phylogeny.fr in “one click” mode. "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."

