

The International Committee on Taxonomy of Viruses

Taxonomy Proposal Form, 2024

**Part 1a: Details of taxonomy proposals**

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| **Title:** | Create one new family (Mycoalphaviridae) including two new genus (Alphasclernavirus, Betasclernavirus) and seven new species | |
| **Code assigned:** | 2024.007F.Uc.v2.Mycoalphaviridae\_newfam |

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**Part 1b: Taxonomy Proposal Submission**

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| **ICTV Subcommittee:** | | | |
| Animal DNA Viruses and Retroviruses |  | Bacterial viruses |  |
| Animal minus-strand and dsRNA viruses |  | Fungal and protist viruses | X |
| Animal positive-strand RNA viruses |  | Plant viruses |  |
| Archaeal viruses |  | General - |  |

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| **List the ICTV Study Group(s) that have seen or have been involved in creating this proposal:** |
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| **Optional – complete only if formally voted on by an ICTV Study Group:** | | | |
| **Study Group** | **Number of members** | | |
| **Votes in support** | **Votes against** | **No vote** |
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| **Submission date:** | DD/MM/YYYY |

**Part 1c: Feedback from ICTV Executive Committee (EC) meeting**

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| **Executive Committee Meeting Decision code:** | **X** |
| A – Accept |  |
| Ac – Accept subject to revision by relevant subcommittee chair. No further vote required |  |
| U – Accept without revision but with re-evaluation and email vote by the EC |  |
| Uc – Accept subject to revision and re-evaluation and email vote by the EC |  |
| Ud – Deferred to the next EC meeting, with an invitation to revise based on EC comments |  |
| J - Reject |  |
| W - Withdrawn |  |

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| **Comments from the Executive Committee:** |
| The EC found the proposal technically sound but with deficiencies in the naming the proposed taxa. Therefore, recommendations are to consider changing names of the proposed genera as they are complicated/difficult to pronounce. In addition, please notice that ICTV adopted binomial format for the species names. Please try to address these nomenclatural issues and resubmit the proposal. |

**Part 1d: Revised Taxonomy Proposal Submission**

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| **Response of proposer:** |
| We have changed the originally proposed nomenclature to address the EC feedback and adopted binomial species nomenclature. |

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| **Revision date:** | 18/10/2024 |

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

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| **Name of accompanying Excel module:** |
| 2024.007F.Uc.v2.Mycoalphaviridae\_newfam.xlsx |

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| **Taxonomic changes proposed:** | | | |
| Establish new taxon | **X** | Split taxon |  |
| Abolish taxon |  | Merge taxon |  |
| Move taxon |  | Promote taxon |  |
| Rename taxon |  | Demote taxon |  |
| Move and rename |  |

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| **Is any taxon name used here derived from that of a living person:** | | **Y/N** |
| **Taxon name** | **Person from whom the name is derived** | **Attached X** |
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| **Abstract of Taxonomy Proposal:** |
| *Taxonomic rank(s) affected*: *Hepelivirales*  *Description of current taxonomy*: The order including four families and twenty-seven species.  *Proposed* *taxonomic change(s):* Create one new family (Mycoalphaviridae) including two new genera (Alphasclernavirus, Betasclernavirus) and seven new species.  *Justification*: Members in the proposed family Mycoalphaviridae have a single-stranded positive-sense RNA genome ranging from 6.0 to 10.1 kb and encoding either one or more open reading frames. Members of the proposed family are only identified in fungi and oomycetes. The RNA-depended RNA polymerase of viruses in the family Mycoalphaviridae has the closest similarity to viruses of the order *Hepelivirales*, though the identity is lower than 20%. These low-level amino acid sequence identities, the different host ranges, and the result of phylogenetic analysis both support the establishment of the new family. The proposed family Mycoalphaviridae includes two proposed genera Alphasclernavirus and Betasclernavirus that accommodate three and seven species, respectively. The identity between genus and between species is lower than 26% and 50%, respectively, in the family. |

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| **Text of Taxonomy proposal:** |
| *Taxonomic rank(s) affected*: *Hepelivirales*  *Description of current taxonomy*: The order including four families and twenty-seven species.  *Proposed* *taxonomic change(s)*: Create one new family (Mycoalphaviridae) including two new genera (Alphasclernavirus, Betasclernavirus) and seven new species.  *Demarcation criteria:*  *Justification*: The proposed family Mycoalphaviridae includes two proposed genera Alphasclernavirus and Betasclernavirus ("sclernavirus" originates from " Sclerotinia sclerotiorum RNA virus L") which accommodate two and five species, respectively. The first member of the proposed family Mycoalphaviridae is Sclerotinia sclerotiorum RNA virus L (SsRV-L), identified in *Sclerotinia sclerotiorum* strain Ep-PN. SsRV-L is phylogenetically related to the human pathogen hepatitis E virus and rubi-like viruses in the order *Hepelivirales*. Many related viruses have been characterized in filamentous fungi and oomycetes, including *Rhizoctonia solani*, *Fusarium graminearum,* and *Botrytis cinerea [1, 6, 7, 9]*.  **Biological and molecular characterization:** The viruses in the proposed family Mycoalphaviridae have a single-stranded positive-sense RNA genome ranging from 6.0 to 10.1 kb. The complete genomes of mycoalphaviruses have one or more open reading frames with poly(A) tails at the 3’-end. According to the genome and polygenetic analysis, the proposed family Mycoalphaviridae was divided into two proposed genera Alphasclernavirus and Betasclernavirus. The alphasclernaviruses have one large ORF and encode the polyprotein, which includes three conserved domains: viral methyltransferase (Mtr), helicase (Hel), and RNA-depended RNA polymerase (RdRp) (Fig 1). On the other hand, the betasclernaviruses have two to five ORFs and the largest ORF encodes the polyprotein. Except for Fusarium graminearum alphavirus-like virus 1(FgALV1), other polyproteins of viruses in the proposed genus Betasclernavirus contain three conserved domains: Mtr, Hel and RdRp, which are similar to the alphasclernaviruses (Fig 1).  SsRV-L is the first well characterized mycoalphavirus and the proposed representative of the type species of the proposed genus Alphasclernavirus with RNA genome size of 6,043 nt in length (Fig 1). A putative large ORF was found, and it putatively encoded a 2,435 aa polyprotein. This polyprotein contains three conserved domains, including Mtr, Hel, and RdRp. And, SsRV-L only has a slight impact on growth and virulence of its host [4].  FgALV1 was the proposed representative of the type species of the genus Betasclernavirus. The complete genome of FgALV1 is 7,501 nt long and contains four putative ORFs (Fig 1). ORF 2 is the largest ORF and predicted to encode a 1,987 aa protein with a calculated molecular mass of 223.54 kDa and contain two conserved domain including Hel and an RdRp. No conserved domains were detected in the putative proteins encoded by ORF1, ORF3, and ORF4 [9].  **Phylogenetic analysis:** Phylogenetic analysis demonstrated the viruses in the proposed family Mycoalphaviridae were grouped together, and clustered with the members in the order *Hepelivirales* (Fig. 2). The phylogenetic analysis supported that these viruses have a far distance relationship to the known members of the order *Hepelivirales* and form an independent phylogenetic branch(Fig. 2).  **Justification for the creation of the new family, two genera and seven species:** All viruses in order *Hepelivirales* comprise at least two ORFs and encode coat protein. SsRV-L and other related viruses in fungi and oomycetes comprise one to five ORFs and encode the polyprotein but lack coat protein gene. Besides, different from the members in the order *Hepelivirales*, which infected vertebrates, invertebrates and plants, SsRV-L and other related viruses are only identified in fungi and oomycetes. Based on the unique genetic organization features and phylogenetic relationship of SsRV-L and other related viruses, we propose to establish a new family Mycoalphaviridae (from ‘myco’ = fungus in Greek + ‘alpha-like virus’) to accommodate these viruses.  The proposed family Mycoalphaviridae includes two proposed genera Alphasclernavirus and Betasclernavirus, which accommodate two and five species, respectively. According to the results of the pairwise distance matrix, the identity of RdRp amino acid sequence ≤ 50% was identified as a new species, the seventeen members in the proposed family Mycoalphaviridae represented seven different species, that infected *Sclerotium rolfsii*, *Botrytis cinerea, Fusarium graminearum, Fusarium sacchari,* and *Sclerotinia sclerotiorum* (Fig. 3).  **Etymology:**  Family: **Mycoalphaviridae**, from ‘myco’ = fungus in Greek + ‘alpha-like virus’ (denoting the host and type of genomic organization)  Genera: “…**sclernavirus**” derives from the name of a first identified virus of this group, Sclerotinia sclerotiorum RNA virus L  Information of the exemplars representing the seven proposed species in the family Mycoalphaviridae is listed in Table 1.   |  | | --- | | Realm: *Riboviria* | | Kingdom: *Orthornavirae* | | Phylum: *Kitrinoviricota* | | Class: *Alsuviricetes* | | Order: *Hepelivirales* | | Family: *Mycoalphaviridae* | | Genus: *Alphasclernavirus* | | Genus: *Betasclernavirus* |  |  |  |  | | --- | --- | --- | | **New species** | **Exemplar isolate** | **GenBank sequence accession number(s)** | | Genus: ***Alphasclernavirus*** | | | | *Alphasclernavirus alphasclerotiniae* | Sclerotinia sclerotiorum mycoalphavirus virus 1 | MT706025 | | *Alphasclernavirus betasclerotiniae* | Sclerotinia sclerotiorum RNA virus L | EU779934 | | Genus: ***Betasclernavirus*** | | | | *Betasclernavirus alphafusarii* | Fusarium graminearum alphavirus-like virus 1 | MN400076 | | *Betasclernavirus botrytidis* | Botrytis cinerea alpha-like virus 1 | MN625250 | | *Betasclernavirus betafusarii* | Fusarium sacchari alphavirus-like virus 1 | MN295968 | | *Betasclernavirus alphasclerotii* | Sclerotium rolfsii alphavirus-like virus 1 | MH766488 | | *Betasclernavirus betasclerotii* | Sclerotium rolfsii alphavirus-like virus 3 | MH766490 | |

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| **References:** |
| 1. Bartholomaus A, Wibberg D, Winkler A, Puhler A, Schluter A, Varrelmann M (2016) Deep sequencing analysis reveals the mycoviral diversity of the virome of an avirulent isolate of *Rhizoctonia solani* AG-2-2 IV. PLoS One 11:e0165965  2. Chiapello M, Rodríguez-Romero J, Ayllón MA, Turina M (2020) Analysis of the virome associated to grapevine downy mildew lesions reveals new mycovirus lineages. Virus Evol 6:veaa058  3. Gilbert KB, Holcomb EE, Allscheid RL, Carrington JC (2019) Hiding in plain sight: New virus genomes discovered via a systematic analysis of fungal public transcriptomes. PLoS One 14:e0219207  4. Liu HQ, Fu YP, Jiang DH, Li GQ, Xie J, Peng YL, Yi XH, Ghabrial SA (2009) A novel mycovirus that is related to the human pathogen Hepatitis E Virus and Rubi-like viruses. J Virol 83:1981-1991  5. Mu F, Li B, Cheng S, Jia J, Jiang D, Fu Y, Cheng J, Lin Y, Chen T, Xie J (2021) Nine viruses from eight lineages exhibiting new evolutionary modes that co-infect a hypovirulent phytopathogenic fungus. PLoS Pathog 17:e1009823  6. Picarelli M, Forgia M, Rivas EB, Nerva L, Chiapello M, Turina M, Colariccio A (2019) Extreme Diversity of Mycoviruses Present in Isolates of *Rhizoctonia solani* AG2-2 LP From Zoysia japonica From Brazil. Front Cell Infect Microbiol 9:244  7. Ruiz-Padilla A, Rodriguez-Romero J, Gomez-Cid I, Pacifico D, Ayllon MA (2021) Novel mycoviruses discovered in the mycovirome of a necrotrophic fungus. mBio 12:e03705-e03720  8. Yao Z, Zou C, Peng N, Zhu Y, Bao Y, Zhou Q, Wu Q, Chen B, Zhang M (2020) Virome Identification and Characterization of *Fusarium sacchari* and *F. andiyazi*: Causative Agents of Pokkah Boeng Disease in Sugarcane. Front Microbiol 11:240  9. Zhang X, Zhang H, Ma D, Chen H, Li W (2020) Novel positive-sense single-stranded RNA virus related to alphavirus-like viruses from *Fusarium graminearum*. Arch Virol 165:487-490  10. Zhu J, Zhu H, Gao B, Zhou Q, Zhong J (2018) Diverse, novel mycoviruses from the virome of a hypovirulent *Sclerotium rolfsii* strain. Front Plant Sci 9:1738 |

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| **Tables, Figures:** |

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Table1 proposed family *Mycoalphaviridae.*

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| **Genus** | **Species** | **Strain** | | | | | | **Genome** | | **Genome length**  **(nt)** | | **Accession number** | | **Reference** | |
| **Virus name** | **a** | **Acronym** | **Isolate** | **Host** |  | |  | |  | |  | |
| *Alphasclernavirus* | *Alphasclernavirus alphasclerotiniae* | Sclerotinia sclerotiorum mycoalphavirus virus 1 | ■ | SsMAV1 | SX276 | Fungi | complete | | 7586 | | MT706025 | | [5] | |
| *Alphasclernavirus betasclerotiniae* | Sclerotinia sclerotiorum RNA virus L | ■ | SsRV-L | Ep-1PN | Fungi | complete | | 6043 | | EU779934 | | [4] | |
|  | Rhizoctonia solani alphavirus-like 3 |  | RsALV3 | IBRS23 | Fungi |  | | 6752 | | [MK507786](https://www.ncbi.nlm.nih.gov/nuccore/MK507786.1) | | [6] | |
| Rhizoctonia solani RNA virus 3 |  | RsRV3 | DC17 | Fungi |  | | 924 | | [KX349066](https://www.ncbi.nlm.nih.gov/nuccore/KX349066.1) | | [1] | |
| Rhizoctonia solani RNA virus 2 |  | RsRV2 | DC17 | Fungi |  | | 924 | | [KX349067](https://www.ncbi.nlm.nih.gov/nuccore/KX349067.1) | | [1] | |
| Rhizoctonia solani RNA virus 1 |  | RsRV1 | DC17 | Fungi |  | | 924 | | [KX349068](https://www.ncbi.nlm.nih.gov/nuccore/KX349068.1) | | [1] | |
| Rhizoctonia solani alphavirus-like 2 |  | RsALV2 | IBRS23, IBRS11, IBRS19 | Fungi |  | | 3396 | | MK507792 | | [6] | |
| Rhizoctonia solani alphavirus-like virus 1 |  | RsALV1 | IBRS19 | Fungi |  | | 2414 | | [MK507793](https://www.ncbi.nlm.nih.gov/nuccore/MK507793.1) | | [6] | |
| Rhizoctonia solani alphavirus-like 4 |  | RsALV4 | 7388 | Fungi |  | | 7838 | | MW596323 | |  | |
| *Betasclernavirus* | *Betasclernavirus alphafusarii* | Fusarium graminearum alphavirus-like virus 1 | ■ | FgALV1 | CF16158 | Fungi | complete | | 7501 | | [MN400076](https://www.ncbi.nlm.nih.gov/nuccore/MN400076.1) | | [9] | |
| Plasmopara viticola lesion associated alpha-like virus 1 |  | PvLaAlpha-LV1 |  | Oomycetes |  | | 7412 | | [MN551115](https://www.ncbi.nlm.nih.gov/nuccore/MN551115.1) | | [2] | |
| *Betasclernavirus botrytidis* | Botrytis cinerea alpha-like virus 1 | ■ | BcALV1 | BCI2 | Fungi |  | | 8008 | | [MN625250](https://www.ncbi.nlm.nih.gov/nuccore/MN625250.1) | | [7] | |
| *Betasclernavirus betafusarii* | Fusarium sacchari alphavirus-like virus 1 | ■ | FsALV1 | FJ-FZ04 | Fungi |  | | 7685 | | [MN295968](https://www.ncbi.nlm.nih.gov/nuccore/MN295968.1) | | [8] | |
| *Betasclernavirus betasclerotii* | Sclerotium rolfsii alphavirus-like virus 3 | ■ | SraLV3 | BLH-1 | Fungi |  | | 7888 | | [MH766490](https://www.ncbi.nlm.nih.gov/nuccore/MH766490.1) | | [10] | |
| *Betasclernavirus alphasclerotii* | Sclerotium rolfsii alphavirus-like virus 1 | ■ | SraLV1 | BLH-1 | Fungi |  | | 7550 | | [MH766488](https://www.ncbi.nlm.nih.gov/nuccore/MH766488.1) | | [10] | |
|  | Morchella importuna RNA virus 1 |  | MiRV1 | SCYDJ1-A1 | Fungi |  | | 10099 | | [MK279480](https://www.ncbi.nlm.nih.gov/nuccore/MK279480.1) | | [3] | |
|  | Sclerotium rolfsii alphavirus-like virus 2 |  | SraLV2 | BLH-1 | Fungi |  | | 7591 | | [MH766489](https://www.ncbi.nlm.nih.gov/nuccore/MH766489.1) | | [10] | |

a: square indicates an exemplary virus for each species.



Fig 1 The genome organization of seven species in proposed family Mycoalphaviridae. Open reading frames (ORFs) are shown as boxes, the conserved motifs are represented in different colors.



Figure 2 Phylogenetic analysis of viruses in proposed familyMycoalphaviridae. The phylogenetic tree of members in the proposed family Mycoalphaviridae and other related families was constructed based on the alignment of the RdRp sequence by a maximum likelihood method. The number at the node represents the bootstrap values (%) obtained with 1000 replicates, and branch lengths correspond to genetic distance.



Figure 3 Comparison of members in proposed familyMycoalphaviridae. Percent identity matrix, generated by Clustal-Omega 2.1 of viruses in proposed familyMycoalphaviridae. The top half of the matrix, in the green scale, is the percent identity of the RdRp coding sequence. For clarity, the 100% identity along the diagonal has been removed.



Fig 4 Conserved RdRp motifs in the mycoalphaviruses and other related viruses in order *Hepelivirales*. The amino acid sequences of viruses were aligned using the MAFFT 7.0 software.