

This form should be used for all taxonomic proposals. Please complete all those modules that are applicable (and then delete the unwanted sections). 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; you can copy the modules to create more than one genus within a new family, for example.

MODULE 1: TITLE, AUTHORS, etc

| Code assigned: | 2014.006b-eD | | | (to be completed by ICTV officers) | | |
|---|-------------------------------|-------------|------------|------------------------------------|---------|---------|
| Short title: Establish the genus <i>Cyclovirus</i> in the family <i>Circoviridae</i> ; creating 28 new species in the family (e.g. 6 new species in the genus <i>Zetavirus</i>) Modules attached $1 \times 2 \times 3 \times 4 \times 5 \times 6 \times 100$ (modules 1 and 10 are required) $1 \times 2 \times 3 \times 4 \times 5 \times 100$ | | | | | | |
| Author(s): | | | | | | |
| Mya Breitbart and Arvind Vars | sani on behalf o | of the Circ | oviridae S | SG; email | – mya@u | ısf.edu |
| Corresponding author with e | -mail address: | ; | | | | |
| Mya Breitbart on behalf of the | Circoviridae S | G; email - | - mya@us | f.edu | | |
| List the ICTV study group(s) | that have seen | n this pro | posal: | | | |
| A list of study groups and contacts http://www.ictvonline.org/subcomm in doubt, contact the appropriate schair (fungal, invertebrate, plant, pvertebrate viruses) | mittees.asp . If subcommittee | Circovi | ridae Stud | y Group | | |
| ICTV Study Group comments (if any) and response of the proposer: | | | | | | |
| | | | | | | |
| Date first submitted to ICTV: | | | | 15, 2015 | | |
| Date of this revision (if different to above): June 29, 2015 | | | | | | |
| ICTV-EC comments and response of the proposer: | | | | | | |
| ICTV-EC comments and response of the proposer: | | | | | | |

MODULE 2: NEW SPECIES

creating and naming one or more new species.

| Code 201 | 2014.006bD (assigned by ICTV officers) | | | | |
|--|--|---|--|--|--|
| To create 28 new species within: | | | | | |
| Genus: Cyclovirus (new) Subfamily: Family: Circoviridae Order: | | If the higher tax created (in a lat "(new)" after its If no genus is s | Fill in all that apply. If the higher taxon has yet to be created (in a later module, below) write "(new)" after its proposed name. If no genus is specified, enter "unassigned" in the genus box. | | |
| Name of new species: | | Representative isolate: (only 1 per species please) | GenBank sequence accession number(s) | | |
| Bat cyclovirus Bat faeces associated cyclovirus 1 Bat faeces associated cyclovirus 2 Bat faeces associated cyclovirus 3 Bat faeces associated cyclovirus 4 Bovine cyclovirus Chimpanzee faeces associated cyclovirus Dragonfly cyclovirus 1 Dragonfly cyclovirus 2 Dragonfly cyclovirus 3 Dragonfly cyclovirus 5 Dragonfly cyclovirus 5 Dragonfly cyclovirus 6 Dragonfly cyclovirus 7 Dragonfly cyclovirus 8 Florida wood cockroach cyclovirus Gallus cyclovirus Human cyclovirus 1 Human cyclovirus 2 Human faeces associated cyclovirus 1 Human faeces associated cyclovirus 2 Human faeces associated cyclovirus 3 Human faeces associated cyclovirus 4 Human faeces associated cyclovirus 5 Human faeces associated cyclovirus 5 Human faeces associated cyclovirus 6 Human faeces associated cyclovirus 5 Human faeces associated cyclovirus 6 Human faeces associated cyclovirus 8 | | bat cyclovirus (BaCyV) bat faeces associated cyclovirus 1 (BaFCyV-1) bat faeces associated cyclovirus 2 (BaFCyV-2) bat faeces associated cyclovirus 3 (BaFCyV-3) bat faeces associated cyclovirus 4 (BaFCyV-4) bovine cyclovirus (BoCyV) chimpanzee faeces associated cyclovirus (ChmFCyV) dragonfly cyclovirus 1 (DfCyV-1) dragonfly cyclovirus 2 (DfCyV-2) dragonfly cyclovirus 3 (DfCyV-3) dragonfly cyclovirus 5 (DfCyV-3) dragonfly cyclovirus 6 (DfCyV-4) dragonfly cyclovirus 7 (DfCyV-7) dragonfly cyclovirus 7 (DfCyV-7) dragonfly cyclovirus 8 (DfCyV-8) Florida wood cockroach cyclovirus (FWCasCyV) gallus cyclovirus (GaCyV) goat cyclovirus (GhCyV-1) human cyclovirus 1 (HuCyV-1) human cyclovirus 3 (HuCyV-2) human faeces associated cyclovirus 2 (HuFCyV-2) human faeces associated cyclovirus 3 (HuFCyV-3) human faeces associated cyclovirus 4 (HuFCyV-4) human faeces associated cyclovirus 5 (HuFCyV-5) human faeces associated cyclovirus 6 (HuFCyV-6) human faeces associated cyclovirus 6 (HuFCyV-6) human faeces associated cyclovirus 8 (HuFCyV-6) human faeces associated cyclovirus 8 (HuFCyV-6) | HQ738637 HM228874 JF938079 JF938081 JF938082 HQ738634 GQ404849 JX185419 JX185422 JX185424 JF938080 JX185426 KC512918 KC512919 KC512920 JX569794 HQ738643 HQ738636 KF031466 KC771281 KF726984 GQ404847 GQ404844 GQ404846 GQ404858 GQ404855 | | |

Reasons to justify the creation and assignment of the new species:

- Explain how the proposed species differ(s) from all existing species.
 - o If species demarcation criteria (see module 3) have previously been defined for the genus, **explain how the new species meet these criteria**.
 - o If criteria for demarcating species need to be defined (because there will now be more than one species in the genus), please state the proposed criteria.
- Further material in support of this proposal may be presented in the Appendix, Module 9

We have analyzed all the full genomes of cycloviruses available in public databases using SDT v1.2 (Muhire et al., 2014). Our analysis (Figures 1-3) shows that 80% pairwise identity species cut off is best suited for cycloviruses (a similar one is proposed for circoviruses) and maintains a uniform classification in the family *Circoviridae*. Hence viruses with <80% pairwise identities coupled with phylogenetic support (Figures 2) should be considered as new species.

MODULE 3: NEW GENUS

creating a new genus

Ideally, a genus should be placed within a higher taxon.

| Code | 2014 | 4.006cD | (assigned by ICTV officers) | | |
|--|---------|--------------|-----------------------------|---|--|
| To create a new genus within: Fill in all that apply. | | | | | |
| Subfar | mily: | | | If the higher taxon has yet to be created | |
| Far | mily: (| Circoviridae | | (in a later module, below) write "(new)" after its proposed name. | |
| 0 | order: | | | If no family is specified, enter "unassigned" in the family box | |

naming a new genus

| Code | 2014.006dD | (assigned by ICTV officers) |
|-----------|--------------------------|-----------------------------|
| To name t | he new genus: Cyclovirus | |

Assigning the type species and other species to a new genus

| Code | 2014.006eD | (assigned by ICTV officers) | | |
|--|--------------------------------|---|--|--|
| To design | gnate the following as the typ | e species of the new genus | | |
| Human cyclovirus 1 Every genus must have a type species. This sh be a well characterized species although not necessarily the first to be discovered | | | | |
| are being | | new species created and assigned to it (Module 2) and any that 7b). Please enter here the TOTAL number of species genus will contain: | | |

Reasons to justify the creation of a new genus:

Additional material in support of this proposal may be presented in the Appendix, Module 9

The genus *Cyclovirus* will accommodate newly discovered viruses whose replication associated proteins (Rep) are most closely related to those of circoviruses (Figures 4-6); yet they have distinct genome organization and characteristics (Rosario et al. 2012b).

Both circoviruses and cycloviruses have ambisense genomes and phylogenetically similar Reps; however, phylogenetic analyses show that circoviruses and cycloviruses form distinct clades and there are consistent differences in the length of the intergenic region located at the 3' end of the major ORFs (Figure 5). Also, in contrast to circoviruses, the Rep of cycloviruses is not encoded on the same strand as the canonical nonanucleotide motif.

Origin of the new genus name:

"Cyclo" is derived from Latin "cyclus" and relates to the circular nature of the DNA genome

Reasons to justify the choice of type species:

Isolated from human cerebrospinal fluid of patients with acute central nervous system infections

Species demarcation criteria in the new genus:

If there will be more than one species in the new genus, list the criteria being used for species demarcation and explain how the proposed members meet these criteria.

Analysis of the percentage pairwise identity of the genome sequences (one minus Hamming distances of pairwise aligned sequences with pairwise deletion of gaps, Figure 1) revealed a gap located at approximately 76-85%. We have used a similar approach to the one used for geminiviruses, in particular the *Mastrevirus*, *Curtovirus*, *Becurtovirus* and *Turncurtovirus* genera (Muhire et al. 2012; Varsani et al. 2014a; 2014b). Based on genome-wide pairwise identities isolates that share <80% genome wide pairwise nucleotide identity coupled with phylogenetic support (Figure 2) are considered distinct species in the genus *Cyclovirus*. We note that this threshold holds true for pairwise comparisons of either the capsid protein gene sequences or the replication associated protein gene sequences. However, only full genomes of cycloviruses will be considered for assignment of new species in the event that share <80% pairwise identity to any classified cyclovirus.

MODULE 10: **APPENDIX**: supporting material Table 1: Details of identified cycloviruses

| bast laeces associated cyclovirus 2 | Species | Acronym | GenBank Accession # | Isolation source | Reference |
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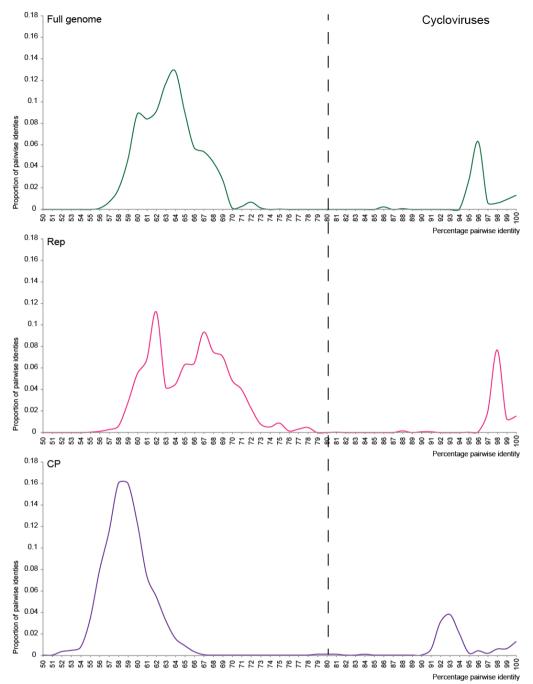


Figure 1: Distribution pairwise identity of cyclovirus genomes. The pairwise identities were calculated using SDT V1.2 (Muhire et al., 2014) with MUSCLE alignment algorithm (Edgar, 2004). Rep = replication associated protein gene; CP = capsid protein gene.

Within the cyclovirus genus we have assigned 28 species (Table 1) based on the 80% species threshold coupled with phylogenetic support (Figure 5). We have retained the original name of the virus sequence deposited by researchers where possible. We have also doubled checked to make sure that variants within a species share >80% pairwise identity (Figure 6).

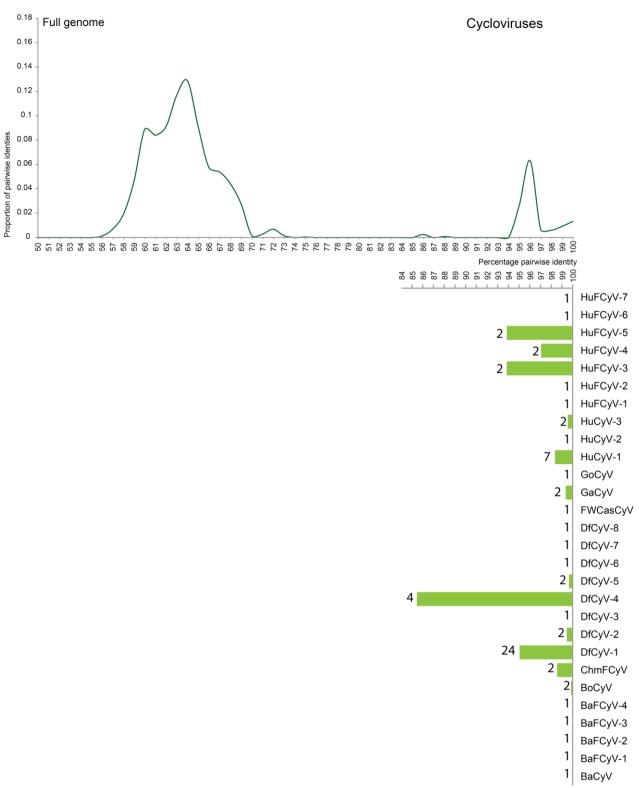


Figure 2: Diversity of variants within each assigned species in the genus cyclovirus. Numbers associated with bars refer to number of isolates within the species.

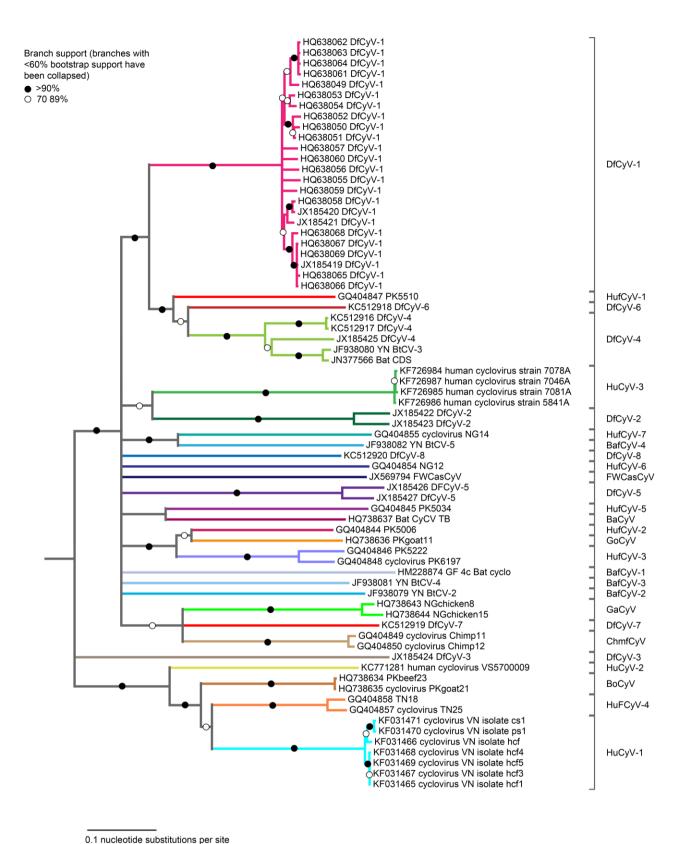


Figure 3: Neighbor-joining (with Jukes-Cantor model) phylogenetic tree (1000 bootstrap) inferred from full genome alignments. Branches with <60% support have been collapsed.

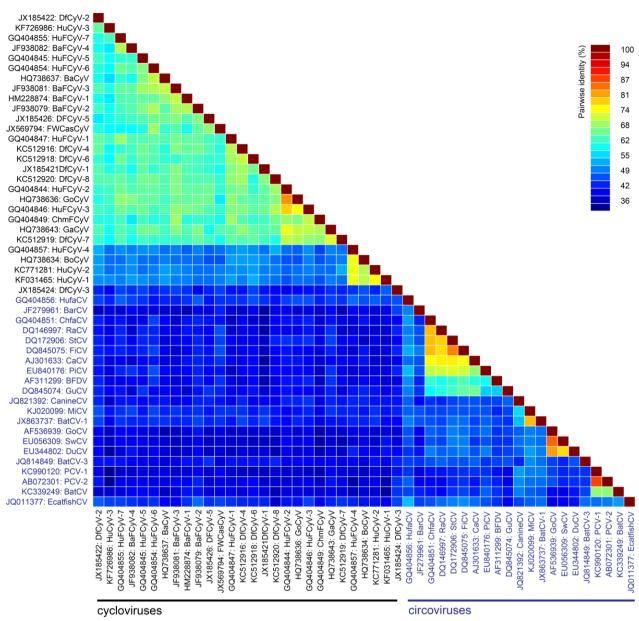


Figure 4: Two directional pairwise identity matrix of the replication associated protein amino acid sequences of representative circoviruses and cycloviruses.

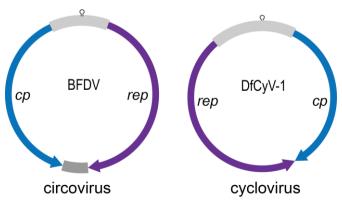


Figure 5: Genome organisation of circoviruses (e.g., beak and feather disease virus; BFDV) and cycloviruses (e.g., dragonfly cyclovirus 1; DfCyV-1). Replication-associated protein (*rep*) and capsid protein (cp) genes as well as intergenic regions (highlighted in grey) are shown.

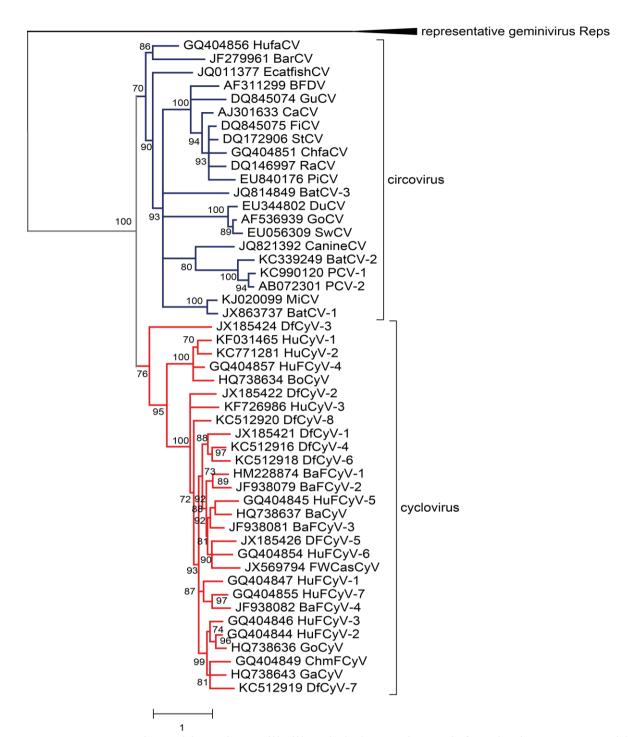


Figure 6: Rep amino acid maximum likelihood phylogenetic tree inferred using PHYML with aLRT branch support. Branches <70% support have been collapsed. The tree is rooted with Rep sequences of geminiviruses.

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