

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:	2010.014	2010.014aV			(to be completed by ICTV officers)		
Short title: New species Anati	d herpesvirus 1	in genu	s Mardiv	irus, subfai	mily		
Alphaherpesvirinae, family He	rpesviridae						
(e.g. 6 new species in the genus Zetavirus)						_	
Modules attached			2 🖂	3 <u> </u>	4 🔛	5 📙	
(modules 1 and 9 are required)		6 🗌	7 📙	8 🗌	9 🖂		
Author(s) with e-mail address(es) of the proposer:							
Andrew Davison (a.davison@mrcvu.gla.ac.uk) & Duncan McGeoch							
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			Herpesvirales Study Group				
in doubt, contact the appropriate subcommittee chair (fungal, invertebrate, plant, prokaryote or			Therpesyltales Study Group				
vertebrate viruses)							
ICTV-EC or Study Group comments and response of the proposer:							
This proposal has had a full cy	cle of discussion	n and ha	s been a	pproved wi	thout disse	ent by the	
Herpesvirales Study Group.						-	
Date first submitted to ICTV:			to Study Group Chair Feb. 3, 2010				
			CO	mmunicate	d to SG Fe	eb. 9, 2010	
			Fii	nal SG vote	complete	d April 7,	
			20	10.			
Date of this revision (if different to above).			Im	June 4 2010			

MODULE 2: **NEW SPECIES**

creating and naming one or more new species.

If more than one, they should be a group of related species belonging to the same genus. All new species must be placed in a higher taxon. This is usually a genus although it is also permissible for species to be "unassigned" within a subfamily or family.

Code 20	10.014aV	(assigned by ICTV officers)				
To create 1	new species within:					
		Fill in all that apply. If the higher taxon has yet to be				
Genus		created (in a later module, below) write				
Subfamily: Alphaherpesvirinae Family: Herpesviridae		"(new)" after its proposed name.				
		If no genus is specified, enter				
Order	:: Herpesvirales	"unassigned" in the genus box.				
And name the new species:						
Anatid herpesvirus 1						
•						

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

- Explain how the proposed species differ(s) from all existing species.
 - 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.
- Provide accession numbers for genomic sequences
- Further material in support of this proposal may be presented in the Appendix, Module 9

The complete genome sequence (158091 bp) of duck enteritis virus (DEV), also known as duck plague virus, has been published (EU082088; Li *et al.*, 2009). DEV has for decades been positioned informally as an unclassified virus in the family *Herpesviridae* under the name anatid herpesvirus 1 (denoted by convention after the host family, Anatidae). Herpesviruses are defined as distinct species if (a) their genomes differ in a readily assayable and distinctive manner across the entire genome (e.g. restriction endonuclease cleavage site patterns obtained with many enzymes) and not merely at a specific site (e.g. small number of genes or small number of restriction endonuclease sites) and (b) if the virus can be shown to have distinct epidemiologic and biologic characteristics. DEV fulfils these criteria.

Li *et al.* (2009) concluded from genomic and phylogenetic analyses that DEV should be classified in the subfamily *Alphaherpesvirinae*, but were unable to decide on a genus. They described the taxonomic position of DEV as 'osculant' (situated between and connecting two groups of organisms), a term that we take in this context to be a euphemism for 'undetermined'. The important findings were as follows.

- 1. DEV has a genome structure (in terms of repeated and unique sequences) that has been found to date among members of the genera *Varicellovirus* and *Iltovirus*.
- 2. In U_L , the gene complement includes two genes (encoding protein LORF4 and lipase) that are specific to the genus *Mardivirus* and none that are specific to any other genus.
- 3. In U_S , the gene complement includes one gene (SORF3) that is found only in members of the genera *Mardivirus* and *Iltovirus*, which contain all classified bird herpesviruses. It also includes genes US4 and US5, which are absent from viruses of the genus *Mardivirus* and present in the other genera in the subfamily *Alphaherpesvirinae*.
- 4. In neighbour-joining (NJ) trees based on single gene analyses, the DNA polymerase, thymidine kinase and deoxyuridine triphosphatase were most closely related to members of the genus *Mardivirus*. However, ICP4 grouped with members of the genus *Varicellovirus*, as did glycoprotein D (although the resolution was not convincing). A more recent analysis of glycoproteins D, E and J, again conducted by an NJ method, indicated grouping of DEV with members of the genus *Mardivirus* (Zhao and Wang, 2010).

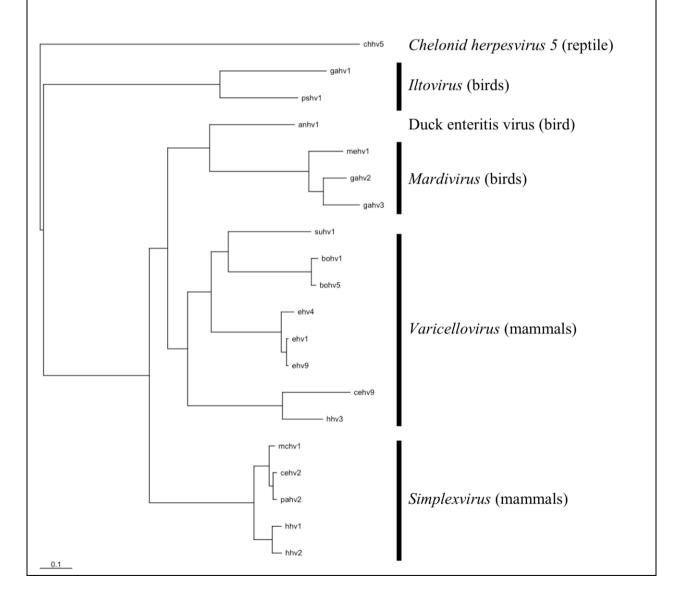
In light of these findings, the uncertainty about assigning DEV to a genus is understandable. However, assignments of herpesvirus species to genera are based primarily on molecular phylogeny and not other genome features. To generate a discriminating phylogeny, we made amino acid sequence alignments for six large, well conserved genes from DEV and 19 members of the subfamily *Alphaherpesvirinae*. The genes were those encoding the single-stranded DNA-binding protein, glycoprotein B, the major capsid protein, DNA polymerase and two subunits of the DNA packaging terminase (genes UL29, UL27, UL19, UL30, UL15 and UL28, respectively). The alignments were trimmed, degapped and concatenated, yielding a final length of 5104 residues. From previous experience, this is a substantial length of alignment, appropriate to deriving a robust phylogenetic tree.

An initial NJ tree (not shown) yielded the standard branching pattern for the subfamily *Alphaherpesvirinae* (McGeoch *et al.*, 2000) with the exception that the branching order of the genera *Simplexvirus* and *Mardivirus* was reversed. This exception affects neither the current classification nor this proposal. In the tree, DEV fell into a clade representing the genus *Mardivirus*. Bootstrap values (100 replicates) supported all nodes maximally except that defining the branching order of genera *Simplexvirus* and *Mardivirus*. A compute-intensive Bayesian Monte Carlo Markov chain (program MrBayes) was then conducted for one million generations, sampling every 100 and imposing a burn-in

value of 5001. The topology of this tree (Figure 1) was the same as that of the NJ tree, with all nodes exhibiting maximal posterior probability and a turtle virus (chelonid herpesvirus 5) falling as the outgroup. The distance between DEV and its closest relatives among members of the genus *Mardivirus* is similar to that between members of another genus, *Varicellovirus*.

We recommend that DEV should be classified as a new species *Anatid herpevirus 1* in the genus *Mardivirus*, being added to the existing members *Gallid herpesvirus 2*, *Gallid herpesvirus 3* and *Meleagrid herpesvirus 1*. We do not think that there is a case for classifying DEV into any other genus or a new genus.

Figure 1. Midpoint-rooted Bayesian phylogenetic tree of DEV (anatid herpesvirus 1) and 19 members of the subfamily *Alphaherpesvirinae*. The prefixes for herpesvirus names are: ch, chelonid; ga, gallid; ps, psittacid; an, anatid; me, meleagrid; su, suid; bo, bovine; e, equid; ce, cercopithecine; h, human; mc, macacine; pa, papiine. The virus, genus or species names are shown, and the host groups that the viruses infect are indicated in parentheses.



MODULE 9: APPENDIX: supporting material

additional material in support of this proposal

References:

- Li, Y., Huang, B., Ma, X., Wu, J., Li, F., Ai, W., Song, M. & Yang, H. (2009). Molecular characterization of the genome of duck enteritis virus. Virology 391, 151-161.
- McGeoch, D. J., Dolan, A. & Ralph, A. C. (2000). Toward a comprehensive phylogeny for mammalian and avian herpesviruses. J. Virol. 74, 10401-10406.
- McGeoch, D. J. & Gatherer, D. (2005). Integrating reptilian herpesviruses into the family Herpesviridae. J. Virol. 79, 725-731.
- Zhao, Y. & Wang, J.W. (2010). Characterization of duck enteritis virus US6, US7 and US8 gene. Intervirology 53, 141-145.

Annex:

Include as much information as necessary to support the proposal, including diagrams comparing the old and new taxonomic orders. The use of Figures and Tables is strongly recommended but direct pasting of content from publications will require permission from the copyright holder together with appropriate acknowledgement as this proposal will be placed on a public web site. For phylogenetic analysis, try to provide a tree where branch length is related to genetic distance.