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:	***2013.011a-dP***	(to be completed by ICTV officers)

Short title: Create 1 new species in the genus *Benyvirus* and assign the genus to the new family *Benyviridae*
(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:

David Gilmer (gilmer@unistra.fr) Claudio Ratti (claudio.ratti@unibo.it) Tetsuo Tamada (tyrotamada@oboe.ocn.ne.jp) Ida Bagus Andika (idabagusyf@yahoo.com) Hideki Kondo (hkondo@rib.okayama-u.ac.jp)

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)	Virgaviridae and Benyvirus Study Group

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

Date first submitted to ICTV: 19 June 2013
Date of this revision (if different to above):

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	***2013.011aP***	(assigned by ICTV officers)

To create 1 new species within:

Genus:	***Benyvirus***
Subfamily:	
Family:	***Benyviridae (new)***
Order:	

Fill in all that apply.

- If the higher taxon has yet to be created (in a later module, below) write "**(new)**" after its proposed name.
- If no genus is specified, enter "**unassigned**" in the genus box.

And name the new species:	**GenBank sequence accession number(s) of reference isolate:**
Burdock mottle virus	AB818898 - AB818899

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

The genus *Benyvirus* within the proposed new family *Benyviridae* contains viruses with multipartite ssRNA+ genomes encapsidated in rigid rod shaped particles. The viruses are vectored by fungoid protists in the genus *Polymyxa*. Although some members of the *Virgaviridae* share these properties, benyviruses are unique in having a single ORF on RNA1 that encodes the replication-associated protein (terminating in a Sindbis-like RdRp domain) and in having a capped and polyadenylated multipartite RNA genome. The RdRp (and the entire replication-associated polyprotein) is only distantly related to those possessed by members of the *Virgaviridae*. Benyvirus RNAs share sequence similarities within the last 70 nts preceding the polyA sequence. Benyvirus RNA2 components show similar genomic organization and contain 6 ORFs. The first is the major coat protein of about 21 kDa, the second corresponds to the minor coat protein component produced from a read-through of the first ORF stop codon leading to a CP-RT protein of about 75 kDa. The three following ORFs constitute the "triple gene block" required for cell-to cell movement. The last ORF encodes a cysteine rich protein. *Beet necrotic yellow vein virus* (type species, BNYVV) and *Beet soil-borne mosaic virus* (BSBMV) possess supplemental genomic RNAs. RNA3, RNA4 are smaller RNAs each encoding a single protein with roles in transmission and symptom development. Some BNYVV isolates possess a fifth RNA (RNA5).

The genus currently contains three species. Two of them naturally infect sugar beet, *Beet necrotic yellow vein virus* (type species) and *Beet soil-borne mosaic virus*. These have less than 60% amino acid identity between their coat proteins and about 84% identity between their replication-associated proteins. The third species (*Rice stripe necrosis virus*) infects monocots.
The formal species demarcation criteria are that distinct species should have coat protein

sequences less than 90% identical, distant serological relations and distinct host ranges.

Burdock mottle virus (BdMoV)

BdMoV was first isolated from burdock plants (*Arctium lappa* L.) showing faint leaf chlorosis or mottling in Japan in 1970. It has a narrow host range but can be transmitted by sap inoculation to several commonly used indicator plants. The mode of transmission in the field is not known. The virus has rod-shaped particles and is associated with viroplasm-like inclusions in the cytoplasm of infected plant cells (Inouye, 1973; Hirano et al., 1999).

The complete sequences of RNA1 and RNA2 (probably the complete genome) of an isolate of BdMoV have been determined and these show an organization and expression strategy similar to *Beet necrotic yellow vein virus*, the type member of the genus *Benyvirus*. The genome organization for BdMoV is shown in Annex Figure 1.

Comparative sequence analyses show that the proposed new species, which has a distinct host range, significantly exceeds the molecular demarcation criteria. The most conserved domains within the replication protein are only 62-81% identical to those of beet necrotic yellow vein virus. Phylogenetic trees for the core RdRp domain (Figure 2a) and the CP (Figure 2b) show that BdMoV consistently groups with the other benyviruses. Comparisons amongst the proteins encoded by RNA2 are summarized in Table 1.

Genome organization and some protein comparisons were reported by Hirano et al., 1999 but the sequences have only recently been deposited in GenBank and a peer-reviewed publication is being prepared.

MODULE 5: **NEW FAMILY**

creating and naming a new family

Code	***2013.011bP***	(assigned by ICTV officers)

To create a new family containing the subfamilies and/or genera listed below within the Order: *unassigned*

If there is no Order, write "**unassigned**" here.
If the Order has yet to be created (in Module 6) please write **"(new)"** after the proposed name.

Code	***2013.011cP***	(assigned by ICTV officers)

To name the new family: *Benyviridae*

assigning subfamilies, genera and unassigned species to a new family

Code	***2013.011dP***	(assigned by ICTV officers)

To assign the following genera to the new family:
You may list several genera here. For each genus, please state whether it is new or existing.

- If the genus is new, it must be created in Module 3
- If the genus already exists, please state whether it is currently unassigned or is to be removed from another family. If the latter, complete Module 7 to 'REMOVE' it from that family

***Benyvirus* (currently an unassigned genus)**

The new family will also contain any other new species created and assigned to it (Module 3) and any that are being moved from elsewhere (Module 7b). **Please enter here the TOTAL number of unassigned species that the family will contain (those NOT within any of the genera or subfamilies listed above):**

0

Reasons to justify the creation of the new family:
Additional material in support of this proposal may be presented in the Appendix, Module 9

The genus *Benyvirus* is not currently assigned to any family. As explained in Module 2, there are similarities to viruses in the family *Virgaviridae* in virion morphology and to some virgaviruses in vector transmission and in phylogenetic analyses of the coat protein and triple gene block movement proteins. However, the genus is excluded from the family because of several key differences. In particular, the multipartite genome is capped and polyadenylated (which virgaviruses are not) and there is a single ORF on RNA1 that encodes a distinctive replication-associated protein terminating in a Sindbis-like RdRp domain. In phylogenetic analyses, the benyvirus replication protein is more closely related to those of a number of animal virus genera than it is to the virgaviruses. Not only do the replication-associated proteins of virgaviruses form a quite separate group in phylogenetic analysis but there is a different translation strategy: the RdRp domain of virgaviruses is expressed by translational readthrough of a smaller protein.

Altogether these features favour the creation of a new family containing one genus and four species (See Figure 3A, B and C).

Origin of the new family name:

From the name of the genus *Benyvirus*

MODULE 9: **APPENDIX**: supporting material

additional material in support of this proposal

References:

Inouye, T. (1973). Host range and electron microscopy of burdock mottle virus, a rod-shaped virus from *Arctium lappa* L. : Studies on the viruses of plants in Compositae in Japan. Ber. Ohara Inst. Landw.Biol., Okayama University 15:207-218.

Hirano, S., Kondo, H., Maeda, T., and Tamada, T. (1999). Burdock mottle virus has a high genome similarity to beet necrotic yellow vein virus. In: Proceedings of the Fourth Symposium of the International Working Group on Plant Viruses with Fungal Vectors, Eds. J. L. Sherwood & C. M. Rush, pp. 33-36. Denver: American Society of Sugar Beet Technologists.

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.

Table 1. Comparative amino acid sequence identity analyses of the proteins encoded by RNA2 of current and proposed members of the genus *Benyvirus*. The amino acid sequence identities between their RdRp domains (produced from the RNA1 polyprotein) are also presented.

Viral protein	Identity (%)		
	BdMoV/ BNYVV	BdMoV/ BSBMV	BdMoV/ RSNV
CP	34.6	29.8	28.4
CP-readthrough domain	21.5	20	18.3
TGB1	44	42.1	41.3
TGB2	48.3	50	35.6
TGB3	28.8	25.4	22.1
Cysteine-rich protein	15	16.5	11.6
RdRp	69.8	69.4	60.1

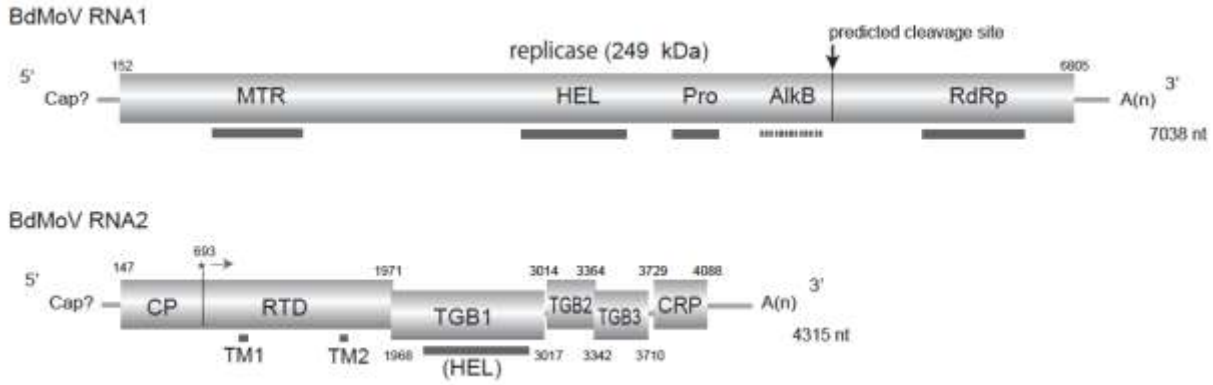
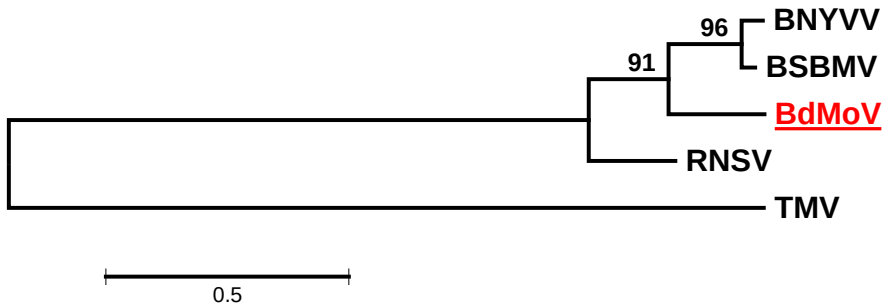
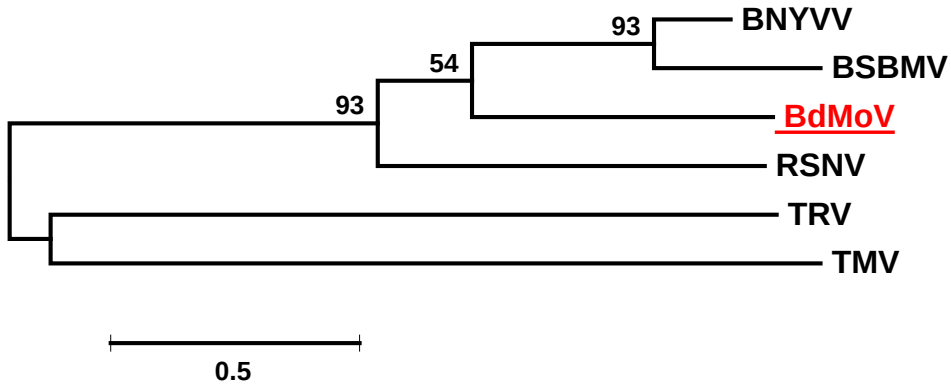


Figure 1: BdMoV genome (From Kondo et al, in preparation). Cap, M^7GpppG; MTR, Methyltransferase; HEL, Helicase; Pro, Protease; AlkB, AlkB-like domain; RdRp, RNA dependent RNA polymerase; CP, Coat protein; RTD, Read-through domain; TGB, triple gene block; CRP, cysteine rich protein.

a RdRp



b CP



c. TGBp1

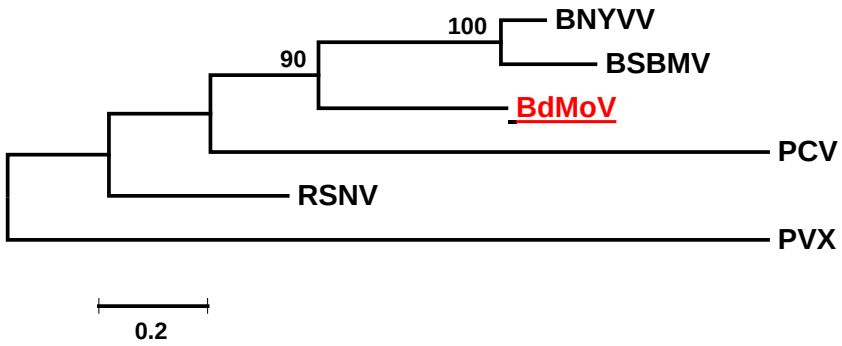
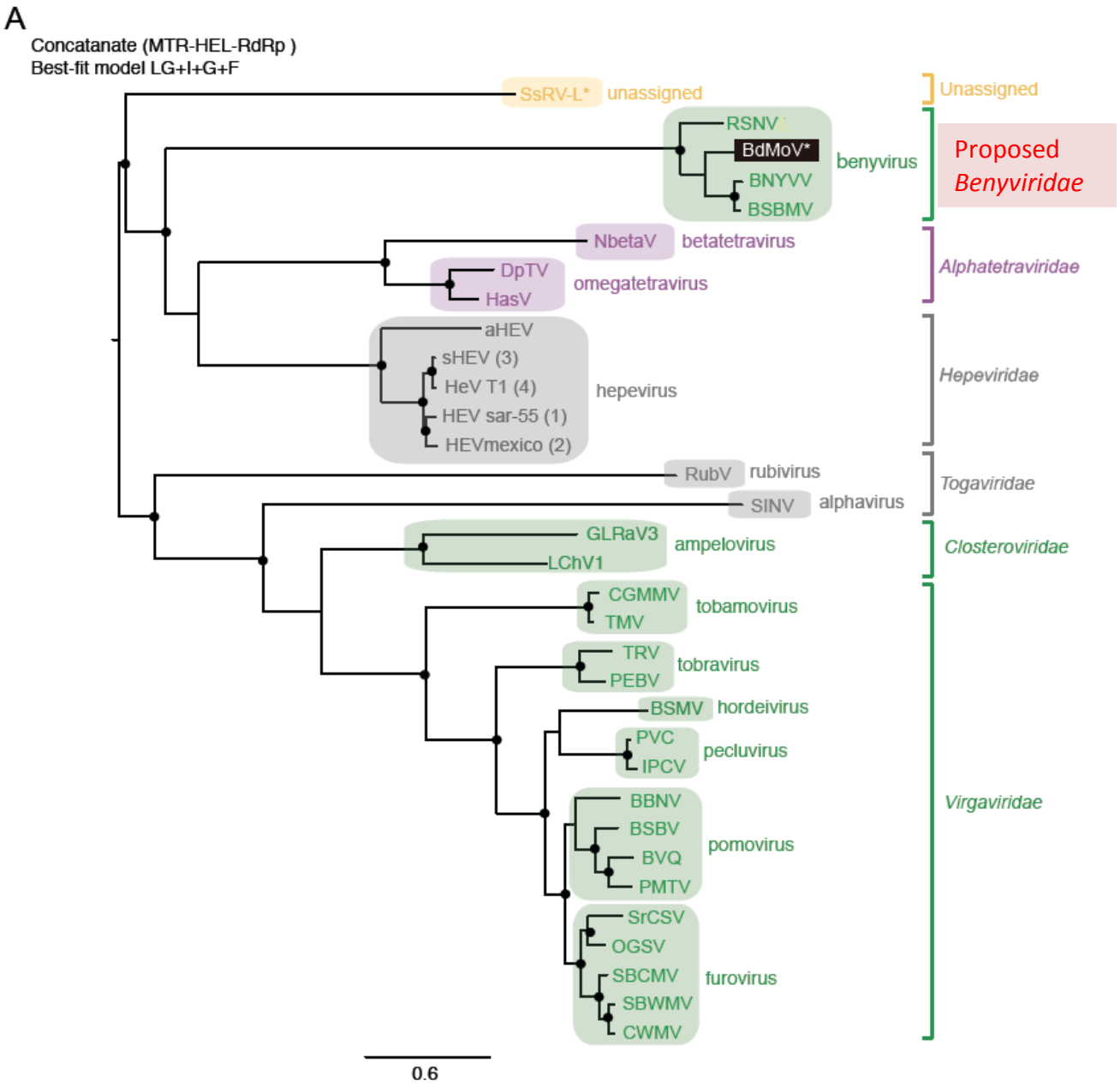


Figure 2: Maximum likelihood analyses of benyviruses based on amino acid sequences of the RdRp motif, CP and triple gene block protein 1 (TGBp1). Amino acid sequences of the core RdRp motif and CP from isolates of beet necrotic yellow vein virus (BNYVV; respective accessions D84410 and D84411), beet soil-borne mosaic virus (BSBMV; AF280539 and AF061869) and rice stripe necrosis virus (RSNV; EU099844 and EU099845) and Burdock mottle virus (BdMoV; AB818898 and AB818899) were aligned with those of tobacco mosaic virus (TMV; genus *Tobamovirus*; NC_001367) and (for the CP) that of tobacco rattle virus (TRV, genus *Tobravirus*; AF034621). TGBp1 proteins were aligned with those of peanut clump virus (PCV; genus *Pecluvirus*; L07269) and potato virus X (PVX; genus *Potexvirus*; YP_002332930). The trees clearly indicate the association of BdMoV to the genus *Benyvirus*. Trees were prepared in MEGA5.2 (JTT model with 1,000 bootstrap replicates).



Legend next page

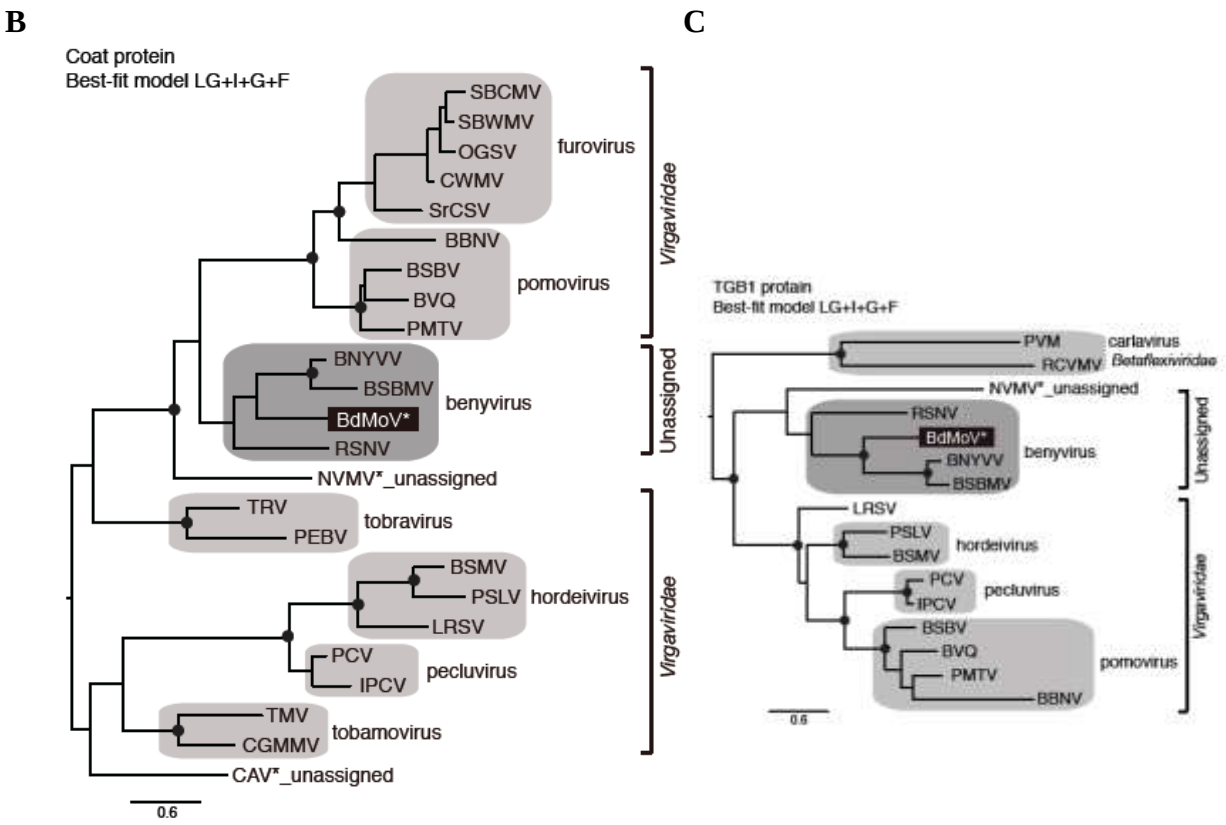


Figure 3 (from Kondo et al., in preparation): Phylogenetic trees calculated from the amino acid sequences of **A:** three concatenated domains (Met–Hel–RdRp) in the replicase proteins of BdMoV and benyviruses together with some other related plant and animal RNA viruses; **B and C:** the coat proteins (B) and TGB1 proteins (C) of BdMoV and benyviruses together with selected rod-shaped viruses included in the family *Virgaviradae*. The alignments of the amino acid sequences were generated with MAFFT and trees were constructed by the ML method using PhyML 3.0. The branch support values were estimated using the approximate likelihood ratio test (aLRT) with a Shimodaira–Hasegawa-like (SH-like) algorithm. The nodes with filled circles are supported by aLRT values greater than 0.9. The scale bars represent the amino acid distances. The sequence data of viruses used for analysis are as follows. **Replicase**: *Virgaviridae*, *Furovirus*, soil-borne wheat mosaic virus (SBWMV): AAA48492, soil-borne cereal mosaic virus (SBCMV): CAB56599, Chinese wheat mosaic virus (CWMV): CAB41769, oat golden stripe virus (OGSV): CAB57882, sorghum chlorotic spot virus (SrCMV): NP_659020, *Pomovirus*, potato mop-top virus (PMTV): CAB58364, beet virus Q (BVQ): CAA11457, beet soil-borne virus (BSBV): CAB10764, broad bean necrosis virus (BBNV): BAA34692, *Pecluvirus*, peanut clump virus (PCV): NP_620047, Indian peanut clump virus (IPCV): NP_835282, *Hordeivirus*, barley stripe mosaic virus (BSMV): AAA46336, AAA66600, *Tobravirus*, tobacco rattle virus (TRV): AAC02066, pea early browning virus (PEBV): CAB37343, *Tobamovirus*, tomato mosaic virus (TMV): NC_001367, cucumber green mottle mosaic virus (CGMMV): NC_001801, *Hepeviridae*, *Hepevirus*, hepatitis E virus (HEV sar-55, Subgroup 1): AAL50058, (HEV mexico, Subgroup 2): AAA45730, (HEV T1, Subgroup 4): CAB83209, swine hepatitis E virus (sHEV, Subgroup 3): ABB88699, avian hepatitis E virus (aHEV): AAS45830; *Togaviridae*, *Rubivrus*, rubella virus (RubV):NP_062883; *Tetraviridae*, *Betatetravirus*, nudaurelia capensis beta virus (NbetaV):NP_048059, *Omegatetravirus*, dendrolimus punctatus virus (DpTV): AAT27317, helicoverpa armigera stunt virus (HasV): AAC98529; unassigned virus, Sclerotinia sclerotiorum RNA virus L (SsRVL) : ACE88957. **CP and TGB1**: *Virgaviridae*, *Furovirus*, SBWMV: NC_002042, SBCMV: NC_002330, CWMV: NC_002356, OGSV: NC_002357, SrCMV: NC_004015, *Pomovirus*, PMTV: NC_003724, NC_003725, BVQ: NC_003511, NC_003512, BSBV: NC_003518, NC_003519, BBNV: NC_004424 NC_004425, *Peculvirus*, PCV: NC_003668, IPCV: NC_004730, *Hordevirus*, BSMV: NC_003481, Poa semilatent virus (PSLV): M81486, Lychnis ringspot virus (LRSV): Z46351. *Tobravirus*, TRV: NC_003811, PEBV: NC_001368, *Tobamovirus*, TMV: NC_001367, CGMMV: NC_001801, *Tymovirales*, *Betaflexiviridae*, *Carlavirus*, potato virus M (PVM): NC_001361, red clover vein mosaic virus (RCVMV): NC_012210; unassigned virus, Nicotiana velutina mosaic virus (NVMV) D00906 Chara australis virus (CAV): JF824737.