This form should be used for all taxonomic proposals. Please complete all those modules that are applicable.

For guidance, see the notes written in blue and the separate document “Help with completing a taxonomic proposal”

Please try to keep related proposals within a single document.

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

| **Code assigned:** | ***2017.011S*** | | | | (to be completed by ICTV officers) |
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| **Short title:** Proposal for a new virus family, *Polycipiviridae*. | | | | | |
| **Modules attached**  (Modules 1, 4 and either 2 or 3 are required. | | **1**  **2**  **3**  **4** | | | |
| **Author(s):** | | | | | |
| Ingrida Olendraitė, Nina I. Lukhovitskaya, Sanford D. Porter, Steven M. Valles,  Andrew E. Firth | | | | | |
| **Corresponding author with e-mail address:** | | | | | |
| Andrew E. Firth, email: [aef24@cam.ac.uk](mailto:aef24@cam.ac.uk) | | | | | |
| **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:** | | | | | |
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| Date first submitted to ICTV: | | | | 19 May 2017 | |
| Date of this revision (if different to above): | | | | 23 November 2017 | |

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| **ICTV-EC comments and response of the proposer:** |
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**Part 2**: **PROPOSED TAXONOMY**

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| Present the proposed new taxonomy on accompanying spreadsheet |
| **Name of accompanying spreadsheet:** TP\_Template\_Excel\_module\_2017.v2\_polycipiviridae |

Please display the taxonomic changes you are proposing on the accompanying spreadsheet module 2017\_TP\_Template\_Excel\_module. Submit both this and the spreadsheet to the appropriate ICTV Subcommittee Chair.

**Part 4:** **APPENDIX**: supporting material

| additional material in support of this proposal |
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| **References:** |
| **Edgar RC**. MUSCLE: Multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Res* 2004; 32: 1792–1797.  **Koonin EV, Wolf YI, Nagasaki K, Dolja VV**. The Big Bang of picorna-like virus evolution antedates the radiation of eukaryotic supergroups. *Nat Rev Microbiol* 2008; 6:500 925–939.  **Le Gall O, Christian P, Fauquet CM, King AMQ, Knowles NJ, et al**. Picornavirales, a proposed order of positive-sense single-stranded RNA viruses with a pseudo-T = 3 virion architecture. *Arch Virol* 2008; 153: 715–727.  **Olendraite I, Lukhovitskaya NI, Porter SD, Valles SM, Firth AE**. Polycipiviridae: a proposed new family of polycistronic picorna-like RNA viruses. *J Gen Virol,* 2017; 98:2368-2378.  **Ronquist F, Teslenko M, Van Der Mark P, Ayres DL, Darling A, et al**. MrBayes 3.2: Efficient bayesian phylogenetic inference and model choice across a large model space. *Syst Biol* 2012; 61:539–542.  **Valles SM, Strong CA, Hashimoto Y**. A new positive-strand RNA virus with unique genome characteristics from the red imported fire ant, Solenopsis invicta. *Virology* 2007; 365: 457–463. |

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| **Annex:**  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, try to provide a tree where branch length is related to genetic distance. |

Members of the order *Picornavirales* are characterized by (i) a positive-sense RNA genome, usually with a 5′ covalently linked VPg ("virus protein, genome linked") and a 3′ poly(A) tail, (ii) a polyprotein gene expression strategy with cleavage mainly mediated by viral protease(s), (iii) a structural protein module containing three jelly-roll capsid protein domains which form small non-enveloped icosahedral virions with pseudo T = 3 symmetry, and (iv) a non-structural protein module containing a superfamily III helicase (or NTPase), a 3C-like chymotrypsin-like protease, and a superfamily I RNA-dependent RNA polymerase (RdRp) encoded sequentially in that order (Le Gall et al., 2008).

Solenopsis invicta virus 2 (SINV-2) is a positive-sense single-stranded RNA virus that infects the red imported fire ant, *Solenopsis invicta* Buren (Valles et al., 2007). The genome is monopartite, polyadenylated, and ~11 kb in length. Isometric particles with a diameter of ~33 nm were found only in ants testing positive for SINV-2 by RT-PCR (Valles et al., 2007). Fourteen other SINV-2-like sequences have now been identified, mostly from ants and all from arthropods (Olendraite et al.; submitted). All sequences exhibit a characteristic polycistronic genome organization with four consecutive 5′ proximal ORFs and one long 3′ ORF (Figure 1). ORFs 1, 3 and 4 are predicted to encode jelly-roll fold picornavirus-like capsid proteins, while ORF2 encodes a protein of unknown function. Ant-infecting members of the group apparently have an additional 5′ ORF (ORF2b) overlapping the 5′ end of ORF2 and encoding a small protein containing a predicted transmembrane domain. The polypeptide encoded by the 3′ ORF5 contains motifs characteristic of a superfamily III helicase, 3C-like chymotrypsin-related protease, and a superfamily I RdRp, encoded in that order. The 5′ UTRs (where sequenced) range up to 366 nt in length, while 3′ UTR lengths range from 385 to 479 nt, excluding the poly(A) sequence. Thus, these viruses have the characteristic protein complement of members of the order *Picornavirales*. In contrast to other members of the order, the SINV-2-like virus structural protein module is further split into separate ORFs rather than depending on polyprotein expression of multiple jelly-roll domains from a single ORF. We obtained amino acid sequences for the RdRp region from the 15 SINV-2-like sequences and representative members of the order *Picornavirales*, aligned the sequences with MUSCLE (Edgar, 2004), and generated a Bayesian Markov chain Monte Carlo based phylogenetic tree using MrBayes (Ronquist et al., 2012) (Figure 2). The SINV-2-like sequences form a distinct clade.

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| **Figure 1. Genome map of SINV-2, a typical polycipivirus.** The genome is represented by a black line. ORFs are indicated in pale blue with vertical offsets indicating reading frame (−1, 0, +1) relative to ORF5. IGR denotes the intergenic region. (Figure reproduced from Olendraite et al., 2017.) |
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| **Figure 2. Phylogenetic tree for polycipiviruses and representative members of the order *Picornavirales*.** Core RdRp amino acid sequences from representative *Picornavirales* viruses were obtained from Koonin et al. (2008), combined with the equivalent regions from the 15 sequences in Table 1 with RdRp coverage, aligned with MUSCLE, and a Bayesian Markov chain Monte Carlo based phylogenetic tree produced. Posterior probabilities are indicated for family root nodes, and elsewhere if *p* < 1.00. Abbreviations: ALSV – apple latent spherical virus; BBWV1 – broad bean wilt virus 1; CPMV – cowpea mosaic virus; CRLV – cherry rasp leaf virus; CrPV – cricket paralysis virus; DCV – Drosophila C virus; DWV – deformed wing virus; EMCV – encephalomyocarditis virus; FMDV – foot-and-mouth disease virus; GFLV – grapevine fanleaf virus; HaRNAV – Heterosigma akashiwo RNA virus; HAV – hepatitis A virus; HplV-81 – Hubei picorna-like virus 81; HplV-82 – Hubei picorna-like virus 82; HRV1A – human rhinovirus 1A; IFV – infectious flacherie virus; LneV-1 – Lasius neglectus virus 1; LniV-1 – Lasius niger virus 1; MsaV-1 – Myrmica scabrinodis virus 1; NIMV – navel orange infectious mottling virus; PnPV – Perina nuda picorna-like virus; PV – polio virus; PYFV – parsnip yellow fleck virus; RTSV – rice tungro spherical virus; SDV – satsuma dwarf virus; ShiV-8 – Shuangao insect virus 8; SINV-2 – Solenopsis invicta virus 2; SINV-4 – Solenopsis invicta virus 4; TRSV – tobacco ringspot virus; TrV – Triatoma virus; TSV – Taura syndrome virus. (Figure reproduced from Olendraite et al., 2017.) |

The unusual genome organization and phylogenetic distinctness of this group of viruses suggests they should be classified into a new virus family for which we propose the name *Polycipiviridae* (**polyci**stronic **pi**corna-like viruses). The characteristic complement of picornavirus-like protein domains suggests that the *Polycipiviridae* should be included within the *Picornavirales* order.

Although the polycipiviruses form a distinct clade, there is considerable diversity among the 15 available sequences (some ORF5 pairwise amino acid identities as low as 17%, Table 1). Thus, we suggest that the family should be split into a number of genera. A phylogenetic analysis of the polycipivirus ORF5 polypeptide sequences showed three distinct groups (Figure 3). We propose the following groupings of the currently available sequences: *Sopolycivirus* (from **So**lenopsis invicta **polyci**pivirus) to include SINV-2, SINV-4 and nine related sequences all of which contain ORF2b and all but one (Shuangao insect virus 8) are ant-associated; *Hupolycivirus* (from **Hu**bei picorna-like virus 81 **polyci**pivirus) to include the two Hubei picorna-like virus 81 sequences; and *Chipolycivirus* (from **Chi**ronomid riparius virus 1 **polyci**pivirus) to include the *C. riparius* TSA and Hubei picorna-like virus 82, both of which contain a GDD to ADD substitution in RdRp motif VI.

**Species Demarcation Criteria**

We propose that, for the time-being, isolates and strains are defined to be of the same species if they have above 90% amino acid sequence identity in the ORF5 polyprotein (Table 1 lists pairwise identities for current members).

Type species are: *Polycipiviridae* and *Sopolycivirus* - *Solenopsis invicta virus 2* (as the first and best characterized member of the family and genus); *Hupolycivirus* - *Hubei picorna-like virus 81* (as the only species in the genus); and *Chipolycivirus* - *Chironomid riparius virus 1* (as the host organism is less certain for the only other species in the genus).

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| **Figure 3. Phylogenetic tree of the proposed family *Polycipiviridae*.** ORF5 amino acid sequences were aligned with MUSCLE, and a Bayesian Markov chain Monte Carlo based phylogenetic tree was produced with MrBayes. All posterior probabilities were equal to 1.00. The tree was mid-point rooted and visualized with FigTree. TSA indicates sequences obtained from the NCBI Transcriptome Shotgun Assembly database. Proposed *Polycipiviridae* genera – *Chipolycivirus*, *Hupolycivirus* and *Sopolycivirus* – are delineated with boxes. For sopolyciviruses, the host ant subfamilies are indicated at right. (Figure reproduced from Olendraite et al., 2017.) |

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| **Table 1. Pairwise identities (%) of ORF5 amino acid sequences of proposed family *Polycipiviridae* members** |