

Sequencing

Argelia Cuenca

	Amp. I	Amp. II	Amp. III	Amp. V	Amp. VII	Amp. VIII	Amp. IX
	VHSV, Ia	IHNV, E (M) + IPNV, 5	SVCV, Ia	EHNV	ISAV, HPRΔ	SAV3 + ISAV, HPRΔ	KHV, CyHV-3
No. of participants performing sequencing	33	35	25	42	34	33	24
No. of participants getting full score	28	23	23	42	29	16	24
No. of correct sequences provided without genotype assigned or incomplete sequence	3	10	0	1	3	15	0
No. of incorrect genotype provided	2	2	2	0	2	2	0

Genotype
Genogroup
Deleted or not (ISAV)

Ampoule number	Pathogen Identification	Amplicon sequenced (ref and primers)	Genotype	Sequence	Possible isolates:
Ampoule III	IHNV	Mid G gene Upstream Primer 5'-AGA-GAT-CCC-TAC-ACC-AGA-GAC-3'; Downstream Primer 5'-GGT-GGT-GTT-GTT-TCC-GTG-CAA-3'. Emmenegger E.J., Meyers T.R., Burton T.O & Kurath G. (2000). Genetic diversity and epidemiology of infectious hematopoietic necrosis virus in Alaska.	U	TTTTATTGGAGGAA AATGTACCAAATCA CCCTGCCAGACTCA TTGGTCCA ACGTAGTTTGGATG GGTGATGCAGGGAT ACCAGCCTGTGATT	DQ164100.1 Infectious hematopoietic necrosis virus isolate BLk94 glycoprotein (G) gene, 100% 645bps. Infectious hematopoietic necrosis virus gene for glycoprotein, complete cds, strain: ChAb76 643/645 99%
Ampoule VII	ISAV HPR-deleted	HA gene Mjaaland et al (2002). Virology, 304:379-391 Klon1EGFP-F1 5'- GGGCTAGCATGGCAGCATTATAATT-3' Klon1EGFP-R1 5'- GGGGTACCGTAGCAACAGACAGGCTCGAT-	HPR deleted	CCAATGACTGCACT GACGGACTACTGA CATGATCATCCCAA CTTCGATG ACACTGGACAACGC GGCAAGGGAGCTGT ACCTGGGAGCATGC	ISAV4(90/09/400) (Genbank Accession DQ785248.1)
Only fill in the Genotype ↑					
No isolate name, serotype, etc. ↓					
Ampoule III	IHNV	Emmenegger et al. (2000) Sequence (5' → 3') IHN-GF1 AGA GAT CCC TAC ACC AGA GAC IHN-GR1 GGT GGT GTT GTT TCC GTG CAA Enzmann et al. (2005) Sequence (5' → 3') IG1 ATG GAC ACC ATG ATC ACC	BLK94, genogroup U, subtype P	GTGCAATCCGTGAA AGCCCTCCCACTCAT CCCCAAAGGGTCGT TCCCATTTCTGTGAA GCTGGTAGCGCGAT GGGCCCTGTACGTC GTCCTGICCTIGGA	100% query cover and 665nt 100% identical with: DQ164100.1 - Infectious hematopoietic necrosis virus isolate BLk94 glycoprotein (G) gene, complete cds
Ampoule VII	ISAV (HPRdel)	HPR of segment 6 (HE gene); Markussen T, Jonassen CM, Numanovic S, Braaen S, Hjortaas M, Nilsen H, Mjaaland S. Evolutionary mechanisms involved in the virulence of infectious salmon anaemia virus (ISAV), a piscine orthomyxovirus. Virology. 2008 May	HPR group 2/ EU-G2 group	TGACCAGACAAGCT TAGGTAACACAGAC ACACTTATCATGAG GGAGGTAGCATTGC ATAAGGAGATGATC AGTAAACTTCAGAG GAACATCACAGATG	ISAV4 90/09/400; DQ785248 Markussen, T., Jonassen, C.M., Numanovic, S., Braaen, S., Hjortaas, M., Nilsen, H. and Mjaaland, S. Evolutionary mechanisms involved in the virulence of infectious salmon anaemia virus (ISAV), a piscine orthomyxovirus. Virology 374 (2), 515-527 (2008) ISAV F72h/02: Isolate from Nova Scotia, belonging to the F11-E subgroup within the F11-G2

To be inserted in “Concluding results” for Ampoule I-IX	To be inserted in “Genotype” for Sequencing results.
VHSV	I (a-e), II, III, IV (a-d)
IHNV	U, M, L, E, J
EHNV	EHNV
Ranavirus – NOT EHNV	Not EHNV
SVCV	Genogroup 1 (a-d), 2, 3, 4
Birnavirus II (Telinavirus)	-
Perch Rhabdovirus	-
IPNV	Genogroup 1, 2, 3, 4, 5 Optionally: within genogroup 1 – genotype (1-4)
HPR-deleted ISAV or HPRO ISAV	HPR-deleted / HPRO
KHV	CyHV (1-3)
SAV	1, 2, 3, 4, 5, 6

VHSV Isolate DK-9895174 (Ia)

<input checked="" type="checkbox"/>	Viral hemorrhagic septicemia virus strain DK-9895174, complete genome	Viral hemorrhagi...	2815	2815	100%	0.0	100.00%	11159	MK829413.1
<input checked="" type="checkbox"/>	Viral hemorrhagic septicemia virus viral cRNA for glycoprotein (G gene), isolate DK-9895390	Viral hemorrhagi...	2815	2815	100%	0.0	100.00%	1524	LN876846.1
<input checked="" type="checkbox"/>	Viral hemorrhagic septicemia virus viral cRNA for glycoprotein (G gene), isolate DK-9895391	Viral hemorrhagi...	2804	2804	100%	0.0	99.87%	1524	LN876847.1
<input checked="" type="checkbox"/>	Viral hemorrhagic septicemia virus isolate DK-9895024, glycoprotein (G) gene, complete cds	Viral hemorrhagi...	2804	2804	100%	0.0	99.87%	1524	AY546599.1



Viral hemorrhagic septicemia virus viral cRNA for glycoprotein (G gene), isolate DK-9895390

GenBank: LN876846.1

[FASTA](#) [Graphics](#)

REFERENCE 1
 AUTHORS Cieslak, M., Mikkelsen, S.S., Skall, H.F., Baud, M., Diserens, N., Engelsma, M.Y., Haenen, O.L., Mousakhani, S., Panzarin, V., Wahli, T., Olesen, N.J. and Schutze, H.
 TITLE Phylogeny of the Viral Hemorrhagic Septicemia Virus in European Aquaculture
 JOURNAL PLoS ONE 11 (10), E0164475 (2016)
 PUBMED [27760205](#)



FEATURES	Location/Qualifiers
source	1..1524 /organism="Viral hemorrhagic septicemia virus" /mol_type="viral cRNA" /isolate="DK-9895390" /host="Oncorhynchus mykiss" /db_xref="taxon:11287" /country="Denmark" /collection_date="1998" /collected_by="EU RL Fish Diseases, Copenhagen, Denmark, N. J. Olesen"
gene	<div style="border: 1px solid red; padding: 2px;">/note="Genogroup: I; Sub-Genogroup: a; isolate name synonym: V04-990K"</div> 1..1524 /gene="G"

Tree Diagram

Download tree in Newick format

Tree type :

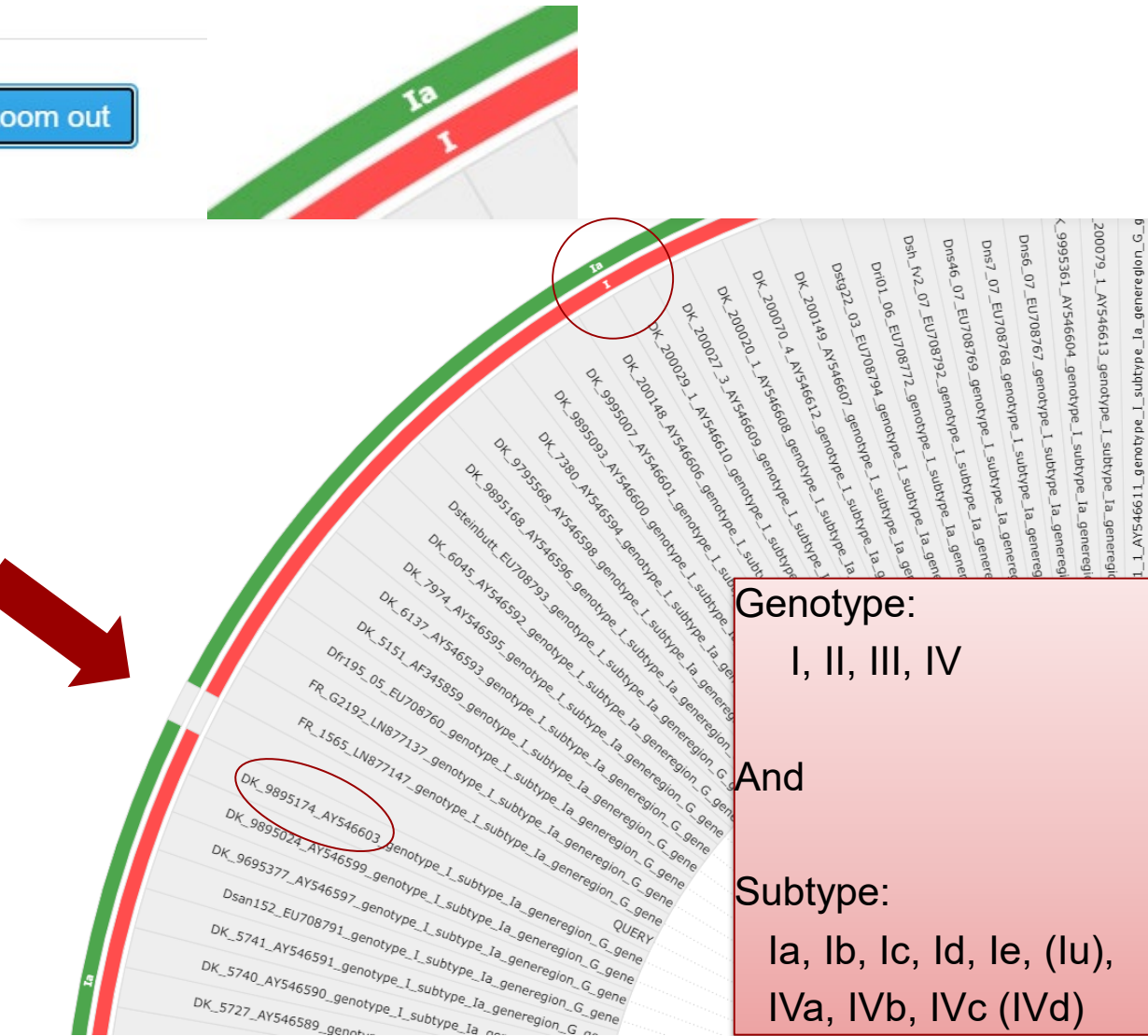
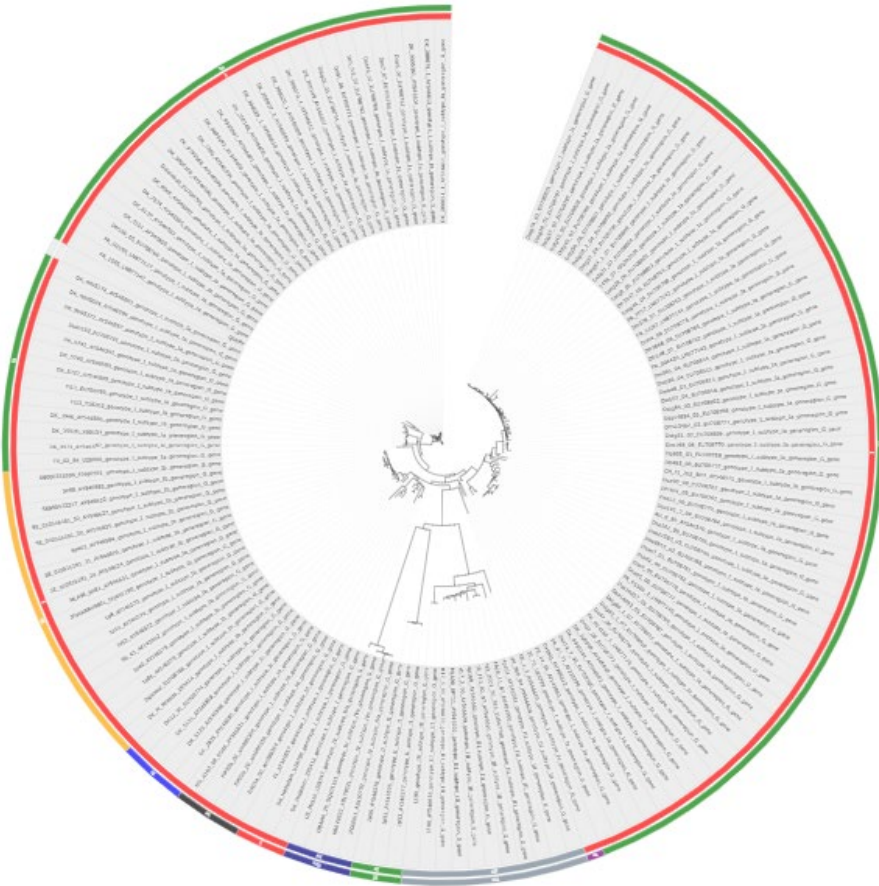
Circular

Rectangular

Zoom level :

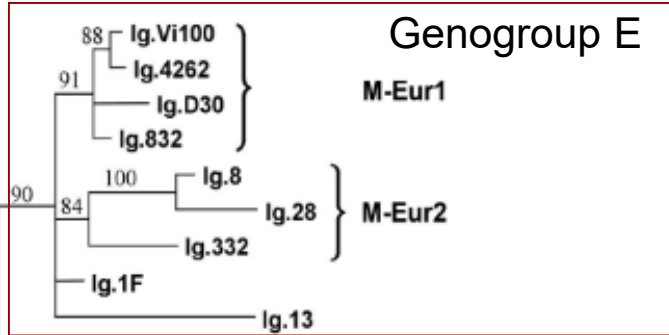
+ zoom in

- zoom out



Genotype:
I, II, III, IV
And
Subtype:
Ia, Ib, Ic, Id, Ie, (Iu),
IVa, IVb, IVc (IVd)

Ampule II- IHNV - Isolate 32/87 genogroup E



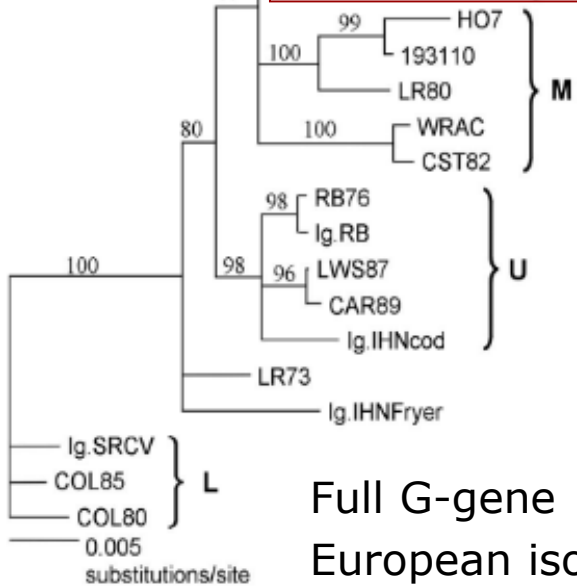
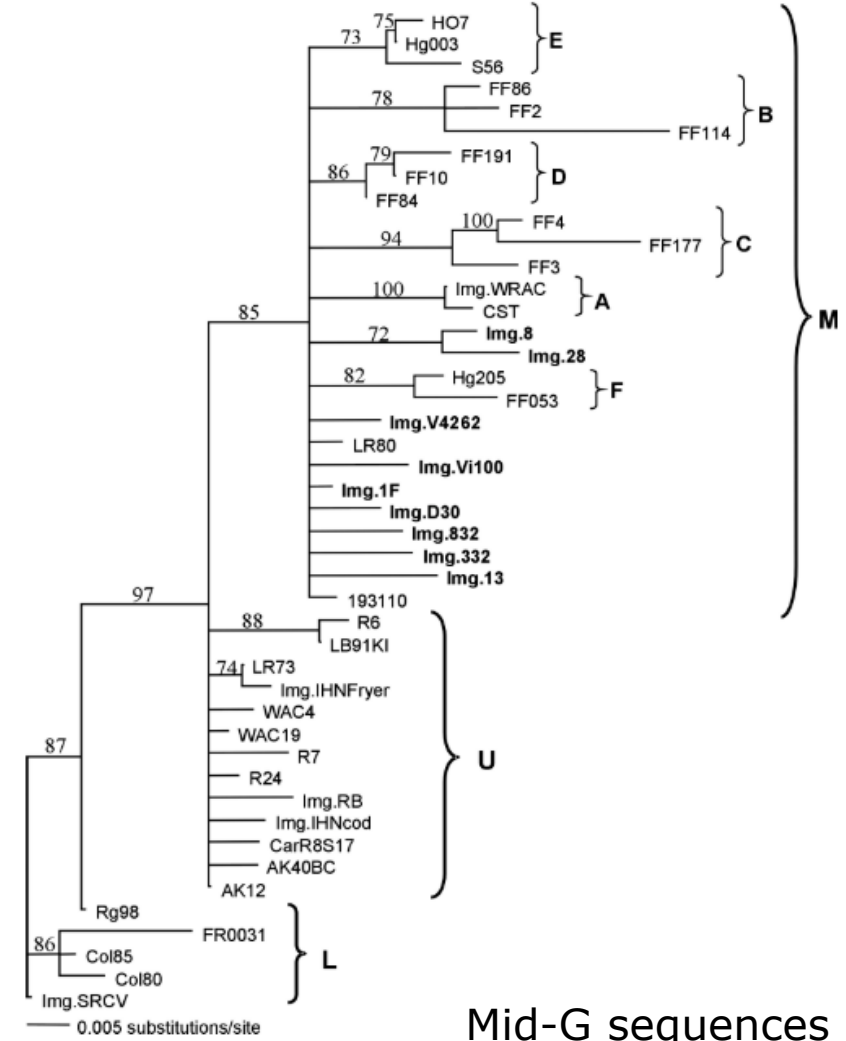
Genogroup M or E?

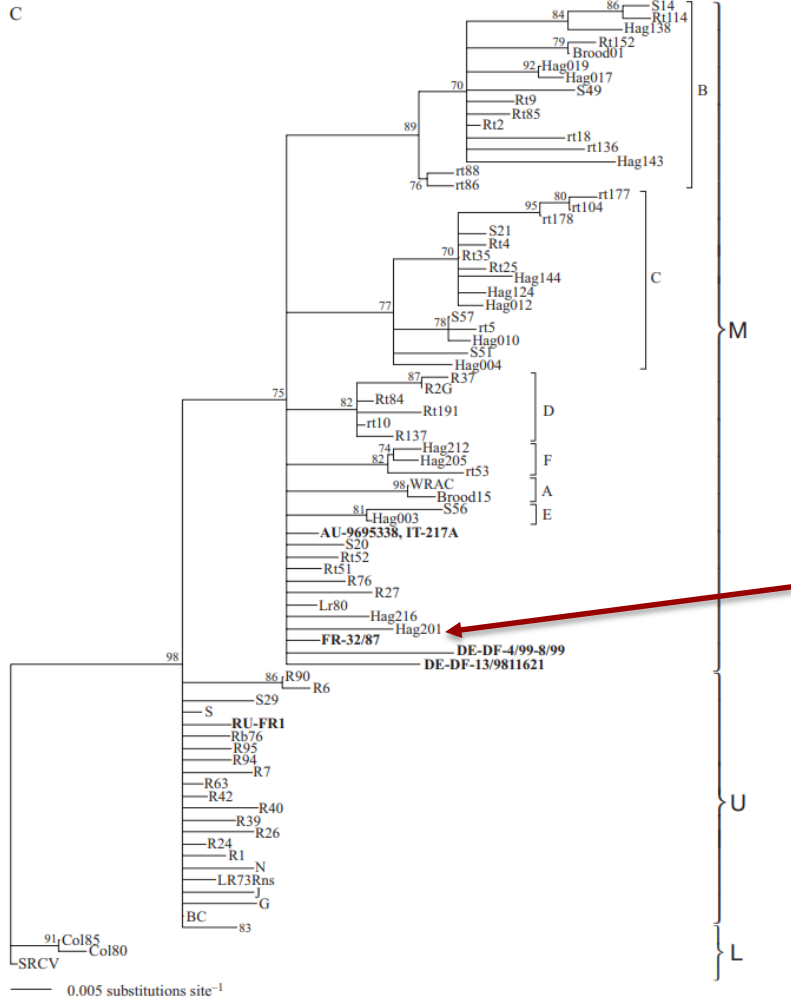
Vol. 66: 187–195, 2005	DISEASES OF AQUATIC ORGANISMS Dis Aquat Org	Published September
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Infectious hematopoietic necrosis virus: monophyletic origin of European isolates from North American Genogroup M

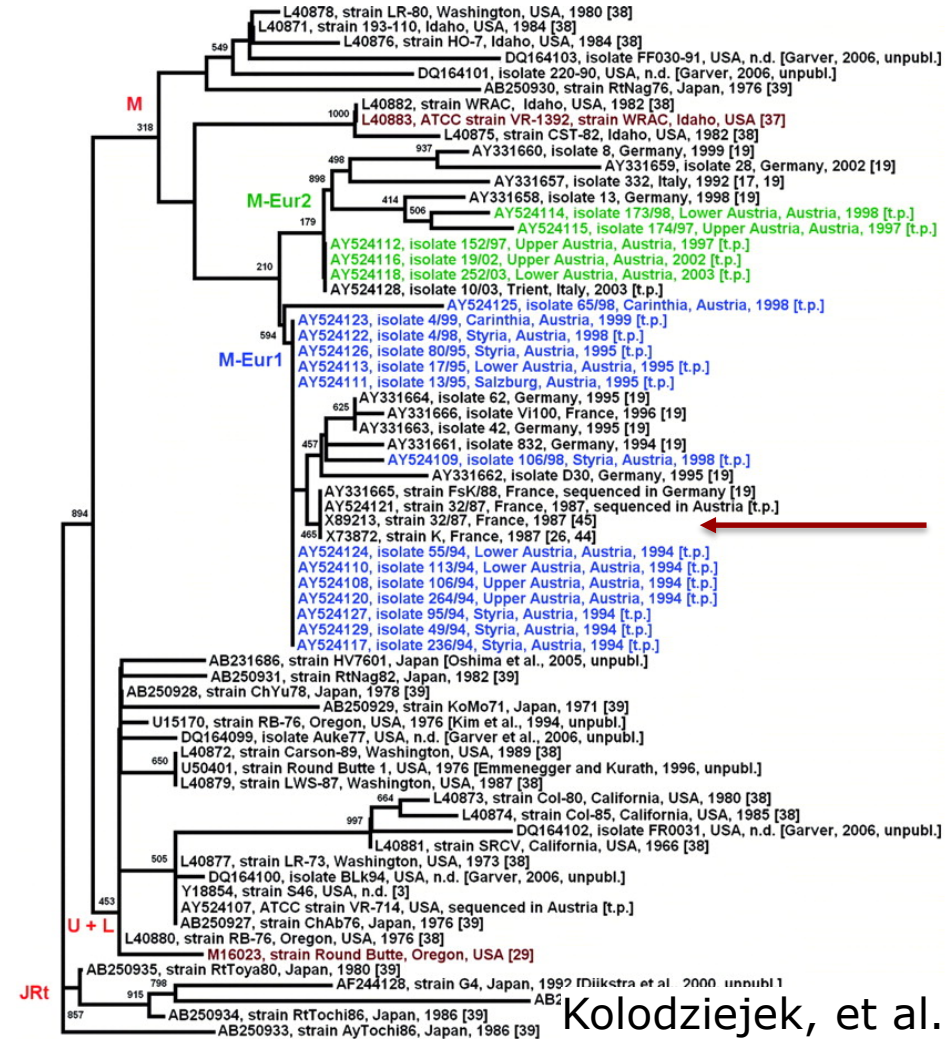
P.-J. Enzmann^{1,*}, G. Kurath², D. Fichtner³, S. M. Bergmann³

Full G-gene
European isolates in bold
(1987 to 2002)





Johansson et al. 2009
doi: 10.3354/dao02108



Kolodziejek, et al. 2008

DOI: 10.1128/JCM.00566-07

Ampule II- IPNV - Isolate Sp genogroup 5

Vol. 45: 89-102, 2001

DISEASES OF AQUATIC ORGANISMS
Dis Aquat Org

Published June 20

Phylogenetic relationships of aquatic birnaviruses based on deduced amino acid sequences of genome segment A cDNA

S. Blake¹, J.-Y. Ma², D. A. Caporale³, S. Jairath¹, B. L. Nicholson^{1,*}

¹Department of Biochemistry, Microbiology and Molecular Biology, University of Maine, Orono, Maine 04469, USA

²Millennium Pharmaceuticals, Inc. Cambridge, Massachusetts 02139, USA

³Department of Biology, University of Wisconsin, Stevens Point, Wisconsin 54481, USA

Genogroup 1 to 5

Optional

Genotype 1 to 4 (in genogroup 1)

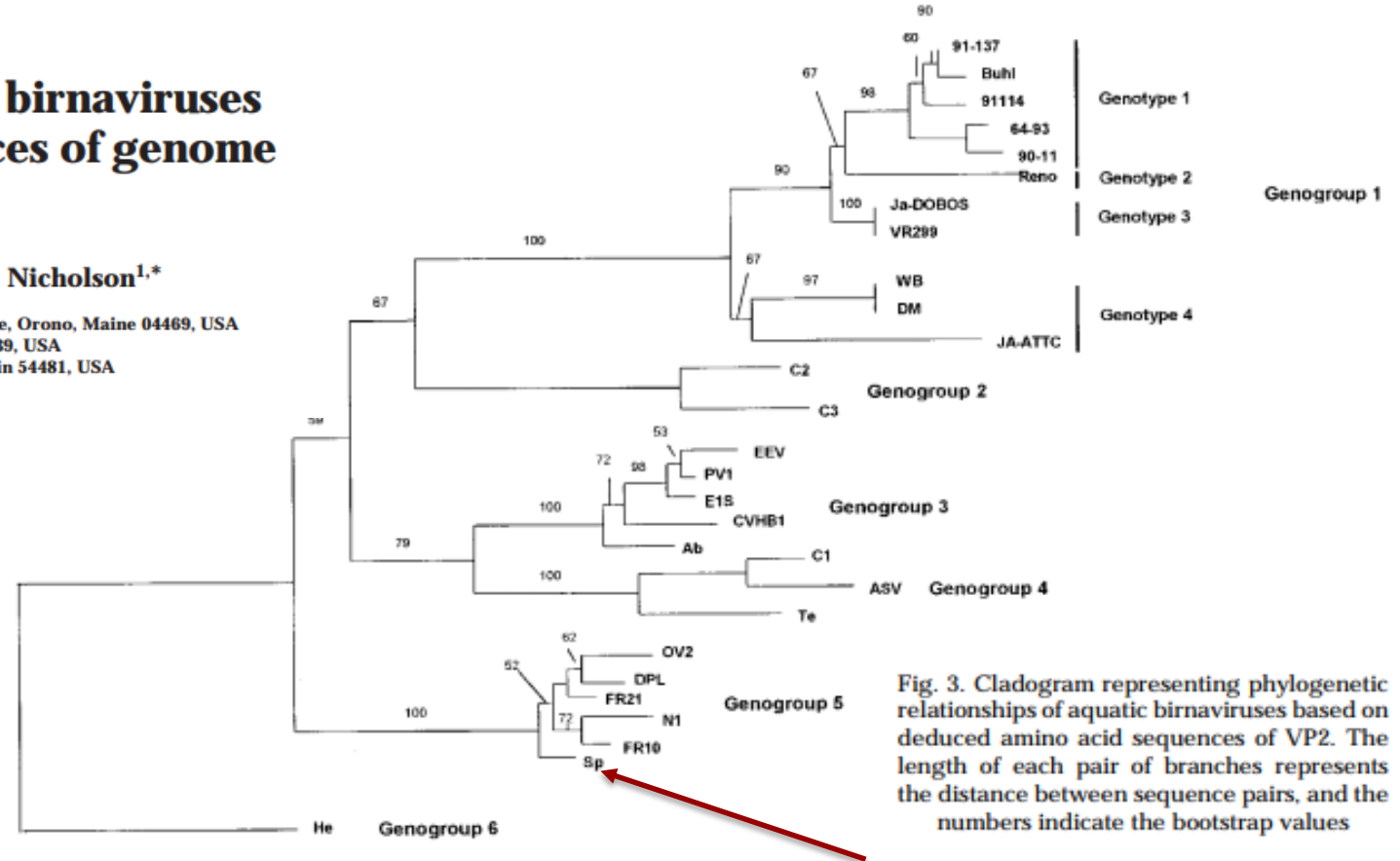
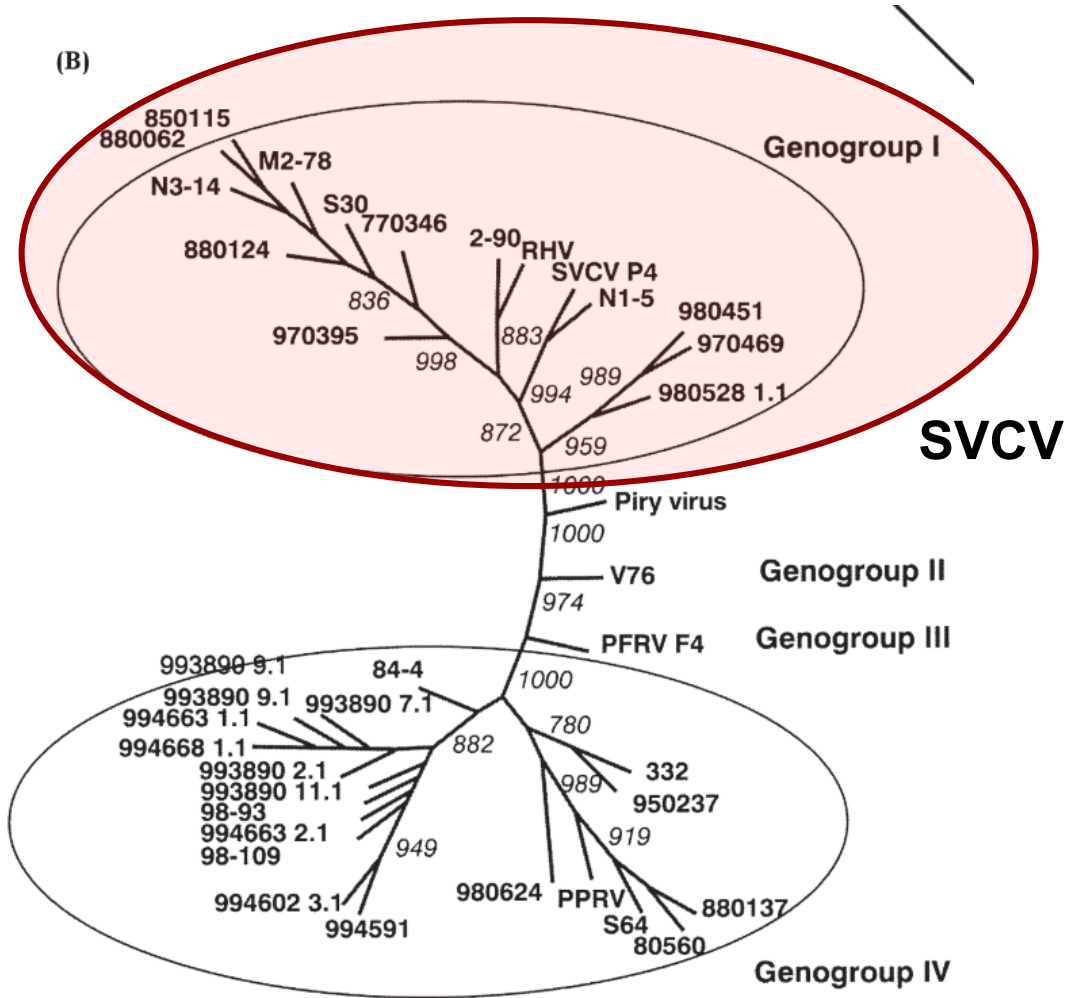


Fig. 3. Cladogram representing phylogenetic relationships of aquatic birnaviruses based on deduced amino acid sequences of VP2. The length of each pair of branches represents the distance between sequence pairs, and the numbers indicate the bootstrap values

Partial VP2 sequences

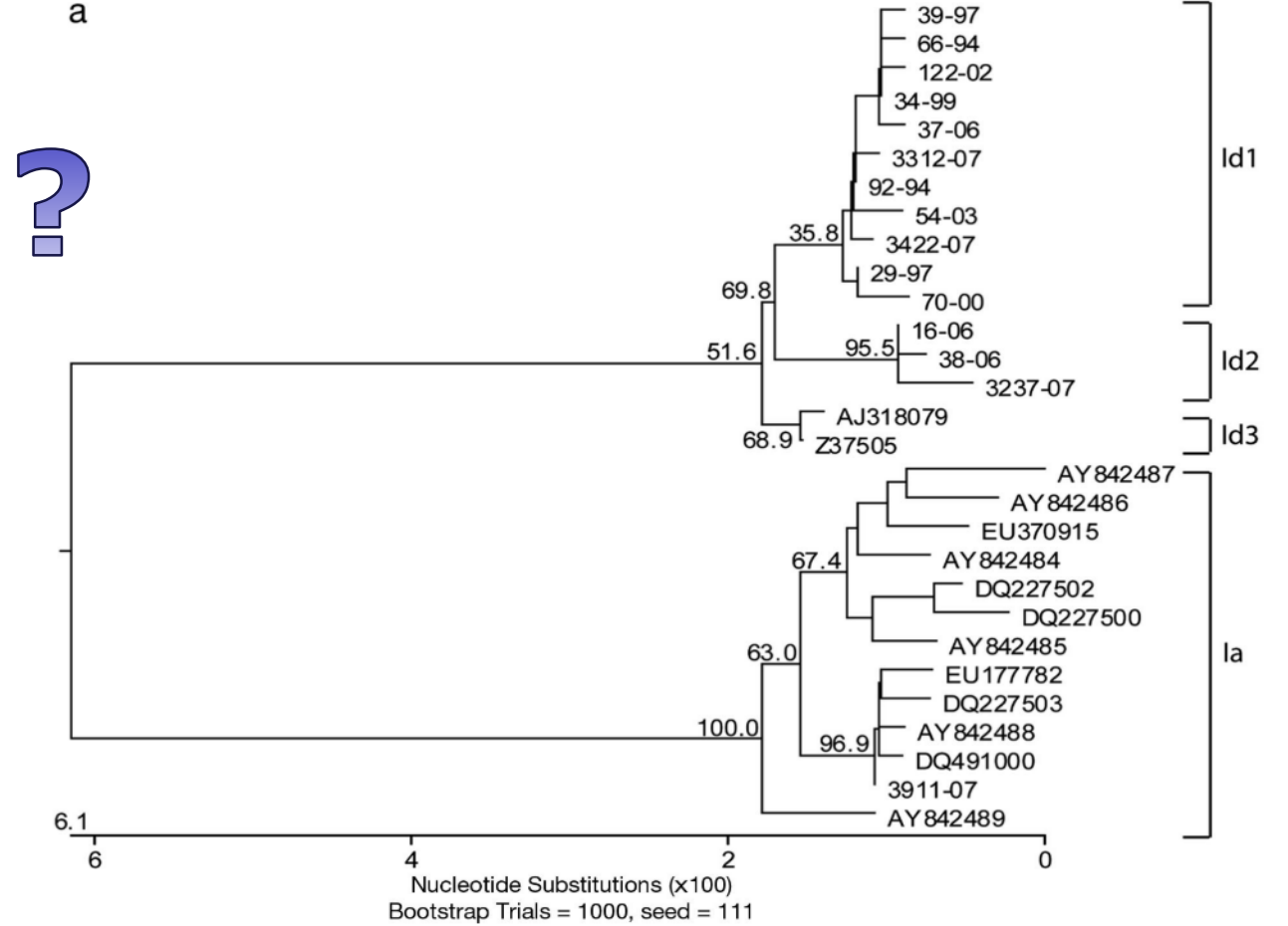
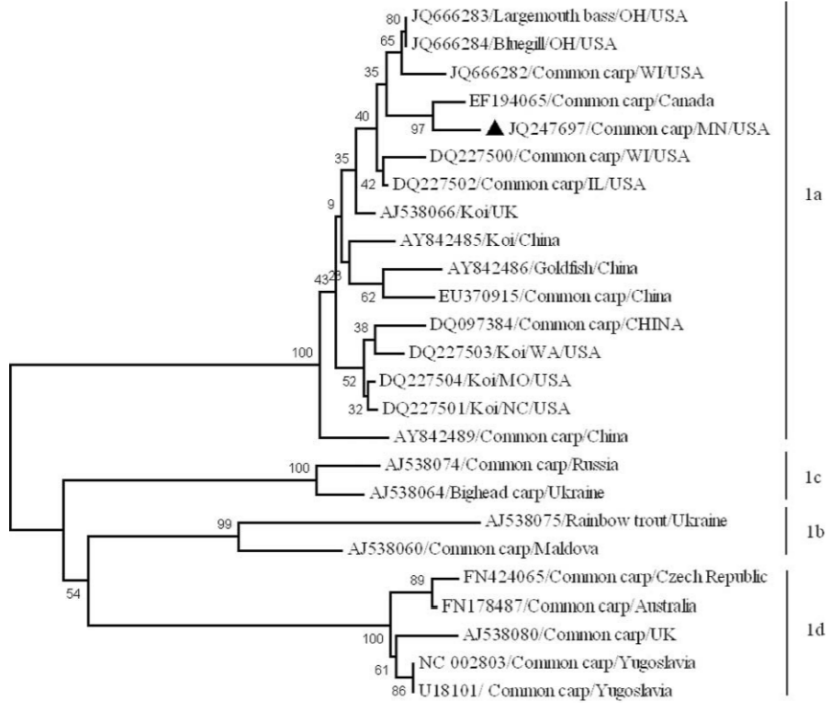
Ampule II- SVCV

Vol. 53: 203–210, 2003	DISEASES OF AQUATIC ORGANISMS Dis Aquat Org	Published February 27
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Nucleotide sequence analysis of the glycoprotein gene of putative spring viraemia of carp virus and pike fry rhabdovirus isolates reveals four genogroups

D. M. Stone^{1,*}, W. Ahne², K. L. Denham¹, P. F. Dixon¹, C. T.-Y. Liu¹, A. M. Sheppard¹, G. R. Taylor¹, K. Way¹

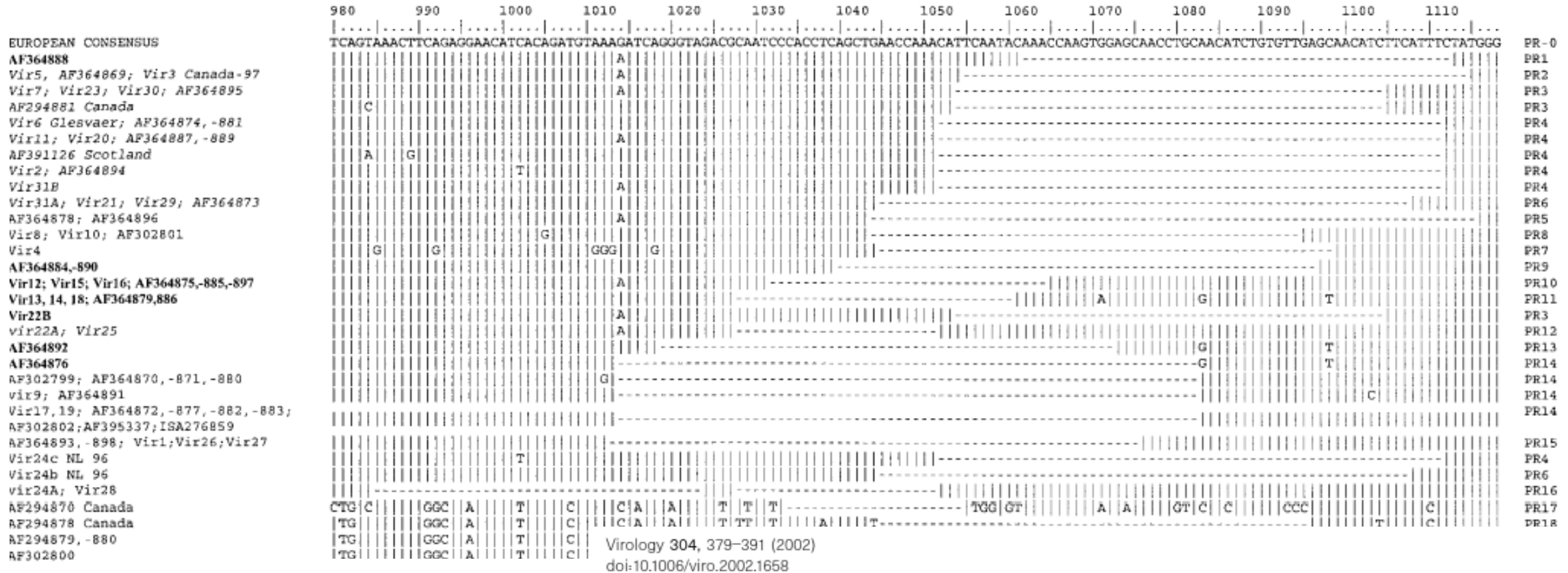


Phelps et al. 2012.
DOI: 10.1080/08997659.2012.711267

Basic et al. 2009

Genogroup I (a-d)

Ampule VIII - Infectious Salmon Anaemia Virus ISAV 390/98, HPRΔ



Highly polymorphic region
Hemagglutinating gene
Segment 6

Polymorphism in the Infectious Salmon Anemia Virus Hemagglutinin Gene: Importance and Possible Implications for Evolution and Ecology of Infectious Salmon Anemia Disease

Siri Mjaaland,^{*1} Olav Hungnes,[†] Ann Teig,^{*} Birgit H. Dannevig,[‡] Kristin Thorud,[§] and Espen Rimstad^{*}

	320	330	340	350	360	370	
AF364888 47/99 SF	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTFNT	-----	-----	-----	-----	MGVAGF PR1
Vir3 Canada-97	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	GVAGF PR2
Vir5 SF-96	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	GVAGF PR2
AF364869 18/96 SF	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	GVAGF PR2
Vir11 TR-96	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
Vir20 TR-98	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
Vir2 Ho-91	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
AF364894 6/91 Ho	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
Vir6 Glesvaer Ho-90	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
AF364874 38/98 NT	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
AF364881 37/98 Tr	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
AF364887 33/98 Tr	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
AF364889 22/96 Tr	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
AF391126 1490/98 Scotland	REVALHKEMIRKRQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
Vir7 NL-93	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	FISMGVAGF PR3
Vir23 NL-96	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	FISMGVAGF PR3
Vir30 NL-96	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	FISMGVAGF PR3
AF294881 U5575-1 Canada	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	FISMGVAGF PR3
AF364895 9/93 NL	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	FISMGVAGF PR3
Vir31B Ro-99	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	MGVAGF PR4
Vir31A Ro-99	REVALHKEMISKLQRNITDVKIRVDAI	PPQL	-----	-----	-----	-----	ISMGVAGF PR6
Vir21 SF-95	REVALHKEMISKLQRNITDVKIRVDAI	PPQL	-----	-----	-----	-----	ISMGVAGF PR6
Vir29 England-99	REVALHKEMISKLQRNITDVKIRVDAI	PPQL	-----	-----	-----	-----	ISMGVAGF PR6
AF364873 14/95 MR	REVALHKEMISKLQRNITDVKIRVDAI	PPQL	-----	-----	-----	-----	ISMGVAGF PR6
AF364878 48/99 SF	REVALHKEMISKLQRNITDVKIRVDAI	PPQL	-----	-----	-----	-----	GVAGF PR5
AF364896 46/99 MR	REVALHKEMISKLQRNITDVKIRVDAI	PPQL	-----	-----	-----	-----	GVAGF PR5
AF302801 10/93 TR	REVALHKEMISKLQRNITDVKIRVDAI	PPQL	-----	-----	-----	-----	SNIFISMGVAGF PR8
Vir8 TR-94	REVALHKEMISKLQRNITDVKIRVDAI	PPQL	-----	-----	-----	-----	SNIFISMGVAGF PR8
Vir10 TR-93	REVALHKEMISKLQRNITDVKIRVDAI	PPQL	-----	-----	-----	-----	SNIFISMGVAGF PR8
Vir4 NL-89	REVALHGEMISELRRNITDVGIGVDAI	PPQL	-----	-----	-----	-----	NIFISMGVAGF PR7
AF364884 54/00 SF	REVALHKEMISKLQRNITDVKIRVDAI	PP	-----	-----	-----	-----	RNIFISMGVAGF PR9
AF364890 57/00 SF	REVALHKEMISKLQRNITDVKIRVDAI	PP	-----	-----	-----	-----	RNIFISMGVAGF PR9
Vir16 ST-97	REVALHKEMISKLQRNITDVKIRVDAI		-----	-----	-----	-----	QVEQPATSVLSNIFISMGVAGF PR10
AF364875 28/97 ST	REVALHKEMISKLQRNITDVKIRVDAI		-----	-----	-----	-----	QVEQPATSVLSNIFISMGVAGF PR10
AF364885 25/97 ST	REVALHKEMISKLQRNITDVKIRVDAI		-----	-----	-----	-----	QVEQPATSVLSNIFISMGVAGF PR10
AF364897 27/97 ST	REVALHKEMISKLQRNITDVKIRVDAI		-----	-----	-----	-----	QVEQPATSVLSNIFISMGVAGF PR10
vir12 ST-97	REVALHKEMISKLQRNITDVKIRVDAI		-----	-----	-----	-----	QVEQPATSVLSNIFISMGVAGF PR10
vir15 ST-97	REVALHKEMISKLQRNITDVKIRVDAI		-----	-----	-----	-----	QVEQPATSVLSNIFISMGVAGF PR10
vir13 ST-96	REVALHKEMISKLQRNITDVKIRVDA		-----	-----	-----	-----	NQVEQPATSVLSNIFISMGVAGF PR11
vir14 ST-97	REVALHKEMISKLQRNITDVKIRVDA		-----	-----	-----	-----	NQVEQPATSVLSNIFISMGVAGF PR11
Vir18 ST 97	REVALHKEMISKLQRNITDVKIRVDA		-----	-----	-----	-----	NQVEQPATSVLSNIFISMGVAGF PR11
AF364886 21/96 ST	REVALHKEMISKLQRNITDVKIRVDA		-----	-----	-----	-----	NQVEQPATSVLSNIFISMGVAGF PR11
Vir22B NL-96	REVALHKEMISKLQRNITDVKIRVDAI	PPQLNQTL	-----	-----	-----	-----	FISMGVAGF PR3
vir22A NL-96	REVALHKEMISKLQRNITDVKIRVDA		-----	-----	-----	-----	FNTNQVEQPATSVLSNIFISMGVAGF PR12

HPR0
or
HPRΔ

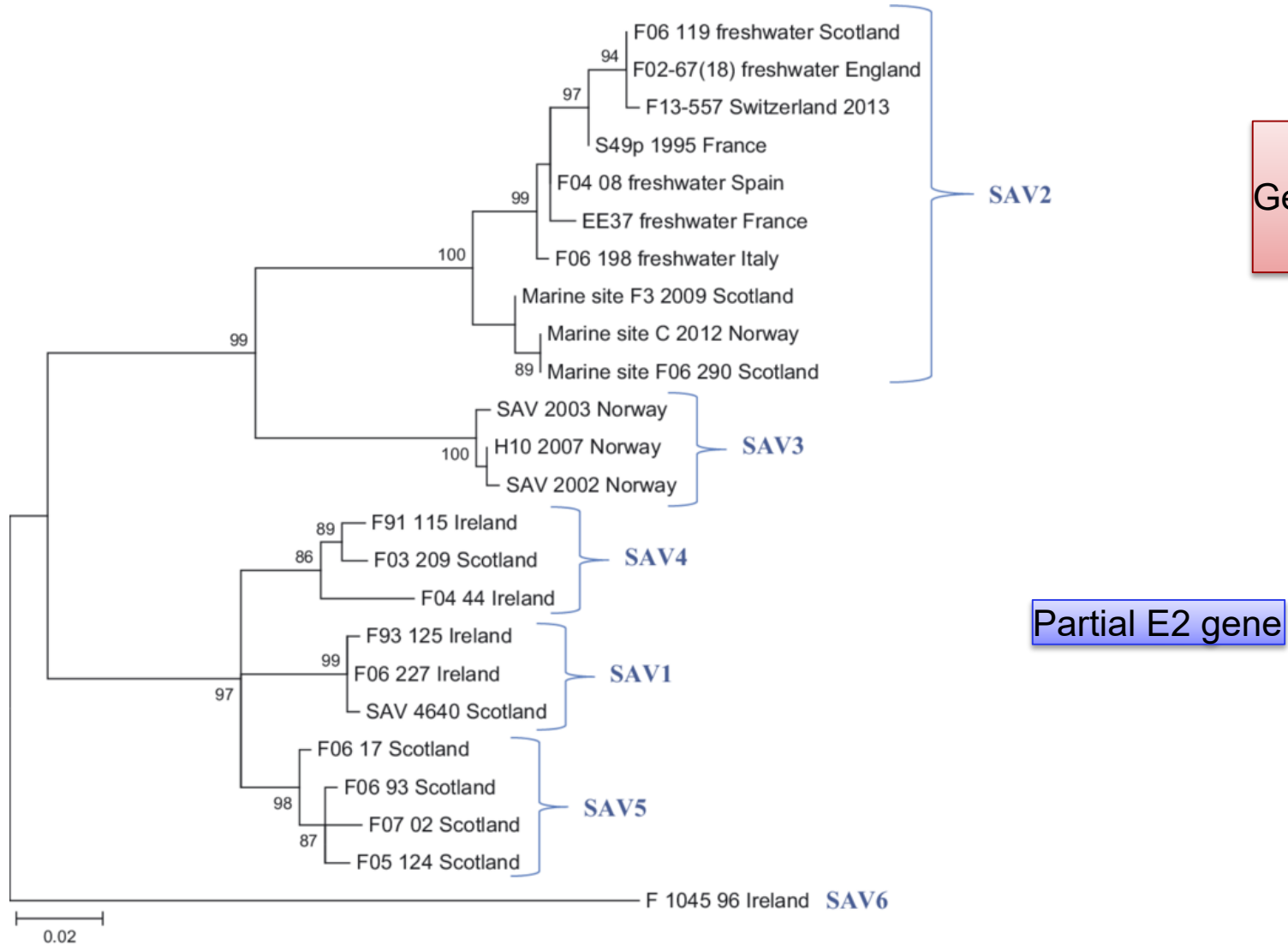
HPR = highly polymorphic region



Notes:

- It is not necessary to find the exact isolate to find out the genotype/genogroups
- Fishpathogen database has only been maintained for VHSV, and records are curated
- Sometimes genotype is written in genbank record – and usually is written in associated publications
- There is no simple recipe to find out genotypes/genogroups
- Possible to keep in house small databases with relevant isolates representing all genotypes/genogroups of a virus.

SAV



Genotypes SAV1 to SAV6

Partial E2 gene

Schmidt-Posthaus et al. 2014
doi: 10.3354/dao02766