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:	***2013.004aP***	(to be completed by ICTV officers)

Short title: One new species in the genus *Cheravirus*
(e.g. 6 new species in the genus *Zetavirus*)

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

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

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and in consultation with Ian Adams (ian.adams@fera.gsi.gov.uk)

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)	Secoviridae

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

Date first submitted to ICTV:	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	**2013.004aP**	(assigned by ICTV officers)

To create 1 new species within:

Genus:	***Cheravirus***
Subfamily:	-
Family:	***Secoviridae***
Order:	***Picornavirales***

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:**
Arracacha virus B	JQ437415 (RNA1) JQ581051 (RNA2)

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

Arracacha virus B virus (AVB) was first reported in 1979 (Kenten and Jones, 1979). The recent full-length genome sequence of the virus (Adams et al., 2013) was obtained for the oca strain of AVB (Jones, 1981; Jones and Kenten, 1981) originally isolated from potato (*Solanum tuberosum*). A total RNA preparation enriched in viral RNA was prepared as follows. *Nicotiana benthamiana* plants were infected with the oca strain of AVB. Clarified extracts were prepared and virus particles were precipitated with a PEG-NaCl solution. Total RNA was extracted from this partially purified virus preparation. 454 sequencing (Roche) was used to sequence cDNA derived from this RNA preparation from oca strain-infected *N. benthamiana*. Sequences were assembled from two separate sequencing runs with no variation in the assembled nucleotide sequence between the two runs, providing good confidence in the validity of the assembly. Comparison of assembled sequences with the cheraviruses *Apple latent spherical virus* (ALSV) and *Cherry rasp leaf virus* (CRLV) produced consensus sequences of 7253 and 3482 nucleotides for RNA1 and 2, respectively. RNA1 is predicted to encode a single polyprotein of 2314 amino acids (259kDa), while RNA2 is predicted to encode a single polyprotein of 102kDa.

Phylogenetic analysis using the deduced amino acid sequence of the RdRp and CP grouped this new virus with members of the genus *Cheravirus* (Adams et al., 2013). To provide further evidence for the grouping of the virus, we have produced a phylogenetic tree by the neighbor-joining method of the MEGA5 software (Tamura et al., 2011) using the deduced amino acid sequence of the Pro-Pol region (region between the conserved CG motif of the proteinase and the conserved GDD motif of the polymerase) (Fig. 1). The Pro-Pol sequence was previously shown to be a useful indicator of taxonomy within the order *Picornavirales* (Le Gall et al., 2008) and within the family *Secoviridae* (Sanfacon et al., 2009). In this analysis, we included all available sequences from the type isolates of members of the family *Secoviridae*. We also included the corresponding sequences from representatives of other families in the order *Picornavirales* and from *Potato virus Y*, a member of the family *Potyviridae*, which was used as an outgroup. This analysis, along with Maximum likelihood and Bayesian derived trees derived from the same dataset (not shown), confirmed the grouping of the virus with other members of the genus *Cheravirus* (Fig. 1). Current species demarcation criteria within the family *Secoviridae* (as defined in the Ninth Report) are: CP amino acid (aa) sequence with less than 75% identity and Pro-Pol region aa sequence with less than 80% identity. For this virus the highest sequence identities with the CP (using a C-terminal 611 amino acid sequence starting at aa 318 of the RNA2 polyprotein and based on the predicted R/G cleavage site) and Pro-Pol regions are 25% and 47%, respectively. A neighbor-joining phylogenetic tree also provided significant evidence (bootstrap >70%) for AVB being monophyletic with the ALSV and CRLV (Adams et al., 2013).

No vectors are known to transmit AVB, although it is easily transmitted by mechanical inoculation and also through true seed and pollen of potato. It enters most tubers produced by infected plants (Jones, 1982). There are two strains described in the literature: Oca and T. They vary in host range, symptomatology in indicator hosts and are serologically only distantly related (Jones and Kenten, 1981).

Virus particles are isometric with an approximate diameter of 26nm. $CsCl_2$ gradient centrifugation of purified particles identified two components consisting of equal quantities of RNA, and protein of two resolvable sizes, 20 and 26kDa; the latter being three times more abundant (Kenten and Jones, 1979). The three cheraviruses so far identified, ALSV, CRLV and *Stocky prune virus* (StPV) all code for three coat proteins. Alignment of the RNA2 polyproteins of ALSV and CRLV with AVB suggest comparable predicted cleavage products with three CPs of predicted 25.8, 20.6 and 21.5 kDa; the discrepancy with the information obtained by electrophoresis being most probably due to low resolution of the gel as previously observed for other cheraviruses.

Taken together, this information indicates that AVB should be considered a distinct species within the genus *Cheravirus*.

MODULE 9: **APPENDIX**: supporting material

additional material in support of this proposal

References:

Adams, I.P., Glover, R., Souza-Richards, R., Bennett, S., Hany, U., Boonham, N., 2013. Complete genome sequence of arracacha virus B: a novel cheravirus. Arch Virol 158(4), 909-913.

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References:

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Jones, R.A.C., Kenten, R.H., 1981. A Strain of Arracacha Virus-B Infecting Oca (Oxalis-Tuberosa, Oxalidaceae) in the Peruvian Andes. Phytopathol Z 100(1), 88-95.

Kenten, R.H., Jones, R.A.C., 1979. Arracacha virus B, a second isometric virus infecting arracacha (Arracacia xanthorrhiza, Umbelliferae) in the Peruvian Andes. Ann Appl Biol 93, 31-36.

Le Gall, O., Christian, P., Fauquet, C.M., King, A.M., Knowles, N.J., Nakashima, N., Stanway, G., Gorbalenya, A.E., 2008. Picornavirales, a proposed order of positive-sense single-stranded RNA viruses with a pseudo-T = 3 virion architecture. Arch Virol 153(4), 715-727.

Maddison, W.P., Maddison, D.R., 2010. Mesquite: a modular system for evolutionary analysis. Version 2.73.

Sanfacon, H., Wellink, J., Le Gall, O., Karasev, A., van der Vlugt, R., Wetzel, T., 2009. Secoviridae: a proposed family of plant viruses within the order Picornavirales that combines the families Sequiviridae and Comoviridae, the unassigned genera Cheravirus and Sadwavirus, and the proposed genus Torradovirus. Arch Virol 154(5), 899-907.

Tamura, K., Peterson, D., Peterson, N., Stecher, G., Nei, M., Kumar, S., 2011. MEGA5: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Mol Biol Evol 28(10), 2731-2739.

Figure 1. Neighbor-joining tree of the Pro-Pol amino acid sequences of members of the Picornavirales

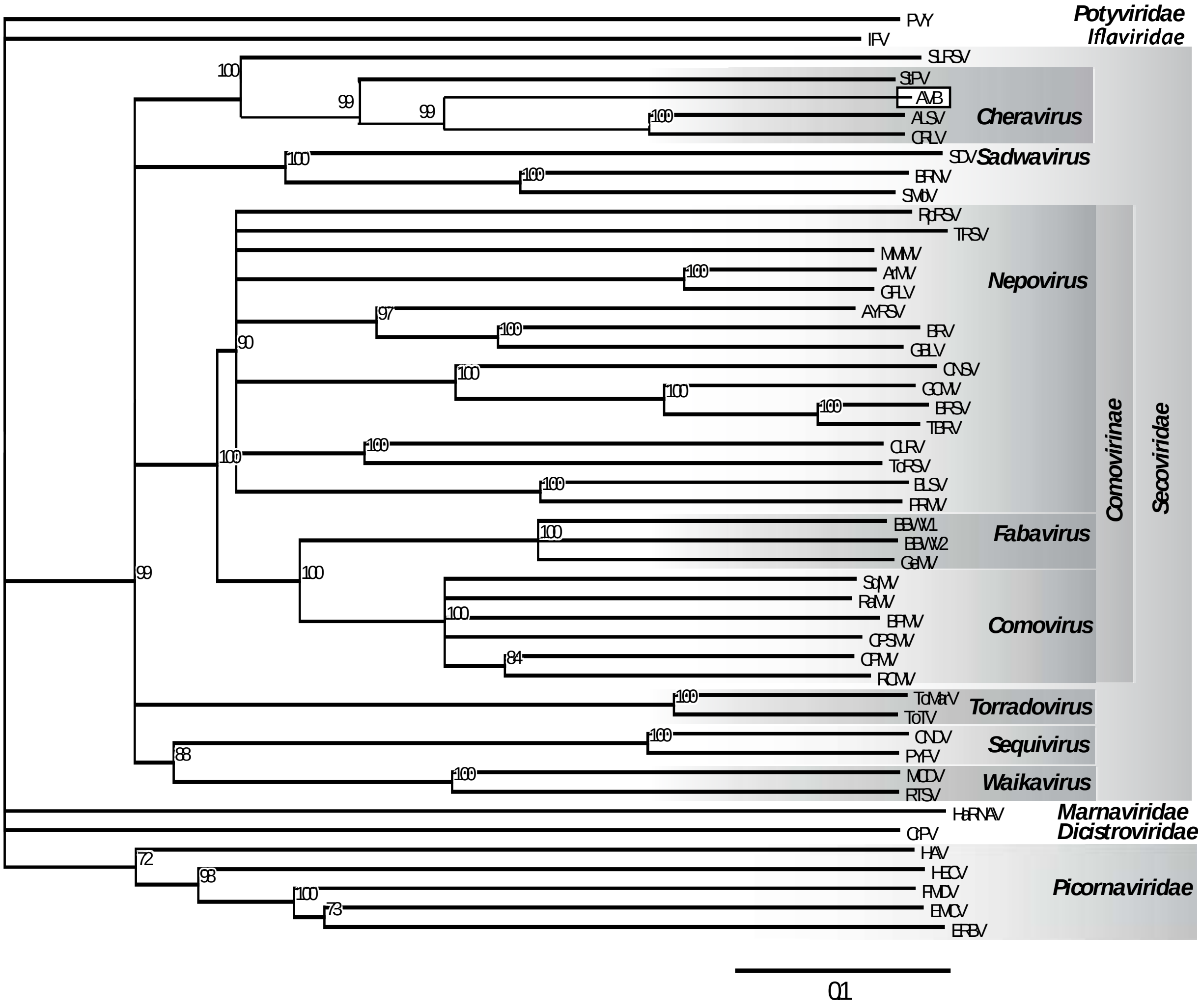

Fig1. Neighbor-joining tree of members of the order *Picornavirales* based on the amino acid sequences of the conserved domains between the “CG” motif of the 3C-like proteinase and the “GDD” motif of the polymerase (Pro-Pol region). The alignment and the tree were generated using MEGA5 (Tamura et al., 2011) and Mesquite (Maddison and Maddison, 2010). Potato virus Y (PVY) a member of the family *Potyviridae* was used as an outgroup. Representative sequences were included for the family *Secoviridae* while only representative members of other families within the order *Picornavirales* were included. The families and genera are delineated on the right. Numbers on nodes show bootstrap values (1000 replicates) above 70%. The bar represents a P distance of 0.1. The GenBank accession numbers used for each virus are as follows: potato virus Y (PVY, NC_001616 = X12456), infectious flacherie virus (IFV, NC_003781 = AB000906), hepatitis A virus (HAV, NC_001489 = M14707), human enterovirus C (HECV, NC_002058.3 = V01149), foot-and-mouth disease virus-type C (FMDV, NC_002554 = AF274010), encephalomyocarditis virus (EMCV, NC_001479 = M81861), equine rhinitis B virus 1 (ERBV, NC_003983 = X96871), cricket paralysis virus (CrPV, NC_003924 = AF218039), *Heterosigma akashiwo* RNA virus (HaRNV, NC_005281 = AY337486), parsnip yellow fleck virus (PYFV, NC_003628 = D14066), carrot necrotic dieback virus (CNDV, EU980442), maize chlorotic dwarf virus (MCDV, NC_003626 = U67839), rice tungro spherical virus (RTSV, NC_001632 = M95497), tomato torrado virus (ToTV, NC_009013 = DQ388879), tomato marchitez virus (ToMarV,

NC_010987 = EF681764), strawberry latent ringspot virus (SLRSV, NC_006964 = AY860978), stocky prune virus (StPV, DQ143874), apple latent spherical virus (ALSV, NC_003787 = AB030940), cherry rasp leaf virus (CRLV, NC_006271 = AJ621357), satsuma dwarf virus (SDV, NC_003785 = AB009958), strawberry mottle virus (SMoV, NC_003445 = AJ311875), black raspberry necrosis virus (BRNV, NC_008182 = DQ344639), raspberry ringspot virus (RpRSV, NC_005266 = AY303787), peach rosette mosaic virus (PRMV, AF016626), Blueberry latent spherical virus (BLSV, AB649296 = BAL04700) tobacco ringspot virus (TRSV, NC_005097 = U50869), melon mild mottle virus (MMMoV, AB518485 = BAJ16223), arabis mosaic virus (ArMV, NC_006057 = AY303786), grapevine fanleaf virus (GFLV, NC_003615 = D00915), artichoke yellow ringspot virus (AYRSV, AM087671), blackcurrant reversion virus (BRV, NC_003509 = AF368272), grapevine Bulgarian latent virus (GBLV, NC_015492 = FN691934), cycas necrosis stunt virus (CNSV, NC_003791 = AB073147), grapevine chrome mosaic virus (GCMV, NC 003622 = X15346), beet ringspot virus (BRSV, NC_003693 = D00322), tomato black ring virus (TBRV, NC_004439 = AY157993), tomato ringspot virus (ToRSV, NC_003840 = L19655), cherry leaf roll virus (CLRV, NC_015414 = FR851461), squash mosaic virus (SqMV, NC_003799 = AB054688), radish mosaic virus (RaMV, NC_010709 = AB295643), cowpea mosaic virus (CPMV, NC_003549 = X00206), red clover mottle virus (RCMV, NC_003741 = X64886), bean pod mottle virus (BPMV, NC_003496 = U70866), cowpea severe mosaic virus (CPSMV, NC_003545 = M83830), broad bean wilt virus 1 (BBWV1, NC_005289 = AB084450), BBWV2 (NC_003003 = AF225953), gentian mosaic virus (GeMV, BAD99001).