

**Part 1:** **TITLE, AUTHORS, APPROVALS, etc**

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| **Code assigned:** | ***2022.005M*** |  |
| **Short title:** Create two new species in genus *Coguvirus* (*Bunyavirales*: *Phenuiviridae*) | | |
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**Author(s) and email address(es)**

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| --- | --- |
| Cao M, Zhang S, Tian X, Navarro B, Di Serio F | caomengji@cric.cn;  qq371260@email.swu.edu.cn; 395485918@qq.com; beatriz.navarro@ipsp.cnr.it; francesco.diserio@ipsp.cnr.it |

**Author(s) institutional address(es) (optional)**

|  |
| --- |
| Southwest University, Beibei, China [MC; SZ, XT]; Istituto per la Protezione Sostenibile delle Piante, CNR, Italy [FDS, BN] |

**Corresponding author**

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| Di Serio F |

**List the ICTV Study Group(s) that have seen this proposal**

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| ICTV *Phenuiviridae* Study Group |

**ICTV Study Group comments and response of proposer**

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| Approved. |

**ICTV Study Group votes on proposal**

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| **Study Group** | **Number of members** | | |
| **Votes support** | **Votes against** | **No vote** |
| ICTV *Phenuiviridae* Study Group | 9 | 0 | 0 |

**Authority to use the name of a living person**

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| --- | --- |
| **Is any taxon name used here derived from that of a living person (Y/N)** | N |

|  |  |  |
| --- | --- | --- |
| **Taxon name** | **Person from whom the name is derived** | **Permission attached (Y/N)** |
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**Submission dates**

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| --- | --- |
| Date first submitted to SC Chair | May 27, 2022 |
| Date of this revision (if different to above) |  |

**ICTV-EC comments and response of the proposer**

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**Part 2:** **NON-TAXONOMIC PROPOSAL**

**Text of proposal**

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**Part 3:** **TAXONOMIC PROPOSAL**

**Name of accompanying Excel module**

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| 2022.005M.N.v1.Coguvirus\_2nsp.xlsx |

**Abstract**

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| Watermelon crinkle leaf-associated virus 1 (WCLaV-1) and watermelon crinkle leaf-associated virus 2 (WCLaV-2) fulfill the criteria (based on biological, molecular, and phylogenetic analyses) to be classified in genus *Coguvirus* (family *Phenuiviridae*) as representative members of two novel species*.* Here, it is proposed to create the species *Coguvirus citrulli and Coguvirus henanense* to classify WCLaV-1 and WCLaV-2, respectively. |

**Text of proposal**

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| --- | --- |
| |  | | --- | | Watermelon crinkle leaf-associated virus 1 (WCLaV-1) and watermelon crinkle leaf-associated virus 2 (WCLaV-2) have been recently identified in watermelon plants (*Citrullus lanatus* (Thunb.) Matsum. & Nakai, 1916) in China. They were reported as negative-sense RNA viruses with a genome composed of three segments (Xin et al., 2017) coding for one protein each. The putative large protein (L) containing the RNA-directed RNA polymerase (RdRp) domain, movement protein (MP) and nucleocapsid protein (NP) encoded by WCLaV-1 and WCLaV-2 share 58.8%, 44.6%, and 46.7% with each other, respectively. The proteins encoded by these viruses share the highest amino acid (aa) sequence identity (ranging from 39.7% to 62.9%) with citrus concave gum-associated virus (CCGaV) and citrus virus A (CiVA), the two current members of the phenuivirid genus *Coguvirus* (Navarro et al., 2018a; 2018b). Recently, a reassessment of the full genome sequences of WCLaV-1 and WCLaV-2 provided evidence that these viruses are actually bipartite viruses with RNA1 encoding the putative L (GenBank #MW842780 and MW842781 for WCLaV-1 and WCLaV-2, respectively) and RNA 2 containing two open reading frames (ORFs) in ambisense orientation encoding the putative MP and NP (GenBank #MW751425 and MW751424 for WCLaV-1 and WCLaV-2, respectively). In both WCLaV1 and WCLaV2, the two ORFs of RNA2 are separated by an AU-rich intergenic region (IR) adopting a hairpin conformation. This genomic organization is identical to that of CCGaV and CiVA (Navarro et al., 2018a; 2018b), with which WCLaV-1 and WCLaV-2 also share almost identical terminal ends (up to 15 nt) of the genomic fragments. Maximum likelihood phylogenetic trees generated using the putative L, MP, and NP amino acid sequences of these viruses and those of representative members in the order *Bunyavirales* always clustered WCLaV-1 and WCLaV-2 in the same clade together with the two currently classified coguviruses (CCGaV and CiVA). High bootstrap values (100 for the node of the clade and 82–99 for the nodes within the clade) in the phylogenetic trees strongly support the close phylogenetic relationships between the viruses in this clade.  Since species demarcation for the genus *Coguvirus* has been established as <95% identity in the amino acid (aa) sequence of the RdRp (TaxoProp 2019.026M), and since WCLaV-1 and WCLaV-2 RdRps share aa sequence identity of 58.8 % with each other and even lower with those of other coguviruses (Table 1), we propose to classify these two viruses in two novel species. | |

**Supporting evidence**

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| **Table 1.** Comparison of the amino acid sequences of the RNA-directed RNA polymerase of WCLaV-1, WCLaV-2, CCGaV and CiVA. Similarity and identity scores (%) are shown on white and grey background, respectively. | | | | |
|  |  |  |  |  |
|  | WCLaV-1 | WCLaV-2 | CCGaV | CiVA |
| RDRP |  |  |  |  |
| WCLaV-1 | 100.0 | 58.8 | 62.5 | 62.9 |
| WCLaV-2 | 76.0 | 100.0 | 57.5 | 57.0 |
| CCGaV | 79.3 | 76.2 | 100.0 | 76.6 |
| CiVA | 78.4 | 75.6 | 88.3 | 100.0 |



**FIGURE 1.** Phylogenetic tree of the RNA-directed RNA polymerase (RdRp) conserved core domain of watermelon crinkle leaf-associated virus 1 and watermelon crinkle leaf-associated virus 2, representative members of genera of the family *Phenuiviridae* and of genera *Orthotospovirus* and *Emaravirus*. Maximum likelihood method adopting the LG + G + F amino acid substitution model was used to infer the phylogenetic tree. Bootstrap probability values (500 replicates) above 50% are shown at branch nodes. Tree branches are proportional to the genetic distances, with the scale bar indicating substitutions per amino acid site. The cluster including current members of genus *Coguvirus* and the members of the new proposed species is reported on orange background (modified from Zhang et al., 2021).



**FIGURE 2.** Maximum likelihood phylogenetic tree inferred from the nucleocapsid protein (NP) sequences of watermelon crinkle leaf-associated virus 1 and watermelon crinkle leaf-associated virus 2 (blue arrows), members of the genus *Coguvirus* and of representative members of all the genera of the family *Phenuiviridae*, with the NP of European mountain ash ringspot-associated virus (family *Fimoviridae*) used as outgroup. Information of bootstrap values, distances, and other symbols are as reported in the legend to Figure 1 (Zhang et al., 2021).

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**FIGURE 3**. Maximum likelihood phylogenetic tree inferred from the movement protein (MP) sequences of watermelon crinkle leaf-associated virus 1 and watermelon crinkle leaf-associated virus 2 (blue arrows), and of representative plant-infecting viruses of the genera *Coguvirus, Emaravirus* (family *Fimoviridae*), *Ophiovirus* (family *Aspiviridae*), *Orthotospovirus* (family *Tospoviridae*), *Tenuivirus*,and *Rubodvirus* (family *Phenuiviridae*), and *Nucleorhabdovirus, Cytorhabdovirus* and *Varicosavirus* (family *Rhabdoviridae*). Information of bootstrap values, distances, and other symbols are as reported in the legend to Figure 1 (Zhang et al., 2021).

**References**

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3. Navarro B, Zicca S, Minutolo M et al (2018b) A negative-stranded RNA virus infecting citrus trees: the second member of a new genus within the order *Bunyavirales.* Front Microbiol 9: 2340. doi: 10.3389/fmicb.2018.02340. PMID: 30333811; PMCID: PMC6176071.
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