This form should be used for all taxonomic proposals. Please complete all those modules that are applicable.

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.

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

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Code assigned:** | ***2017.006S*** | | | | (to be completed by ICTV officers) |
| **Short title:** Create 2 new species (*Kunsagivirus B* & *Kunsagivirus C*) in the genus *Kunsagivirus*  (e.g. 6 new species in the genus *Zetavirus*) | | | | | |
| **Modules attached**  (Modules 1, 4 and either 2 or 3 are required. | | **1**  **2  3  4** | | | |
| **Author(s):** | | | | | |
| Roland Zell, Eric Delwart, Alexander E. Gorbalenya, Tapani Hovi, Andrew M.Q. King, Nick J. Knowles, A. Michael Lindberg, Mark A. Pallansch, Ann C. Palmenberg, Gabor Reuter, Peter Simmonds, Tim Skern, Glyn Stanway and Teruo Yamashita | | | | | |
| **Corresponding author with e-mail address:** | | | | | |
| Roland Zell ([roland.zell@med.uni-jena.de](mailto:roland.zell@med.uni-jena.de)) | | | | | |
| **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 (there are six virus subcommittees: animal DNA and retroviruses, animal ssRNA-, animal ssRNA+, fungal and protist, plant, bacterial and archaeal) | | | *Picornaviridae* Study Group | | |
| **ICTV Study Group comments (if any) and response of the proposer:** | | | | | |
|  | | | | | |
|  | | | | | |
| Date first submitted to ICTV: | | | | 02 June 2017 | |
| Date of this revision (if different to above): | | | |  | |

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

**Part 2**: **PROPOSED TAXONOMY**

|  |
| --- |
| Present the proposed new taxonomy on accompanying spreadsheet |
| **Name of accompanying spreadsheet:** 2017.006S.N.v1.Kunsagivirus\_2sp |

Please display the taxonomic changes you are proposing on the accompanying spreadsheet module 2017\_TP\_Template\_Excel\_module. Submit both this and the spreadsheet to the appropriate ICTV Subcommittee Chair.

**Part 4:** **APPENDIX**: supporting material

| additional material in support of this proposal |
| --- |
| **References:** |
| **European roller kunsagivirus:**  Boros A, Kiss T, Kiss O, Pankovics P, Kapusinszky B, Delwart E, Reuter G. 2013. Genetic characterization of a novel picornavirus distantly related to the marine mammal-infecting aquamaviruses in a long-distance migrant bird species, European roller (Coracias garrulus). J. Gen. Virol. 94(9):2029-2035.  **Bat kunsagivirus:**  Yinda CK, Zell R, Deboutte W, Zeller M, Conceicao-Neto N, Heylen E, Maes P, Knowles NJ, Ghogomu SM, Van Ranst M, Matthijnssens J. 2017. Highly diverse population of *Picornaviridae* and other members of the *Picornavirales* in Cameroonian fruit bats. BMC Genomics 18(1):249.  **Bakunsavirus:**  Buechler CR, Bailey AL, Lauck M, Heffron A, Johnson JC, Campos Lawson C,  Rogers J, Kuhn JH, O'Connor DH. 2017. Genome sequence of a novel kunsagivirus  (*Picornaviridae*: *Kunsagivirus*) from a wild baboon (*Papio cynocephalus*). Genome  Announc. 5(18). pii: e00261-17. doi: 10.1128/genomeA.00261-17. |

|  |
| --- |
| **Annex:**  Please explain the reasons for the taxonomic changes you are proposing and provide evidence to support them. The following information should be provided, where relevant:   * **Species demarcation criteria**: Explain how new species differ from others in the genus and demonstrate that these differences meet the criteria previously established for demarcating between species. If no criteriahave previously been established, and if there will now be more than one species in the genus, please state the demarcation criteria you are proposing. * **Higher taxa**:   + There is no formal requirement to state demarcation criteria when proposing new genera or other higher taxa. However, a similar concept should apply in pursuit of a rational and consistent virus taxonomy.   + Please indicate the **origin of names** assigned to new taxa at genus level and above.   + For each new genus a **type species** must be designated to represent it. Please explain your choice. * **Supporting evidence**: The use of Figures and Tables is strongly recommended (note that copying from publications will require permission from the copyright holder). For phylogenetic analysis, try to provide a tree where branch length is related to genetic distance. |

**Create 2 new species (*Kunsagivirus B* & *Kunsagivirus C*) in the genus *Kunsagivirus***

A novel picornavirus was detected in faeces from the fruit bat *Eidolon helvum* in Cameroon. Its genome sequence (KX644936) shows significant similarity to that of the kunsagivirus detected in a faecal sample of the European roller (*Coracias garrulus*); the amino acid identities of both the 3CD protein and the P1 polyprotein equals c. 50% (compare Suppl. Tables 1 and 2).

Another kunsagi-like virus, bakunsavirus (KY670597), was detected in a wild baboon (*Papio cynocephalus*) from Mikumi National Park (Tanzania). Its P1 aa identity with roller kunsagivirus and bat kunsagivirus is 50% and 52%, respectively.

A detailed analysis of P1, P2 and P3 regions as well as of the processed proteins VP0CP, VP3CP, VP1CP, 2A2 (2A4), 2B, 2CHel, 3A, 3BVPg, 3CPro, 3DPol suggest that bat kunsagivirus and bakunsavirus belong to distinct species of the *Kunsagivirus* genus (compare Table 1).

**Table 1: Divergence of kunsagivirus species (number of aa and nt differences, respectively, per site)**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Protein** | **Species** | ***Kunsagivirus A*** | ***Kunsagivirus B*** | ***Kunsagivirus C*** |
| VP0 | *Kunsagivirus A* | - | 0.498 | 0.503 |
|  | *Kunsagivirus B* | 0.538 | - | 0.481 |
|  | *Kunsagivirus C* | 0.575 | 0.548 | - |
|  |  |  |  |  |
| VP3 | *Kunsagivirus A* | - | 0.436 | 0.411 |
|  | *Kunsagivirus B* | 0.452 | - | 0.418 |
|  | *Kunsagivirus C* | 0.404 | 0.500 | - |
|  |  |  |  |  |
| VP1 | *Kunsagivirus A* | - | 0.461 | 0.588 |
|  | *Kunsagivirus B* | 0.502 | - | 0.588 |
|  | *Kunsagivirus C* | 0.718 | 0.709 | - |
|  |  |  |  |  |
| 2A2\* | *Kunsagivirus A* | - | 0.502 | 0.562 |
|  | *Kunsagivirus B* | 0.567 | - | 0.585 |
|  | *Kunsagivirus C* | 0.718 | 0.679 | - |
|  |  |  |  |  |
| 2B | *Kunsagivirus A* | - | 0.441 | 0.476 |
|  | *Kunsagivirus B* | 0.486 | - | 0.455 |
|  | *Kunsagivirus C* | 0.582 | 0.518 | - |
|  |  |  |  |  |
| 2C | *Kunsagivirus A* | - | 0.420 | 0.476 |
|  | *Kunsagivirus B* | 0.439 | - | 0.476 |
|  | *Kunsagivirus C* | 0.539 | 0.559 | - |
|  |  |  |  |  |
| 3A | *Kunsagivirus A* | - | 0.550 | 0.551 |
|  | *Kunsagivirus B* | 0.667 | - | 0.624 |
|  | *Kunsagivirus C* | 0.726 | 0.762 | - |
|  |  |  |  |  |
| 3B | *Kunsagivirus A* | - | 0.375 | 0.375 |
|  | *Kunsagivirus B* | 0.250 | - | 0.333 |
|  | *Kunsagivirus C* | 0.250 | 0.208 | - |
|  |  |  |  |  |
| 3C | *Kunsagivirus A* | - | 0.572 | 0.546 |
|  | *Kunsagivirus B* | 0.710 | - | 0.538 |
|  | *Kunsagivirus C* | 0.650 | 0.654 | - |
|  |  |  |  |  |
| 3D | *Kunsagivirus A* | - | 0.449 | 0.470 |
|  | *Kunsagivirus B* | 0.434 | - | 0.499 |
|  | *Kunsagivirus C* | 0.528 | 0.530 | - |
|  |  |  |  |  |
|  |  |  |  |  |
| P1 | *Kunsagivirus A* | - | 0.468 | 0.458 |
|  | *Kunsagivirus B* | 0.501 | - | 0.451 |
|  | *Kunsagivirus C* | 0.498 | 0.517 | - |
|  |  |  |  |  |
| P2 | *Kunsagivirus A* | - | 0.451 | 0.512 |
|  | *Kunsagivirus B* | 0.483 | - | 0.506 |
|  | *Kunsagivirus C* | 0.619 | 0.591 | - |
|  |  |  |  |  |
| P3 | *Kunsagivirus A* | - | 0.491 | 0.495 |
|  | *Kunsagivirus B* | 0.527 | - | 0.519 |
|  | *Kunsagivirus C* | 0.573 | 0.579 | - |

\* The equivalent protein of *Kunsagivirus C* is 2A4

**Creation of two new kunsagivirus species requires definition of species demarcation criteria:**

**-** The presently known kunsagiviruses share an essentially identical genome layout with a type IV IRES structure.

- All processed proteins of the various kunsagiviruses are orthologous. The lengths of the polyproteins vary from 2218 to 2248 aa.

- The capsid protein VP0 remains uncleaved.

- There are 1-3 aphthovirus-like 2A proteins with NPGP sequence motif plus an additional 2A protein with unknown function (no protease, no H-box/NC motif, no AIG1 type guanine binding domain of a P-loop NTPase with GxxGxGKS motif).

- The overall aa identities of the P1/P2/P3 polyproteins are >45/>35/>40% (compare Table 1: divergence <55/<65/<60%). However, aa identities of the processed protein range from 28% (2A2) to 80% (3B). The overall nt identities of the P1/P2/P3 genome regions are >50/>45/>45% (compare Table 1: divergence <50/<55/<55%).

The aligned polyprotein sequences of the three viruses suggest a 3-4-4 genome layout for bat kunsagivirus and a 3-6-4 genome layout for bakunsavirus, respectively, due to an uncleaved VP0 capsid protein and 2 to 4 putative 2A proteins (i.e., 1 and 3 aphthovirus-like polypeptides with NPGP sequence motif, respectively, plus an additional 2A protein with unknown function; compare Figure 1):

*Kunsagivirus A* and *B*: European roller kunsagivirus, bat kunsagivirus

VPg+5'UTRIRES-IV[1AB-1C-1D-2A1NPG↓P/2A2-2B-2CHel/3A-3BVPg-3CProt-3DPol]3'UTR-poly(A)

*Kunsagivirus C*: bakunsavirus

VPg+5'UTRIRES-IV[1AB-1C-1D-2A1NPG↓P/2A2NPG↓P/2A3NPG↓P/2A4-2B-2CHel/3A-3BVPg-3CProt-3DPol]3'UTR-poly(A)

The 5'-untranslated region of all known kunsagiviruses have a HCV-like type IV IRES.

**Distinctive features when comparing the three species:**

(i) 1 to 3 copies of an aphthovirus-like 2A protein with NPGP motif and variable lengths,

(ii) a N-terminal deletion of c. 30 aa of 3A protein of bakunsavirus,

(iii) a longer 3C protease of roller kunsagivirus (206 aa vs. 187/183 aa),

(iv) a longer 3D polymerase of bat kunsagivirus (509 aa vs. 476/468 aa), and

(v) a longer 3'-untranslated region of bakunsavirus (180 nt vs. 28/45 nt) (compare Table 2)

**Table 2: Comparison of 5'-, 3'-NTR und processed proteins**

|  |  |  |  |
| --- | --- | --- | --- |
| Genome | *Kunsagivirus A* | *Kunsagivirus B* | *Kunsagivirus C* |
| region | (roller kunsagivirus) | (bat kunsagivirus) | (bakunsavirus) |
| 5-UTR | 500 nt | 393 nt | 532 nt |
| VP0 | 315 aa | 299 aa | 295 aa |
| VP3 | 228 aa | 228 aa | 235 aa |
| VP1 | 240 aa | 238 aa | 40 aa |
| 2A1NPGP | 55 aa | 23 aa | 17aa, 49 aa, 64 aa |
| 2A2 | 165 aa | 175 aa | 157 aa |
| 2B | 111 aa | 111 aa | 110 aa |
| 2C | 296 aa | 295 aa | 296 aa |
| 3A | 132 aa | 129 aa | 101 aa |
| 3B | 24 aa | 24 aa | 24 aa |
| 3C | 206 aa | 187 aa | 183 aa |
| 3D | 476 aa | 509 aa | 468 aa |
| 3'-UTR | 28 nt | 45 nt | 180 nt |

Phylogenetic analyses reveal clustering of the three known kunsagiviruses in the *Aquamavirus/Avihepatovirus/Avisivirus/Kunsagivirus/Pasivirus/Parechovirus* supergroup (compare Figs. 2 and 3). [Note: the supergroup concept does not imply a taxonomic entity but reflects phylogenetic clustering of the respective genera observed in different tree inference methods (NJ, ML, Bayesian MCMC)]. Closest relative of kunsagiviruses is seal picornavirus (*Aquamavirus A1*) which has an identical genome layout but lower aa identity.

**Genome organisation:**



**Figure 1:** Comparison of *Kunsagivirus A, B, C* genome organisation (schematic depiction). The open reading frames are indicated by boxes. Positions of putative aa cleavage sites and the lengths of the deduced proteins are shown as proposed by Boros et al. (2013) and Yinda et al (2017). Arrows indicate the putative processing sites. The 5'-NTR of bat kunsagivirus is incomplete.



**Figure 2:** Phylogenetic analyses of picornavirus **3CD** using Bayesian tree inference (MrBayes 3.2). Twenty-five picornavirus sequences of the *Aquamavirus/Avihepatovirus/Avisivirus/Kunsagivirus/ Pasivirus/Parechovirus* supergroup were retrieved from GenBank; the newt ampivirus sequence served as outgroup [Note: the supergroup concept does not imply a taxonomic entity but reflects phylogenetic clustering of the respective genera observed in different tree inference methods (NJ, ML, Bayesian MCMC)]. Presented are GenBank accession numbers, ***genus*** ***names***, *species names* and *types* (underlined). If available, common names and designations of isolates [in square brackets] are also given. Yet unassigned viruses are printed in blue. Proposed names are printed in red and indicated by a dot (●). Numbers at nodes indicate posterior probabilities obtained after 2,000,000 generations. The optimal substitution model (GTR+G+I) was determined with MEGA 5. The scale indicates substitutions/site.



**Figure 3:** Phylogenetic analyses of picornavirus **P1** capsid protein precursor using Bayesian tree inference (MrBayes 3.2). Twenty-six picornavirus sequences of the *Aquamavirus/Avihepatovirus/ Avisivirus/Kunsagivirus/Pasivirus/Parechovirus* supergroup were retrieved from GenBank; the newt ampivirus sequence served as outgroup [Note: the supergroup concept does not imply a taxonomic entity but reflects phylogenetic clustering of the respective genera observed in different tree inference methods (NJ, ML, Bayesian MCMC)]. Presented are GenBank accession numbers, ***genus*** ***names***, *species names* and *types* (underlined). If available, common names and designations of isolates [in square brackets] are also given. Yet unassigned viruses are printed in blue. Proposed names are printed in red and indicated by a dot (●). Numbers at nodes indicate posterior probabilities obtained after 2,000,000 generations. The optimal substitution model (GTR+G+I) was determined with MEGA 5. The scale indicates substitutions/site.

**Suppl. Table 1. Estimates of Evolutionary Divergence of 3CD Protein between Sequences**

[ 1] #KC935379\_Kunsagivirus\_A1\_strain\_roller/SZAL6-KuV/2011/HUN

[ 2] #KX644936\_Kunsagivirus\_B1\_Bat\_Kunsagivirus

[ 3] #KY670597\_Kunsagivirus\_C1\_Bakunsa\_virus\_strain\_baboon/M27-KuV/1986/TAN\_P3

[ 4] #EU142040\_Aquamavirus\_AV-A\_SePV-1\_HO-02-21

[ 5] #JQ316470\_Pasivirus\_A1\_swine/France/2011

[ 6] #AB937989\_Crohivirus\_1\_shrew/ZM54/Zambia/2012

[ 7] #KX644937\_Bat\_crohivirus\_clone\_Bat/CAM/CroV-P25/2013

[ 8] #L02971\_Parechovirus\_A1\_HPeV-1\_Harris

[ 9] #AF327920\_Parechovirus\_B1\_LV-1\_87-012

[10] #HF677705\_Parechovirus\_C1\_Sebokele\_virus\_1\_strain\_An\_B\_1227\_d

[11] #KF006989\_Parechovirus\_D1\_Ferret\_parechovirus\_strain\_ferret/MpPeV1/NL

[12] #KC465954\_Avisivirus\_A1\_strain\_turkey/M176-TuASV/2011/HUN

[13] #KC614703\_Avisivirus\_A1\_isolate\_turkey/USA/IN1/2010

[14] #KF979333\_Avisivirus\_B1\_chicken\_picornavirus\_2\_isolate\_44C

[15] #KF979334\_Avisivirus\_C1\_chicken\_picornavirus\_3\_isolate\_45C

[16] #KT880669\_Avisivirus\_C1\_Pf-CHK1/AsV

[17] #DQ249299\_Avihepatovirus\_DHAV-1\_03D

[18] #KJ000696\_Aalivirus\_A1\_duck/GL/12/China/2012

[19] #KM203656\_Orivirus\_1\_strain\_chicken/Pf-CHK1/2013/HUN

[20] #KT880667\_Orivirus\_2\_Pf-CHK1/OrV-A2

[21] #KJ641698\_bat\_picornavirus\_isolate\_bat/BtMf-PicoV-1/SAX2011

[22] #KC843627\_Potamipivirus\_A1\_Eel\_picornavirus\_F15-05

[23] #JX134222\_Limnipivirus\_A1\_BGPV-1\_04-032

[24] #KF306267\_Limnipivirus\_B1\_CarpPV

[25] #KF183915\_Limnipivirus\_C1\_FHMPV-1\_isolate\_fhm/1/MN/USA/2010

[26] #KP770140\_Ampivirus\_A1\_strain\_NEWT/2013/HUN

[ 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 ]

[ 1] -

[ 2] 0.518 -

[ 3] 0.583 0.583 -

[ 4] 0.697 0.703 0.675 -

*[ 5] 0.767 0.751 0.743 0.750 -*

[ 6] 0.746 0.750 0.734 0.748 0.645 -

[ 7] 0.742 0.760 0.723 0.744 0.656 0.541 -

[ 8] 0.750 0.746 0.728 0.720 0.710 0.630 0.622 -

[ 9] 0.749 0.735 0.725 0.693 0.693 0.599 0.581 0.510 -

[10] 0.734 0.735 0.703 0.706 0.686 0.608 0.587 0.539 0.340 -

[11] 0.739 0.733 0.706 0.707 0.673 0.646 0.598 0.591 0.571 0.574 -

[12] 0.745 0.753 0.734 0.747 0.721 0.720 0.737 0.717 0.706 0.702 0.711 -

[13] 0.747 0.750 0.741 0.743 0.722 0.719 0.732 0.713 0.704 0.700 0.709 0.041 -

[14] 0.721 0.721 0.731 0.713 0.698 0.674 0.698 0.701 0.673 0.688 0.688 0.492 0.491 -

[15] 0.735 0.738 0.743 0.735 0.717 0.714 0.724 0.726 0.704 0.703 0.706 0.460 0.459 0.494 -

[16] 0.736 0.739 0.741 0.734 0.721 0.713 0.724 0.726 0.706 0.704 0.706 0.455 0.454 0.495 0.014 -

[17] 0.742 0.729 0.725 0.724 0.717 0.685 0.680 0.675 0.658 0.658 0.679 0.618 0.627 0.598 0.612 0.611 -

[18] 0.721 0.727 0.728 0.732 0.697 0.668 0.651 0.683 0.665 0.665 0.679 0.571 0.567 0.556 0.554 0.552 0.535 -

[19] 0.744 0.744 0.738 0.769 0.732 0.714 0.696 0.713 0.678 0.683 0.702 0.685 0.678 0.664 0.683 0.681 0.618 0.648 -

[20] 0.741 0.743 0.738 0.770 0.743 0.721 0.703 0.708 0.681 0.683 0.707 0.703 0.694 0.679 0.672 0.670 0.623 0.642 0.158 -

[21] 0.742 0.751 0.744 0.766 0.716 0.673 0.668 0.690 0.697 0.702 0.686 0.725 0.716 0.710 0.710 0.709 0.686 0.695 0.695 0.700 -

[22] 0.742 0.734 0.739 0.750 0.718 0.678 0.672 0.703 0.661 0.655 0.678 0.744 0.740 0.721 0.731 0.732 0.685 0.696 0.702 0.700 0.676 -

[23] 0.768 0.790 0.764 0.778 0.760 0.724 0.721 0.692 0.704 0.704 0.718 0.748 0.744 0.721 0.746 0.739 0.713 0.714 0.725 0.717 0.698 0.670 -

[24] 0.760 0.764 0.757 0.785 0.742 0.708 0.729 0.707 0.693 0.697 0.706 0.759 0.757 0.733 0.760 0.760 0.734 0.721 0.732 0.735 0.720 0.660 0.561 -

[25] 0.759 0.777 0.758 0.774 0.750 0.734 0.711 0.710 0.676 0.685 0.716 0.758 0.753 0.738 0.741 0.737 0.719 0.719 0.708 0.708 0.718 0.668 0.566 0.490 -

[26] 0.851 0.841 0.824 0.818 0.835 0.823 0.832 0.824 0.809 0.806 0.833 0.831 0.831 0.834 0.824 0.826 0.825 0.812 0.837 0.840 0.829 0.831 0.836 0.825 0.824 -

The number of amino acid differences per site from between sequences are shown. Standard error estimate(s) are shown above the diagonal. The analysis involved 26 amino acid sequences. The coding data was translated assuming a Standard genetic code table. All ambiguous positions were removed for each sequence pair. There were a total of 847 positions in the final dataset. Evolutionary analyses were conducted in MEGA5 [1].

\_\_\_ within type comparison, \_\_\_ between types/within species comparison,

\_\_\_ between species/within genus comparison, \_\_\_ between genera comparison

**Suppl. Table 2. Estimates of Evolutionary Divergence of P1 Polyprotein between Sequences**

[ 1] #KC935379\_Kunsagivirus\_1\_strain\_roller/SZAL6-KuV/2011/HUN

[ 2] #KX644936\_Kunsagivirus\_B1\_bat\_kunsagivirus

[ 3] #KY670597\_Kunsagivirus\_C1\_Bakunsa\_virus\_baboon/M27-KuV/1986/TAN

[ 4] #EU142040\_Aquamavirus\_A\_SePV-1\_HO-02-21

[ 5] #JQ316470\_Pasivirus\_1\_swine/France/2011

[ 6] #AB937989\_Crohivirus\_strain\_ZM54

[ 7] #KX644937\_Bat\_crohivirus\_clone\_Bat/CAM/CroV-P25/2013

[ 8] #L02971\_Parechovirus\_A1\_HPeV-1\_Harris

[ 9] #AF327920\_Parechovirus\_B1\_LV-1\_87-012

[10] #HF677705\_Parechovirus\_C1\_Sebokele\_virus\_1\_An/B/1227/d

[11] #KF006989\_Parechovirus\_D1\_Ferret\_parechovirus\_isolate\_MpPeV1

[12] #KC465954\_Avisivirus\_A1\_strain\_turkey/M176-TuASV/2011/HUN

[13] #KC614703\_Avisivirus\_A1\_isolate\_turkey/USA/IN1/2010

[14] #KF979333\_Avisivirus\_B1\_Chicken\_picornavirus\_2\_isolate\_44C

[15] #KF979334\_Avisivirus\_C1\_Chicken\_picornavirus\_3\_isolate\_45C

[16] #KT880669\_Avisivirus\_C1\_Pf-CHK1/AsV

[17] #DQ249299\_Avihepatovirus\_A1\_DHAV-1\_03D

[18] #KJ000696\_Aalivirus\_A1\_duck\_picornavirus\_GL/12

[19] #KM203656\_Orivirus\_1\_strain\_chicken/Pf-CHK1/2013/HUN

[20] #KT880667\_Orivirus\_2\_Pf-CHK1/OrV-A2

[21] #KJ641698\_bat\_picornavirus\_isolate\_bat/BtMf-PicoV-1/SAX2011

[22] #JQ814853\_Rhinolophus\_affinis\_picornavirus\_1

[23] #Potamipivirus\_A1\_EelPV\_F15-05

[24] #JX134222\_Limnipivirus\_A1\_Bluegill\_picornavirus\_isolate\_04-032

[25] #KF306267\_Limnipivirus\_B1\_Carp\_picornavirus\_1\_isolate\_F37/06

[26] #KF183915\_Limnipivirus\_C1\_Fathead\_minnow\_picornavirus\_isolate\_fhm/1/MN/USA/2010

[27] #KP770140\_Ampivirus\_A1\_strain\_NEWT/2013/HUN

[ 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 ]

[ 1] -

[ 2] 0.497 -

[ 3] 0.496 0.516 -

[ 4] 0.761 0.753 0.740 -

[ 5] 0.803 0.775 0.803 0.802 -

[ 6] 0.795 0.795 0.784 0.790 0.697 -

[ 7] 0.820 0.778 0.797 0.791 0.679 0.626 -

[ 8] 0.802 0.775 0.774 0.766 0.698 0.708 0.670 -

[ 9] 0.793 0.783 0.795 0.809 0.699 0.691 0.641 0.515 -

[10] 0.795 0.770 0.773 0.794 0.699 0.709 0.672 0.536 0.432 -

[11] 0.786 0.772 0.784 0.795 0.736 0.730 0.681 0.615 0.608 0.632 -

[12] 0.785 0.798 0.789 0.798 0.753 0.752 0.764 0.735 0.763 0.757 0.759 -

[13] 0.788 0.791 0.787 0.807 0.748 0.753 0.770 0.733 0.764 0.757 0.752 0.107 -

[14] 0.806 0.806 0.790 0.792 0.770 0.760 0.763 0.733 0.760 0.760 0.769 0.585 0.579 -

[15] 0.800 0.790 0.777 0.786 0.776 0.762 0.785 0.746 0.740 0.742 0.754 0.555 0.558 0.556 -

[16] 0.797 0.781 0.777 0.790 0.777 0.753 0.773 0.749 0.745 0.740 0.733 0.564 0.562 0.569 0.125 -

[17] 0.789 0.756 0.764 0.795 0.772 0.742 0.759 0.705 0.722 0.728 0.742 0.667 0.671 0.653 0.648 0.644 -

[18] 0.828 0.792 0.797 0.816 0.782 0.775 0.767 0.740 0.736 0.731 0.740 0.691 0.700 0.663 0.668 0.669 0.621 -

[19] 0.816 0.788 0.808 0.801 0.795 0.787 0.786 0.778 0.780 0.782 0.767 0.713 0.714 0.711 0.717 0.717 0.703 0.697 -

[20] 0.812 0.791 0.808 0.809 0.790 0.785 0.786 0.777 0.776 0.783 0.767 0.703 0.705 0.718 0.720 0.720 0.697 0.691 0.100 -

[21] 0.816 0.800 0.771 0.826 0.770 0.763 0.757 0.770 0.732 0.748 0.751 0.785 0.780 0.784 0.773 0.774 0.785 0.781 0.793 0.792 -

[22] 0.796 0.786 0.776 0.802 0.757 0.747 0.765 0.748 0.746 0.755 0.753 0.780 0.775 0.772 0.783 0.791 0.788 0.791 0.801 0.797 0.307 -

[23] 0.806 0.793 0.801 0.775 0.773 0.766 0.762 0.765 0.743 0.765 0.756 0.789 0.785 0.786 0.766 0.767 0.764 0.754 0.805 0.798 0.793 0.794 -

[24] 0.843 0.831 0.831 0.868 0.832 0.804 0.815 0.808 0.825 0.829 0.831 0.841 0.836 0.835 0.840 0.838 0.809 0.815 0.820 0.827 0.846 0.830 0.817 -

[25] 0.843 0.825 0.825 0.844 0.825 0.786 0.799 0.818 0.825 0.811 0.819 0.820 0.820 0.827 0.833 0.827 0.803 0.797 0.818 0.819 0.854 0.842 0.798 0.410 -

[26] 0.844 0.823 0.826 0.847 0.830 0.797 0.818 0.810 0.815 0.816 0.824 0.844 0.841 0.837 0.822 0.826 0.814 0.812 0.820 0.827 0.852 0.839 0.810 0.430 0.298 -

[27] 0.912 0.934 0.924 0.910 0.925 0.911 0.923 0.918 0.919 0.921 0.916 0.911 0.919 0.904 0.912 0.915 0.916 0.909 0.923 0.920 0.910 0.909 0.918 0.921 0.915 0.926 -

The number of amino acid differences per site from between sequences are shown. Standard error estimate(s) are shown above the diagonal. The analysis involved 27 amino acid sequences. The coding data was translated assuming a Standard genetic code table. All ambiguous positions were removed for each sequence pair. There were a total of 1168 positions in the final dataset. Evolutionary analyses were conducted in MEGA5 [1].

\_\_\_ within type comparison, \_\_\_ between types/within species comparison,

\_\_\_ between species/within genus comparison, \_\_\_ between genera comparison