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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

MODULE 1: TITLE, AUTHORS, etc

Code assigned:	2012.001	a-dV		(to be con officers)	mpleted by	ICTV
Short title: create 3 species order <i>Mononegavirales</i> (e.g. 6 new species in the genus A Modules attached (modules 1 and 9 are required)	s and 1 genus, Zetavirus)	, Perhabdo 1 🔀 6 🗌	<i>ovirus,</i> in 2 ⊠ 7 □	the famil 3 ⊠ 8 □	y <i>Rhabdo</i> 4 □ 9 ⊠	viridae, 5 🗌

Author(s) with e-mail address(es) of the proposer:

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 (fungal, invertebrate, plant, prokaryote or vertebrate viruses)	A list of study groups and contacts is provided at <u>http://www.ictvonline.org/subcommittees.asp</u> . If in doubt, contact the appropriate subcommittee chair (fungal, invertebrate, plant, prokaryote or vertebrate viruses).	Rhabdoviridae study group

ICTV-EC or Study Group comments and response of the proposer:

ICTV-EC comments: Modify.

1. Supply the GenBank accession number for Perch rhabdovirus - done.

2. Consider a specific nucleotide sequence divergence range (in a defined gene or

concatenation) to delineate species within the genus - done.

3. Consider an alternative genus name, as Perhavirus is close to Percavirus, which is an established genus in the family Herpesviridae -Done. The genus name has been modified, now *Perhabdovirus*, to reflect the host species and the nomenclature used for the other genera in the family *Rhabdoviridae*.

Date first submitted to ICTV:	May 08, 2012
Date of this revision (if different to above):	18 October 2012

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. Wherever possible, provide sequence accession number(s) for one isolate of each new species proposed.

Code	201	2.001aV	(assigned by ICTV officers)			
To creat	te 3 ne	ew species within:				
				Fill in all that apply.		
Ge	enus:	Perhabdovirus (ne	w)	If the higher taxon has yet to be		
Subfar	mily:			"(new)" after its proposed name		
Far	mily:	Rhabdoviridae		 If no genus is specified, enter 		
0	rder:	Mononegavirales		"unassigned" in the genus box.		
And name the new species: R			Representat accession nu	epresentative isolate(s) and GenBank sequence ccession number(s):		
Perch rhabdovirus Pe		Perch rhabdo	erch rhabdovirus (PRV) JX679246			
Anguillid rhabdovirus Sea trout rhabdovirus		Eel virus Europ Eel virus Amer Lake trout rh	Eel virus European X (EVEX) FN557213, Eel virus American (EVA) JX827265 Lake trout rhabdovirus 903/87 (LTRV-903-87)			
			Swedish sea	wedish sea trout virus 28/97 (SSTV-28/97) AF434991		

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**.
 - 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

Viruses described in the published literature as perch rhabdovirus (PRV), eel virus European X (EVEX) and eel virus American (EVA), and the lake trout rhabdovirus 903/87 (LTRV-903/87) are rhabdoviruses isolated from the perch (*Perca fluviatilis*), eel (*Anguilla anguilla* and *Anguilla rostrata*), European lake trout (*Salmo trutta lacustris*), respectively (Betts et al., 2003; Dorson et al., 1984; Hill et al., 1980, Johansson et al., 2001).

The complete N, P, M, G and L gene sequences are available for the Eel virus European X (FN557213, JX827265) and the original perch rhabdovirus isolate from 1984 (JX679246).

The complete N, P, M, G gene sequences and partial L gene sequence are published for Lake trout rhabdovirus 903/87 (AF434991) and the Swedish sea trout virus 28/97 (AF434992) (Johansson et al., 2001, 2002).

The perch rhabdovirus (PRV), lake trout rhabdovirus 903/87 (LTRV 903/87), Swedish sea trout virus isolate 28/97 (SSTV 28/97), and Anguillid rhabdovirus isolates eel virus European X (EVEX) and eel virus American (EVA) share morphological characteristics, genome organization and sequence similarities with vesiculoviruses

and with viruses in the newly proposed genus *Sprivivirus* (proposal under revision). However, there are sufficient differences in the viral hosts, virus replication temperatures, gene sequences, and phylogenetic placement between these viruses and other viruses currently assigned to the *Vesiculovirus* genus or the newly proposed genus *Sprivivirus* to consider PRV, EVEX and LTRV as representative virus isolates of three new virus species, *Perch rhabdovirus*, *Anguillid rhabdovirus* and *Sea trout rhabdovirus* in a new genus of aquatic rhabdoviruses, *Perhabdovirus*.

LTRV 903/87 shares 72% and 65% N gene nucleotide identity with PRV and EVEX respectively, and PRV and EVEX shares 64% nucleotide identity in the N gene. Similar divergence in nucleotide sequence exists between these viruses in the P, M, G and L gene.

LTRV-903/87 and Swedish sea trout rhabdovirus share the high degree of nucleotide sequence and deduced amino acid similarity throughout the entire genome and they are considered to belong to the same species, *Sea trout rhabdovirus*. Based on partial L gene sequences (216nt) the Eel virus European X (EVEX) and Eel virus American (EVA) share a high degree of nucleotide sequence identity and are considered belonging to the same new species, *Anguillid rhabdovirus* (Figure 1).

The related Siniperca chuatsi rhabdovirus (SCRV; Tao et al., 2007), isolated from mandarin fish or Chinese perch (*Siniperca chuatsi*) shares some sequence similarity throughout the genome and intergenic regions, Sequence analysis of SCRV has revealed a gene encoding a small hydrophobic protein between the M and G genes that is not present in PRV, EVEX or LTRV 903/87. Due to this finding, and a relatively low level of nucleotide sequence identity with PRV, EVEX and LTRV 903/87 in the N gene of <55%, it is suggested that SCRV is not considered a species within the *Perhabdovirus* genus until sequence information from additional virus isolates becomes available.

MODULE 3: NEW GENUS

creating a new genus

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

Code	201	2.001bV	(assigned by ICTV officers)		
To create	a new	genus within:		Fill in all that apply.	
Subfa	mily:			• If the higher taxon has yet to be created	
Fa	mily:	Rhabdoviridae		(In a later module, below) write "(new)" after its proposed name	
С	Order:	Mononegavirales		 If no family is specified, enter "unassigned" in the family box 	

naming a new genus

Code	2012.001	cV	(assigned by ICTV officers)
To name the new genus: <i>Perhabdovi</i>		Perhabdovir	rus

Assigning the type species and other species to a new genus

Code	2012.001dV	(assigned by ICTV officers)				
To designa	To designate the following as the type species of the new genus					
Perch rhabdovirus Every genus must have a type species. This be a well characterized species although no necessarily the first to be discovered		Every genus must have a type species. This should be a well characterized species although not necessarily the first to be discovered				
The new genus will also contain any other new species created and assigned to it (Module 2) and any that are being moved from elsewhere (Module 7b). Please enter here the TOTAL number of species (including the type species) that the genus will contain:						
3						

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 perch rhabdovirus (PRV), lake trout rhabdovirus 903/87 (LTRV 903/87), Swedish sea trout virus isolate 28/97 (SSTV 28/97), and Anguillid rhabdovirus isolates eel virus European X (EVEX) and Eel virus American (EVA) share morphological characteristics, genome organization and sequence similarities with vesiculoviruses and with viruses in the newly proposed genus *Sprivivirus*. However, there are sufficient differences in the viral hosts, virus replication temperatures, gene sequences, and phylogenetic placement between these viruses and other viruses currently assigned to the *Vesiculovirus* genus or the newly proposed genus *Sprivivirus* to justify the creation of another new genus of aquatic rhabdoviruses, *Perhabdovirus*.

Viruses in the genus *Perhabdovirus* (new) have five major structural proteins designated N, P, M, G and L. The genome is approximately 11.1kb in length with a short leader sequence that precedes the N gene and a short non translated region following the L gene.

The transcriptional start and the termination/polyadenylation signals of 3' UUGUC and 3'

AUNC(U)₆₋₇ respectively are similar to those observed for viruses from other genera within the family *Rhabdoviridae*, however, apart from intergenic region between the N and P genes which is a conserved 3' GAAA the remaining intergenic regions are highly variable in length and in sequence (Figure 2)

Similar to other fish viruses the replication temperature range of the perhaviruses is lower than that of mammalian rhabdoviruses, reflecting the aquatic poikilothermic nature of the host species. Fish viruses are typically isolated on cultured fish cell lines at 15-25°C.

Phylogenetic analysis based on protein sequences consistently places the perch, anguillid, and sea trout rhabdoviruses in a monophyletic lineage that is supported by bootstrap values of >88%, distinct from the lineages corresponding to *Novirhabdovirus* and *Vesiculovirus* genera and from the newly proposed genus *Sprivivirus* irrespective of protein sequence compared (Figure 3, 4).

Origin of the new genus name:

Derived from the name of the type species perch rhabdovirus, Perhabdovirus

Reasons to justify the choice of type species:

The perch rhabdovirus (PRV) infects the broadest range of fish species and of the three proposed species within the new genus the PRV is likely to pose the greatest threat to both cultivated and wild fish populations.

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.

Common with many fish viruses there is no single host fish species for the viruses in each of the three proposed species in the genus *Perhabdovirus*. Although the natural host range for the *Anguillid rhabdovirus* appears to be restricted to the European and Japanese eel (*Anguilla anguilla and Anguilla rostrata*), the *Sea trout rhabdovirus* has been isolated from sea trout (*Salmo trutta trutta*), lake trout (*Salmo truttae lacustris*) and perch (*Perca fluviatilis*), and the *Perch rhabdovirus* infects perch, largemouth bass (Micropterus salmoides), grayling (*Thymallus thymallus*) and pike (*Esox lucius*)

Viruses within species within the genus have been distinguished serologically but the species distinction is more likely to be based on both nucleotide sequence divergence and phylogenetic separation of the viruses (Figure 1). For example, based on partial L gene sequences, strains within a virus delineating a species the nucleotide sequence divergence values range from 7.9% for the sea trout rhabdoviruses to 9.3% for the anguillid rhabdoviruses and 14.2% for the perch rhabdoviruses. The partial L gene nucleotide sequence divergence between species ranges from 18-19% between anguillid rhabdoviruses and sea trout rhabdoviruses, 19-23% between anguillid rhabdoviruses. As a general rule, nucleotide sequence divergence of >17% delineates separate species in the genus, while virus isolates that are at least 85% identical are considered to belong to the same species.

MODULE 9: APPENDIX: supporting material

additional material in support of this proposal

References:

Betts A.M., Stone D.M., Way K., Torhy C., Chilmonczyk S., Benmansour A., and de Kinkelin P. (2003). Emerging rhabdovirus infections of fresh water fish. Dis Aquat. Org. 57:201-212

Dorson M, Torchy C, Chilmonczyk S, Kinkelin P de, Michel C (1984) A rhabdovirus pathogenic for perch (Perca fluviatilis L.): isolation and preliminary study. J. Fish. Dis. 7: 241-245

Hill, B.J., Williams, R.F., Smale, C.J., Underwood, B.O. and Brown, F. (1980) Physicochemical and Serological Characterization of Two Rhabdoviruses Isolated from Eels. Intervirology 1980;14:208-212

Johansson, T., Nylund, S., Olesen, N.J. and Bjorklund, H. (2001) Molecular characterisation of the nucleocapsid protein gene, glycoprotein gene and gene junctions of rhabdovirus 903/87, a novelfish pathogenic rhabdovirus. Virus Res. 80 (1-2), 11-22

Johansson, T., Ostman-Myllyoja, L., Hellstrom, A., Martelius, S., Olesen, N.J. and Bjorklund, H (2002) A novel fish rhabdovirus from sweden is closely related to the Finnish rhabdovirus 903/87. Virus Genes 25 (2), 127-138

Tao, J.J., Gui, J.F. and Zhang, Q.Y. ((2007). Isolation and characterization of a rhabdovirus from co-infection of two viruses in mandarin fish, Aquaculture 262 (1), 1-9

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.

Perch rhabdovirus (new)



Figure 1

The phylogenetic relationships of the different isolates of the species in the proposed genus *Perhabdovirus* based on partial L gene sequences (216 nucleotides) The tree includes sequence data for 6 isolates of *perch rhabdovirus*, 7 isolates of *sea trout rhabdovirus*, and 2 isolates of *anguillid rhabdovirus*. Two related viruses Trees were generated using Neighbor-Joining methods and bootstrap values >70% are shown on the trees.

N-P

P-M

M-G

EVEX	AUACUUUUUUU <u>GAAA</u> UUGUC	AUACUUUUUUU <u>ACAA</u> UUGUC	AUACUUUUUUU <u>GA</u> UUGUC
PRV	AUACUUUUUU GAAAUUGUC	AUACUUUUUU GACAAUUGUC	AUACUUUUUUU <u>GAAA</u> UUGUC
LTRV	AUACUUUUUUUGAAAUUGUC	AUGCUUUUUUU <u>GGUCAA</u> UUGUC	AUACUUUUUUUGACGUUGUC

G-L

EVEX	AUACUUUUUU	JGA	UAAG	SUUGUC
PRV	AUACUUUUUU	G	CAA	UUGUC
LTRV	AUACUUUUUU	GU	CGA	UUGUC

Figure 2

Putative transcriptional termination signal, intergenic region (underlined) and the transcription initiation site highlighting the variations in the intergenic region length (2-6 nt) and sequence.

Figure 3 (following 2 pages)

Phylogenetic relationships between rhabdoviruses based on the complete N protein and using the human parainfluenza virus 1 (HPIV-1) as an out group. The rhabdoviruses used in the analysis were vesicular stomatitis Alagoas virus (VSAV), cocal virus (COCV), vesicular stomatitis Indiana virus (VSIV), vesicular stomatitis New Jersey virus (VSNJV), ISFV, chandipura virus (CHPV), grass carp rhabdovirus (GrCRV), tench rhabdovirus (TenRV), pike fry rhabdovirus (PFRV), European and Asian spring viraemia of carp virus (SVCV Fijan and SVCV Asian), siniperca chuatsi rhabdovirus (SCRV), eel rhabdovirus Eurpoean X (EVEX), perch rhabdovirus (PRV), Swedish sea trout virus (SSTV), European lake trout rhabdovirus (LTRV), Adelaide river virus (ARV), bovine ephemeral virus (BEFV), SIGMAV, mokola virus (MOKV), rabies virus (RABV), Australian bat lyssavirus (ABLV), potato yellow dwarf virus (PYDV), sonchus yellow net virus (SYNV), lettuce necrotic virus (LNYV), Northern, cereal mosaic virus (NCMV), viral haemorrhagic septicaemia virus (VHSV), snakehead rhabdovirus (SHRV), infectious haematopoietic necrosis virus (IHNV) and hirame rhabdovirus (HIRRV). Trees were generated using (a) maximum parsimony and (B) neighbor-joining methods and bootstrap values >70% are shown on the trees.



Figure 3B. Complete N protein, neighbor joining



0.2

Figure 4 (following 4 pages)

Phylogenetic relationships between rhabdoviruses based on the complete (A) P protein (B) M protein (C) G protein and (D) L protein Trees were generated using neighborjoining methods and bootstrap values >70% are shown on the trees. Details of the rhabdoviruses used in the analysis are provided in the legend for figure 3.

Fig. 4A. P protein









0.2

Fig. 4D. L protein



0.2