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**

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| **Code assigned:** | ***2017.015D*** | | | | (to be completed by ICTV officers) |
| **Short title:** 2 new species unassigned in family *Hepadnaviridae* | | | | | |
| **Modules attached**  (Modules 1, 4 and either 2 or 3 are required. | | **1**  **2  3  4** | | | |
| **Author(s):** | | | | | |
| Lars Magnius and Heléne Norder | | | | | |
| **Corresponding author with e-mail address:** | | | | | |
| Lars.magnius@gmail.com | | | | | |
| **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) | | | Hepadnaviridae & Hepatitis delta virus Study Group | | |
| **ICTV Study Group comments (if any) and response of the proposer:** | | | | | |
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|  | | | | | |
| Date first submitted to ICTV: | | | | 12 June 2017 | |
| Date of this revision (if different to above): | | | | 22 June 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:** 2017.015D.N.v1.Hepadnaviridae\_2sp |

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 |
| --- |
| **References:** |
| Dill JA, Camus AC, Leary JH, Di Giallonardo F, Holmes EC, Ng TF. Distinct Viral Lineages from Fish and Amphibians Reveal the Complex Evolutionary History of Hepadnaviruses. J Virol. 2016 90(17):7920-33. |

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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. |

Lip tissues from the bluegill fish (*Lepomis macrochirus*) were analysed for viruses due to orocutaneous neoplasms in two fish populations. They were from a mixed fish aquarium in Georgia USA. Metagenomics was performed and hepadnavirus like sequences were identified. These sequences were further confirmed by sequencing with specific primers and the amount of virus was determined by qPCR. The sequences obtained were used as queries in BLAST analysis against whole genome shotgun and transcriptome shotgun assembly databases in GenBank. By this analysis complete hepadnavirus genome was identified from Tibetan frog and partial hepadnavirus genome from African cichlid (Dill et al., 2016). Phylogenetic analysis proved that the virus genomes from the bluegill and Tibetan frog represent new hepadnavirus with all known ORFs apart from the X-gene. The partial genome obtained from the African cichlid proved that only the polymerase gene could be identified. These viruses diverge with more than 30% from all previously known hepadnaviruses based on the polymerase gene. The African cichlid HBV formed a root in the phylogenetic tree based on the polymerase of all Hepadnaviridae members. However, as the full genome is not known this virus is not part of the present species proposals. The above described sequences have been published (Dill et al., 2016).

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HBV-A

HBV-F

WMHBV *Woolly monkey hepatitis B virus*

RLB *Roundleaf bat hepatitis B virus*

PB HBV *Pomona bat hepatitis B virus*

WHV *Woodchuck hepatitis virus*

TMB HBV *Tent-making bat hepatitis B virus*

BGHBV *Bluegill hepatitis B virus* (KX058433)

DHBV *Duck hepatitis B virus*

HHBV *Heron hepatitis B virus*

PHBV *Parrot hepatitis B virus*

TFHBV *Tibetan frog hepatitis B virus* (KX058435)

WSHBV *White sucker hepatitis B virus*

ACHBV African cichlid hepatitis B virus (KX058434)

100

100

98

87

100

100

100

100

88

100

100

94

*Hepatitis B virus*

Fig. 1. Phylogenetic tree based on Maximum likelihood analysis of the polymerase gene of members of genera *Avihepadnavirus* and *Orthohepadnavirus*. Bootstrap values of 1,000 replicas are given at the branches in percentage. The species names are shown in italics, the suggested new species names in red with accession numbers. The accession number of the new African chichlid hepatitis B virus is also shown, but the full genome of this virus is not known yet, thus it is not proposed yet for acceptance as member of a new species.

Table 1. Percent amino acid (upper triangle) and nucleotide (lower triangle) divergence of the polymerase gene of bluegill hepatitis B virus (BGHBV), Tibetan frog hepatitis B virus (TFHBV) and African cichlid hepatitis B virus (ACHBV) compared to the polymerase gene of avihepadnavirus (duck HBV (DHBV), heron HBV (HHBV) and parrot HBV (PHBV)) and orthohepadnavirus genomes (hepatitis B virus genotypes A and F (HBV-A, HBV-F), woolly monkey HBV (WMHBV), roundleaf bat HBV (RLB HBV), pomona bat HBV (PB HBV), tent-making bat HBV (TMB HBV)) and white sucker HBV (WSHBV).

HBV-A HBV-F WMHBV RLB HBV PB HBV WHV TMB HBV **BGHBV** DHBV HHBV PHBV **TFHBV** **ACHBV** WSHBV

HBV-A 17 28 44 48 47 48 **60** 64 64 64 **67** **68** 68

HBV-F 14 29 43 44 46 47 **62** 63 63 65 **67** **69** 67

WMHBV 22 24 45 44 47 48 **61** 63 63 65 **67** **69** 67

RLB HBV 36 36 37 35 44 47 **61** 64 66 66 **67** **70** 67

PB HBV 37 36 37 31 45 47 **62** 68 69 69 **74** **71** 68

WMHBV 39 39 39 39 41 48 **60** 67 67 68 **71** **69** 67

TMB HBV 39 38 39 39 40 41 **61** 65 65 65 **68** **70** 67

**BGHBV 48 48 48 48 49 49 48 63 64 65 65 68 63**

DHBV 49 49 49 51 55 52 51 **47** 30 32 **62** **68** 64

HHBV 50 50 50 51 55 52 51 **48** 23 33 **64** **67** 63

PHBV 51 51 51 51 56 52 50 **48** 24 26 **64** **68** 63

**TFHHBV 55 54 54 54 60 56 54 50 49 51 51 68 67**

**ACHBV 57 57 57 58 59 58 58 57 58 57 57 56 67**

WSHBV 52 52 51 52 54 53 52 **50** 51 51 51 **52** **42**