

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

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| **Code assigned:** | **2020.001M** |  |
| **Short title:** Create one new genus (*Alpharicinrhavirus*) including three new species (*Mononegavirales*: *Rhabdoviridae*) | | |
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**List the ICTV Study Group(s) that have seen this proposal**

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| ICTV *Rhabdoviridae* Study Group |

**ICTV study group comments and response of proposer**

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| Approved by all responding SG members (11 of 14) with minor revisions. |

**Authority to use the name of a living person**

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| --- | --- | --- |
| **Taxon name** | **Person from whom the name is derived** | **Permission attached (Y/N)** |
| Not applicable |  |  |

**Submission dates**

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| Date first submitted to SC Chair |  |
| Date of this revision (if different to above) |  |

**ICTV-EC comments and response of the proposer**

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

**Name of accompanying Excel module**

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| 2020.001M\_014M\_015M\_016M.R.Rhabdoviridae.xlxs |

**Abstract**

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| The new genus *Alpharicinrhavirus* is proposed to accommodate four currently unassigned rhabdoviruses that have been detected in hard ticks (Acari: Ixodidae). The viruses will be assigned to three new species within the new genus: *Bole alpharicinrhavirus*, *Wuhan alpharicinrhavirus* and *Blanchseco alpharicinrhavirus*. Each member virus was detected by metagenomic sequence analysis of ticks. No virus isolates are available at this time. |

**Text of proposal**

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| |  | | --- | | **Distinguishing characteristics of the genus**  The genus *Alpharicinrhavirus* comprises rhabdoviruses detected in hard ticks (Acari: Ixodidae). The viruses form a well-supported and distinct monophyletic clade based on a well-supported Maximum Likelihood tree inferred from complete L sequences. Members of the genus are most closely related to alphanemrhaviruses which that have been detected in nematode worms.  **Proposed species**  ***Bole alpharicinrhavirus*.** Bole tick virus 2(BlTV-2) was detected in hard ticks (*Hyalomma asiaticum*) collected in Bole, Xinjiang Province, China, in 2012. Near-complete genome sequences including complete coding sequences and partial terminal sequences have been determined for BlTV-2 [3].  ***Wuhan alpharicinrhavirus*.** Wuhan tick virus 1(WhTV-1) was detected in hard ticks (*Rhipicephalus microplus*) collected in Wuhan, Hubei Province, China, in 2012. Near-complete genome sequences including complete coding sequences and partial terminal sequences have been determined for WhTV-1 [3].  Rhipicephalus associated rhabdo-like virus (RaRLV) was detected in hard ticks(*Rhipicephalus microplus*) collectedin Yunnan Province, the China, in 2016. Near-complete genome sequences including complete coding sequences and partial terminal sequences have been determined for RaRLV (MH814974).  RaRLV and WhTV-1 are very closely related and are considered to be strains of the same virus.  ***Blanchseco alpharicinrhavirus.*** Blanchseco virus (BCOV) was detected in a pool of hard ticks(*Amblyomma ovale*) collected from dogs from the Caribbean island of Trinidad, in 2017. The near-complete genome sequence including complete coding sequences and partial terminal sequences have been determined for BCOV [5].  **Other likely members of the genus**  Tacheng tick virus 3 (TcTV-3) was isolated from hard ticks (*Dermacentor marginatus*) collected in Tacheng, Xinjiang Province, China, in 2013 [3].  Manly virus (MLYV) was isolated from hard ticks (*Amblyomma moreliae*) collected in Australia, in 2016 [2].  TcTV-3 and MLYV and NyTRV are probable members of the genus but only a partial genome sequences are available, including complete *L* gene sequences for TcTV-3 and MLYV [3]. They are included here for comparative purposes but, as the coding sequence is incomplete, we do not propose formal classification at this time.  Nayun tick rhabdovirus (NyTRV) was detected in hard ticks (*Rhipicephalus* sp.) collected in Yunnan Province, China, in 2013 [7]. Based on the small region of available sequence in the *L* gene, NyTRV (like RaRLV) appears to be a strain of WhTV-1.  No isolates are available for any of these viruses and no other biological data are available.  **Other related viruses**  Taishun tick virus (TsTV)was detected in hard ticks (*Haemaphysalis hystricis*) collected in Taishun, Zhejiang Province, China, in 2013. Near-complete genome sequences including complete coding sequences and partial terminal sequences have been determined for TsTV [3].  Norway mononegavirus 1 (NWMV-1) was detected in hard ticks (*Ixodes ricinus*) collected in Norway, in 2014. Near-complete genome sequences including complete coding sequences and partial terminal sequences have been determined for NWMV-1 [4](MF141072).  Huangpi tick virus 3(HpTV-3) was detected in hard ticks (*Haemaphysalis doenitzi*) collected in the Huangpi District of Wuhan, Hubei Province, China, in 2012. Near-complete genome sequences including complete coding sequences and partial terminal sequences have been determined for HpTV-3 [3].  An unnamed rhabdovirus was detected in hard ticks (*Ixodes scapularis*) collected in Wisconsin, USA, in 2015 [1]. Based on partial *L* gene sequences, it is most closely related to NWMV-1.  American dog tick rhabdovirus 2 was detected in hard ticks (*Dermacentor variabilis*) collected in the USA, in 2016 [6]. Based on partial *L* and *N* gene sequences, it is most closely related to TsTV.  Other rhabdoviruses have been detected in, or isolated from, ticks but they are phylogenetically distant from the proposed alpharicinrhaviruses.  **Genomes**  Alpharicinrhavirus genomes range in length from approximately 10.5 kb to 11.9 kb, containing the five canonical rhabdovirus structural protein genes (*N*, *P*, *M*, *G* and *L*), except for WhTV-1 and RaRLV which lack a gene encoding the G protein (**Figure 1**). The absence of the G gene in two strains of the same virus detected in different locations and at different times appears to indicate this is a genuine gene deletion rather than a sequencing artifact. The *G* genes of other alpharicinrhaviruses encode type I transmembrane glycoproteins displaying 12 conserved cysteine residues that are typical of animal rhabdovirus G proteins (**Figure 2**). Alternative small ORFs (>180 nt) occur in some genes. It is not known if they are expressed but small ORFs in the WhTV-1 and RaRLV *P* and *L* genes are conserved.  **Phylogeny**  Based on ML trees generated from complete L protein sequences, alpharicinrhaviruses form a well-supported monophyletic clade that is distinct from all currently assigned genera and other currently unassigned rhabdoviruses (**Figure 3**).  **Sequence identity**  Amino acid sequence identities between viruses to be assigned to different species are relatively low (<40% in the N and G proteins and <60% in the L proteins) (**Tables 1-3**). WhTV-1 and RaRLV (99.9% identity in the N and L proteins) are considered to be strains of the same virus.  **Species demarcation criteria**  Viruses assigned to different species within the genus *Alpharicinrhavirus* have several of the following characteristics: A) minimum amino acid sequence divergence of 10% in N proteins; B) minimum sequence divergence of 10% in the L proteins; C) minimum amino acid sequence divergence of 15% in G proteins; D) significant differences in genome organization as evidenced by numbers and locations of ORFs; E) can be distinguished in virus neutralisation tests; and F) occupy different ecological niches as evidenced by differences in vertebrate hosts and or arthropod vectors.  All proposed members of the new genus meet demarcation criteria A, B, C and F. WhTV-1 also meets criterion D. As there are no isolates of these viruses, cross-neutralization data are not available (criterion E).  **Derivation of the genus name**  *Alpharicinrhavirus* is the alpha group of tick rhabdoviruses derived from *ricinus* (Latin, tick) and rhabdovirus.  **Type species**  *Bole alpharicinrhavirus* is designated as the type species of the genus as the genome sequence of Bole tick virus 2 has no ambiguous nucleotide assignments and provides clear indications of the locations of all transcriptional control sequences. | |

**Supporting evidence**

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**Figure 1.** Alpharicinrhavirus genome organisations. Arrows indicate the locations of long open reading frames (ORFs), each of which is located within a transcriptional unit bounded by conserved transcription initiation and transcription termination/polyadenylation sequences. Alternative ORFs (shaded grey) of significant length (>180 nucleotides) occur in some genes. N, P, M, G and L represent ORFs encoding the canonical rhabdovirus structural protein genes. Proteins encoded in other ORFs do not share recognisable homology with proteins of other known rhabdoviruses.

BlTV-2\_G MGTVGEVFLLLSLLSFLPLAGS-DPVVKAIAAP---YFFPENLNYAWHPIEVTSLTCPPQ

BCOV\_G M-----FLLFLLLFVVHPLVGTGLPENTTWDFPNHVYFFPRDELYTWKPIRSSELSCPPF

MLYV\_G M------YCLLWVLCLLPCVVSEWKPDVAFAFP-------EDRKYIWKRINVSDMECPAI

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BlTV-2\_G RSIPEQDNGIPVIFETIHPSSLERALVNGYSCYTSTMAVKCSVNFVGWKTLSHQITNKEP

BCOV\_G FPITTGSGKISVTYATPGPMTTESATITGYSCYKSIFSVKCSENFVGWQTISHTIRPTEV

MLYV\_G YNNPSLNPVVRVNLRHPEFTGEFNDVIKGFICTKLKFAVKCSVNFFGWKTYQHSSQDLSP

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BlTV-2\_G SSTTCWEAIKRQEDGAQSPPPTFPAPNCAWWSENWAELDYTLVLKHPARQDPYTEALYDP

BCOV\_G SHRECLDEYRRIKTGSLPAAHEFPAPNCGWWADRWAEKEYITLIHHPVSFDPYSLEFFDP

MLYV\_G VISECTSAVEMYRVNGQTTTPAYPTPNCGWMAEHWAEESFIVLTDHPVHSDPYSGELVDK

\* . . . .. ... :\*:\*\*\*.\* ::.\*\*\* .: : .\*\*. \*\*\*: : \*

BlTV-2\_G LFPGGSCNKAECPLIHDGGIWIQTEPLASICKHWEVLQ-GLTYTGPEIGR-ILFSPEGPP

BCOV\_G LFPGGKCNNRVCSLIHQGGIWIQTEEHVGICENWHS---GIGHVGLVKNELVLFPMIGEV

MLYV\_G AFPGGTCATEHCQTIHHGSLWIPVEKVSPSCKFFKVSTGYLGTTTGAIQRRLLYSTGMGV

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BlTV-2\_G KYLDHSCRMTFCGRRGYRLQDGEFLVFS--SPPAWGVPPVCPAGTLVRAHTPEEEIRWNE

BCOV\_G RSLQGACTLKFCGHIGYRLATGEFFKLD--FYSLPKRIPFCEKGVQVRLDSPRGMTYEIE

MLYV\_G RSLAGICRMSFCGHEGWRLSTGEFVEVPGDFLKFTGTIPNCAPGLKISEPTVEGKLARSH

: \* \* :.\*\*\*: \*:\*\* \*\*\*. . \* \* \* : : . .

BlTV-2\_G ISKMEEADRLMCISRLSVAYATGKVSLELLGSLVPSHGGPGTAYRINNGTLEAAHVKYVP

BCOV\_G EKVMKEEDRLECLTSVALMQSTGHASQYLLSTLVPRHAGPGDAYRLSNGIIEHAHVFYVG

MLYV\_G LEIMDQEARLQCLSTLAIAVSTKKVSPFILSLFTPTHPGKGKAYRLHGSYIEEATVPYIG

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BlTV-2\_G LINSSN-EEGDLIGVGPDGTPILWEYWVLSG--SRIIGPNGVYKSKGR-IIVPNFERRKL

BCOV\_G ITKASTGRDQNVVGIDSKGNPVQWTDWIKDN--GTLIGPNGVTRKPGSPVVIPRFERLAN

MLYV\_G LRTLMTTPHEDNIGIREDGTLVSWTDWVNVTGMSGLSGPNGIIRFPDGKILIPNSDMYSM

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BlTV-2\_G TYDLTIHVFEDLKEIPHPSLVIRSNHTDLLRKVSHNQGVEGDHWASIRLWFSSLWGSFIW

BCOV\_G EYDLSLARVQSLKPIPHPLVTFLANKSDFLESESDNLGRDGSFWEGVRDWFHSMWGSFTW

MLYV\_G RHSLLLTMTQELQDVPHPHREIEKNQTDSITKLNRDLGTDG--APNIGDWFRSPGGIAAI

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BlTV-2\_G TCGLALIGLILICCICRRVRCCCRGCGRP-----QSKEAGG------WESIEMNDL

BCOV\_G SMILVVLGIVGLVFLLRRYSVKLRLPDSPKVEKKKKKSRRGNPVRDPWGDIADTA-

MLYV\_G SIPIVILIAIILFCLLKNAKCTRKTIVVKP----QKRQPNS------WANVATTSV

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**Figure 2.** Clustal W alignment of theamino acid of the sequences G proteins of alpharicinrhaviruses (including Manly virus for comparative purposes). Twelve conserved cysteine residues are highlighted. Predicted transmembrane domains are shaded in grey.

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**Figure 3.** The evolutionary history was inferred from a ClustalW alignment of complete L protein sequences of 140 animal rhabdoviruses currently assigned to species (or proposed to be assigned in other proposals) as well as the proposed alpharicinrhaviruses (Wuhan tick virus 1, Rhipicephalus associated rhabdo-like virus, Bole tick virus 2 and Blanchseco virus) and two possible members of the proposed genus (Tacheng tick virus 3 and Manly virus). The data set also includes three tick rhabdoviruses that are currently unclassified. Phylogenetically informative sites were selected from the alignment using Gblocks resulting in 938 positions in the final dataset. The tree was inferred in MEGA by using the Maximum Likelihood method based on the Whelan And Goldman + Freq. model. The tree with the highest log likelihood (-107344.41) is shown. The percentage of trees in which the associated taxa clustered together is shown next to the branches. Initial tree(s) for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using a JTT model, and then selecting the topology with superior log likelihood value. The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. Bootstrap values (100 iterations) are shown for each node.

**Table 1.** Percentage amino acid sequence identities (p-distance) of a ClustalW alignment of alpharicinrhavirus N proteins.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | BlTV-2 | BCOV | WhTV-1 | RaRLV |
| BlTV-2 |  |  |  |  |
| BCOV | 36.8 |  |  |  |
| WhTV-1 | 25.3 | 26.0 |  |  |
| RaRLV | 25.3 | 26.0 | 99.9 |  |

**Table 2.** Percentage amino acid sequence identities (p-distance) of a ClustalW alignment of alpharicinrhavirus G proteins.

|  |  |  |  |
| --- | --- | --- | --- |
|  | BlTV-2 | BCOV | [MLYV] |
| BlTV-2 |  |  |  |
| BCOV | 39.0 |  |  |
| [MLYV] | 31.5 | 32.7 |  |

**Table 3.** Percentage amino acid sequence identities (p-distance) of a ClustalW alignment of alpharicinrhavirus L proteins.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | BlTV-2 | BCOV | [TcTV-3] | WhTV-1 | RaRLV | [MLYV] |
| BlTV-2 |  |  |  |  |  |  |
| BCOV | 56.6 |  |  |  |  |  |
| [TcTV-3] | 48.4 | 49.1 |  |  |  |  |
| WhTV-1 | 47.2 | 46.7 | 64.4 |  |  |  |
| RaRLV | 47.2 | 46.7 | 64.4 | 99.9 |  |  |
| [MLYV] | 50.2 | 48.4 | 50.5 | 50.5 | 50.5 |  |

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