

## Template for Taxonomic Proposal to the ICTV Executive Committee To create a new SubFamily in an existing Family

Code<sup>†</sup>  To create new subfamilies in the family\*

Code<sup>†</sup>  To name the new subfamilies\*

Code<sup>†</sup>  To designate the following genera as part of the new subfamily:

*Spinareovirinae*  
*Orthoreovirus*  
*Aquareovirus*  
*Coltivirus*  
*Oryzavirus*  
*Fijivirus*  
*Mycoreovirus*  
*Cypovirus*  
*Idnoreovirus*  
*Dinovernavirus*

*Sedoreovirinae*  
*Orbivirus*  
*Rotavirus*  
*Seadornavirus*  
*Phytoreovirus*  
*Cardoreovirus*  
*Mimoreovirus*

<sup>†</sup> Assigned by ICTV officers

\* repeat these lines and the corresponding arguments for each genus created in the family

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## Old Taxonomic Order

Order

Family

*Reoviridae*

SubFamily

Genus

*Orthoreovirus*

*Aquareovirus*

*Oryzavirus*

*Cypovirus*

*Fijivirus*

*Coltivirus*

*Mycoreovirus*

*Rotavirus*

*Phytoreovirus*

*Orbivirus*

*Seadornavirus*

*Idnoreovirus*

*Dinovernavirus*

*Cardoreovirus*

*Mimoreovirus*

## New Taxonomic Order

Order

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*Dinovernavirus*

SubFamily

Genus

*Sedoreovirinae*

*Orbivirus*

*Rotavirus*

*Seadornavirus*

*Phytoreovirus*

*Cardoreovirus*

*Mimoreovirus*

## ICTV-EC comments and response of the SG

Decision: Accept proposals 2007.127-130V.01 and move to 02. Although this proposal was generally welcomed, it was strongly suggested that due to the large genomic differences of the RNA-dependent RNA polymerases (Fig. 2 of proposal), two new families instead of subfamilies be created which at present would still be unassigned with regard to an order. The EC suggested to include the latin word for 'turret' (*i.e.* *turris*) into the name of the *Orthoreovirinae*, and find another name for the *Parareovirinae* that was not considered adequate.

The Study group has reflected on the recommendations of EC and would like to maintain the proposal as a creation of two subfamilies within the family *Reoviridae* rather than subdividing it into two families that are unassigned to an order.

Although there are large genomic differences between the polymerases of the turreted and non-turreted "reoviruses", genetic distances of the same magnitude (or even larger distances) can exist within the turreted and non-turreted groups themselves, and even within single genera within each group (e.g. the cypoviruses, seadornaviruses and rotaviruses). Therefore genetic distances alone do not provide an adequate parameter for dividing the existing family into two new families.

Regardless of the genetic distances (which as stated above can be larger within each group [either the turreted or the non-turreted] than between the two groups) phylogenetic clustering, clearly identifies two major groups of viruses: one turreted and one non-turreted.

The coltiviruses cluster with the turreted viruses and recent studies have shown an evolutionary jump between coltiviruses and aquareoviruses (one of the spiked or turreted reoviruses), involving gene duplication, followed by rearrangement of the duplicated genome segment (Mohd Jaafar *et al.*, 2008). This changed the number of segments from 11 (aquareoviruses) to 12 (coltiviruses). This explains the different number of genome segments but the relatively close phylogenetic relationships between these two genera. Initial studies indicated that the coltiviruses are unturreted, a conclusion that is supported by recent electronmicrographs of coltivirus particles, which have failed to detect turrets on the virus core.

The coltivirus core has a double shelled morphology, similar to that of the rotaviruses, orbiviruses and seadornaviruses (*i.e.* the non-turreted viruses). Cryoelectron-microscopy is now being used to confirm these findings. If confirmed, it will indicate that the coltiviruses represent an intermediate between the non-turreted to turreted viruses (or vice versa), and may help to explain how these different groups and genera of viruses evolved.

Genetic, biochemical, phylogenetic and structural analyses clearly show several different relationships that firmly tie the different genera of reoviruses together within the existing family, but these would still allow their grouping into two major sub-families. The separation of the viruses into two distinct families undervalues and potentially hides the significance of the relationships that have been shown to exist between the different genera of viruses within the current family. The study group therefore supports the proposal for dividing this family into two subfamilies. As requested by the Committee we have considered alternative names.

We wish to keep the syllables 'reovirinae' to reflect the origins of the family name, but suggest Spina (spike : latin) , and Sedo (smooth : latin) as prefixes. We therefore suggest the names *Spinareovirinae* and *Sedoreovirinae* as explained below.

### **Argumentation to create a new subfamily:**

The *Reoviridae* represents the largest family of dsRNA viruses. It contains 15 officially recognized genera. Virus members of this family have genomes composed of 9, 10, 11 or 12 segments of linear dsRNA. Member viruses have been isolated from a wide range of mammals, birds, reptiles, fish, crustaceans, marine protists, insects, ticks, arachnids, plants and fungi and include a total of 75 virus species, with ~30 further tentative species reported to date (Brussaard *et al.*, 2004; Mertens *et al.*, 2005). Virus particles have icosahedral symmetry with a diameter of approximately 60 to 85 nm. Their morphology has been studied intensively by X-ray crystallography and cryo-electron microscopy (Prasad *et al.*, 1988; Yeager *et al.*, 1990, 1994; Grimes *et al.*, 1998; Gouet *et al.*, 1999; Hill *et al.*, 1999; Reinisch *et al.*, 2000; Diprose *et al.*, 2001; Nason *et al.*, 2004) and they can contain one, two or three concentric protein layers, identified as 'subcore', 'core' and 'outer capsid', respectively. The inner-capsid layers and proteins are involved primarily in virus assembly and replication, and show a remarkable degree of structural conservation between different genera, exemplified by the subcore shell, which is constructed from 120 molecules of a single protein (Grimes *et al.*, 1998; Reinisch *et al.*, 2000; Mertens, 2004). In contrast, the outer-capsid proteins, which are involved in virus transmission, cell attachment and penetration, show greater variation, reflecting differences in the targeted host species, as well as responses to immune selective pressure by 'neutralizing' antibodies.

However, based on the core structure, the reoviruses can be subdivided into two groups. The 'turreted' viruses have 12 icosahedrally arranged projections (called turrets or spikes) situated on the surface of the icosahedral core particle, one at each of the five-fold axes (e.g. orthoreoviruses or cypoviruses) (Baker *et al.*, 1999; Hill *et al.*, 1999; Nibert & Schiff, 2001). In the case of orthoreoviruses, each turret is constituted of a pentamer of the capping enzyme. In contrast, cores of the 'non-turreted' viruses have a 'protein bilayer' structure, with a smooth or bristly surface appearance (e.g. rotaviruses or orbiviruses) (Grimes *et al.*, 1998; Baker *et al.*, 1999; Mertens *et al.*, 2005).

Phylogenetic analyses of representative members of the distinct genera, using the amino acid sequences of the RNA-dependent RNA polymerase, have shown that members of the separate genera exhibit amino acid sequence identities of less than 30% (Figure 1). There are 2 exceptions: (i) *Rotavirus B*, shows only 22% identity with other rotaviruses (which may justify the recognition of two distinct genera of rotaviruses at some future point, e.g. ortho and pararotaviruses...this point would need further consultation with rotavirologists); (ii) *Aquareovirus* and *Orthoreovirus*, show an amino acid sequence identity of up to 42%. Overall the phylogenetic tree of the polymerase (shown in figure 2), clearly identifies the turreted viruses and non-turreted viruses as members of separate clades. This genetic (and structural) dichotomy supports the separation of the reoviruses into two subfamilies (see below).

Unpublished phylogenetic analyses confirm previous suggestions that that the "reoviruses" have co-evolved with their respective hosts, and that overall these viruses form a single monophyletic group. However, this group has been evolving for over ~550 million years, which has resulted in sequence divergence to near randomness and clear structural differences. However, the replication mechanisms of these viruses and certain structural parameters of the internal proteins (of the subcore shell and polymerase complexes) remain fundamentally similar. The unpublished evolutionary data suggest that the 'non-turreted' viruses may be the ancestral lineage from which the 'turreted' viruses have evolved.

It is noteworthy that closer relations could be identified from 'signature' sequences or by structural analyses of the proteins, between certain of the genera of the *Reoviridae*. For instance among the non-turreted viruses, closer relationships exist between the rotaviruses and seadornaviruses, which could be identified by sequence analysis of the outer coat proteins, polymerase, capping enzyme and structural analyses of outer coat proteins (Mohd Jaafar et al., 2005a, 2005b and 2005c).

These analyses suggest that seadornaviruses and rotaviruses are genetically linked and that an evolutionary jump has occurred, possibly through gene duplication followed by rearrangement, which would explain the different number of genome segments (11 for the rotavirus and 12 for the seadornaviruses).

Similarly within the turreted group, close relationships can also be identified between members of distinct genera. Particularly the aquareoviruses and the orthoreoviruses which have homologous proteins exhibiting up to 42% amino acid identity (Attoui *et al.*, 2001). Their morphology, as revealed by cryo-electron microscopy, is super-imposable.

Within this group a similar situation to that of the rotaviruses and the seadornaviruses, was identified between the aquareoviruses and coltivirus. Gene duplication, followed by rearrangement may have occurred, changing the number of segments from 11 (aquareoviruses) to 12 (coltivirus) which would again explain their different number of genome segments.

We propose the creation of two subfamilies called *Spinareovirinae* and *Sedoreovirinae*, within the existing family *Reoviridae*. The name *Spinareovirinae* is derived from Spina (Spina; latin for spike: used as an alternative to turreted, particularly in early research with the cypoviruses).

The name *Sedoreovirinae* will be used to identify the subfamily containing the non-turreted virus genera and is derived from the latin word "Sedo" which means smooth to denote the absence of spikes or turrets from the core particles of these viruses which have a relatively smooth morphology.

### **Origin of the proposed subfamily name**

The name *Spinareovirinae* will be used to identify the subfamily containing the spiked or turreted viruses and is derived from 'reovirus' and the latin word 'spina' as a prefix, which means spike, denoting the presence of spikes or turrets on the surface of the core particles. The term 'spiked' is an alternative to 'turreted', that was used in early research to describe the structure of the particle, particularly with the cypoviruses.

The name *Sedoreovirinae* will be used to identify the subfamily containing the non-turreted virus genera and is derived from 'reovirus' and the latin word 'sedo', which means smooth, denoting the absence of spikes or turrets from the core particles of these viruses, which have a relatively smooth morphology.

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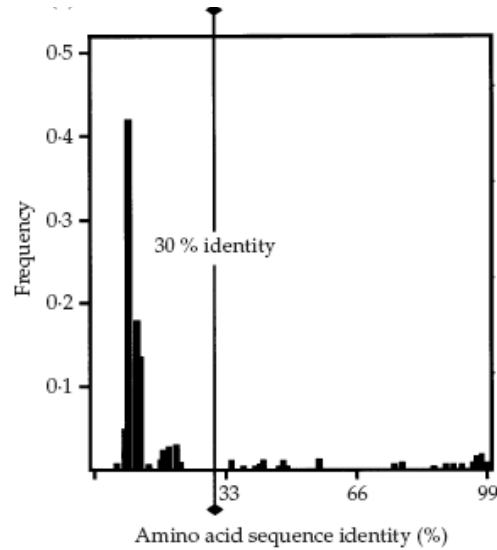
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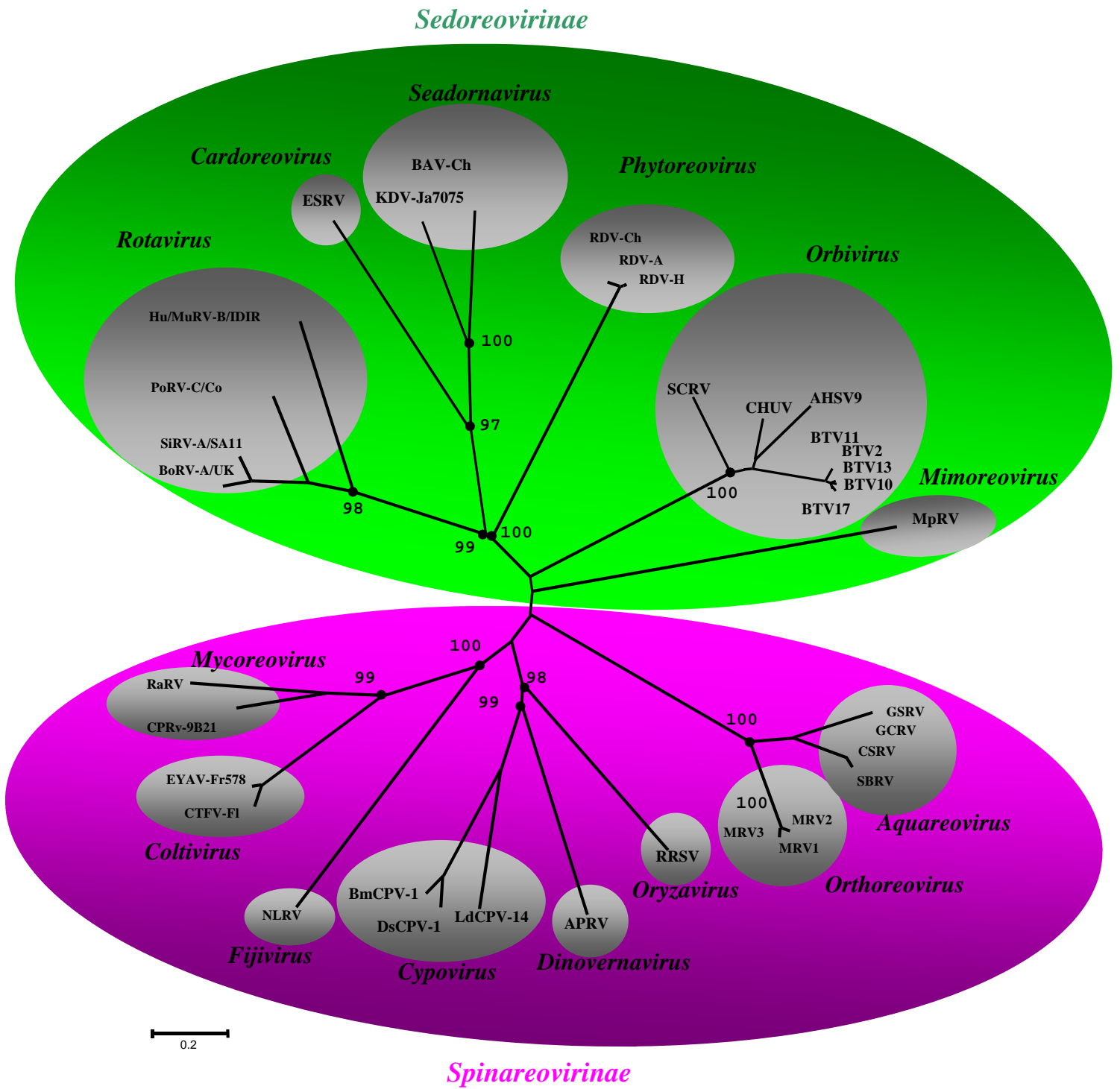
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## Annexes:



**Figure 1:** Frequency distribution histogram of amino acid sequence identities between polymerases of members of the *Reoviridae*. Sequence analysis included polymerase sequences of 34 viruses. The vertical line at 30% amino acid sequence identity indicates the limits of distinction between genera except for *Rotavirus B*, which is 22% identical to other rotaviruses, and aquareoviruses, which show 40–42% amino acid sequence identity to orthoreoviruses.





**Figure 2:** radial neighbour-joining phylogenetic tree showing the turreted and non-turreted clades of family *Reoviridae*. The branching is supported by bootstrap values higher than 97%.