

Transmembrane segment

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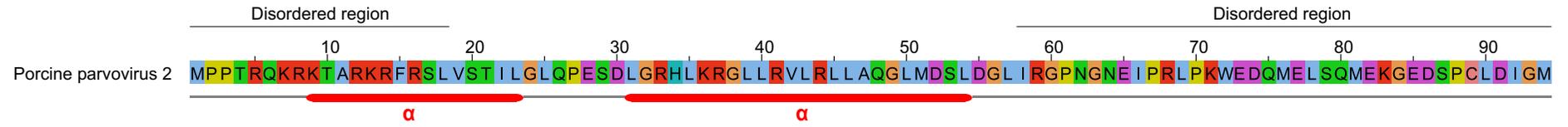
L

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47

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54

Parvovirus B19	1	- - - - - MDSYL T - T P - M P Y H P V A V M Q N L E E K M Q Y Y L - V - K - T Y T S L G K - - - - L - A Y N Y P V L T M L G L A M S Y K L G P R K V L L T V L Q - - - - - G F M T L G I A N W L S W E - - - - -	81
Simian parvovirus	1	- - - - - M D L S Q I S - E K - L R Q N T N P L M N L E E M T Q S L L - K - K - T Y T K G V E - - - - L - V L H Y P L V T T L A L V I S F K L G I P K V W W T L R L - - - - - G F M I L G I L N L L N W E - - - - -	81
Rhesus macaque parvovirus	1	- - - - - M D L S L I S - E S - L R Q N M N P L M N L E E M T Q S L V - K - R - I Y T K G V D - - - - L - V C N Y P S V I T L A L V M S Y R L G P R R V L W M L Q L - - - - - G V M I S G I L N L L S - - - - -	79
Pigtailed macaque parvovirus	1	- - - - - M A L S Q I S - E N - L R L S M R S L M N L E E L T Q S L - L - V - K T Y T M G V Q - - - - L - V S H Y P S L T I L G L G I L Y K L G P Q R T W W M L Q L - - - - - G F T I T G I L N L L S - - - - -	79
Seal parvovirus	1	- - - - - M S L S Q N R S W Q E I L S P - L E - S G L Q T M V I - - - - E - A L H Y P A L T T L G L A I A W K K G P Q L L G W T L R L - - - - - A Y T I L G I V S S R S W A L I P T H T S H P L T R N S S R Q P P E K R V S R Q Q W S R L S L S	103
Chipmunk parvovirus	1	- - - - - M V Q L F L N Q T E T L L Q N N - L N - E A W Q S L I T - - - - G - A S T Y P L T G I L A L A I L W K T A P L L T R L T Q W L - - - - - V F T T L G T L T - - - - -	65
Human parvovirus 4	1	- - - - - M S E S T L L V T A A M K E L I P L - L N - Q T L S G L P E - - - - A - G L L S P G I I M L V L V I L W I V V P L R D Q W M R Q Q - - - - - N I M M N G T Q R - L S M G T S L I Y M V T A L T D - - - - -	83
Porcine hokovirus	1	- - - - - M E P I A T V A T V I K E L V P L - V N - Q T L N A L P S A - - - - P E S W S P G T I M L V L V I L W I M L P L R D Q W M K Q R - - - - - N T M N G T M K C F A M V I C H T S M G E G L I G - - - - -	84
Bovine hokovirus	1	- - - - - M L D Q A T T V A T V I R E L V P L - V N - Q T L S A L P T - - - - A - D S W S P A T I M L V L V I L W I M V P L R D Q W M R Q R - - - - - S T M N G T M K C C L M A T C H M F M V T G Q I D - - - - -	84
Deer tetraparvovirus	1	- - - - - M E S V V T V A T V L E E L I P L - V N - Q T L N A L P K - - - - G - D F L S P V I V M L V L V I L W I M A P L R A Q W T R P L - - - - - N T M T N G T Q I C Y L M G T Y H T S M V M E Q T D L - - - - -	84
Ovine hokovirus	1	- - - - - M A M A E V S T L A T V V Q E L L P L - V N - Q T L N D L P K - - - - G - D F W S P V T I M L V L V I L W I M V P L R D Q W T R Q R - - - - - N I M T N G T M K C Y H M G M S H I Y T D T G L T D - - - - -	85
Rodent tetraparvovirus	1	- - - - - M A D L L L P L S S I M R E L L P A - L N - H T A T E L A R E - - - - G V S W S P G T I M L V L V I L W I M A P L R D R L M R P Q - - - - - R D T M N G T M R C S S M G T C P I C M A M G R M A - - - - -	85
Opposum tetraparvovirus	1	- - - - - M E V S E G L Q L R T I A Q A T - A N - Q T A E A I L P G - - - - A G S L S P A I I M L V L V I L W I M A P L R A Q W M R R L - - - - - N A M T N G M M S C W V M G M C H T - - - - -	76
Eidolon parvovirus 1	1	- - - - - M E E P L L N S A L T S L K V L A P L - V N - Q T I H S L A E - - - - V - V S L S P A T I M M V L V I L W I M A P L R A Q W M R Q L - - - - - A I M T E G M T K C F P T G T S P T - - - - -	77
Bovine parvovirus 3	1	M P P S N T S H L Q G S L P Q I S - - - - H L L S E T E M M N - - - - - L E - K S L E R L I Y K I C L L - A W E L P G V A L L L L L V V G L F - - L R R R W P Q H I L G L I S S C G T W P G M I K C I L V G P G I P H D G S S - - - - -	313

Z protein



S1 Alignment. Codon alignment of all B19V VP1 coding sequences

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GAGGGCGCCGAGAGGGGTGGGAGACGACGAGGGGGGTGGGATTGTGCGGATTAATATACCAATAAAAATG
CAGAGTGCCGACAGCACCAACAGCAACCTCAAGGTCATGGGGGGGACCGAGAGCATCCGGGCATTGGCGC
GCAGGCACCGTGTTCGGGACTCATGGGGTGACGACCACTCAGACGCGAATGGTTATATTGAGCCCTAAGACT
GACTACAAGCCGCTGTTCTGGACGCTGACACCAGTAAATTTGGCTCCGAACCAGGCATGGGGTTCTGACA
CCGTGGCAGTACTTTGATTTAACTGCTACATGAACCACTTACCCGCTCTGATTGGCAAGAGCTAGGGAGA
CGATACGACTCCATTAGGCCCAAATCCCTGAGTATATCAGTAGAGAACGTGGTATCAAAGACGTACATCAG
ACGAACAACGAGACAAACGTGCATGACTCCGGAACAGGTGGCATCTTGATATTCGAAGACTCCGAGTACACC
TTCCGTATGTATTGGGCATGCTCAAGAGGGAAACCCTGGTGCCTGAGTATTGAGTGGTACAACCCTCCC
CAATACGCCTATTTTACGGGTTTTAATCCTATTGCTTGGGACCATGCAAACGGTTCAATTAAGTATCAGGTA
CATCCTAGCGCGACACGGAGTTCTTTGTTCTGGAGGAACACGCTGCCCAAATACTGCGCAGCGGTGATGGT
ACCTCCTTTGCTTACGAGTTTCCAGTCTCGAACCTAAGCGACTGGGCACCCGTATGGGCACGCTGAACCTC
CGTCATAACCCGTTCTGCCAGCAGACTGGCGATTTATTTGGGACAGGATGGGAACAATGCACCTACATTT
TATCAGCTCAGGGGACTGATTTGGACATATTTCTCAGGGGTTTATACCGGGCCCAAGACCGTGTCTGCCG
GTTTCCACACAGCTAAGTGCTAGCAGCGACTTCGATGAGATGTCTGCGATCGCCTACGGTGTATCGACTAGT
AATAATCGGCACTCTCTCATGCCGTTTACCAGACAGGCTACAACCATCTCCACTCAAACCTATAATCGCCAG
GGGGAGGTAGAGAGAAATGTACATTTCCAATTGGGTGATATGGCCTTTGCTCGGTCTCTGCTGAAGATTCT
TTTTATGAAAGATTTGACGAGGACAAAGACTACCGTAATCCTGGTGGTTACGTCAAAAAACCTCGACCTCTG
GTGACAGCAGAGAGAGAAGGTCTTGGAGAAAGACCTGGAGATGCCCTTATGGTCCCCACCTGGGGAGCGAAA
TTACCTGGCAGCAGTACAGGACCTGGAGCCACTAAAACGGAGAAAGTTTCCCTGCCGTTTATTCCCCCATG
CCCGGGGCTTGTGGGACGAGCGCCCCCTTTGTTACGAGGACGATATCTGGTGTAAAGGCCATACACTGAT
TGCTCTTTCATGTCTGAGAAAAATAATCTCGGTGCCTGGGCGCTCGTGGATCCTCCGCCCCAAGTGTTTTTT
CGTATGCAGCCGAGGTGGGTCCGCCCGCGGACTTGGACCAACGCACCTCTCTACCGCTGCTTTGAAT
CAGTATGCCATGTTCACTGTGAGCTACACTATGGAGTGGGTGTGCGAACCGCGGAAGCACACTAGACGGCAT
AATTTGGAACCTCCGCCTCCCATGCCTTACACGGAGTCCGGCGACCCCCGTTTCTCCTTACTCGTTTCGCAT
GTTACCAATGATTACCCTCGATATTCTTTACCTGTGGAAGCGTTTCGCCAGAGGGACGCGCGCATCGTGTA
TAA-----

S5 Alignment. Alignment of the X-like protein of bovine parvovirus 3 with the reference alignment of the X proteins of erythroviruses and tetraparvoviruses, in text format.

We used MAFFT-add to align the X-like protein of bovine parvovirus 3 with the reference alignment of the X protein of erythro- and tetraparvoviruses, which is derived from the alignment of the PLA2 domain and presented in Figure 6 (see main text).

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>ParvovirusB19/1-81
-----MDSYLT-TP-MPYHPVAVMQLNLEEKMQYYL-V-K-TYTSLGK----L-AYNYPVLTMLGLAM
SYKLGPRKVLTLVQL-----GFMTLGIANWLSWE-----
>SimianParvo/1-81
-----MDLSQIS-EK-LRQNTNPLMNLEEMTQSL-L-K-K-TYTKGVE----L-VLHYPLVTTLALVI
SFKLGIPKVVWTLRL-----GFMIILGILNLLNWE-----
>RhesusMacaque/1-79
-----MDLSLIS-ES-LRQNMNPLMNLEEMTQSLV-K-R-IYTKGVD----L-VCNYPVITLALVM
SYRLGPRRVLWMLQL-----GVMISGILNLLS-----
>PigtailedMacaque/1-79
-----MALSQIS-EN-LRLSMRSLMNLEELTQSL-L-V-KTYTMGVQ----L-VSHYPSLTILGLGI
LYKLGPRQRTWMLQL-----GFTITGILNLLS-----
>SealParvo/1-103
-----MSLSQNRSWQEILSP-LE-SGLQTMVI----E-ALHYPALTTLGLAI
AWKKGPRQLLWTLRL-----AYTILGIVSSRSWALIPHTSHPLTRNSSRQPPEKRVSRQQWSRLSLS
>ChipmunkParvo/1-65
-----MWQLFLNQTETLLQNN-LN-EAWQSLIT----G-ASTYPLTGILALAI
LWKTAPLLTRLTQWL-----VFTTLGTLT-----
>Human_parvovirus_4/1-83
-----MSESTLLVTAAMKELIPL-LN-QTLSGLPE----A-GLLSPGIIMLVLVI
LWIVVPLRDQWMRQQ-----NIMMNGTQR-LSMGTSLIYMVTALTD-----
>PorcineHokoHK7/1-84
-----MEPIATVATVIKELVPL-VN-QTLNALPSA----PESWSPGTIMLVLVI
LWIMLPLRDQWMKQR-----NTTMNGTMKCFAMVICHTSMEGLIG-----
>YakHoko/1-84
-----MLDQATTVATVIRELVPL-VN-QTLSALPT----A-DSWSPATIMLVLVI
LWIMVPLRDQWMRQR-----STTMNGTMKCCLMATCHMFMVTGQID-----
>DeerTetraparvo|NC_031670_1_cds_YP_009315887_1_2/1-/1-84
-----MESVVTVATVLEELIPL-VN-QTLNALPK----G-DFLSPVIVMLVVI
LWIMAPLRAQWTRPL-----NTMTNGTQICYLMGTYHTSMVMEQTDL-----
>OvineHoko1/1-85
-----MAMAEVSTLATVVQELLPL-VN-QTLNDLPK----G-DFWSPVTIMLVLVI
LWIMVPLRDQWTRQR-----NIMTNGTMKCYHMGMSHIYTDGLTD-----
>RodentTetraparvo/1-85
-----MADLLLPLSSIMRELLPA-LN-HTATELARE----GVSWSPGTIMLVLVI
LWIMAPLRDRLMRPQ-----RDTMNGTMRCSSMGTCPCICMAMGRMA-----
>OpposumTetraparvo/1-76
-----MEVSEGLQLLRTIAQAT-AN-QTAEAILPG----AGSLSPAIIIMLVLVI
LWIMAPLRAQWMRRL-----NAMTNGMMSCVWVMGCHT-----
>EidolonParvo1/1-77
-----MEEPLLNSALTSLKVLAPL-VN-QTIHSLAE----V-VSLSPATIMWVVI
LWIMAPLRAQWMRQL-----AIMTEGMTKCFPTGTSP-----
>BovineParvovirus3/1-99
MPPSNTSHLQGSPLQIS----HLLSETEMMN-----LE-KSLERLIYKICLL-AWELPGVALLLLLV
VGLF--LRRRWPQHILGLISSCGTTWPGMIKICILVGPPIPHDGSS-----
```

S6 Alignment. The X ORF has a potential AUG start codon in all erythro- and tetraparvoviruses

Codon-based nucleotide alignment of the X ORFs of erythro- and tetraparvoviruses, derived from the reference alignment of the VP1 protein by using TranslatorX, as in Fig 5 of the main text. Numbering corresponds to the VP1 CDS.

Putative AUG start codons of the X ORF are highlighted in yellow. For each species, the putative AUG start codon is conserved in all isolates (not shown in this alignment). Note how the AUGs tend to be in a similar location within erythroparvoviruses (except in chipmunk parvovirus and seal parvovirus), and within tetraparvoviruses.

UAG or **UGA**: stop codon

AUG: putative start codon of the X ORF

Erythroparvoviruses

Parvovirus B19	185	UAG UUGCUCGCAUUAAAAUAACCUUAAAAAUUCUCCAGACUUUAUAGUCAUCAUUUCAAAGUC AUG GACAGUUUUCUGACCACCCCAU-----GCCUUUCAUCCAGUAGCAGUCAUG	301
Simian parvovirus	293	ACCUUAAA---CAACAUUUACAAGACUAUAUAGACAAUCCAGAUAAAGUACACUU UAG ACUUGUCUC AUG GACCUCUCCAGAUUUCAGAGAA-----ACUGAGGCAGAACACAAAUCUUUA	406
Rhesus macaque	299	AUCUACAA---AAACAAAUUGAAAACUAUAAAAUAAUCCAAACAAAUACAUUACAGUUGUCUC AUG GACCUCUCCUGAUUUCAGAGAG-----UCUGAGGCAAAACAUGAAUCCUCUA	412
Pigtailed macaque	194	UGGUUAAU---CGCAUUAAAAAAAU UAA AGACAAUCCUGAUUUUACACUGACUCCUUAGUC AUG GGCGCUCUCCAGAUUUCGAGAA-----UCUAAGGCUGAGCAUGGAAAGUCUA	307
Chipmunk parvovirus	311	CCGUAGCU---AACACAGCCAAGCGGU UAA AAACUGACGAGGAUCCCUA-----UCCUUUGGGGCCCCCCAC UAA CAGAAAACGCCCCGGUUCUCCGUUGCGGAGCCAG AUG UGGCAAUUGUUU	427
Seal parvovirus	338	GGAGGCACUAUGGAUCCUCAAAGGAGCAGGGCGCAGCACCCCGCUGAUCCAGACAU UAG CAGGCCUCCUCUCACAUACAGGGCCCG-----GUGUUGGGGAGC AUG UCCCUAUA	454

Tetraparvoviruses

Human parvovirus 4	467	UUUUUGAA---GACUCGC UAA CGAACUUUGCCAAAGAAGACU UAG ACACCCUGGCAACAACUCCACGAGCAGUUUAUCAAACUCUUACCCU-----CCAG AUG UCGGAGUCCACCUUGUUA	580
Porcine hokovirus	464	UCUUUGAG---GAGAGUGUAUCUCCUUGGUCUGAGGAAGACAAAAAAUU UGA AACAGAUUGAAGGGCAGUUCAGGAAAUUCCAUCCACCCACAGAUACGGAGG AUG GAGCCGAUAGCUACG	586
Yak hokovirus	491	UUUUUCAG---GGCAUGUUUUCUCCCAUCAAACCGAAGAUAGACCUAU UAG ACACCAUACAGAAAGCAGUUUGAGGAGUUUUUUUAUCCUCCUA UAG UGGAAC AUG CUGGACCAGGCGACUACA	613
Deer tetraparvovirus	491	ACUUUGAU---GGCAUGUUGUACCUAUUCCAGUGGAACAACAUCCAAUUG UAG AGCAGA UAA AGGCUCAGUUUU UAA AUAUUUUUUAUCCGCCACCU UAG CUGGAG AUG GAGUCGGUGGUGACG	613
Ovine hokovirus	491	UUUUCAAU---GGUAUGUUGACGCCGUUUGAUUCCGACCAGCGACCCGUGGUCGAGCAGAUUACACAGCAGUUCU UAG ACAUCUUUACCCCCACCAGUGCAUGGCC AUG GCGGAGGUGUCGACA	613
Rodent tetraparvo	467	ACUUUGAC---CAGUUCAUACCCCGUGGACCCCGGGGAGCCCGAGAUUC UAA AGCAGGCCUUCAGCGCAUG UAG AAUUAUUAUCCACCCCGCCCU---CAGG AUG GCGGACCUUCUCCUCC	586
Eidolon parvovirus	458	UUUUUGAU---GAGUCUGUGCUGGGCAGCGUUAGCGGAGACCCGAAAACUUUGCUUUUGUGAAGCAGGCAG UAG UUAGUGCUUUUCAUCCUGCA-----G AUG GAGGAGCCGCUACUGAACAGC	574
Opposum tetraparvo	440	UUUUUACU---GACUCUAUUAGUGCGGGUG UAG CUGGAACA---CCACUCUGGGGAGAUACUGCAGAAGCUCUCAUGUCGGCCCU-----GCAGGGA AUG GAGUAUCAGAGGGU	544