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:	***2011.009a-mP***	(to be completed by ICTV officers)

Short title: Divide the genus *Necrovirus* into 2 new genera, *Alphanecrovirus* and *Betanecrovirus*
(e.g. 6 new species in the genus *Zetavirus*)

Modules attached (modules 1 and 9 are required)	**1** ☒	**2** ☐	**3** ☒	**4** ☐	**5** ☐
	6 ☐	**7** ☒	**8** ☒	**9** ☒	

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

D'Ann Rochon (dann.rochon@agr.gc.ca) on behalf of the Tombusviridae Study Group

List the ICTV study group(s) that have seen this proposal:

	Tombusviridae

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

Date first submitted to ICTV: 5 August 2011
Date of this revision (if different to above):

MODULE 3: **NEW GENUS**

creating a new genus

Code	***2011.009aP***	(assigned by ICTV officers)

To create a new genus within:

Subfamily:	
Family:	***Tombusviridae***
Order:	

naming a new genus

Code	***2011.009bP***	(assigned by ICTV officers)

To name the new genus: *Alphanecrovirus*

Assigning the type species and other species to a new genus

Code	***2011.009cP***	(assigned by ICTV officers)

To designate the following as the type species of the new genus:

Tobacco necrosis virus A

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:

Background:

The current genus *Necrovirus* consists of 7 species: tobacco necrosis virus A (TNV-A; the type member), olive mild mosaic virus (OMMV), olive latent virus-1 (OLV-1), leek white stripe virus (LWSV), tobacco necrosis virus-D (TNV-D), beet black scorch virus (BBSV) and chenopodium necrosis virus (ChNV). Virus particles are ~28nm spheres which encapsidate an approximate 3.7 kb RNA genome. All necroviruses are soil-transmitted and most are transmitted by *Olpidium spp.* Necroviruses differ from other viruses in the family *Tombusviridae* in genome organization and their coat proteins are shorter, lacking a C-terminal protruding domain. Complete genome sequences for all but ChNV are known. ORF 1 encodes the replicase with the polymerase domain being present 3' of a readthrough codon. ORFs 2 and 3 are small centrally located ORFs that encode the movement proteins, MP1 and MP2. ORF 4 lies in the 3' terminal region and encodes the coat protein. A fifth small ORF of unknown function is encoded by some species.

Phylogenetic analyses:

The polymerase, MP1 and MP2 proteins of the 6 necroviruses sequenced fall into two phylogenetically distinct groups; one consisting of TNV-A, OMMV and OLV-1 and the other consisting of LWSV, TNV-D and BBSV (Figs. 1,2), supporting the division of this genus into two separate genera. Unlike the polymerase, MP1 and MP2 proteins, the coat proteins of the 6 viruses group together.

Genetic distance:

1) Polymerase: The polymerases of BBSV, TNV-D and LWSV share 57-69% aa sequence identity with each other but only 32-34% identity with the other 3 sequenced necroviruses. Similarly, the polymerases of OMMV, TNV-A and OLV-1 share 90-92% aa sequence identity with each other but only 32-34% identity with the other necroviruses. The two groups of necroviruses are approximately as distant from each other as they are to other *Tombusviridae* members (Table 1).

2) Movement protein 1 (MP1): The percent aa sequence identity in MP1 sequences further supports the formation of two distinct genera. The MP1 sequences of BBSV, TNV-D and LWSV share 39-49% aa sequence identity with each other but only 6-21% identity with the other 3 necroviruses. Similarly, the MP1 sequences of OMMV, TNV-A and OLV-1 share 86-90% aa sequence identity with each other but only 6-21% identity with the other necroviruses. The MP1 sequences of the two groups of necroviruses are approximately as distant from each other as they are to other *Tombusviridae* members (Table 2).

3) Movement protein 2 (MP2): The percent aa sequence identity in MP2 further supports the formation of two distinct genera from the current necroviruses. MP2 sequences of BBSV, TNV-D and LWSV share 39-47% aa sequence identity with each other but only 10-25% identity with the other 3 necroviruses. Similarly, MP2 sequences of OMMV, TNV-A and OLV-1 share 98-100% aa sequence identity with each other but only 10-25% identity with the other necroviruses. The two groups of necroviruses are approximately as distant from each other as they are to other *Tombusviridae* members (Table 3).

4) Coat protein: The coat proteins of the necroviruses do not fall into separate groups based on aa sequence identity. Percent sequence identity ranges from 26-86%. It is notable that TNV-D in the proposed genus *Betanecrovirus* and OMMV in the proposed genus *Alphanecrovirus* share 86% aa sequence identity and are serologically cross-reactive (Cardoso et al., 2005). On the other hand, OLV-1 (*Alphanecrovirus*) is serologically related to both TNV-A (same genus) and TNV-D (*Betanecrovirus*) (Martelli *et al*. (1996).

Phylogenetic considerations: It is likely that the similarity in coat protein sequences between the 2 groups is a result of recombination events between members of the two groups as suggested for OLV-1 which has been proposed to be a recombinant between OMMV and TNV-D (Cardoso et al., 2005).

The position of ChNV:

This virus was described by Tomlinson et al (1983; Ann. Appl. Biol. 102: 135-147). They felt that the virus might be distinct from existing species because it did not cause local lesions in two hosts that normally give local lesions when inoculated with various TNV isolates and it also went systemic in plants in which TNV isolates often don't go systemic. ChNV was serologically identical to TNV-D but that does not help to determine whether it is an alpha or betanecrovirus. Until sequence data become available, we propose that it should become a species unassigned within the family.

Summary of the proposed changes:

Current species in genus *Necrovirus*	**Proposed new status**
Tobacco necrosis virus A (type)	*Alphanecrovirus* (type)
Olive mild mosaic virus	*Alphanecrovirus*
Olive latent virus 1	*Alphanecrovirus*
Tobacco necrosis virus D	*Betanecrovirus* (type)
Beet black scorch virus	*Betanecrovirus*
Leek white stripe virus	*Betanecrovirus*
Chenopodium necrosis virus	Unassigned *Tombusviridae*

Summary of SG views on this proposal: The high level of sequence diversity in the polymerase, MP1 and MP2 sequences strongly suggests that consideration be given to the proposal that the current genus be divided into 2 genera. This proposal was supported by 4 of the SG members. Two of the SG members did not support this proposal believing that the CP similarities and serological cross-reactivities between the two groups should be given greater weight. In addition, one SG member pointed out that the two groups: (i) do not seem to have host type preferences (i.e. monocots versus dicots; herbaceous versus woody); (ii) share the same ecological niche (soil); (iii) have the same epidemiological behaviour; (iv) when known, have the same type of vector (*Olpidium*). Thus, as a group, necroviruses represent a good example of a biological "*unicus*". This raised the question as to whether the molecular divergence in the polymerase should be given such a preponderant weight as to justify two genera?

Origin of the new genus name:

The existing name *Necrovirus* is retained but with alpha- and beta- added to distinguish the two genera

Reasons to justify the choice of type species:

TNV-A is currently the type member of the genus *Necrovirus* and was the first virus to be sequenced in the proposed new genus

Species demarcation criteria in the new genus:

1) Less than 93% aa sequence identity in the polymerase and
2) Less than 55% aa sequence identity in the coat protein

MODULE 3: **NEW GENUS**

creating a new genus

Code	***2011.009dP***	(assigned by ICTV officers)

To create a new genus within:

Subfamily:	
Family:	***Tombusviridae***
Order:	

naming a new genus

Code	***2011.009eP***	(assigned by ICTV officers)

To name the new genus: *Betanecrovirus*

Assigning the type species and other species to a new genus

Code	***2011.009fP***	(assigned by ICTV officers)

To designate the following as the type species of the new genus:

Tobacco necrosis virus D

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:

See reasons given for genus *Alphanecrovirus* above.

Origin of the new genus name:

The existing name *Necrovirus* is retained but with alpha- and beta- added to distinguish the two genera

Reasons to justify the choice of type species:

TNV-D is the best studied of the viruses and was the first to be sequenced.

Species demarcation criteria in the new genus: .

Alphanecrovirus:
1) Less than 93% aa sequence identity in the polymerase and
2) Less than 55% aa sequence identity in the coat protein

MODULE 7: **REMOVE and MOVE**

Part (a) taxon/taxa to be removed or moved

Code	*2011.009gP*	(assigned by ICTV officers)

To remove the following taxon (or taxa) from their present position:

Tobacco necrosis virus A, Olive mild mosaic virus and Olive latent virus 1

The present taxonomic position of these taxon/taxa:

Genus:	***Necrovirus***
Subfamily:	
Family:	***Tombusviridae***
Order:	

If the taxon/taxa are to be abolished (i.e. not reassigned to another taxon) write "yes" in the box on the right	

Reasons to justify the removal:

These species are currently in the genus *Necrovirus*. As the genus will disappear in the proposed reorganization the species need to be removed and assigned to the appropriate new genus as described below.

Part (b) re-assign to a higher taxon

Code	*2011.009hP*	(assigned by ICTV officers)

To re-assign the taxon (or taxa) listed in Part (a) as follows:

Genus:	***Alphanecrovirus* (new)**
Subfamily:	
Family:	***Tombusviridae***
Order:	
Order:	

Reasons to justify the re-assignment:

See module 3, above

MODULE 7: **REMOVE and MOVE**

Part (a) taxon/taxa to be removed or moved

Code	***2011.009iP***	(assigned by ICTV officers)

To remove the following taxon (or taxa) from their present position:

Tobacco necrosis virus D, Beet black scorch virus and Leek white stripe virus

The present taxonomic position of these taxon/taxa:

Genus:	***Necrovirus***
Subfamily:	
Family:	***Tombusviridae***
Order:	

If the taxon/taxa are to be abolished (i.e. not reassigned to another taxon) write "yes" in the box on the right	

Reasons to justify the removal:

These species are currently in the genus *Necrovirus*. As the genus will disappear in the proposed reorganization the species need to be removed and assigned to the appropriate new genus as described below.

Part (b) re-assign to a higher taxon

Code	***2011.009jP***	(assigned by ICTV officers)

To re-assign the taxon (or taxa) listed in Part (a) as follows:

Genus:	***Betanecrovirus* (new)**
Subfamily:	
Family:	***Tombusviridae***
Order:	
Order:	

Reasons to justify the re-assignment:

See module 3, above

MODULE 7: **REMOVE and MOVE**

Part (a) taxon/taxa to be removed or moved

Code	***2011.009kP***	(assigned by ICTV officers)

To remove the following taxon (or taxa) from their present position:

Chenopodium necrosis virus

The present taxonomic position of these taxon/taxa:

Genus:	***Necrovirus***
Subfamily:	
Family:	***Tombusviridae***
Order:	

If the taxon/taxa are to be abolished (i.e. not reassigned to another taxon) write "yes" in the box on the right	

Reasons to justify the removal:

This species is currently in the genus *Necrovirus*. As the genus will disappear in the proposed reorganization the species need to be removed and re-assigned.

Part (b) re-assign to a higher taxon

Code	***2011.009lP***	(assigned by ICTV officers)

To re-assign the taxon (or taxa) listed in Part (a) as follows:

Genus:	**unassigned**
Subfamily:	
Family:	***Tombusviridae***
Order:	
Order:	

Reasons to justify the re-assignment:

Biological data suggest that Chenopodium necrosis virus is closely related to necroviruses, but without sequence data the only sensible option is to place it unassigned within the family for the present.

MODULE 7: **REMOVE and MOVE**

Part (a) taxon/taxa to be removed or moved

Code	***2011.009mP***	(assigned by ICTV officers)

To remove the following taxon (or taxa) from their present position:

Genus *Necrovirus*

The present taxonomic position of these taxon/taxa:

Genus:	***Necrovirus***
Subfamily:	
Family:	***Tombusviridae***
Order:	

If the taxon/taxa are to be abolished (i.e. not reassigned to another taxon) write "yes" in the box on the right	**YES**

Reasons to justify the removal:
This genus will disappear in the proposed reorganization.

MODULE 9: **APPENDIX**: supporting material

additional material in support of this proposal

References:

Cardoso J.M., Félix, M.R., Clara, M.I., Oliveira, S. (2005) *Archives of Virology* 150: 815-823.

Martelli, G.P., Yilmaz, M.A., Savino, V., Baloglu, S., Grieco, F., *et al*. (1996). *European Journal of Plant Pathology* 102: 527-536.

Rubino, L. and Martelli, G.P. (2008) Necrovirus. Encyclopedia of Virology, 3rd Edition, pg 403-405.

Figure 1. Distance tree of the polymerase of *Tombusviridae* members highlighting the position of the polymerases of necroviruses which are represented in two distinct lineages. Alignments were conducted using ClustalX 2.1 and trees were generated with the Neighbor Joining algorithm using 1000 bootstrap replicates.

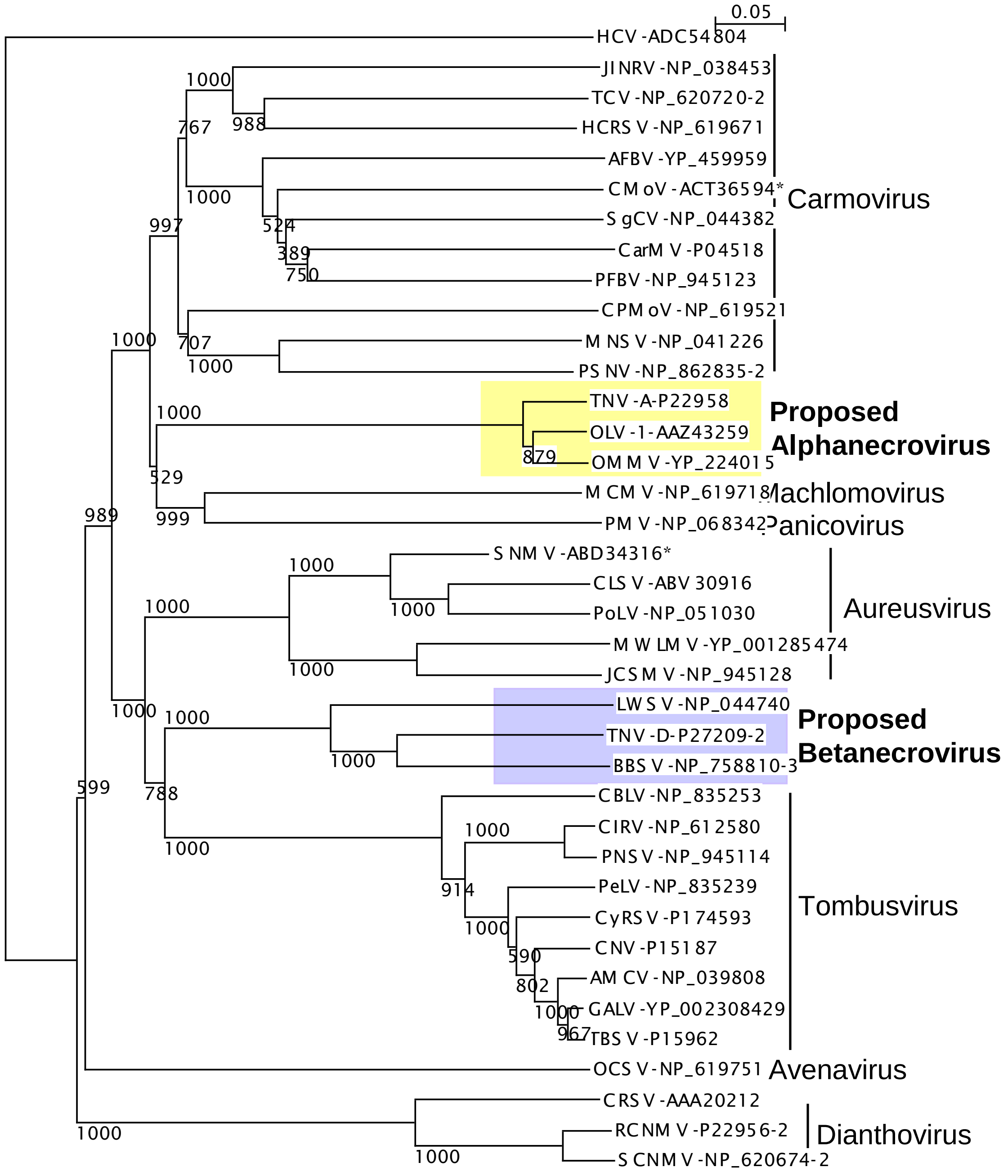

Figure 2. Distance tree of the MP1 (top) and MP2 (bottom) of *Tombusviridae* members highlighting the position of the MPs of necroviruses which are represented in two distinct lineages for both MPs. Alignments were conducted using ClustalX 2.1 and trees were generated with the Neighbor Joining algorithm using 1000 bootstrap replicates.

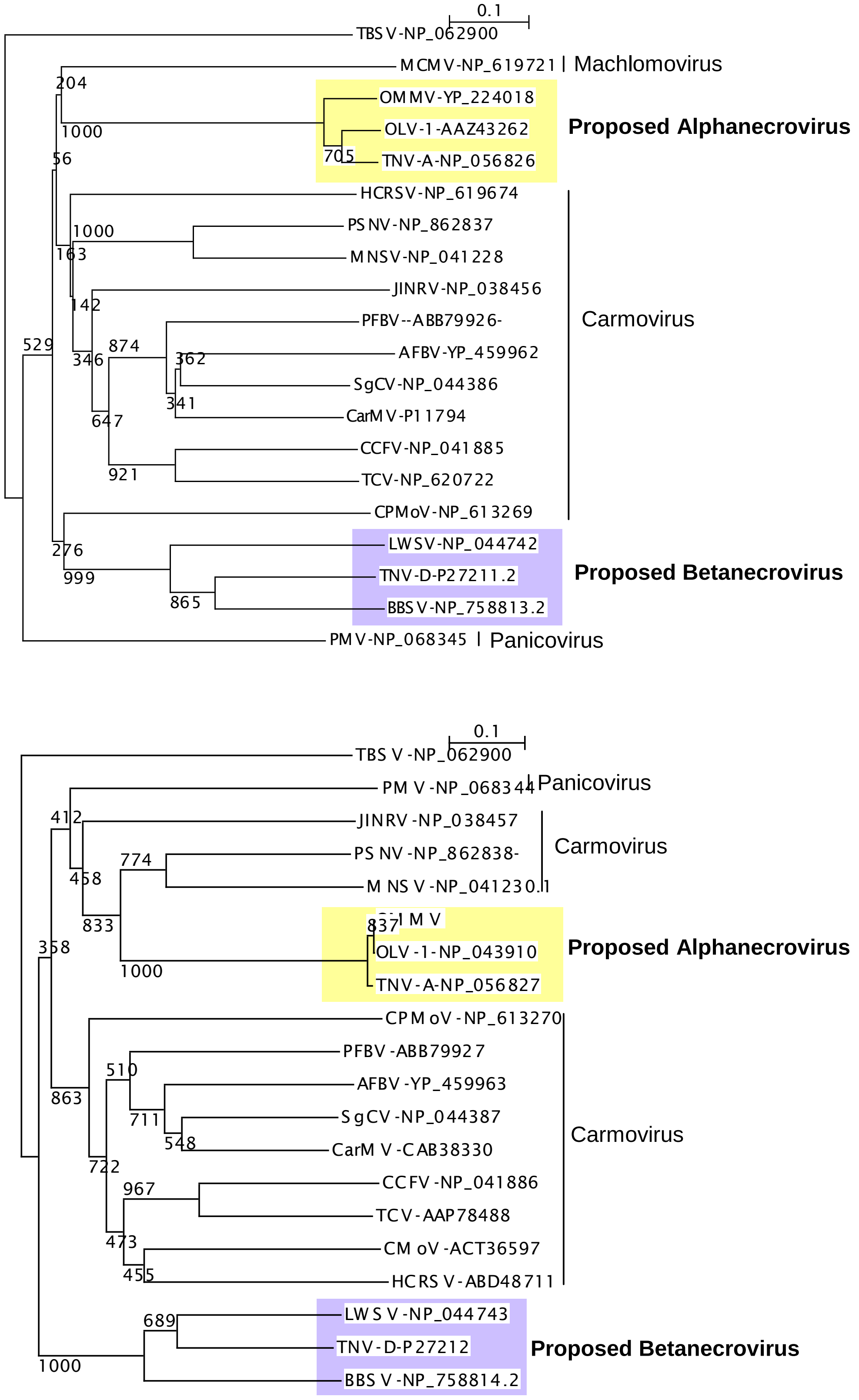

Figure 3. Distance tree of the coat proteins of *Tombusviridae* members highlighting the position of the of necroviruses which cluster in one lineage, unlike what is observed in the polymerase and MP trees. Alignments were conducted using ClustalX 2.1 and trees were generated with the Neighbor Joining algorithm using 1000 bootstrap replicates.

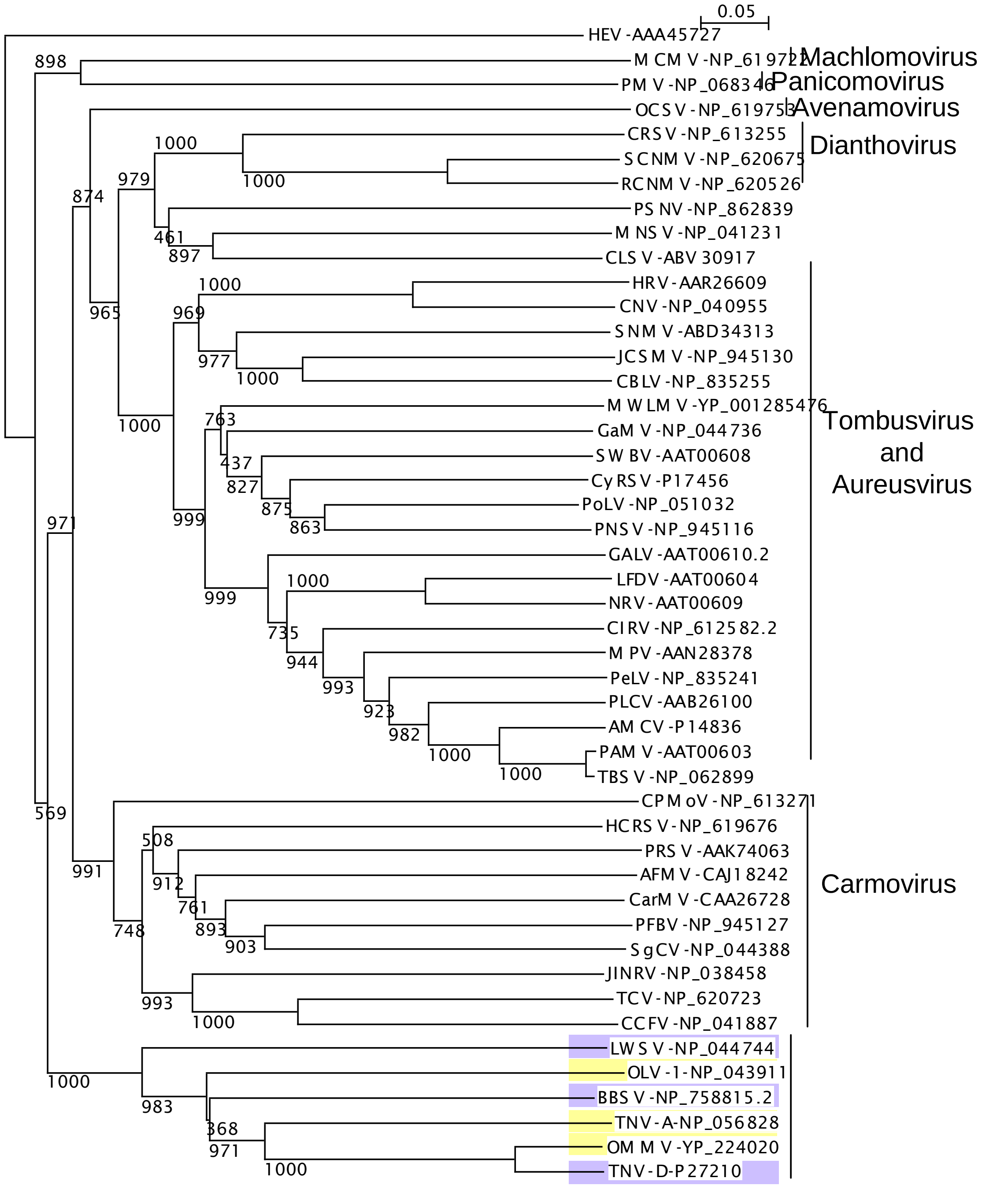

Table 1. Percent amino acid sequence identity between the polymerases of the 6 necroviruses and other *Tombusviridae* members.

	% aa sequence identity in polymerase						
	BBSV	**TNV-D**	**LWSV**	**OMMV**	**TNV-A**	**OLV-1**	**Other Tombus-viridae**
BBSV	-	**69**	**57**	**34**	**34**	**34**	**22-39**
TNV-D		-	**60**	**33**	**34**	**33**	**23-39**
LWSV			-	**32**	**33**	**33**	**22-37**
OMMV				-	**90**	**92**	**25-39**
TNV-A					-	**90**	**25-38**
OLV-1						-	**25-38**
Other Tombus-viridae							-

Table 2. Percent amino acid sequence identity between the MP1 sequences of the 6 necroviruses and those of carmo-, panico and machlomoviruses.

	% aa sequence identity in MP1 movement protein						
	BBSV	**TNV-D**	**LWSV**	**OMMV**	**TNV-A**	**OLV-1**	**Carmo Panico Machlomo**
BBSV	-	**49**	**39**	**20**	**19**	**15**	**9-23**
TNV-D		-	**46**	**21**	**20**	**20**	**8-22**
LWSV			-	**6**	**8**	**8**	**10-23**
OMMV				-	**86**	**87**	**8-22**
TNV-A					-	**90**	**8-32**
OLV-1						-	**11-24**
Carmo Panico Machlomo							-

Table 3. Percent amino acid sequence identity between the MP2 sequences of the 6 necroviruses and those of carmo and panicoviruses

	% aa sequence identity in MP2 movement protein						
	BBSV	**TNV-D**	**LWSV**	**OMMV**	**TNV-A**	**OLV-1**	**Carmo Panico**
BBSV	-	**43**	**39**	**25**	**21**	**25**	**7-26**
TNV-D		-	**47**	**10**	**10**	**10**	**4-21**
LWSV			-	**21**	**21**	**21**	**5-25**
OMMV				-	**98**	**100***	**5-30**
TNV-A					-	**98**	**5-30**
OLV-1						-	**5-30**
Carmo Panico							-

Table 4. Percent amino acid sequence identity between the coat protein sequences of the 6 necroviruses and those of other *Tombusviridae* members.

	% aa sequence identity in coat protein						
	BBSV	**TNV-D**	**LWSV**	**OMMV**	**TNV-A**	**OLV-1**	**Other Tombus-viridae**
BBSV	-	**40**	**34**	**41**	**37**	**38**	**9-21**
TNV-D		-	**26**	**86**	**41**	**38**	**10-19**
LWSV			-	**27**	**31**	**27**	**5-19**
OMMV				-	**45**	**40**	**11-20**
TNV-A					-	**38**	**7-21**
OLV-1						-	**8-18**
Carmo Panico							-