

Supplementary Information

Novel G9 rotavirus strains co-circulate in children and pigs, Taiwan

Fang-Tzy Wu^{1*}, Krisztián Bányai^{2*}, Baoming Jiang³, Luke Tzu-Chi Liu¹, Szilvia Marton², Yhu-Chering Huang⁴, Li-Min Huang⁵, Liao Ming-Hui^{6#}, Chao A. Hsiung^{7#}

¹ Center for Diagnostics and Vaccine Development, Centers for Disease Control, Taipei, Taiwan

² Institute for Veterinary Medical Research, Centre for Agricultural Research, Hungarian Academy of Sciences, Budapest, Hungary

³ Centers for Disease Control and Prevention, Atlanta, Georgia, USA

⁴ Department of Pediatrics, Chang Gung Children's Hospital, Chang Gung Memorial Hospital, Chang Gung University, College of Medicine, Taoyuan, Taiwan

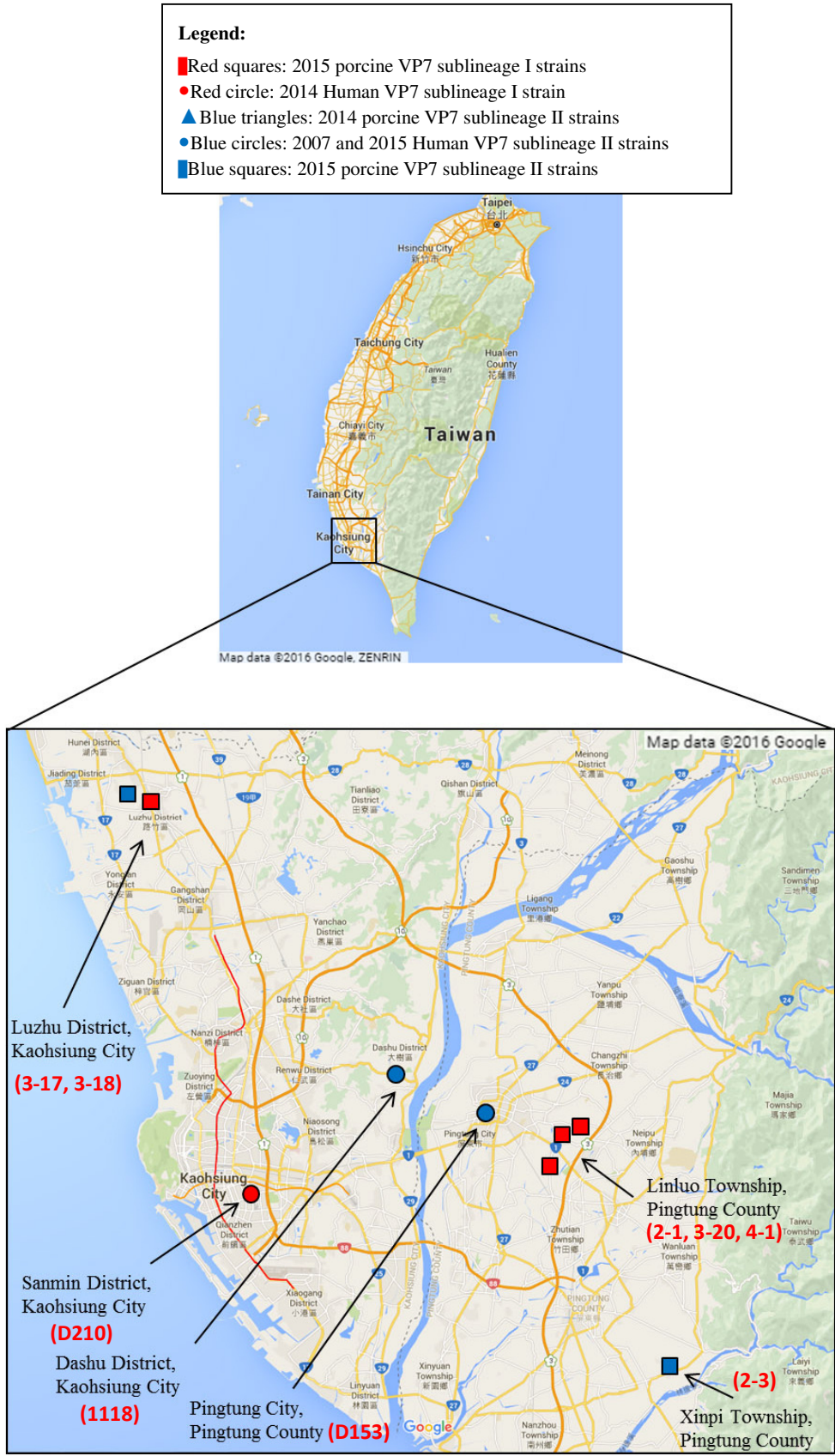
⁵ Department of Pediatrics, National Taiwan University Hospital, College of Medicine, National Taiwan University, Taipei, Taiwan

⁶ College of Veterinary Medicine, National Pingtung University of Science Technology, Taiwan

⁷ Institute of Population Health Sciences, National Health Research Institutes, Zhunan, Taiwan

* # These authors have equally contributed to this work.

Supplementary Figure S1. Close-up map of part of Kaohsiung city and Pingtung county. Strain names (in red) are marked in parentheses adjacent to names of district/township from which G9 positive pig farms and residences of patients are located. Map modified from Google Maps: Map data ©2015 Google, ZENRIN, with Adobe Photoshop CS 8.0 (www.photoshop.com)

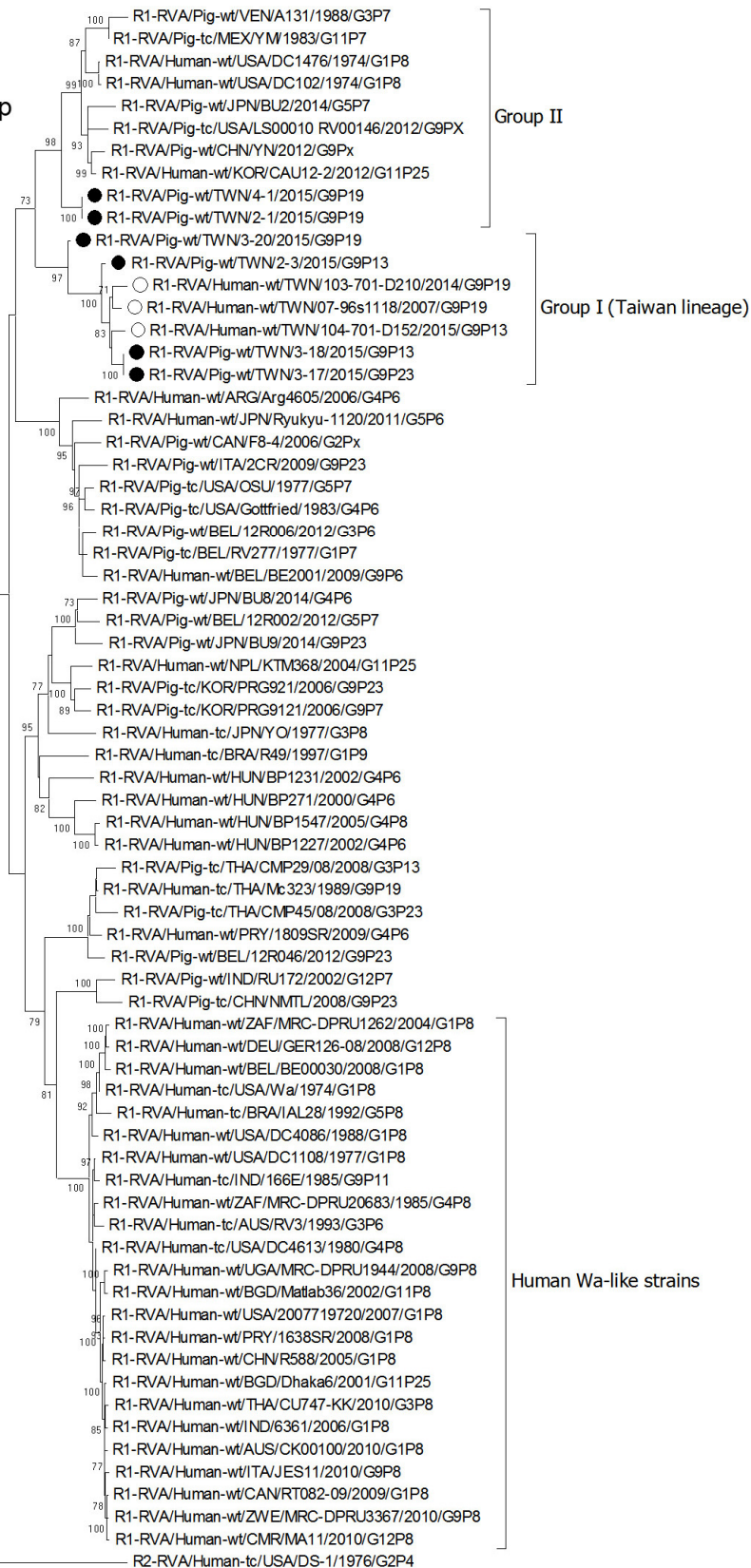


Supplementary Figure S2. Extended phylogenetic trees of the simplified trees in Figure 4 (inserted main text). (A) VP1, (B) VP2, (C) VP3, (D) VP6, (E) NSP1, (F) NSP2, (G) NSP3, (H) NSP4, and (I) NSP5.

Refer to Fig. 4 for explanation of methods used and legends. MEGA6 was used to generate neighbor-joining (maximum composite likelihood) or maximum likelihood trees with 1,000 bootstrap replicates using the best-fit substitution models determined by the built-in model test application:

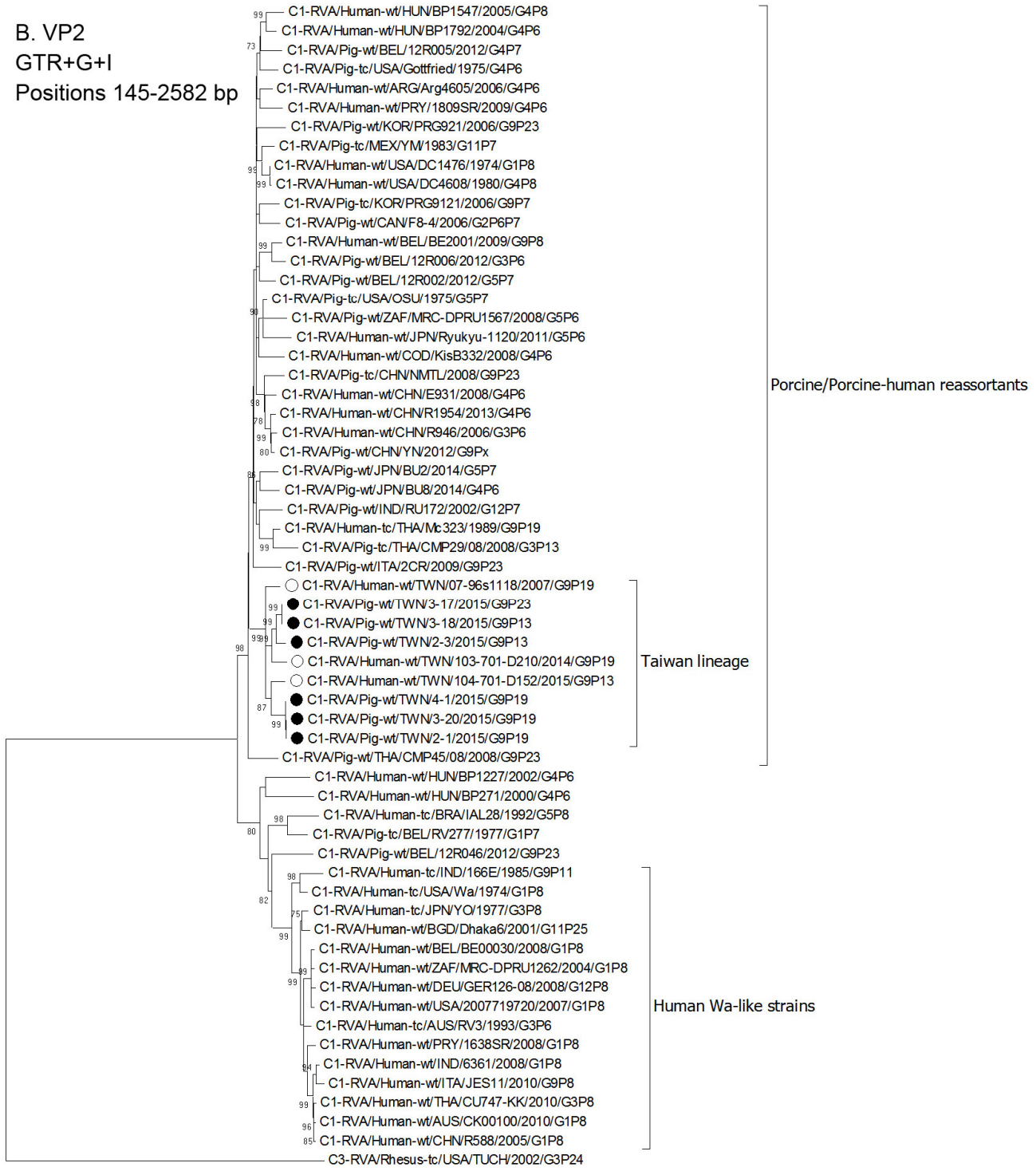
GTR=General Time Reversible, T92=Tamura 92, HKY=Hasegawa-Kishino-Yano 85, TN93=Tamura-Nei 93, G=gamme sites, I=invariant sites. The region within each of the gene segment used for each analysis is also shown. Open circles indicate Taiwanese human strains, closed circles denote Taiwanese porcine strains from 2015, open diamonds denote Taiwanese porcine strains from 2014.

A. VP1
GTR+G+I
Positions 19-3270 bp

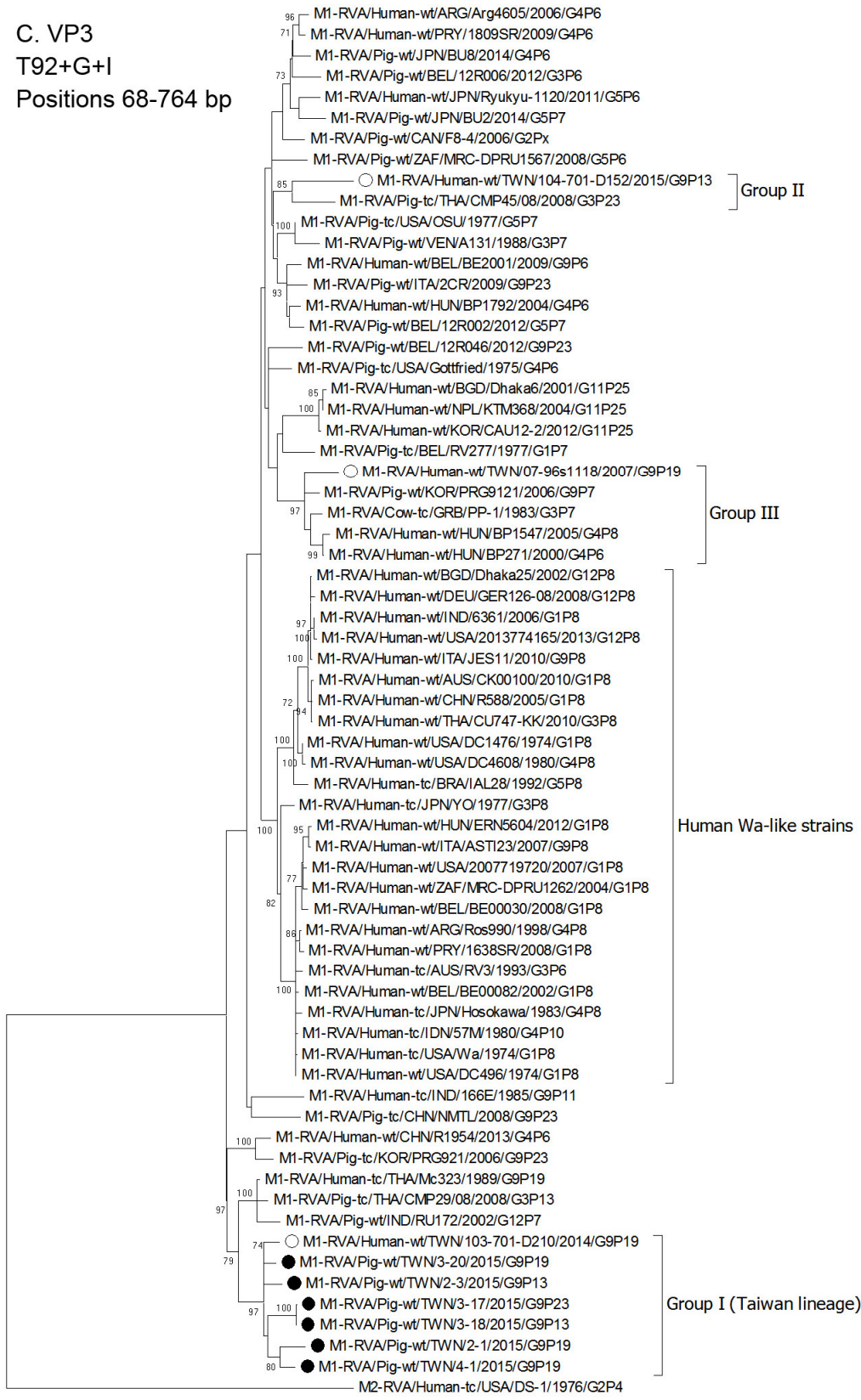


0.1

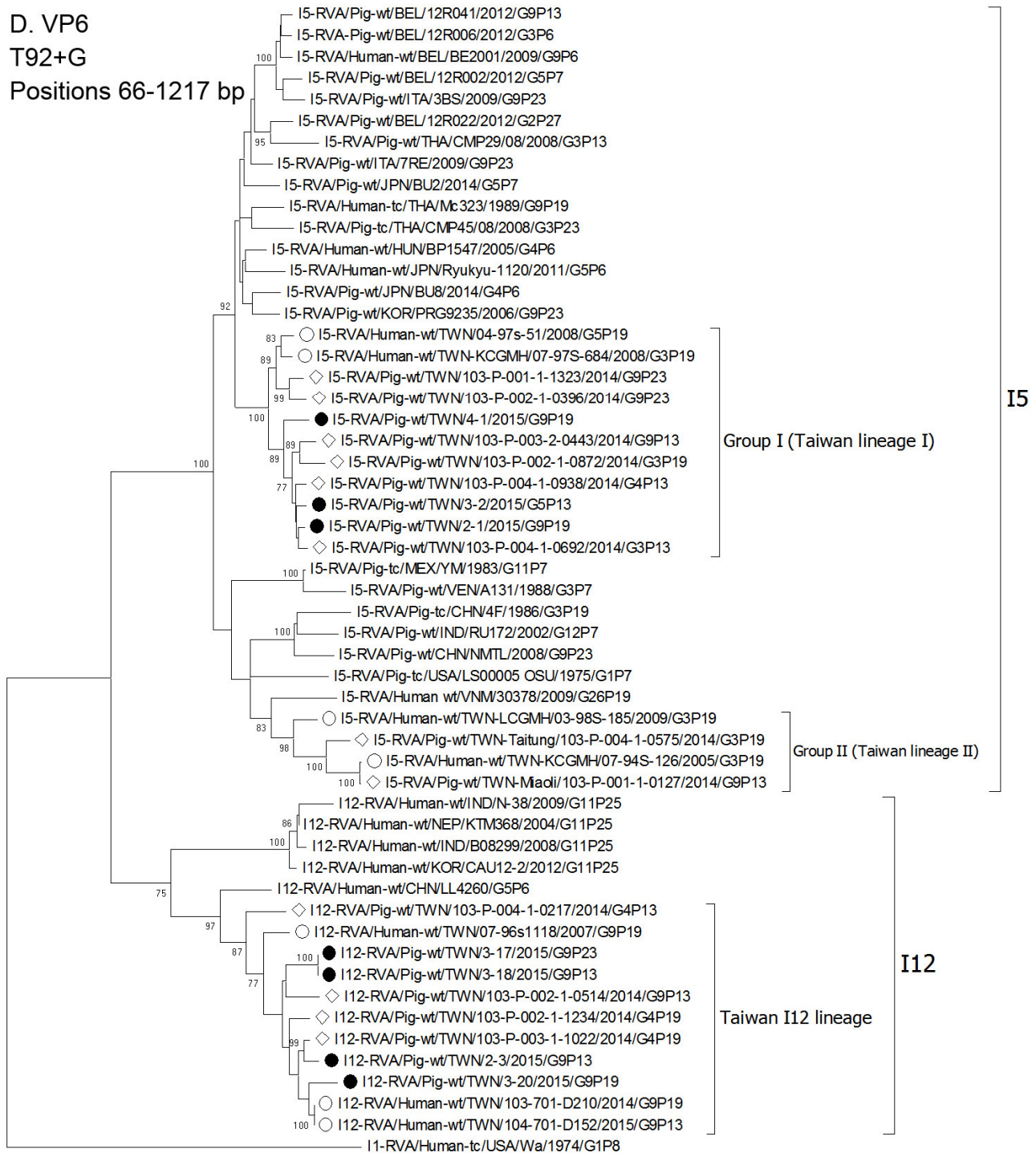
B. VP2
GTR+G+I
Positions 145-2582 bp



C. VP3
T92+G+I
Positions 68-764 bp

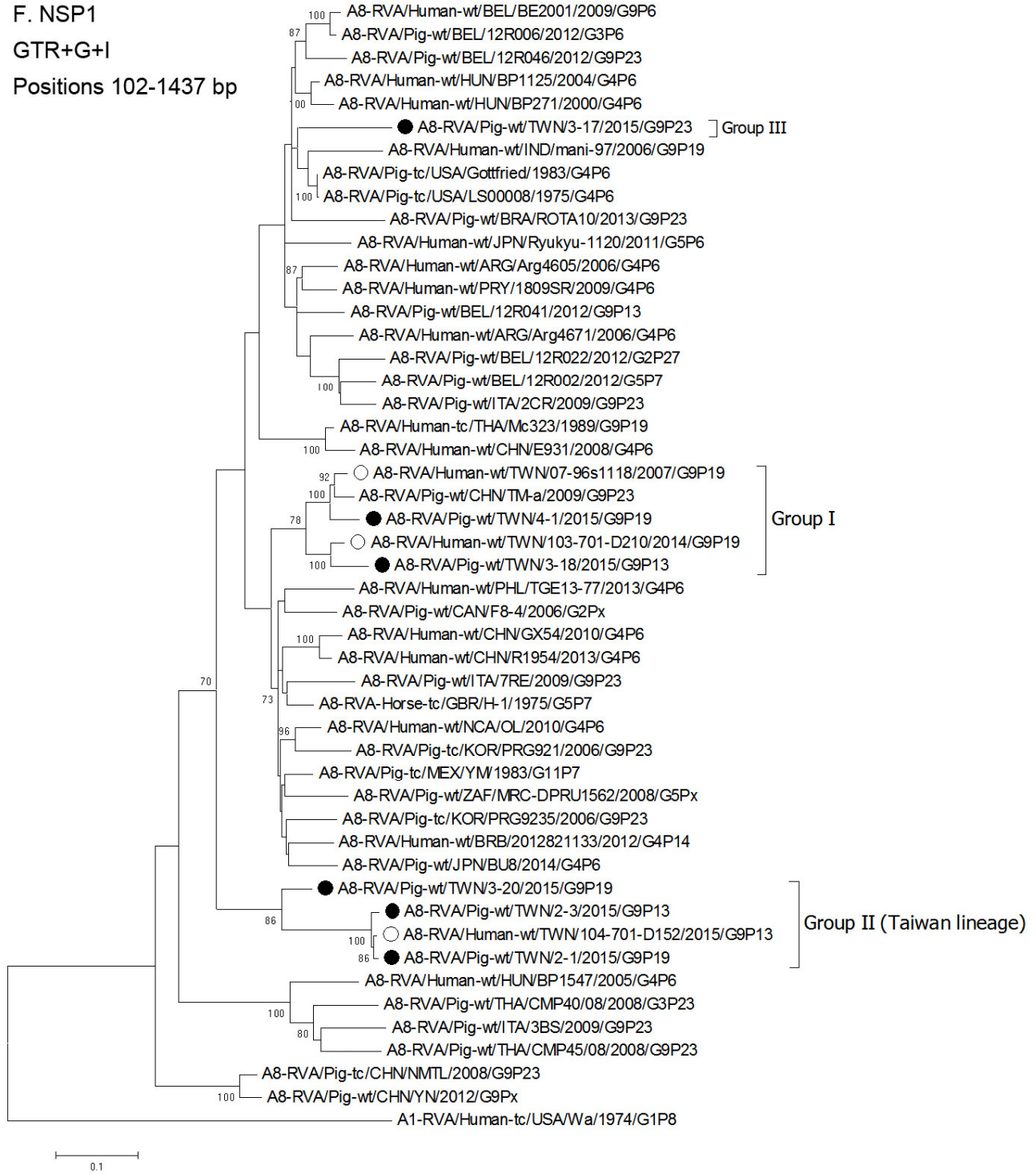


D. VP6
T92+G
Positions 66-1217 bp



0.05

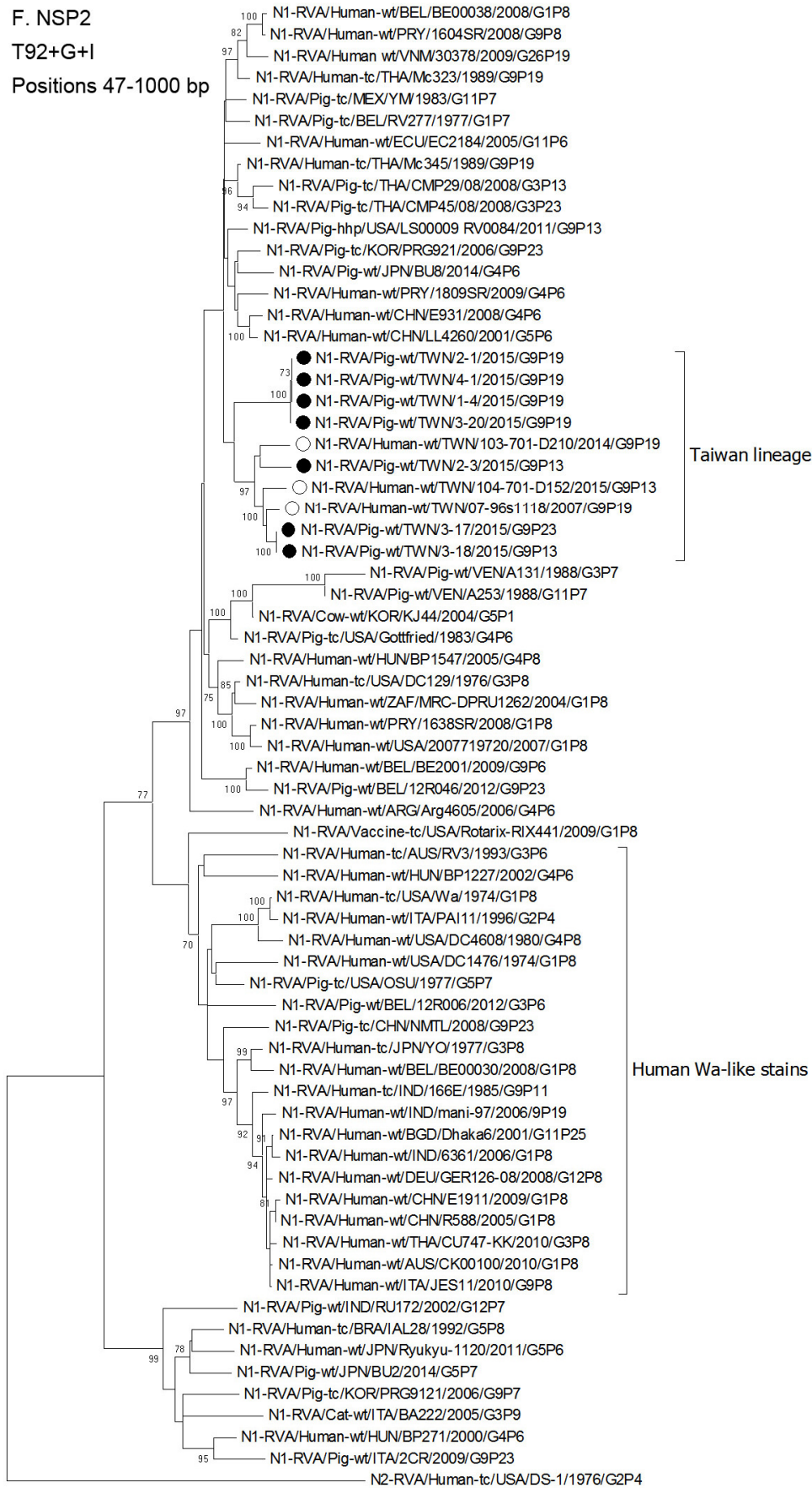
F. NSP1
 GTR+G+I
 Positions 102-1437 bp



F. NSP2

T92+G+I

Positions 47-1000 bp

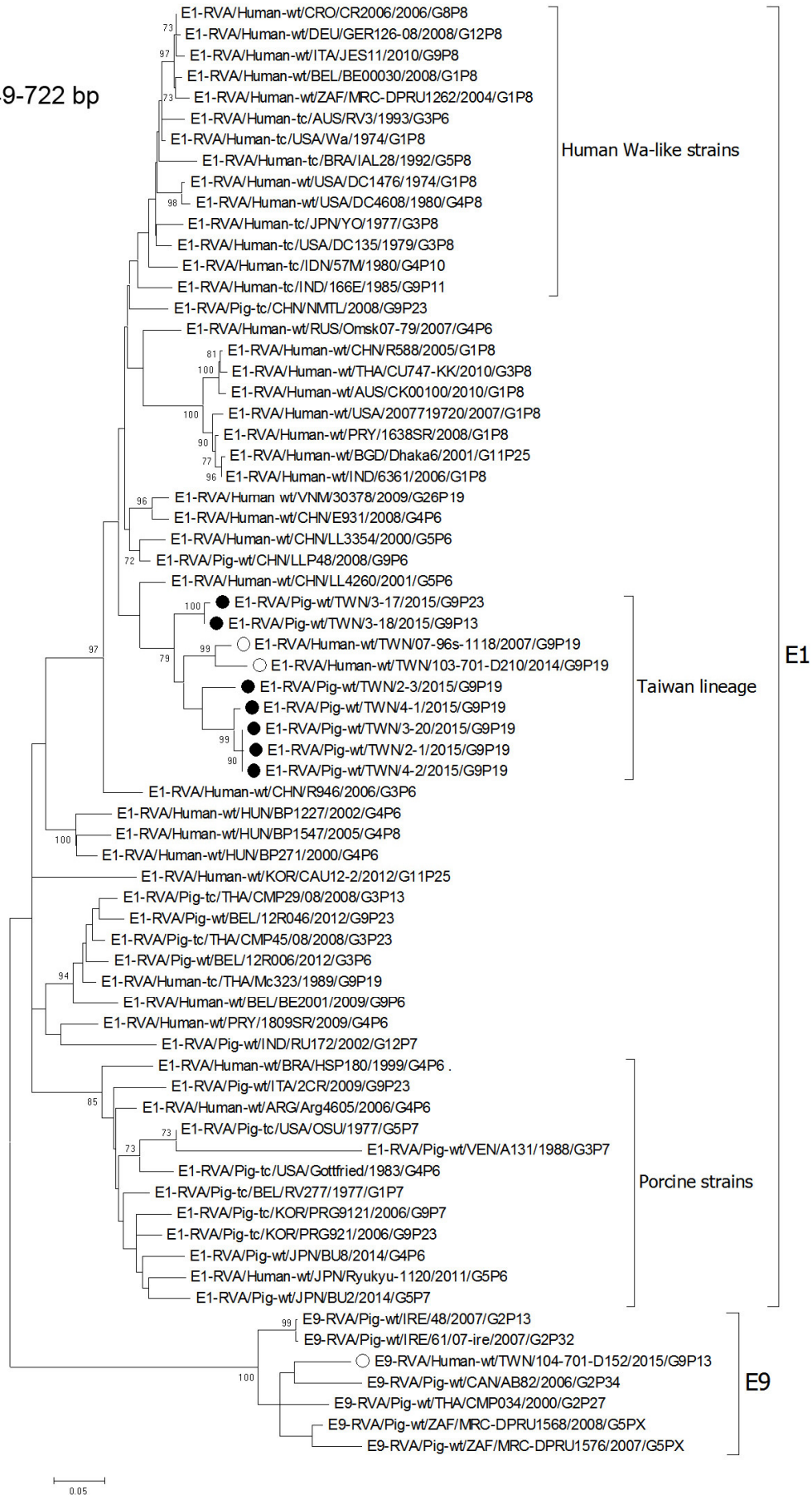


G. NSP3
T92+G
Positions 26-977 bp

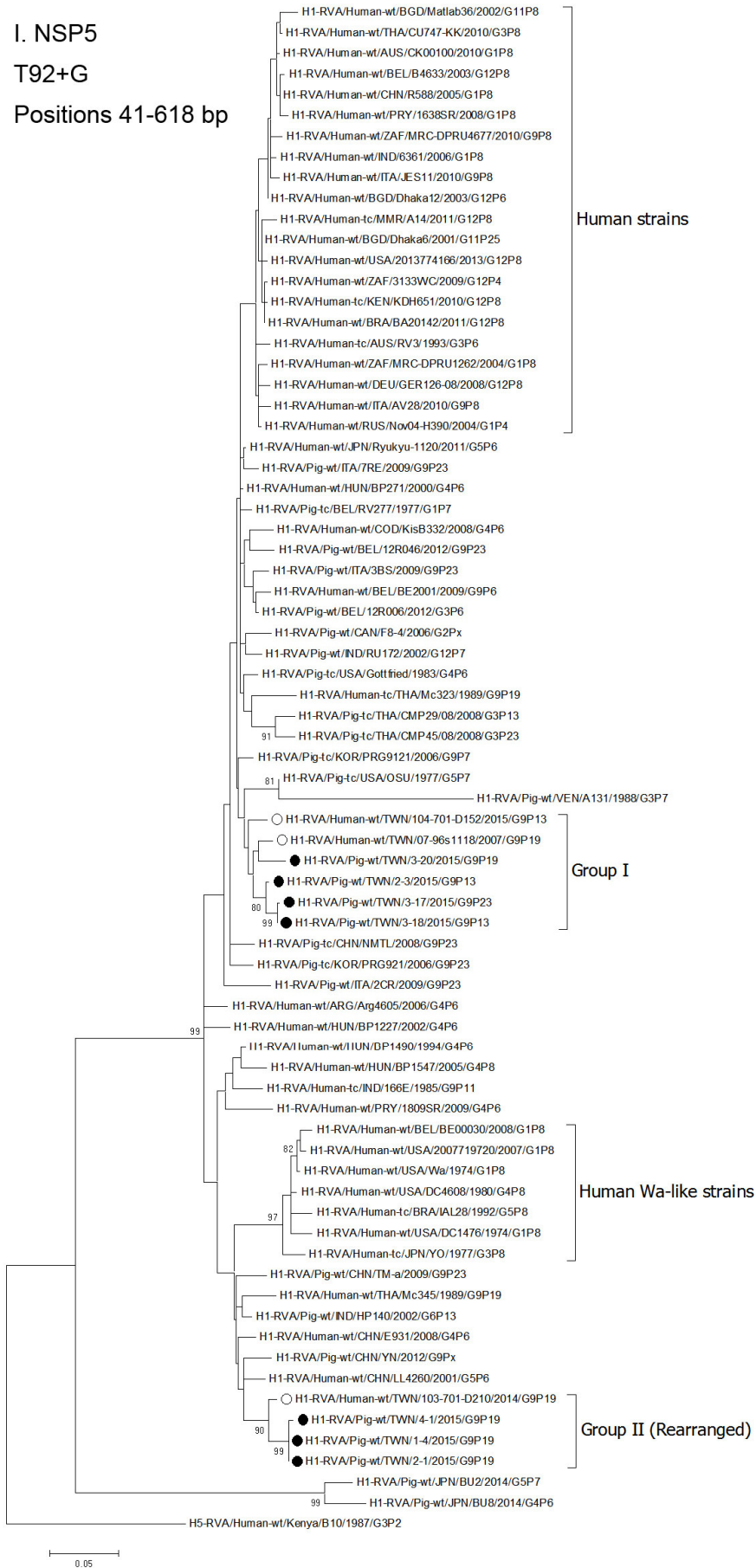


0.1

H. NSP4
T92+G
Positions 49-722 bp



I. NSP5
T92+G
Positions 41-618 bp



Supplementary Table S1 List of primers used in RT-PCR and sequencing

Target	Name	Sequence	Position (ref accession)	Reference
VP1	GEN_VP1Fb	GGC TAT TAA AGC TRT ACA ATG GGG AAG	1-27(DQ490539)	4
	VP1-771f	AGT YGC ATT AAT WGA YAT TAA YGG TAC	771-797(DQ490539)	5
	VP1-955r	CAA CTA ACC AYT CTT GAA TCA TTC	955-932(DQ490539)	5
	VP1-1375f	GAT GTT CCT GGA CGA AGA AC	1375-1394(DQ490539)	5
	VP1-1619R	CCC ATW ATT ATT CCT TTY CTR AA	1641-1619(DQ490539)	This study
	VP1-1650r	CAT ATC TAA ACC CAT AAT TAT TCC	1650-1627(DQ490539)	5
	VP1-2216f	TAA CTA AAA TTA TAC ARA TGA CAT CTG	2216-2242(DQ490539)	5
	VP1-2294r	GTW GTT ACW CGT TCT GAY GG	2294-2272(DQ490539)	5
	GEN_VP1Rb	GGT CAC ATC TAA GCG YTC TAA TCT TG	3302-3277(DQ490539)	4
VP2	GEN_VP2Fc	GGC TAT TAA AGG YTC AAT GGC GTA CAG	1-27 (X14942)	4
	VP2-m719f	GAT ATG AGA CAA CAA GTT CAR G	719-740 (X14942)	5
	VP2-852r	TTA TTC AAT GGT TCA ACT ART TG	880-858 (X14942)	5
	VP2-1389f	GAA TGC AAA GAA TGC ATT ATA G	1368-1389 (X14942)	5
	VP2-1443r	AAA TTT TGT ATT TGC TGT TCT GCT A	1443-1419 (X14942)	5
	VP2-2114f	GAG CAG ATT GAA CGA GCT TCA	2114-2134 (X14942)	5
	VP2-2202r	CCA TAC ATC TCA TCT CTT TC	2202-2183 (X14942)	5
	GEN_VP2Rbc	GTC ATA TCT CCA CAR TGG GGT TGG	2716-2693 (X14942)	4
VP3	GEN_VP3Fe	GGC TWT TAA AGC ART ATT AGT AGT G	1-25 (AY267335)	4
	VP3-m758f	CAT ACT ATY AAA TTG AAK CAR GA	758-780 (AY267335)	5
	VP3-m913r	AGC YGA ACC AAG CAT RTA TAY KA	913-891 (AY267335)	5
	VP3-m1550f	AAT TTY AAR AAT ATT TAY GAY TGG AC	1550-1575 (AY267335)	5
	VP3-m1704r	TTT ATT ATR AAT AAA TGR TTR TTT CC	1704-1679 (AY267335)	5
	GEN_VP3Rc	GGT CAC ATC ATG ACT AGT GTG	2591-2570 (AY267335)	4
VP4	VP4 F primer	GGC TAT AAA ATG GCT TCG CT	1-20 (JX406750)	6
	VP4-5a	AAT GAT TAT CAG ACT CCA AT	1438-1457 (JX406750)	7
	Con3	TGG CTT CGC CAT TTT ATA GAC A	11-32 (JX406750)	8
	VP4F	TAT GCT CCA GTN AAT TGG	142-159 (JX406750)	15
	793F	TGG AAA GAA ATG CAR TAY AA	799-818 (JX406750)	9
	Con2	ATT TCG GAC CAT TTA TAA CC	887-868 (JX406750)	8
	VP4R	ATT GCA TTT CTT TCC ATA ATG	805-785 (JX406750)	15
	1583R	CCH GAR AAC ATN GAR AAC ATA TC	1604-1582 (JX406750)	9
	VP4 R primer	GGG GGT CAC ATC CTC	2359-2348 (+3) (JX406750)	6
NSP1	GEN_NSP1F	GGC TTT TTT TTA TGA AAA GTC TTG	1-24 (JX406751)	4
	GEN_NSP1R	GGT CAC ATT TTA TGC TGC C	1531-1547 (HQ650120)	4
NSP2	GEN_NSP2F	GGC TTT TAA AGC GTC TCA G	1-19 (JX406754)	4
	GEN_NSP2R	GGT CAC ATA AGC GCT TTC	1059-1042 (JX406754)	4
NSP3	NSP3 F primer	GGC TTT TAA TGC TTT TCA GTG GTT G	1-25 (JX406753)	6

	NSP3 R primer	GGT CAC ATA ACG CCC CTA TAG	1074-1054 (JX406753)	6
NSP4	GEN_NSP4F	GGC TTT TAA AAG TTC TGT TCC	1-21 (JX406756)	4
	JRG30	GGC TTT TAA AAG TTC TGT	1-18 (JX406756)	10
	JRG31	ACC ATT CCT TCC ATT AAG	738-721 (JX406756)	10
	GEN_NSP4R	GGW YAC RYT AAG ACC RTT CC	750-731 (JX406756)	4
NSP5	GEN_NSP5F	GGC TTT TAA AGC GCT ACA G	1-19 (JX406757)	4
	GEN_NSP5R	GGT CAC AAA ACG GGA GT	664-648 (JX406757)	4
VP7	Beg9	GGC TTT AAA AGA GAG AAT TTC CGT CTG G	1-28 (JX406755)	11
	VP7-39F	GCT CYT TTT RAT GTA TGG TAT TGA ATA TAC CAC	39-71 (JX406755)	9
	9con1	TAG CTC CTT TTA ATG TAT GG	37-56 (JX406755)	12
	VP7-406R	CTT TAA AAT ANA DGA DCC WRT YGG CCA	406-379 (JX406755)	9
	9con2	GTA TAA AAT ACT TGC CAC CA	941-922 (JX406755)	12
	End9	GGT CAC ATC ATA CAA TTC TAA TCT AAG	1062-1036 (JX406755)	11
VP6	GEN_VP6F	GGC TTT WAA ACG AAG TCT TC	1-20 (JX406752)	4
	JRG7	GGC TTT AAA ACG AAG TCT TC	1-20 (JX406752)	14
	VP6-F	GAC GGV GCR ACT ACA TGG	747-764 (JX406752)	13
	VP6-R	GTC CAA TTC ATN CCT GGT GG	1126-1107 (JX406752)	13
	JRG8	GGT CAC ATC CTC TCA CTA CAT	1356-1336 (JX406752)	14
	GEN_VP6R	GGT CAC ATC CTC TCA CT	1356-1340 (JX406752)	4

Supplementary Table S2 Percentage identity of each gene segment with reference and closest BLAST hit strains in GenBank

		Human D152 G9P13				Human D210 G9P19				Human 1118 G9P19			
	Cutoff	Genotype	Reference strain %	GenBank strain %	Genotype	Reference strain %	GenBank strain %	Genotype	Reference strain %	GenBank strain %			
VP1	83	R1	86% (Wa)	87% (H-1)	R1	86% (Wa)	88% (BU8)	R1	86% (Wa)	88% (BU8)			
VP2	84	C1	89% (Wa)	93% (E931)	C1	89% (Wa)	92% (E931)	C1	88% (Wa)	93% (E931)			
VP3	81	M1	87% (Wa)	90% (YM1)	M1	86% (Wa)	90% (Mc345)	M1	89% (Wa)	92% (PRG9121)			
VP4	80	P13	87% (HP140)	95% (CMP213)	P19	87% (NIV929893)	87% (NIV929893)	P19	88% (NIV929893)	88% (NIV929893)			
VP6	85	I12	86% (KTM368)	92% (LL4260)	I12	86% (KTM368)	92% (LL4260)	I12	88% (KTM368)	92% (LL4260)			
VP7	80	G9	90% (W161)	92% (G2275)	G9	91% (W161)	93% (OM46)	G9	89% (W161)	92% (OM46)			
NSP1	79	A8	83% (Gottfried)	85% (YN)	A8	86% (Gottfried)	92% (TM-a)	A8	86% (Gottfried)	96% (TM-a)			
NSP2	85	N1	89% (Wa)	94% (Mc345)	N1	88% (Wa)	95% (Mc345)	N1	89% (Wa)	96% (Mc345)			
NSP3	85	T1	90% (Wa)	94% (E931)	T1	90% (Wa)	94% (E931)	T1	90% (Wa)	94% (E931)			
NSP4	85	E9	94% (CMP034)	94% (CMP034)	E1	92% (Wa)	94% (Omsk07-79)	E1	93% (Wa)	94% (LL4260)			
NSP5	91	H1	94% (Wa)	98% (Ryukyu-1120)	H1	96% (Wa)	97% (TM-a)	H1	94% (Wa)	98% (PRG9121)			
		Porcine 2-3 G9P13				Porcine 3-18 G9P13				Porcine 2-1 G9P19			
	Cutoff	Genotype	Reference strain %	GenBank strain %	Genotype	Reference strain %	GenBank strain %	Genotype	Reference strain %	GenBank strain %			
VP1	83	R1	87% (Wa)	89% (BP1231)	R1	86% (Wa)	89% (BP1231)	R1	87% (Wa)	92% (CAU12-2)			
VP2	84	C1	89% (Wa)	92% (OSU)	C1	89% (Wa)	92% (E931)	C1	88% (Wa)	92% (E931)			
VP3	81	M1	87% (Wa)	90% (Mc345)	M1	87% (Wa)	89% (Mc345)	M1	87% (Wa)	91% (Mc345)			
VP4	80	P13	87% (HP140)	94% (CMP178)	P13	86% (HP140)	93% (12R041)	P19	87% (NIV929893)	87% (Mc345)			
VP6	85	I12	86% (KTM368)	91% (LL4260)	I12	87% (KTM368)	92% (LL4260)	I5	89% (OSU)	93% (BU8)			
VP7	80	G9	90% (W161)	93% (OM46)	G9	91% (W161)	93% (OM46)	G9	91% (W161)	92% (OM46)			
NSP1	79	A8	84% (Gottfried)	85% (ROTA05)	A8	85% (Gottfried)	91% (TM-a)	A8	84% (Gottfried)	85% (ROTA05)			
NSP2	85	N1	89% (Wa)	95% (Mc345)	N1	89% (Wa)	96% (Mc345)	N1	89% (Wa)	94% (Mc345)			
NSP3	85	T1	89% (Wa)	94% (GX54)	T1	89% (Wa)	94% (E931)	T1	89% (Wa)	94% (E931)			
NSP4	85	E1	93% (Wa)	95% (LL3354)	E1	93% (Wa)	95% (LL4260)	E1	93% (Wa)	94% (E931)			
NSP5	91	H1	94% (Wa)	98% (Ryukyu-1120)	H1	93% (Wa)	98% (Ryukyu-1120)	H1	95% (Wa)	97% (LL4260)			
		Porcine 4-1 G9P19				Porcine 3-20 G9P19				Porcine 3-17 G9P23			
	Cutoff	Genotype	Reference strain %	GenBank strain %	Genotype	Reference strain %	GenBank strain %	Genotype	Reference strain %	GenBank strain %			
VP1	83	R1	87% (Wa)	92% (CAU12-2)	R1	87% (Wa)	89% (DC1476)	R1	86% (Wa)	89% (BP1231)			
VP2	84	C1	88% (Wa)	92% (E931)	C1	88% (Wa)	92% (E931)	C1	89% (Wa)	92% (E931)			
VP3	81	M1	87% (Wa)	91% (Mc323)	M1	87% (Wa)	91% (Mc323)	M1	86% (Wa)	89% (Mc323)			
VP4	80	P19	87% (NIV929893)	87% (Mc345)	P19	87% (NIV929893)	87% (Mc345)	P23	88% (A34)	90% (GUB71)			
VP6	85	I5	90% (OSU)	95% (BU8)	I12	86% (KTM368)	91% (LL4260)	I12	87% (KTM368)	92% (LL4260)			
VP7	80	G9	91% (W161)	93% (OM46)	G9	90% (W161)	92% (OM46)	G9	90% (W161)	92% (OM46)			
NSP1	79	A8	86% (Gottfried)	94% (TM-a)	A8	84% (Gottfried)	89% (TM-a)	A8	90% (Gottfried)	90% (Gottfried)			
NSP2	85	N1	89% (Wa)	94% (Mc345)	N1	89% (Wa)	94% (Mc345)	N1	90% (Wa)	96% (Mc345)			
NSP3	85	T1	89% (Wa)	93% (GX54)	T1	89% (Wa)	93% (GX54)	T1	90% (Wa)	95% (E931)			
NSP4	85	E1	92% (Wa)	94% (LLP48)	E1	92% (Wa)	94% (LL3354)	E1	93% (Wa)	94% (LLP48)			
NSP5	91	H1	95% (Wa)	96% (LL4260)	H1	93% (Wa)	98% (Ryukyu-1120)	H1	93% (Wa)	98% (Ryukyu-1120)			

Technical Appendix References

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