

## **Supporting Online Material**

### **Antigenic and Genetic Characteristics of the Early Isolates of Swine-Origin 2009 A(H1N1) Influenza Viruses Circulating in Humans**

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**Table S1.** GenBank accession numbers of nucleotide sequences of 2009 A(H1N1) viruses used in genetic analyses.

Strain designation	Passage Level	GENBANK PB2	GENBANK PB1	GENBANK PA	GENBANK HA	GENBANK NP	GENBANK NA	GENBANK M	GENBANK NS
A/ARIZONA/01/2009	MX/C1				GQ117067	GQ117063	GQ117064	GQ117066	GQ117065
A/ARIZONA/02/2009	C1	GQ117076	GQ117075		GQ117079	GQ117074	GQ117077	GQ117078	
A/CALIFORNIA/04/2009	C2	FJ969516		FJ969515	GQ117044	FJ969512	FJ969517	FJ969513	FJ969514
A/CALIFORNIA/04/2009	CX	FJ969079	FJ966080	FJ969081	FJ966082	FJ966083	FJ966084	FJ966085	FJ966086
A/CALIFORNIA/05/2009	C1	FJ969595	FJ969598	FJ969597	FJ966952	FJ966953	FJ966956	FJ966954	
A/CALIFORNIA/06/2009	M1	FJ969663	FJ969665	FJ969664	FJ966960	FJ966961	FJ9671075	FJ966962	FJ9671074
A/CALIFORNIA/06/2009	M1/C1								
A/CALIFORNIA/07/2009	C1	FJ969530	FJ969531	FJ969529	FJ981613			FJ969527	FJ969528
A/CALIFORNIA/07/2009	E2	FJ984387		FJ969539	FJ969540	FJ969536	FJ984386	FJ969537	FJ969538
A/CALIFORNIA/07/2009	M1	FJ969678	FJ969678	FJ969677	FJ966974			FJ966975	
A/CALIFORNIA/07/2009	M1/C1								
A/CALIFORNIA/08/2009	C1							FJ969518	FJ969519
A/CALIFORNIA/08/2009	E2	FJ984365	FJ984367	FJ984368	FJ971076	FJ984366		FJ969532	FJ969533
A/CALIFORNIA/09/2009	Original				FJ966971		FJ966973	FJ966972	FJ969538
A/CALIFORNIA/10/2009	Original				FJ969511			FJ969510	
A/CALIFORNIA/11/2009	C1				GQ160579				
A/CALIFORNIA/12/2009	C1				GQ160506				
A/CALIFORNIA/13/2009	C1				GQ160607				
A/CALIFORNIA/14/2009	C1	GQ117035	GQ117034	GQ117037	GQ117040	GQ117033	GQ117036	GQ117039	GQ117038
A/COLORADO/03/2009	C1		GQ168884		GQ117119	GQ117117	GQ117118		
A/INDIANA/09/2009	C1	GQ168870	GQ117093	GQ117095	GQ117097	GQ117092	GQ117094	GQ117096	GQ168871
A/MASSACHUSETTS/06/2009	C1		GQ168847		GQ117043	GQ117041	GQ117042	GQ168849	GQ168848
A/MASSACHUSETTS/07/2009	C1		GQ168873		GQ117103	GQ117101	GQ117102	GQ168874	GQ168874
A/MEXICO/395/2009	E2				GQ162194		GQ162193		
A/MEXICO/395/2009	Original								
A/MEXICO/108/2009	C1/C1	GQ162180		GQ149653	GQ149662	GQ149660	GQ149659	GQ149657	GQ149658
A/MEXICO/108/2009	C1			GQ149686	GQ149689	GQ149685	GQ149688	GQ149690	GQ149687
A/MEXICO/108/2009	Original				GQ149651		GQ149650	GQ149649	
A/MEXICO/108/2009	C1-IR		GQ149652	GQ149661	GQ149654	GQ149655	GQ149656	GQ162179	
A/MEXICO/108/2009	E1				GQ149668	GQ149667		GQ149666	
A/MEXICO/108/2009	E2				GQ162170		GQ162169		
A/MEXICO/115/2009	C1	GQ162199			GQ149692	GQ149693		GQ149691	GQ149694
A/MEXICO/415/2009	Original								
A/MEXICO/417/2009	C1/C1				GQ162182		GQ162181		
A/MEXICO/4269/2009	C1/C1				GQ162172		GQ162171		
A/MEXICO/4482/2009	Original				GQ149671		GQ149670		
A/MEXICO/4482/2009	C1-IR		GQ149675	GQ149676	GQ149677	GQ149678		GQ162192	
A/MEXICO/4482/2009	E2				GQ162190		GQ162189		
A/MEXICO/4482/2009	C1				GQ149674	GQ149673	GQ149672		
A/MEXICO/4486/2009	C1/C1	GQ162177	GQ162176	GQ149646	GQ149647	GQ149645	GQ149644	GQ149642	GQ149643
A/MEXICO/4486/2009	C1	GQ149617	GQ149618	GQ149620	GQ149623	GQ149619	GQ149622	GQ149624	GQ149621
A/MEXICO/4486/2009	Original				GQ149665		GQ149664	GQ149663	
A/MEXICO/4486/2009	C1-IR		GQ162186		GQ162185	GQ162184	GQ162187	GQ162188	
A/MEXICO/4486/2009	E2				GQ162202		GQ162201		
A/MEXICO/4502/2009	C1/C1				GQ162195				
A/MEXICO/4516/2009	C1					GQ149648		GQ162178	
A/MEXICO/4575/2009	C1/C1				GQ162197		GQ162196		
A/MEXICO/4593/2009	C1/C1				GQ162174		GQ162173		
A/MEXICO/4595/2009	C1/C1				GQ162191				
A/MEXICO/4603/2009	C1/C1	GQ162198	GQ149683		GQ149684	GQ149682	GQ149681	GQ149679	GQ149680
A/MEXICO/4603/2009	C1	GQ162168	GQ162167	GQ149627	GQ149630	GQ149628	GQ149629	GQ149626	GQ149625
A/MEXICO/4603/2009	C1/E2				GQ149641	GQ149639	GQ149640	GQ162175	
A/MEXICO/4604/2009	C1	GQ149632	GQ149633	GQ149636	GQ149634	GQ149637	GQ149631	GQ149638	GQ149635
A/MEXICO/4627/2009	C1/C1				GQ162183				
A/MEXICO/4635/2009	C1/C1				GQ162200				
A/MEXICO/4646/2009	C1/C1				GQ162204			GQ162203	
A/MICHIGAN/02/2009	C1		GQ168880	GQ117109	GQ117112	GQ117107	GQ117108	GQ117111	GQ117110
A/MINNESOTA/10/2009	C1	GQ117070	GQ117069		GQ117068	GQ117071	GQ117072	GQ117073	GQ117072
A/NEBRASKA/02/2009	C1		GQ168875		GQ168875	GQ117104	GQ117105		GQ117106
A/NEBRASKA/03/2009	E2	GQ160597	GQ160596		GQ160599	GQ160595			GQ160598
A/NEW YORK/04/2009	C1				GQ160568				
A/NEW YORK/05/2009	E2				GQ160542				
A/NEW YORK/06/2009	C1		GQ168850		GQ168851	FJ984341	FJ984340	FJ984338	FJ984339
A/NEW YORK/08/2009	E2				GQ168850		GQ168848	GQ168849	
A/NEW YORK/09/2009	E2				GQ168855		GQ168854		
A/NEW YORK/09/2009	C1		GQ117120		GQ168856				
A/NEW YORK/10/2009	C1	GQ168876	FJ984373	FJ984374	FJ984375	FJ984372	FJ984371	FJ984369	FJ984370
A/NEW YORK/11/2009	C1		GQ168852	FJ984346	FJ984347	FJ984345	FJ984344	FJ984342	FJ984343
A/NEW YORK/11/2009	E2				GQ168652		GQ168651		
A/NEW YORK/12/2009	C1		GQ168844		FJ984337	FJ984336	FJ984335	GQ168845	FJ984334
A/NEW YORK/12/2009	E2				GQ168673		GQ168672		
A/NEW YORK/13/2009	C1		GQ168881	GQ117115	GQ117116	GQ117113	GQ117114	GQ168883	GQ168882
A/NEW YORK/13/2009	C1				GQ168668		GQ168666	GQ168667	
A/NEW YORK/14/2009	E2				GQ168657		GQ168656		
A/NEW YORK/15/2009	C1	GQ168878	GQ168877	FJ984380		FJ984379	FJ984378	FJ984376	FJ984377
A/NEW YORK/18/2009	C1	FJ984351	FJ984353	FJ984354	FJ984355	FJ984352	FJ984350	FJ984348	FJ984349
A/NEW YORK/18/2009	E2				GQ160556				
A/NEW YORK/19/2009	C1		FJ984392	FJ984393	FJ984394	FJ984391	FJ984390	FJ984388	FJ984389
A/NEW YORK/19/2009	Original				FJ969509				
A/NEW YORK/20/2009	C1				GQ117082	GQ117080	GQ117081	GQ168863	GQ168862
A/NEW YORK/20/2009	E2		GQ168867		GQ117086	GQ117083	GQ117084	GQ117085	GQ168868
A/NEW YORK/20/2009	Original				FJ969542		FJ969541		
A/NEW YORK/22/2009	C1	GQ117021	GQ117020		GQ117024	GQ117019	GQ117022	GQ117023	GQ168846
A/NEW YORK/23/2009	C1		GQ168864	GQ168865	FJ984364	FJ984363	FJ984362	GQ168866	FJ984361
A/NEW YORK/23/2009	E2				GQ168628		GQ168627		
A/NEW YORK/24/2009	C1				GQ160601			GQ160600	
A/NEW YORK/24/2009	E2						GQ168669		
A/NEW YORK/26/2009	E2				GQ168617				
A/NEW YORK/27/2009	E2				GQ168659			GQ168658	
A/NEW YORK/28/2009	E2				GQ168645				
A/NEW YORK/30/2009	C1				GQ160566				
A/NEW YORK/31/2009	C1			FJ984359	FJ984360	FJ984358	FJ984357	GQ168859	FJ984356
A/NEW YORK/31/2009	E2				GQ168661			GQ168660	
A/NEW YORK/34/2009	C1				GQ160567				
A/NEW YORK/35/2009	C1				GQ160543				
A/NEW YORK/37/2009	C1				GQ160527				
A/NEW YORK/39/2009	C1				GQ160602				
A/OHIO/07/2009	C1-NP	GQ160545	GQ160544	FJ984400	FJ984401	GQ117098	GQ117099	FJ984398	FJ984399
A/OHIO/07/2009	E2				GQ117100		GQ117098		GQ168872
A/OHIO/07/2009	C1		GQ168887		FJ984397	FJ984396		FJ984395	
A/OHIO/07/2009	Original				FJ969521		FJ969520		
A/OHIO/07/2009	Original-NP WASH				FJ969535		FJ969534		
A/SOUTH CAROLINA/09/2009	C1		GQ168854		GQ117056	GQ117052	GQ117053	GQ117055	GQ117054
A/TEXAS/04/2009	X/C1	GQ168885	FJ981616	FJ981619	FJ981615	FJ981618	FJ981614	FJ981617	FJ981620
A/TEXAS/04/2009	X	FJ969525	FJ969526	FJ969524	FJ981612	FJ969679	FJ969681	FJ969680	
A/TEXAS/04/2009	Original				FJ966982			FJ966983	
A/TEXAS/05/2009	X		FJ969522	FJ969670		FJ969667	FJ969669	FJ969668	FJ969666
A/TEXAS/05/2009	C1		GQ168860	FJ981610	GQ168861	FJ981609		FJ981608	FJ981611
A/TEXAS/05/2009	Original				FJ966959				
A/TEXAS/05/2009	X/C1								
A/TEXAS/06/2009	C1	GQ168879			FJ984385	FJ984384	FJ984383	FJ984381	FJ984382
A/TEXAS/07/2009	C1	GQ117089	GQ117088		GQ117091	GQ117087		GQ117090	GQ168869
A/TEXAS/08/2009	C1	GQ117047	GQ117046	GQ117049	GQ117051	GQ117045	GQ117048	GQ117050	GQ168853
A/TEXAS/08/2009	E2				GQ168633		GQ168632		
A/TEXAS/09/2009	C1	GQ117027	GQ117026	GQ117029	GQ117032	GQ117025	GQ117028	GQ117031	GQ117030
A/TEXAS/09/2009	E2				GQ168671		GQ168670		
A/TEXAS/15/2009	C1		GQ122094		G				

**Table S2.** Geographic isolation of five early genome variants of the 2009 A(H1N1) virus. Con. indicates consensus sequence.

Genome variants	Location	HA	NA	NP	NS1	PA
i	Mexico, Arizona, California, Colorado, Texas	Con.	Con.	Con.	Con.	Con.
ii	Mexico, Arizona, California, Indiana, Minnesota, Nebraska, Texas	Con.	Con.	T373I	Con.	M582L
iii	Mexico, Michigan	Con.	V106I, N247D	V100I	Con.	Con.
iv	California, Massachusetts, Nebraska, New York, Ohio	S206T	V106I, N247D	V100I	I123V	Con.
v	California	S91P, V323I	Con.	Con.	Con.	S224P

**Table S3.** Hemagglutination Inhibition (HI) assay (Kendal et al, 1982) of the 2009 A(H1N1) outbreak isolates A/California/4/2009 and A/California/5/2009; together with reference antigens and contemporary swine H1 antigens. Swine influenza viruses previously isolated from humans and sera raised to those viruses are shown in blue. Human seasonal influenza viruses and sera raised to those viruses are shown in green. 2009 A(H1N1) viruses are shown in red.

## HEMAGGLUTININ INHIBITION REACTIONS OF INFLUENZA SPECIMENS (04/23/2009)

STRAIN DESIGNATION	REFERENCE FERRET ANTISERA									DATE COLLECTED	PASSAGE
	WI/10	OH/2	IL/9/07	SW/IA	SW/MN	SW/IN	BR/59	BR/10			
A/WISCONSIN/10/98 <sup>a</sup>	2560	5120	1280	1280	1280	1280	5	5		31/12/98	C3/C3E2)
A/OHIO/2/2007 <sup>a</sup>	1280	5120	2560	2560	2560	2560	5	5		17/8/07	C1
A/ILLINOIS/9/2007 <sup>a</sup>	640	5120	5120	2560	512	5120	5	5		1/9/07	MX/C3
A/SWINE/IOWA/2006	320	5120	640	2560	1280	640	5	5		1/11/06	C2/C1
A/SWINE/MINNESOTA/2002	640	5120	640	1280	2560	2560	5	5		UNKNOWN	SIVCX/C3
A/SWINE/INDIANA/2000	320	5120	640	640	640	1280	5	5		UNKNOWN	SIVCX/C3
A/BRISBANE/59/2007 <sup>b</sup>	5	5	5	5	5	5	5	640	5	1/7/07	E2/E2
A/BRISBANE/10/2007 <sup>c</sup>	5	5	5	5	5	5	5	640	640	6/2/07	E2/E2)
<b>TEST ANTIGENS</b>											
A/CALIFORNIA/4/2009	640	5120	640	2560	1280	1280	5	5		1/4/09	C1(4/22/09)
A/CALIFORNIA/4/2009	640	5120	1280	320	5120	2560	5	5		1/4/09	C1/C1(4/22/09)
A/CALIFORNIA/5/2009	320	5120	640	1280	1280	1280	5	5		30/3/09	C1(4/22/09)

- a. Triple reassortant swine-like H1N1 virus  
b. Seasonal H1N1 reference strain  
c. Seasonal H3N2 reference strain

**Table S4.** HI assay table of 2009 A(H1N1) outbreak isolates from the USA and from Mexico. Swine influenza viruses previously isolated from humans and sera raised to those viruses are shown in blue. 2009 A(H1N1) viruses and sera raised to those viruses are shown in red.

## HEMAGGLUTINATION INHIBITION REACTIONS OF INFLUENZA H1N1 SWINE LIKE VIRUSES(05/0709)

STRAIN DESIGNATION	REFERENCE FERRET ANTISERA							DATE COLLECTED	PASSAGE	
	SW/IA/30	NJ/8/76	OH/2	WI/10	IL/9	CA/04	CA/05			
A/SWINE/IOWA/1930	320	40	5	5	5	5	5	20	UNK	XEXE6
A/NEWJERSEY/8/1976	80	160	5	10	5	5	5	40	UNK	SpfE6
A/OHIO/2/2007	80	1280	5120	640	5120	2560	5120	2560	08/18/11	C1
A/WISCONSIN/10/98	5	2560	5120	1280	2560	640	2560	640	01/01/03	C3/C3E2
A/ILLINOIS/9(33304)/2007	80	1280	5120	1280	5120	2560	5120	2560	09/02/11	C1
A/California/04/2009	10	320	5120	320	640	1280	2560	640	04/02/13	C2
A/California/05/2009	40	320	5120	320	1280	1280	5120	1280	03/31/13	C2)
A/Swine/Minnesota/2002	40	640	5120	640	2560	1280	5120	2560	UNK	SIVCX/C3
<b>TEST ANTIGENS</b>										
A/California/06/2009	80	640	5120	640	2560	1280	5120	1280	04/17/13	M1/C1
A/California/07/2009	320	1280	5120	1280	5120	2560	5120	2560	04/10/13	C1
A/California/07/2009	80	640	5120	320	1280	2560	5120	1280	04/10/13	E2
A/California/08/2009	160	1280	5120	640	2560	2560	5120	2560	04/10/13	C1
A/California/08/2009	160	320	5120	320	1280	2560	5120	1280	04/10/13	E2
A/Kansas/2/2009	160	640	5120	640	2560	2560	5120	1280	04/25/13	C1
A/Kansas/3/2009	40	320	2560	320	640	1280	5120	640	04/25/13	C1
A/Ohio/07/2009	80	640	2560	640	1280	1280	5120	640	04/25/13	E2
A/Ohio/07/2009	80	640	5120	320	1280	1280	5120	640	04/25/13	C1
A/New York/07/2009	40	320	2560	320	1280	640	2560	1280	UNK	E2
A/New York/7/2009	40	320	2560	320	640	640	2560	640	UNK	C1
A/New York/08/2009	80	320	2560	320	1280	1280	2560	640	04/26/13	E2
A/New York/8/2009	40	320	2560	160	640	640	2560	640	04/26/13	C1
A/New York/18/2009	160	1280	2560	640	2560	2560	5120	2560	UNK	E2
A/New York/20/2009	160	640	5120	640	2560	5120	5120	1280	UNK	E2
A/New York/23/2009	160	640	2560	320	1280	1280	5120	1280	04/25/13	C1
A/New York/23/2009	80	640	5120	640	1280	640	5120	1280	04/25/13	E2
A/New York/31/2009	40	320	2560	320	640	640	2560	640	04/25/13	C1
A/New York/31/2009	80	640	5120	640	1280	1280	5120	1280	04/25/13	E2
A/Texas/04/2009	20	160	1280	160	640	640	1280	640	04/15/13	X/C1
A/Texas/05/2009	320	1280	5120	640	2560	2560	5120	2560	04/16/13	X/C1
A/Texas/06/2009	80	640	5120	320	2560	1280	2560	1280	04/24/13	C1
A/Texas/7/2009	40	320	2560	320	1280	1280	2560	1280	04/26/13	C1
A/Texas/08/2009	40	320	5120	320	1280	1280	2560	640	04/25/13	E2
A/Texas/8/2009	40	320	2560	320	1280	1280	2560	640	04/25/13	C1
A/Texas/09/2009	80	640	2560	320	1280	1280	5120	640	04/26/13	E2
A/Texas/9/2009	20	160	1280	160	640	640	1280	640	04/26/13	C1
A/Indiana/9/2009	40	640	5120	320	1280	1280	2560	640	04/23/13	C1
A/Minnesota/02/2009	80	640	5120	640	2560	2560	5120	1280	UNK	C1
A/Georgia/01/2009	80	640	5120	320	1280	1280	2560	640	04/28/13	E1
A/South Carolina/09/2009	80	640	5120	640	2560	2560	5120	1280	04/27/13	C1
A/Nebraska/02/2009	320	1280	5120	1280	2560	2560	5120	1280	UNK	C1
A/Colorado/03/2009	320	640	5120	640	2560	2560	5120	1280	04/28/13	C1
A/Arizona/02/2009	160	640	5120	640	1280	2560	2560	1280	04/27/13	C1
A/Delaware/02/2009	160	640	5120	640	1280	2560	5120	1280	04/29/13	E1
A/Delaware/03/2009	320	1280	5120	1280	2560	5120	5120	2560	04/29/13	E1
A/Mexico/4482/2009	320	1280	5120	640	2560	2560	5120	2560	04/15/13	E2
A/Mexico/4486/2009	160	1280	5120	1280	2560	2560	5120	1280	04/15/13	C1
A/Mexico/4486/2009	160	1280	2560	640	1280	2560	5120	1280	04/15/13	E2
A/Mexico/4108/2009	320	1280	5120	1280	2560	2560	5120	2560	04/04/13	C1
A/Mexico/4108/2009	80	320	5120	320	1280	1280	2560	1280	04/04/13	E1
A/Mexico/3955/2009	160	640	5120	640	2560	2560	5120	2560	04/04/13	E2
A/Mexico/4108/2009	160	640	5120	320	2560	1280	5120	640	04/04/13	C1/C1
A/Mexico/4486/2009	160	640	5120	640	1280	1280	2560	1280	04/15/13	C1/C1
A/Mexico/4516/2009	40	640	2560	320	1280	2560	5120	1280	04/17/13	C1/C1
A/Mexico/4603/2009	80	320	2560	320	1280	1280	2560	640	04/20/13	E2
A/Mexico/4595/2009	320	640	5120	640	2560	2560	5120	2560	04/21/13	C1/C1
A/Mexico/4596/2009	160	640	5120	640	2560	2560	5120	2560	04/21/13	C1/C1
A/Mexico/4627/2009	80	640	2560	640	640	1280	5120	1280	04/21/13	C1/C1
A/Mexico/4635/2009	160	640	5120	640	2560	2560	5120	1280	04/21/13	C1/C1
A/Mexico/4646/2009	160	640	5120	320	1280	1280	5120	1280	04/21/13	C1/C1
A/Mexico/4176/2009	160	640	2560	640	1280	1280	5120	1280	04/14/13	C1/C1
A/Mexico/4482/2009	320	1280	5120	640	2560	2560	5120	2560	04/15/13	C1
A/Mexico/4603/2009	160	640	5120	640	1280	1280	5120	1280	04/20/13	C1/C1



**Table S6.** GenBank accession numbers of nucleotide sequences of swine influenza viruses used in genetic analyses shown in Fig. S2.

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**Fig. S1.** Phylogenetic trees for each sequenced gene of outbreak strains as of 16 May 2009, and selected other strains. Sequences were chosen, from among all avian, swine, and human influenza sequences in GenBank to be representative of the relevant lineages. Trees made from all swine relevant swine sequences are shown in Fig S2. Lineage labels to the right of the trees indicate to the lineage to which the gene segment belongs. Nucleotide sequences of each gene segment were aligned using Mafft (available from <http://www.ebi.ac.uk/mafft/>). MODELTEST was run on the aligned sequences and GTR+I+ $\Gamma$ 4 was determined to be the most appropriate evolutionary model for phylogenetic tree construction. Initial tree construction was performed using PhyML v2.4.5 under the GTR+I+ $\Gamma$ 4 model. Global optimization of branch topology was then performed on the tree with the best likelihood score from PhyML using Garli v0.96b8 under the model parameters determined by MODELTEST. Garli was run for 300,000 generations. **A.** PB2, **B.** PB1, **C.** PA, **D.** HA, **E.** NP, **F.** NA, **G.** M, **H.** NS.

Fig. S1A PB2

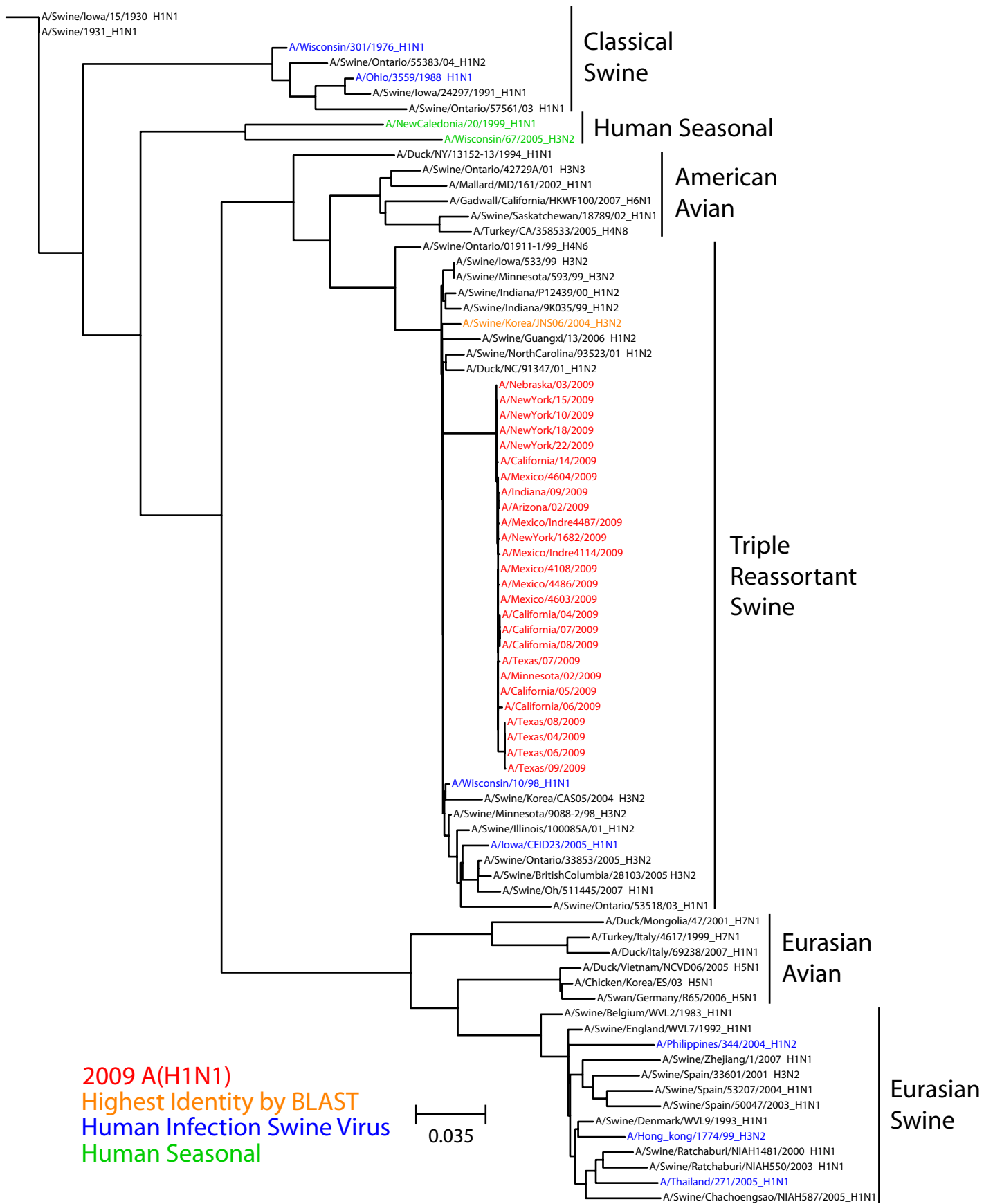




Fig.S1C PA

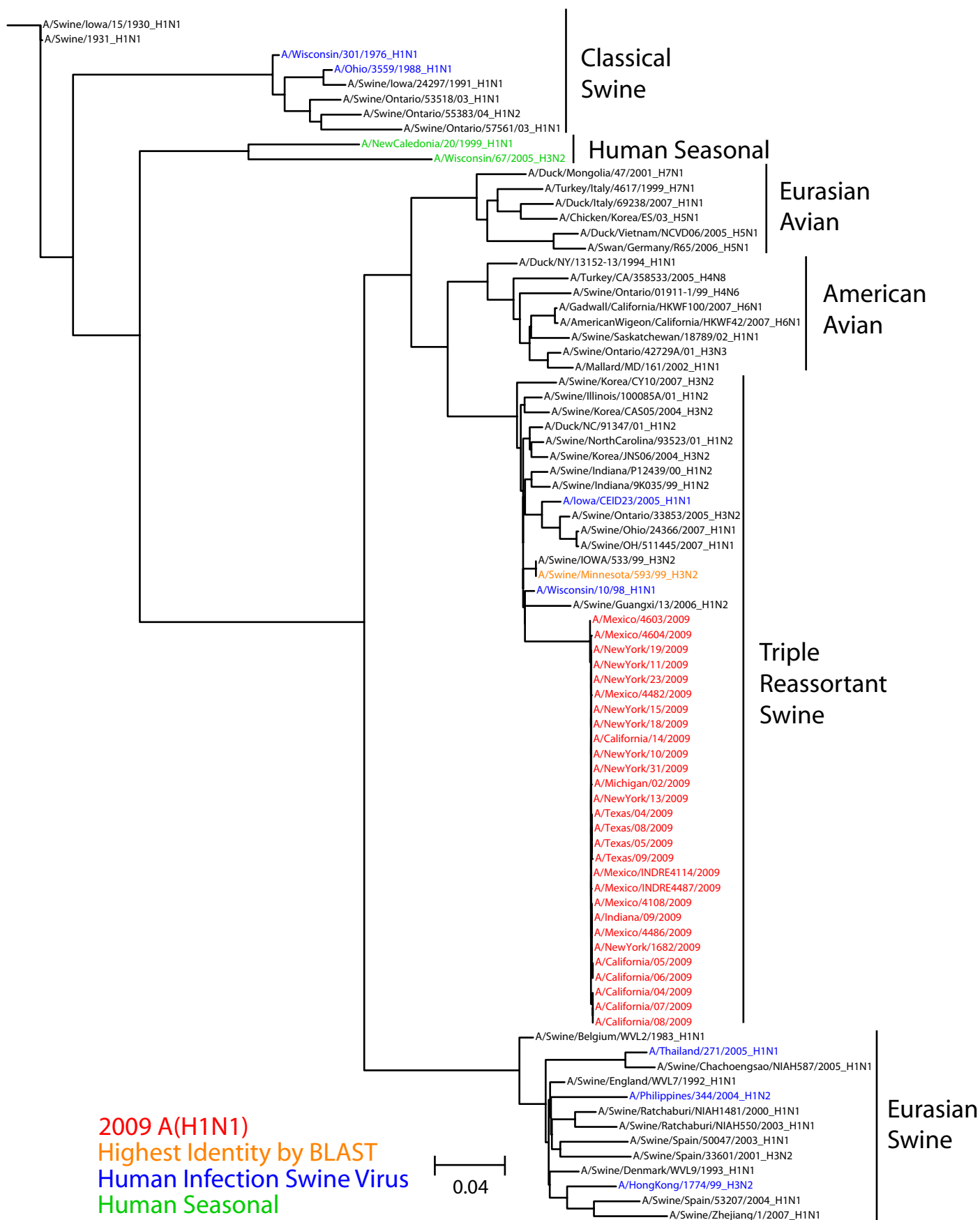


Fig.S1D HA



Fig.S1E NP

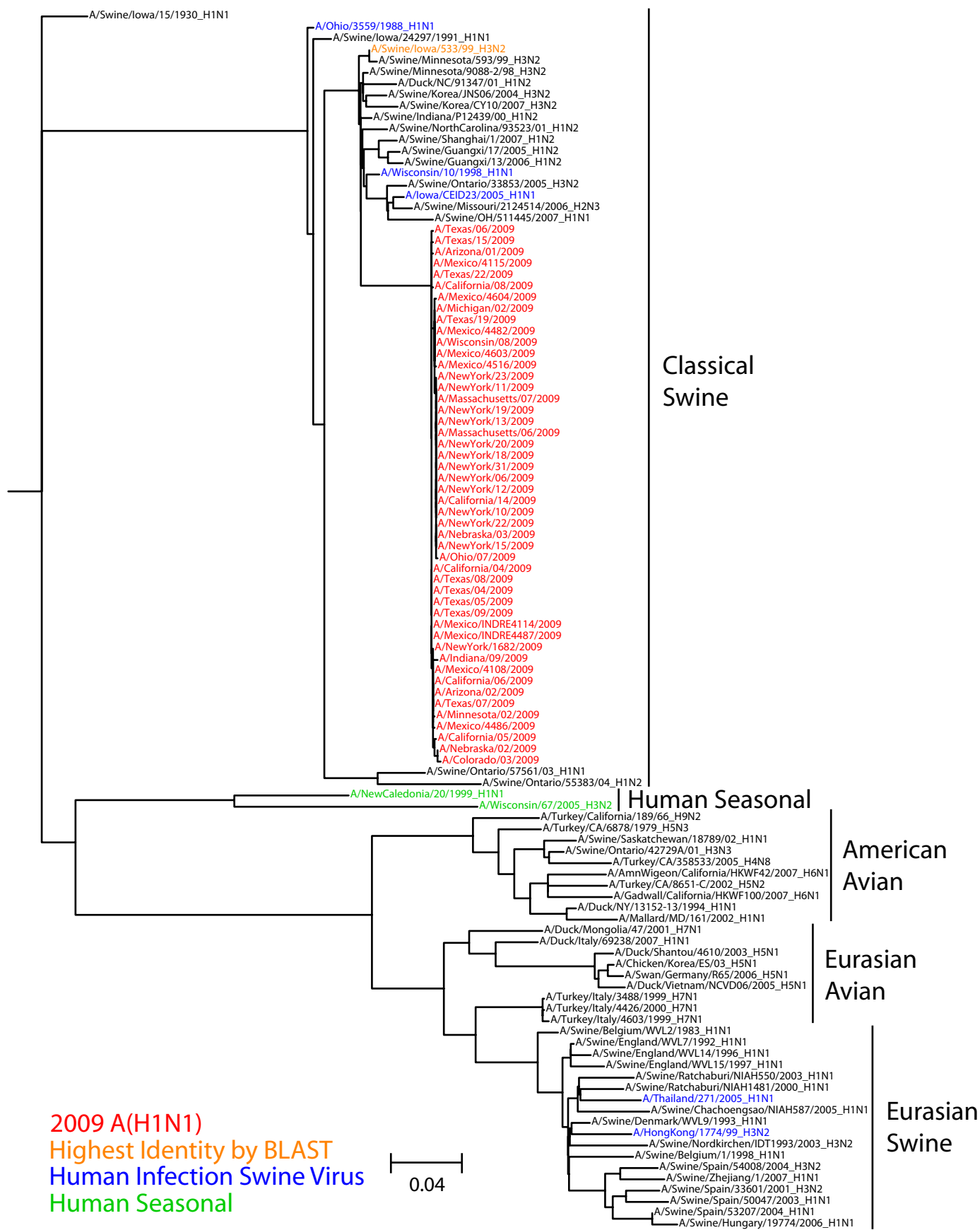




Fig.S1F NA

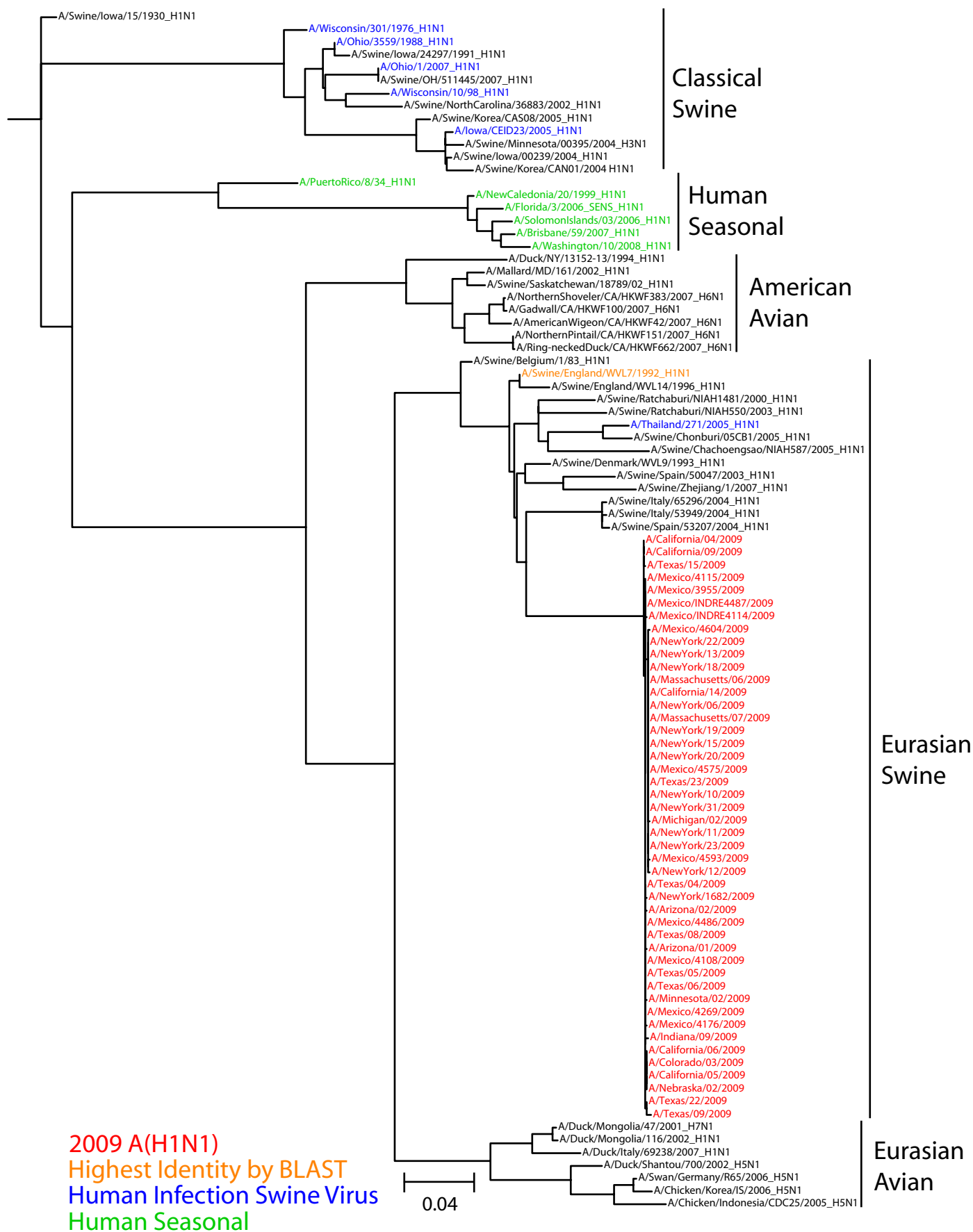
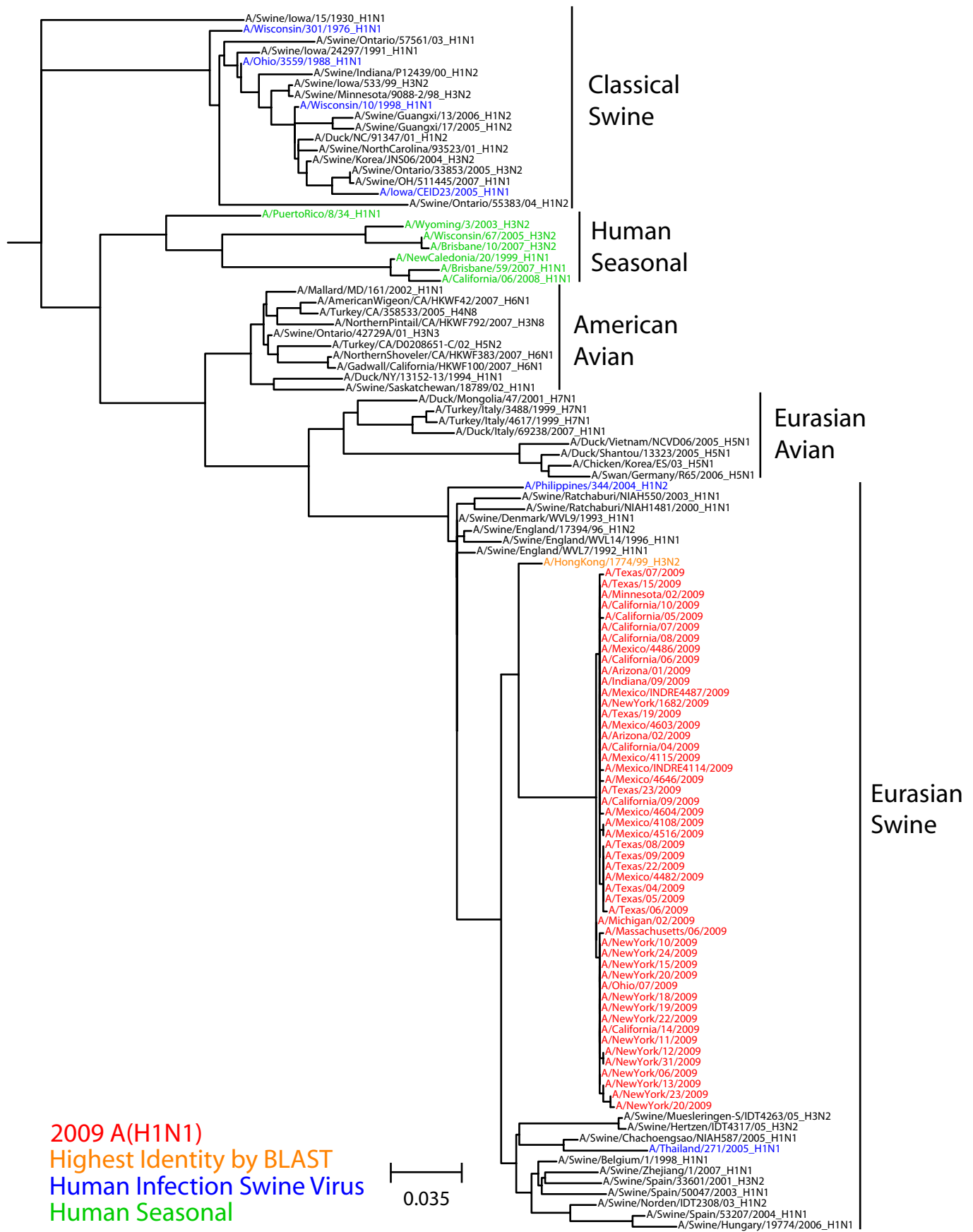
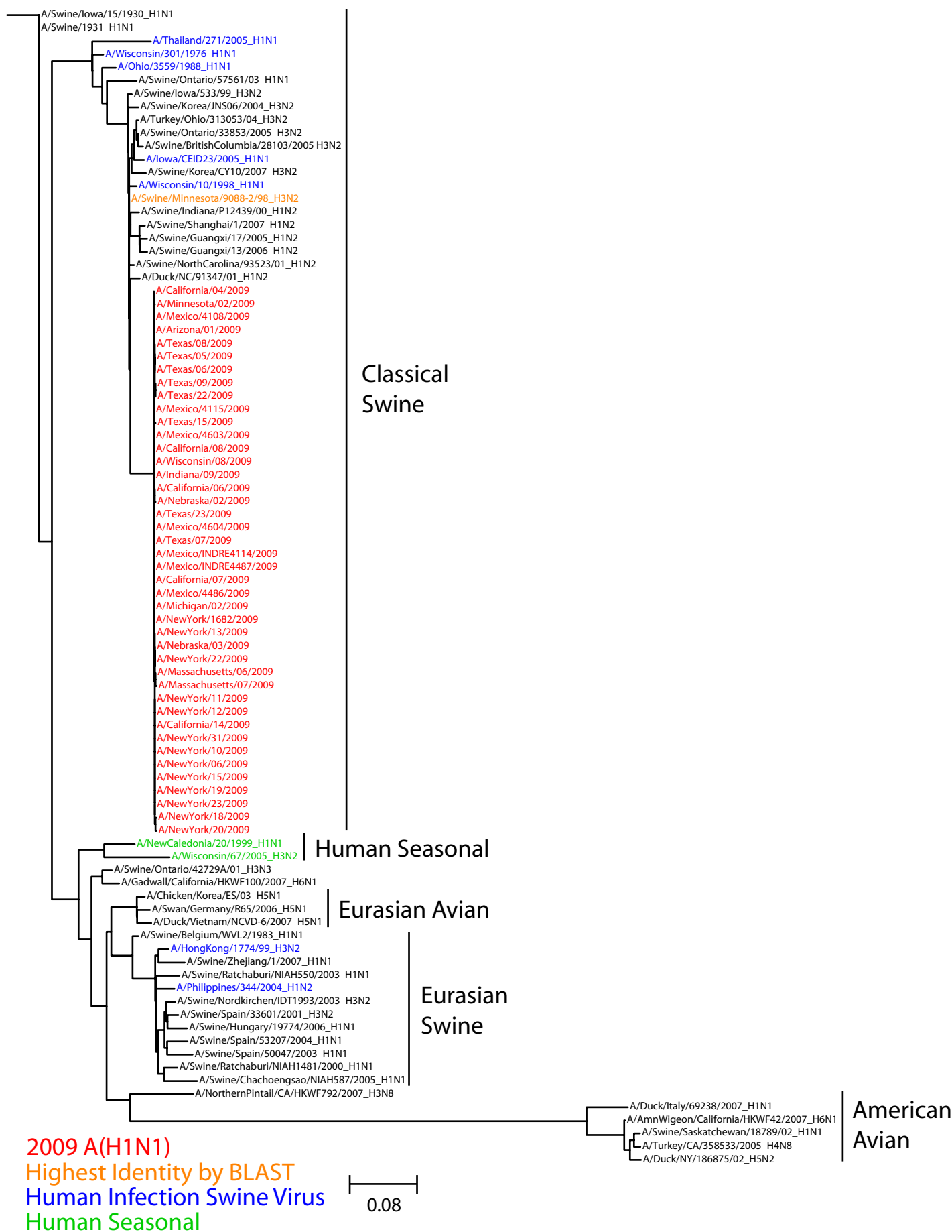


Fig. S1G M





**Fig. S2.** Phylogenetic trees for each gene segment of the 2009 A(H1N1) strains as of 17 May 2009, and all relevant swine sequences (HA all swine H1 sequences longer than 900 base pairs (bp), NA all swine N1 sequences longer than 900 bp, other gene segments all swine viruses (PB2, PB1 and PA longer than 1000 bp; NP longer than 900 bp; M longer than 800 bp; NS longer than 700 bp) in GenBank. 2009 A(H1N1) strains show in red. Nucleotide sequences of each segment were aligned using Mafft (available from <http://www.ebi.ac.uk/mafft/>). MODELTEST was run on the aligned sequences and GTR+I+ $\Gamma$ 4 was determined to be the most appropriate evolutionary model for phylogenetic tree construction. Initial tree construction was performed using PhyML v2.4.5 under the GTR+I+ $\Gamma$ 4 model. Global optimization of branch topology was then performed on the tree with the best likelihood score from PhyML using Garli v0.96b8 under the model parameters determined by MODELTEST. Garli was run for 300,000 generations. At least 52 branches in the Classical Swine and Triple Reassortant Lineages of the trees of the 8 gene segments are approximately as long or longer than the branch to the 2009 A(H1N1) strains (shown in red) for each segment (PB2 7 branches, PB1 5, PA 5, HA 9, NP 6, M 6, NS 9). As with the 2009 A(H1N1) viruses, all 8 segments of two swine H1N1 viruses, A/Swine/Chachoengsao/NIAH587/2005 and A/Swine/Chonburi/NIAH589/2005, appear on long branches with no close relatives. An H1N2 virus, A/Swine/Miyazaki/1/2006, shows a similar pattern for all but its NA sequence which was excluded from the analyses because it is not an N1. **A.** PB2, **B.** PB1, **C.** PA, **D.** HA, **E.** NP, **F.** NA, **G.** M, **H.** NS.



Fig. S2B PB1

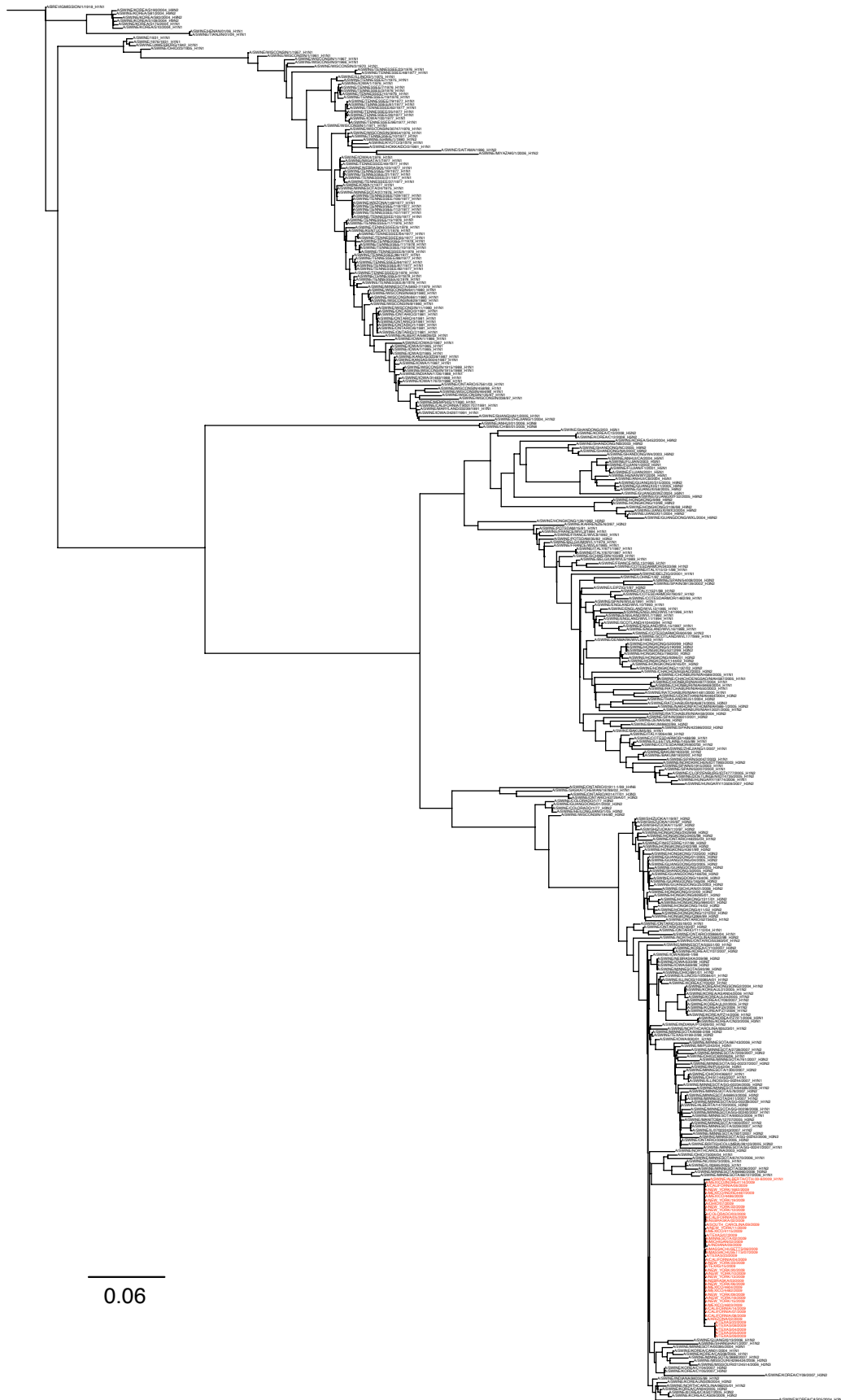




Fig. S2D HA

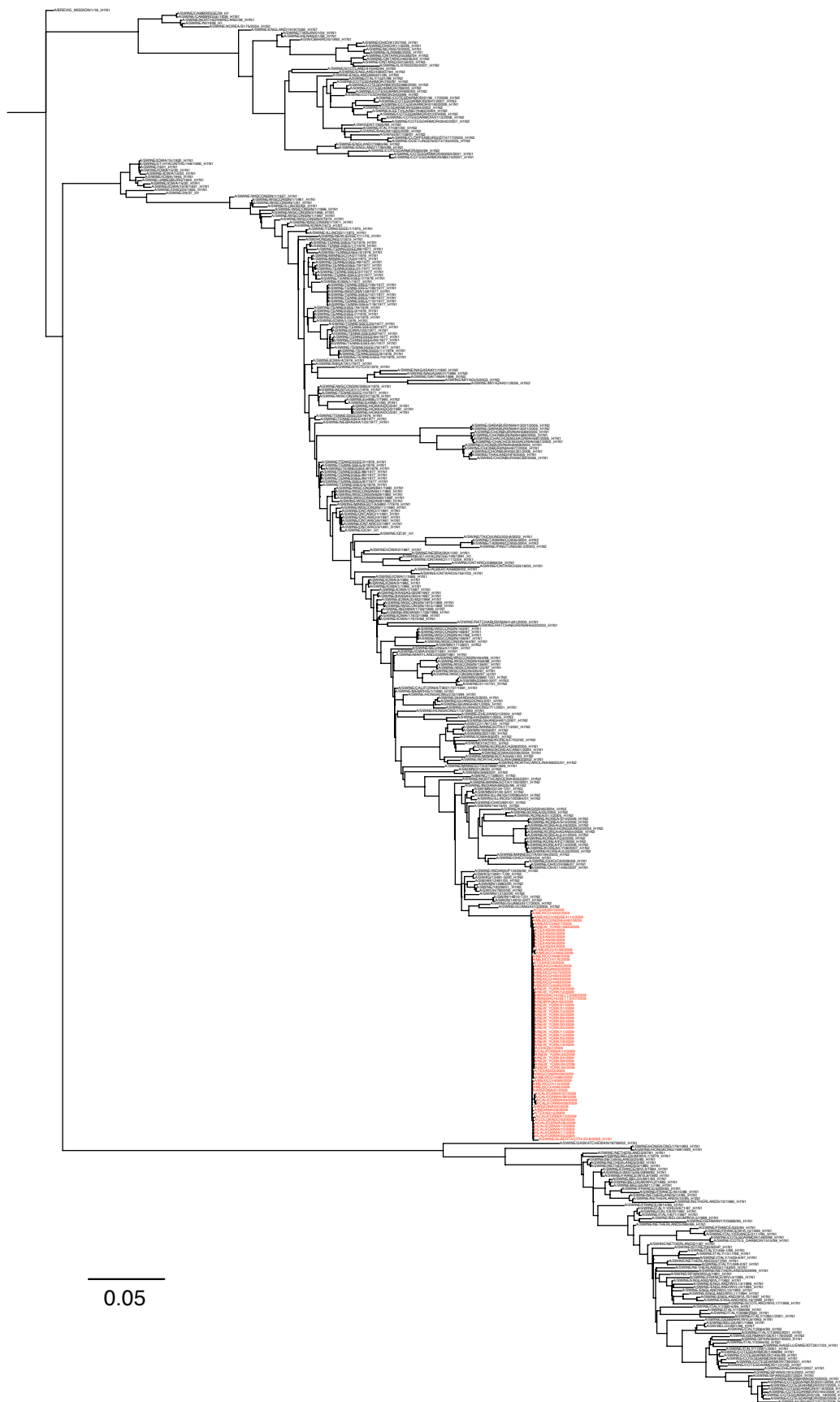




Fig. S2E NP





Fig. S2G M

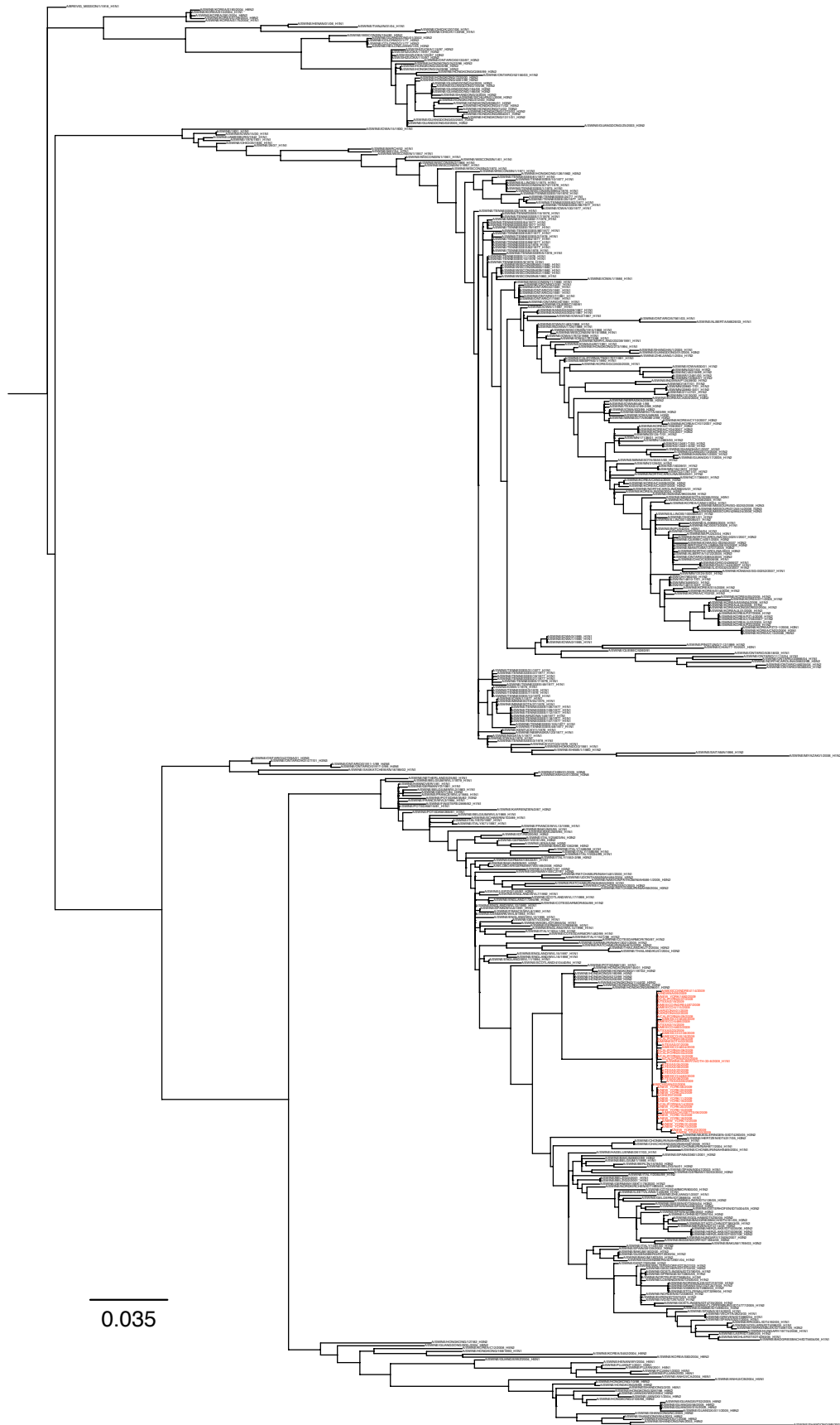


Fig. S2H NS



**Fig. S3.** Version of Fig. 3 from the main manuscript with names of each antigen and antiserum, and as a zoomable PDF to be able to make names legible.

