

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:	2016.007aP		(to be completed by ICTV officers)			
Short title: Create two species in genus <i>Macluravirus</i> , family <i>Potyviridae</i>						
(e.g. 6 new species in the genus a <b>Modules attached</b> (modules 1 and 10 are required)	Zetavirus)	1 × 6 □	2 × 7 □	3	4	5 ☐ 10 ⊠
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List the ICTV study group(s) that have seen this proposal:						
A list of study groups and contacts is provided at <a href="http://www.ictvonline.org/subcommittees.asp">http://www.ictvonline.org/subcommittees.asp</a> . If in doubt, contact the appropriate subcommittee chair (fungal, invertebrate, plant, prokaryote or vertebrate viruses)		Potyviridae				
ICTV Study Group comments (if any) and response of the proposer:						
Date first submitted to ICTV:  July 2016						
Date of this revision (if different to above):						

EC comment:	
SG response:	

## **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	6.007aP	(assigned by ICTV officers)		
To create	2 new	species within:			
				Fill in all that apply.	
(	Genus:	Macluravirus		If the higher taxon has yet to be	
Subf	amily:			created (in a later module, below) write "(new)" after its proposed	
F	amily:	Potyviridae		name.	
	Order:			<ul> <li>If no genus is specified, enter "unassigned" in the genus box.</li> </ul>	

		Genbank
Name of species	Exemplar Isolate	Accession no.
Broad-leafed dock virus A	broad-leafed dock virus A isolate ab032	KU053507
	Auckland	
Yam chlorotic mosaic virus	yam chlorotic mosaic virus isolate YS	KT724961

# 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.
  - o 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 family *Potyviridae* consists of plant viruses with a single stranded, positive sense RNA genome and flexuous, filamentous particles. Genomes have a VPg covalently linked to the 5'-end and the 3'-terminus is polyadenylated. Genomes encode a large precursor polyprotein (except in the case of genus *Bymovirus*, which is bipartite and encodes two polyproteins) that is self-cleaved into a set of functional proteins that is strongly conserved throughout the family. The family is divided into eight genera: *Brambyvirus*, *Bymovirus*, *Ipomovirus*, *Macluravirus*, *Poacevirus*, *Potyvirus*, *Rymovirus*, and *Tritimovirus*. Macluraviruses resemble members of the genus *Potyvirus* in their transmission by aphids but virions are slightly shorter. They form a distinct group in phylogenetic analyses and have different polyprotein consensus cleavage sites (Adams *et al.*, 2011).

According to criteria published in the ICTV 9th Report (Adams *et al.*, 2011), species are distinguished by the following criteria:

- Genome sequence relatedness: different species have CP as sequence identity less than ca. 80%; and nt sequence identity less than 76% either in the CP or over the whole genome. There are also differences in polyprotein cleavage sites.
- Host range and key host reactions; lack of cross protection.
- Different inclusion body morphology.
- Antigenic properties: serological relatedness may help in distinguishing species.

The two viruses described below both appear to represent new species in the genus *Macluravirus*. For each, there is at least one complete or nearly-complete genome sequence in the public domain

and these show the expected ORFs, conserved motifs and cleavage sites. The molecular criteria to create new species have been satisfied. For yam chlorotic mosaic virus there is a peer-reviewed publication giving the provenance of the virus and additional biological data. For broad-leafed dock virus A the paper describes the provenance of the host and symptoms it exhibited, but no attempt was made to satisfy Koch's postulates so symptom expression is uncertain. Phylogenetic analysis of the complete polyprotein coding sequences (Annex Figure 1) supports the placement of these viruses as distinct species within genus *Macluravirus*.

#### Yam chlorotic mosaic virus

Partial genomes of four virus isolates described as yam chlorotic necrotic mosaic virus (YCNMV) were isolated from Dioscorea parviflora and Dioscorea zingbierensis in Yunnan Province, China, by Wang et al., 2009. The complete genome of a fifth isolate, YS, from Dioscorea parviflora, and also from Yunnan, was published by Zhang et al 2016. The complete sequence of YCNMV-YS was obtained using degenerate primers with RT-PCR and 5'RACE (Zhang et al., 2016). YCNMV-YS resembled the other members of the genus *Macluravirus* in two key respects: (1) the P1 proteinase found at the N-terminus in the other monopartite viruses of the family is absent, and (2) the typical potyvirus aphid-transmission motifs DAG in the CP and PTK and KITC in the HC-Pro are also absent. However, the conserved motifs GSGKSXXXP, DEXH, NGTS, AFDF, GDD and WCANNGTSSE are present. The seven predicted proteinase cleavage sites contain the motif LQ(E)/M(A), which is typical of members of the genus, with the leucine being strictly conserved. The YCNMV-YS sequence most closely resembles that of the macluravirus Chinese yam necrotic mosaic virus (ChYNMV) isolates PES3, BRI and FX1 (Kondo, 2001; Kondo and Fujita, 2012). Although closely related, we consider that YCNMV and ChYNMV are distinct species. They infect the same host genus, Dioscorea, but YCNMV-YS was identified in D. parviflora, an indigenous species from Yunnan Provence and locally cultivated, while the three isolates of ChYNMV were isolated from *D. polystachya* and *D. oppositifolia*. Sequence identities between the genomes of YCNMV and ChYNMV isolates PES3 (AB710145) and FX1 (KJ789135) are 72.6 % and 72.9 %, and polyproteins share 80.3 % and 80.7 % identities, respectively. The nucleotide identities are below the species demarcation limit for the family, but the amino acid identities are marginally above. Protease cleavage sites on the polyprotein differ between YCNMV-YS and ChYNMV isolates in all but one site (NIb/CP). Taken together, this evidence indicates that YCNMV is distinct from ChYNMV (Annex Figure 1). We have suggested the shorter name Yam chlorotic mosaic virus for the species and confirmed that this is perfectly acceptable to the authors of the respective papers.

#### Broad-leafed dock virus A

A complete sequence of broad-leafed dock virus A (BLDVA) was obtained using immunocapture of double-stranded RNA from a composite sample from eight symptomatic broad-leafed dock (*Rumex obtusifolius*) plants followed by high-throughput sequencing (Blouin *et al.*, 2016). The dock plants had symptoms ranging from mild mottle to general necrosis. Linking symptoms on host plants with presence of virus was not done. Analysis of the high-throughput sequencing data revealed that one or more of the eight dock leaves that made up the composite sample was also infected with an isolate of cherry leaf roll virus (CLRV; genus *Nepovirus*), and so it is uncertain if the CLRV isolate was responsible for symptoms. Blouin *et al.*, (2016) provisionally named the new virus Rumex virus Y. The letter Y is generally used to designate members of the genus *Potyvirus*, so we suggest the species name *Broad-leafed dock virus A*.

BLDVA isolate ab032 Auckland is a monopartite RNA virus of 8174 nucleotides extending from the 5' UTR (154 nucleotides) to the 3' UTR (208 nucleotides) that encodes a single polyprotein of 2603 amino acids. The closest relative to this virus is *Chinese yam necrotic mosaic virus* with 52% amino acid identity over the polyprotein. The new virus genome encodes the *Potyviridae* 

motifs listed in the NCBI's conserved motif database, and the P1 cistron is absent. A phylogenetic tree based on the full-length polyproteins of other members of the *Potyviridae* shows that broadleafed dock virus A is a member of genus *Macluravirus* (Annex Figure 1).

## MODULE 10: APPENDIX: supporting material

additional material in support of this proposal

#### References:

Adams MJ, Antoniw JF, Fauquet CM (2005). Molecular criteria for genus and species discrimination within the family *Potyviridae*. *Arch Virol* 150:459–479.

Adams MJ, Zerbini FM, French R, Rabenstein F, Stenger DC, Valkonen JPT (2011). Family *Potyviridae*. In: Virus Taxonomy, Ninth Report of the International Committee on Taxonomy of Viruses (A.M.Q. King, M.J. Adams, E.B. Carstens & E.J. Lefkowitz, eds), pp. 1069-1089. Elsevier Academic Press, London.

Blouin AG, Ross HA, Hobson-Peters J, O'Brien CA, Warren B, MacDiarmid R (2016). A new virus discovered by immunocapture of double-stranded RNA, a rapid method for virus enrichment in metagenomic studies. *Molecular Ecology Resources*. DOI:10.1111/1755-0998.12525

Kondo T (2001). The 3' terminal sequence of Chinese yam necrotic mosaic. *Arch Virol* 146:1527-1535.

Kondo T, Fujita T (2012). Complete nucleotide sequence and construction of an infectious clone of Chinese yam necrotic mosaic virus suggest that macluraviruses have the smallest genome among members of the family *Potyviridae*. *Arch Virol* 157:2299-2307.

Wang JG, Zou XJ, Zheng HY, Adams MJ, Chen HR, Chen JP (2009). Molecular characterization of a new macluravirus from yam in Yunnan, China. *Arch Virol* 154:1379-1380.

Zhang P, Peng J, Guo H, Chen J, Chen S, Wang J (2016). Complete genome sequence of yam chlorotic necrotic mosaic virus from *Dioscorea parviflora*. *Arch Virol* 161:1715-1717.

#### 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.

#### Figure 1.

Neighbor-joining tree generated from an alignment of complete polyprotein sequences showing estimated phylogeny of polyprotein amino acid sequences deduced from completely sequenced isolates of representative members of the family *Potyviridae*. Representative members of genera *Brambyvirus*, *Bymovirus*, *Ipomovirus*, *Macluravirus*, *Poacevirus*, *Potyvirus*, *Rymovirus*, *Tritimovirus*, and one virus, rose yellow mosaic virus, for which a genus is currently unassigned, were used. Three new proposed members of genus *Potyvirus* (see separate proposal) are indicated with a red dot, and the two new members of genus *Macluravirus* proposed here are indicated with a green dot. The tree was deduced in Mega v6.06 after alignment by Muscle using Neighbor-

joining based on the Poisson model with 1,000 bootstrap replicates. Branch lengths are proportional to genetic distance in units of amino acid substitutions per site. Bootstrap support for branches >60% is shown at the junctions of branches.

Accession codes corresponding to the nucleotide sequence of each virus genome sequence used in the tree are: Agropyron mosaic virus, NC\_005903; Algerian watermelon mosaic virus, NC 010736; Apium virus Y, NC 014905; arracacha mottle virus, NC 018176; artichoke latent virus, NC\_026759; Asparagus virus 1, NC\_025821; banana bract mosaic virus, NC\_009745; barley mild mosaic virus (RNA1), D83408; Basella rugose mosaic virus, NC 009741; bean common mosaic necrosis virus, NC 004047; bean common mosaic virus, NC 004047; bean yellow mosaic virus, NC 003492; beet mosaic virus, NC 005304; bidens mosaic virus, NC\_023014; bidens mottle virus, NC\_014325; blackberry virus Y, NC\_008558; blue squill virus A, NC\_019415; broad-leafed dock virus A, KU053507; brome streak mosaic virus, NC\_003501; Brugmansia mosaic virus, NC 020105; Brugmansia suaveolens mottle virus, NC 014536; Caladenia virus A, NC\_018572; calla lily latent virus, NC\_021196; Canna yellow streak virus, NC 013261; carrot thin leaf virus, NC 025254; Catharanthus mosaic virus, KP742991; celery mosaic virus, NC\_015393; chilli ringspot virus, NC\_016044; chilli veinal mottle virus, NC\_005778; Chinese yam necrotic mosaic virus, NC\_018455; clover yellow vein virus, NC\_003536; cocksfoot streak virus, NC\_003742; Colombian datura virus; NC\_020072; cowpea aphid-borne mosaic virus, NC 004013; Cyrtanthus elatus virus A, NC 017977; Daphne mosaic virus, NC\_008028; Dasheen mosaic virus, NC\_003537; donkey orchid virus A, NC\_021197; East Asian Passiflora virus, NC 007728; Freesia mosaic virus, NC 014064; fritillary virus Y, NC\_010954; Gloriosa stripe mosaic virus, EF427894; habenaria mosaic virus, EF427894; Hardenbergia mosaic virus, NC 015394; Hippeastrum mosaic virus, NC 017967; Hordeum mosaic virus, NC\_005904; Japanese yam mosaic virus, NC\_000947; jasmine virus T, KT222674; johnsongrass mosaic virus, NC 003606; keunjorong mosaic virus, NC 016159; konjac mosaic virus, NC\_007913; leek yellow stripe virus, NC\_004011; lettuce Italian necrotic virus; KP769852; lettuce mosaic virus, NC\_003605; lily mottle virus, NC\_005288; Lupinus mosaic virus, NC 014898; maize dwarf mosaic virus, NC 003377; Moroccan watermelon mosaic virus, NC\_009995; Narcissus degeneration virus, NC\_008824; Narcissus late season yellows virus, NC 023628; Narcissus yellow stripe virus, NC 011541; oat mosaic virus (RNA1), NC 004016; oat necrotic mottle virus, NC\_005136; onion yellow dwarf virus, NC\_005029; Ornithogalum mosaic virus, NC 019409; Panax virus Y, NC 014252; papaya leaf distortion mosaic virus, NC\_005028; papaya ringspot virus, NC\_001785; passion fruit woodiness virus, NC\_014790; pea seed-borne mosaic virus, NC\_001671; peanut mottle virus, NC\_002600; Pennisetum mosaic virus, NC 007147; pepper severe mosaic virus, NC 008393; pepper veinal mottle virus, NC\_011918; pepper yellow mosaic virus, NC\_014327; Peru tomato mosaic virus, NC\_004573; plum pox virus, NC\_001445; pokeweed mosaic virus, NC\_018872; potato virus A, NC\_004039; potato virus V, NC\_004010; potato virus Y, NC\_001616; rose yellow mosaic virus, NC\_019031; ryegrass mosaic virus, NC\_001814; scallion mosaic virus, NC\_003399; shallot yellow stripe virus, NC\_007433; sorghum mosaic virus, NC\_004035; soybean mosaic virus, NC\_002634; squash vein yellowing virus, NC\_010521; sugarcane mosaic virus, NC\_003398; sugarcane streak mosaic virus, NC\_014037; sunflower chlorotic mottle virus, NC\_014038; sunflower mild mosaic virus, NC 021065; sweet potato feathery mottle virus, NC 001841; sweet potato latent virus, NC\_020896; sweet potato mild mottle virus, NC\_003797; sweet potato virus 2, NC\_017970; sweet potato virus C; NC\_014742; sweet potato virus G, NC\_018093; tamarillo leaf malformation virus, NC\_026615; Telosma mosaic virus, NC\_009742; thunberg fritillary mosaic virus, NC 007180; tobacco etch virus, NC 001555; tobacco vein banding mosaic virus, NC 009994; tobacco vein mottling virus, NC\_001768; tomato necrotic stunt virus, NC\_017824; Triticum mosaic virus, NC\_012799; turnip mosaic virus, NC\_002509; vanilla distortion mosaic virus,

NC\_025250; Verbena virus Y, NC\_010735; wheat yellow mosaic virus (RNA1), FJ361765; wild potato mosaic virus, NC\_004426; wild tomato mosaic virus, NC\_009744; Wisteria vein mosaic virus, NC\_007216; yambean mosaic virus, NC\_016441; yam chlorotic mosaic virus, KT724961; yam mild mosaic virus, NC\_019412; yam mosaic virus, NC\_004752; Zantedeschia mild mosaic virus, NC\_011560; Zea mosaic virus, NC\_018833; zucchini shoestring virus, KU355553;

zucchini tigre mosaic virus, NC\_023175; zucchini yellow mosaic virus, NC\_003224.

