

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

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| **Code assigned:** | ***2022.013B*** |  |
| **Short title:** Create one new genus (*Bocovirus*) including one new species (*Caudoviricetes*) | | |
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**Author(s) and email address(es)**

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

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| Bacterial Viruses Subcommittee  *Caudoviricetes* Study Group |

**ICTV Study Group comments and response of proposer**

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**ICTV Study Group votes on proposal**

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| **Study Group** | **Number of members** | | |
| **Votes support** | **Votes against** | **No vote** |
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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 2022 |
| Date of this revision (if different to above) |  |

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

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**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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| 2022.013B.N.v1.Bocovirus\_ng.xlsx |

**Abstract**

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| A novel podovirus vB\_SInP-X14, was isolated from surface coastal waters of Qingdao, China, which can infect a marine bacterium *Shewanella inventionis*. Here, we combined the results from phylogenetic analysis by VipTree and VICTOR, network analysis by vConTACT 2.0, and to propose a new genus *Bocovirus* with containing a single species, vB\_SInP-X14. |

**Text of proposal**

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| |  | | --- | | **Species demarcation criteria:** Two phages are assigned to the same species if theirgenomes are more than 95% identical over their genome length for isolates. These values can be calculated by a number of tools, such as BLASTn [1]–usually calculated using intergenomic distance calculator VIRIDIC [2].  **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. [3] | |

**Supporting evidence**

Shewanella phage vB\_SInP-X14 was isolated from surface coastal waters of Qingdao, China (36.063°N, 120.320°E), using Shewanella inventionisZQ14 as the host. Phage vB\_SInP-X14 encapsulates a double-strand DNA genome with a length of 36,515 bp, G+C content of 44.06%, and coding density of 89.9%. A total of 39 ORFs were predicted. The complete genome of vB\_SInP-X14 has been deposited in the GenBank database under the accession number MK796797.

Firstly, vB\_SInP-X14 was compared to other bacteriophage genomes using ViPTree (<https://www.genome.jp/viptree>, version 2.0). In the result, vB\_SInP-X14 clustered with other podoviruses, supported by the TEM micrograph (Figure 1).

After searching against the entire NCBI database, we found four homologous phages to vB\_SInP-X14, which were unclassified at genus level. Through searching against the IMG/VR database [4], we detected 14 non-redundant homologous UViGs closely related to vB\_SInP-X14. A total of 4406 reference genomic sequences of isolated *Caudoviricetes* were downloaded from NCBI-RefSeq, and combined with the above homologous sequences to vB\_SInP-X14 in NCBI and IMG/VR. All proteins were compared using all-verses-all DIAMOND BLASTp (Evalue ≤1E-10, coverage ≥50%, amino acid identity ≥30%). Then vConTACT 2.0 was used to calculate the similarity score between genomes. In the network diagram, vB\_SInP-X14 has linkages between six other isolated phages. Among them, vB\_SInP-X14, four isolated phages without genus-level taxonomy information (Thalassomonas phage BA3, Vibrio phage 1.183.O, Vibrio phage 1.184.A, Vibrio phage 1.211.A) and 14 UViGs from IMG/VR were classified into the same viral cluster using vConTACT2. Although Vibrio phage VvAW1 and Rhodoferax phage P26218 have linkage with vB\_SInP-X14, they were not classified in VC 0\_0 and were assigned as outliers (Figure 2B). According to the results, we suggest the members of VC 0\_0 could represent a new genus, named here as *Bocovirus*. In addition, the VC 0\_0 is a relatively independent cluster away from other defined viral genera (Figure 2A).

The whole-genome phylogenetic tree including 82 viral isolates was constructed by the Virus Classification and Tree Building Online Resource (VICTOR) [2]. vB\_SInP-X14 and its homologue sequences form a distinct clade in the VICTOR phylogenetic tree. The unclassified branch represented by vB\_SInP-X14 is distant from other classified viruses (Figure 4). According to the whole-genome phylogeny and OPTSIL classification, 20 phages and UViGs represented by vB\_SInP-X14 can be divided into the same subfamily, while 19 of them except Vibrio phage VvAW1 can be grouped into a novel genus [5]. In addition, the whole-genome Average Nucleotide Identity (ANI) was computed by fastANI [1], then visualized by pheatmap in R. The FastANI heatmap showed that the ANI values of the 19 phages and UViGs ranged from 76.39% to 91.26% (Figure 4), which is higher than the threshold ANI values (> 70%) for the classification of the same genus by the International Committee on Taxonomy of Viruses (ICTV) [3].

All analyses showed that the vB\_SInP-X14 is distinct from other bacteriophages and represents a new clade of bacterial viruses. Based on the above result, we proposed here a new bacteriophage genus, *Bocovirus* in the class *Caudoviricetes*.

 

**Figure 1**. Phylogenetic and comparative genomic analysis of vB\_SInP-X14. The left part was the phylogenetic tree of vB\_SInP-X14 and 1009 reference virus genomes. The coloured segments of the outer rings represents the virus family (inner ring) and host group (outer ring). The right part was the transmission electron micrograph (TEM) of Shewanella phage vB\_SInP-X14.

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**Figure 2** Gene-content-based viral network of Shewanella phage vB\_SInP-X14, including bacterial viruses from the NCBI-Ref database, and related UViGs from IMG/VR dataset. In order to highlight the key result, we only showed the nodes associated with a podovirus morphology classified by the ICTV and removed the singleton nodes. The network was visualized using Gephi version 0.9.2. (A)The Fruchterman Reingold layout (Gephi) explores the clustering status of the groups represented by vB\_SInP-X14 globally. (B) Using force atlas layout, the local network diagram in which the nodes have a direct linkage to vB\_SInP-X14 were shown. The edges represent the similarity scores between genomes based on shared gene content.

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**Figure 3** Whole-genome-based phylogenetic tree constructed by VICTOR with the formula d6. The phylogenic tree consists of 82 phage genomes, including 56 podoviruses classified by ICTV, with 1-2 representative strains selected from each genus. ICTV and OPTSIL clusters at the genus and family levels. Each genus is indicated by a unique color. vB\_SInP-X14 were shown in red. The bootstrap value of ≥50 were shown

**References**

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