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:** | ***2019.019S*** | |  |
| **Short title:** Create one new species (*Tremovirus B*) in the genus *Tremovirus* | | | |
|  | | | |
| **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 | |
| Zell R, Gorbalenya AE, Hovi T, Knowles NJ, Lindberg M, Oberste S, Palmenberg AC, Reuter G, Simmonds P, Skern T, Tapparel C, Wolthers K, Woo P | | roland.zell@med.uni-jena.de; a.e.gorbalenya@lumc.nl; tapani.hovi@thl.fi; nick.knowles@pirbright.ac.uk; michael.lindberg@lnu.se; soberste@cdc.gov; acpalmen@wisc.edu; reuter.gabor@gmail.com; peter.simmonds@ndm.ox.ac.uk; timothy.skern@meduniwien.ac.at; caroline.tapparel@unige.ch; k.c.wolthers@amc.uva.nl; pcywoo@hkucc.hku.hk | |
| **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]) | | Jena University Hospital [RZ]  Leiden University Medical Center [AEG]  National Institute for Health and Welfare [TH]  The Pirbright Institute [NJK]  Linnaeus University Kalmar [ML]  Centers for Disease Control and Prevention [SO]  University of Wisconsin [ACP]  University of Pécs [GR]  University of Oxford [PS]  Medical University of Vienna [TS]  University of Geneve [CT]  Universiteit van Amsterdam [KW]  University of Hong Kong [PW] | | | | |
| **Corresponding author** | | | |
| **Roland Zell** (roland.zell@med.uni-jena.de) | | | |
| **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) | |  | |
| **ICTV Study Group comments (if any) and response of the proposer:** | | | |
|  | | | |
|  | | | |
| Date first submitted to ICTV: | | | 21/05/2019 |
| 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:** 2019.019S.A.v1.1newsp\_Tremovirus\_B.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. |

**Create one new species (*Tremovirus B*) in the genus *Tremovirus***

The genus *Tremovirus* presently consists of only one species, *Tremovirus A*, with one type. Novel, tremovirus-like picornaviruses were detected in organ pools (gut, liver, lungs) of Chinese softshell turtles (*Pelodiscus sinensis*) and Chinese broad-headed pond turtles (*Mauremys megalocephala*) from China (Shi et al., 2018). These viruses differ from the pemapiviruses (see accompanying proposal) which were detected in the same hosts. These viruses presumably represent two types of a second tremovirus species, designated **tremovirus B1** (Chinese softshell turtle picornavirus 2) and **tremovirus B2** (Chinese broad-headed pond turtle picornavirus 2). A virus closely related to the Chinese softshell turtle picornavirus 2 was also described by Pan et al. (KY432472; unpublished).

**Relation to tremovirus A1 (avian encephalitis virus 1) and other picornaviruses:**

- Genome layout of tremovirus B1 and B2:

5'-UTR[1A-1B-1C-1D-/2AH-box-NC-2B-2Chel/3A-3BVPg-3Cpro-3Dpol]3'-UTR

(compare Fig. 1 of supporting material)

- Tremovirus B1 and B2 have typical hallmarks of picornaviruses:

- **capsid proteins** 1B, 1C, 1D have **rhv** domains with drug-binding site,

- **2A** has a **H-box/NC**-motif,

- **2Chel** with **GxxGxGKS** motif of helicases,

- **3BVPg** peptide with **Y-3** residue,

- **3Cpro** with **GxCGx14GxH** motif,

- **3Dpol** with **KDE**, **PSG** and **YGDD** motifs; the **FLKR** motif is modified: **FLSR** and **YLSR**,

respectively.

- The P1 and 3CD sequences cluster with tremovirus A1 in **phylogenetic analyses** (compare Figs. 2 & 3 of supporting material).

**Distinguishing features of tremovirus B1 and B2 compared to tremovirus A1:**

- Tremoviruses B1 and B2 have long 2AH-box/NC proteins (303-305 aa vs. 154 aa of tremovirus A1.

- **Sequence divergence** (uncorrected p-distance) of complete genome suggest a distinct tremovirus species with 2 types:

[ 1 2 3 4 ]

[1] AJ225173, Tremovirus A1

[2] KY432472, Tremovirus B1 0.569

[3] MG600107, Tremovirus B1 0.564 0.038

[4] MG600110, Tremovirus B2 0.582 0.451 0.449

- **Sequence divergences** (uncorrected p-distances) of orthologous proteins in pairwise comparisons of tremoviruses B with representative picornaviruses of supergroup 5 (*Hepatovirus*/*Tremovirus*/unclassified craheliviruses, roheliviruses and fipiviruses) justify creation of a new species (compare Table 1).

**Table 1: Amino acid divergence\***

tremovirus B1 vs. member of ... P1 VP1 2Chel 3Cpro 3Dpol

*Within-genus* comparisons:

*Tremovirus* *Tremovirus B* (tremovirus B2) 0.296 0.311 0.435 0.449 0.499

*Tremovirus A* 0.482 0.543 0.641 0.615 0.56.6

*Between-genus* comparisons:

*Crahelivirus*† *Crahelivirus A*† 0.647 0.708 0.618 0.620 0.651

*Gruhelivirus*† *Gruhelivirus A*† 0.614 0.655 0.588 0.693 0.664

*Hepatovirus* *Hepatovirus A* 0.517 0.556 0.627 0.644 0.629

*Hepatovirus B*  0.511 0.570 0.615 0.620 0.617

*Hepatovirus C*  0.514 0.579 0.642 0.644 0.624

*Hepatovirus D*  0.530 0.569 0.654 0.648 0.640

*Hepatovirus E*  0.513 0.560 0.635 0.644 0.626

*Hepatovirus F*  0.515 0.567 0.617 0.671 0.632

*Hepatovirus G*  0.511 0.572 0.621 0.644 0.624

*Hepatovirus H*  0.527 0.569 0.615 0.616 0.620

*Hepatovirus I*  0.520 0.597 0.621 0.667 0.649

*Rohelivirus\*\** *Rohelivirus A*† 0.728 0.840 0.681 0.775 0.683

*Fipivirus*† *Fipivirus A*† 0.718 0.824 0.718 0.845 0.738

*Fipivirus B*†0.724 0.824 0.736 0.791 0.745

*Fipivirus C*†0.715 0.821 0.748 0.790 0.742

*Fipivirus D*†0.708 0.835 0.698 0.794 0.747

*Fipivirus E*† 0.692 0.803 0.723 0.806 0.756

\* number of amino acid differences per site

† proposed taxa

**Exemplar:**

*Tremovirus B*, tremovirus B1 (Pelodiscus sinensis picornavirus 1) strain CNSR2011, GenBank acc. no. KY432472

**Species demarcation criteria:**

Members of a species of the genus *Tremovirus*:

- are less than 50% divergent in polyprotein aa sequence,

-are less than 35% divergent in P1 aa sequence,

-are less than 50% divergent in 2C+3CD aa sequence,

- share a common genome organization,

| **References:** |
| --- |
| 1. Shi M, Lin XD, Chen X, Tian JH, Chen LJ, Li K, Wang W, Eden JS, Shen JJ, Liu L, Holmes EC, Zhang YZ. 2018. The evolutionary history of vertebrate RNA viruses. Nature 556:197-202.  2. Pan et al., unpublished |

**Supporting Material**



**Figure 1:** Genome of the tremoviruses (schematic depiction). The open reading frame is indicated by a box. Positions of putative 3Cpro cleavage sites are indicated by a ▼. The VP0 processing site is indicated by a hash (#). The names and lengths of the deduced proteins are presented. The 5'-UTR may be incomplete.



**Legend to Figure 2:**  Phylogenetic analysis of picornavirus **P1** using Bayesian tree inference (MrBayes 3.2). Thirty-five picornavirus sequences of the *Hepatovirus/Tremovirus* supergroup were retrieved from GenBank; the cardiovirus sequence served as outgroup. [Note: the supergroup does not imply a taxonomic entity but reflects phylogenetic clustering of the respective genera observed in different tree inference methods (NJ, ML, Bayesian MCMC).] Presented are GenBank accession numbers, ***genus*** ***names***, *species names*, type and—if available—common names in round brackets. Designations of isolates are given in square brackets. Yet unassigned viruses are printed in blue. The proposed names are printed in red and indicated by a dot (●). Numbers at nodes indicate posterior probabilities obtained after 2,000,000 generations. The optimal substitution model (GTR+G+I) was determined with MEGA 5. The scale indicates substitutions/site.



**Legend to Figure 3:**  Phylogenetic analysis of picornavirus **3CD** using Bayesian tree inference (MrBayes 3.2). Thirty-seven picornavirus sequences of the *Hepatovirus/Tremovirus* supergroup were retrieved from GenBank; the cardiovirus sequence served as outgroup. [Note: the supergroup does not imply a taxonomic entity but reflects phylogenetic clustering of the respective genera observed in different tree inference methods (NJ, ML, Bayesian MCMC).] Presented are GenBank accession numbers, ***genus*** ***names***, *species names*, type and—if available—common names in round brackets. Designations of isolates are given in square brackets. Yet unassigned viruses are printed in blue. The proposed name is printed in red and indicated by a dot (●). Numbers at nodes indicate posterior probabilities obtained after 2,000,000 generations. The optimal substitution model (GTR+G+I) was determined with MEGA 5. The scale indicates substitutions/site.