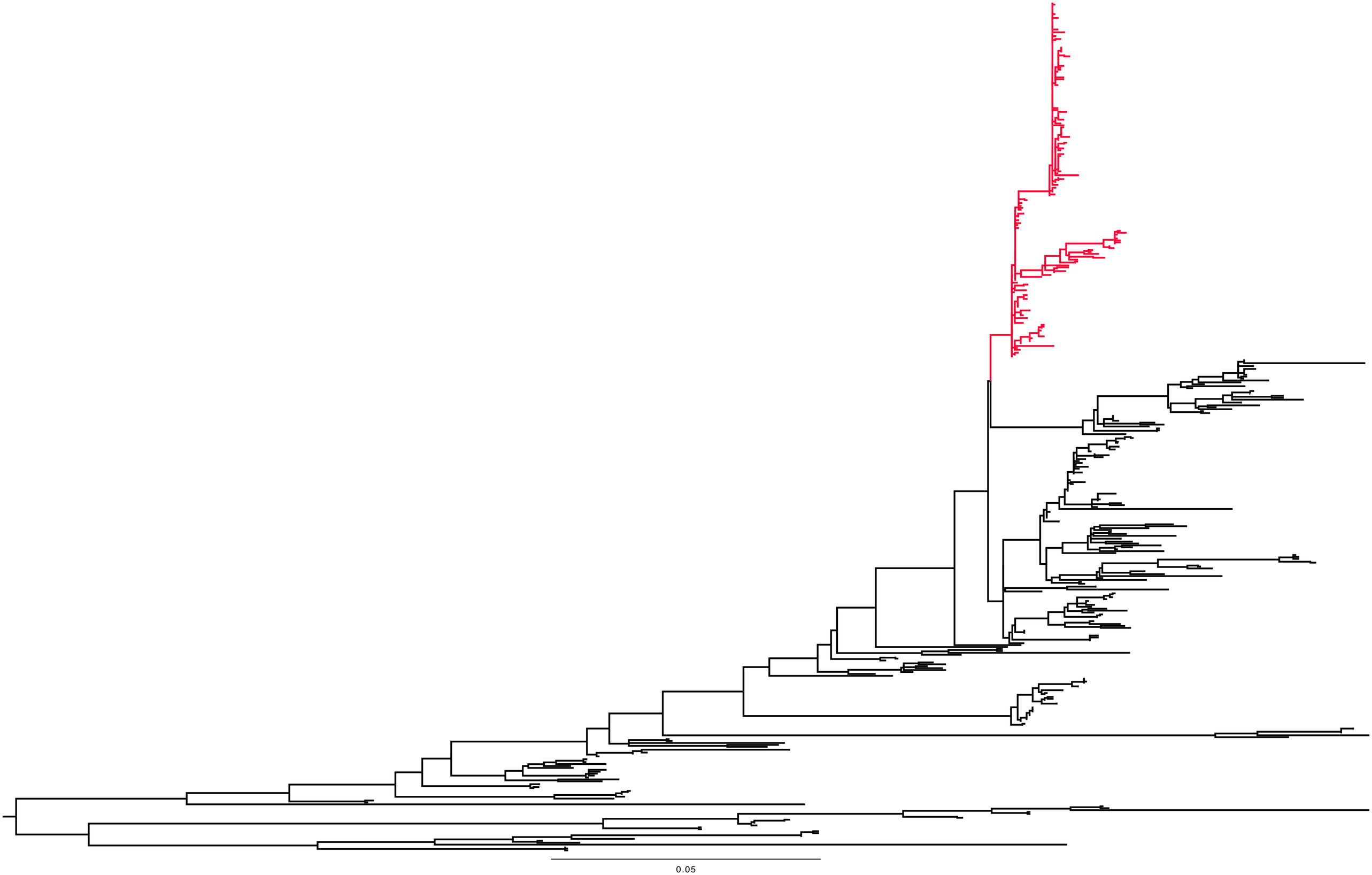
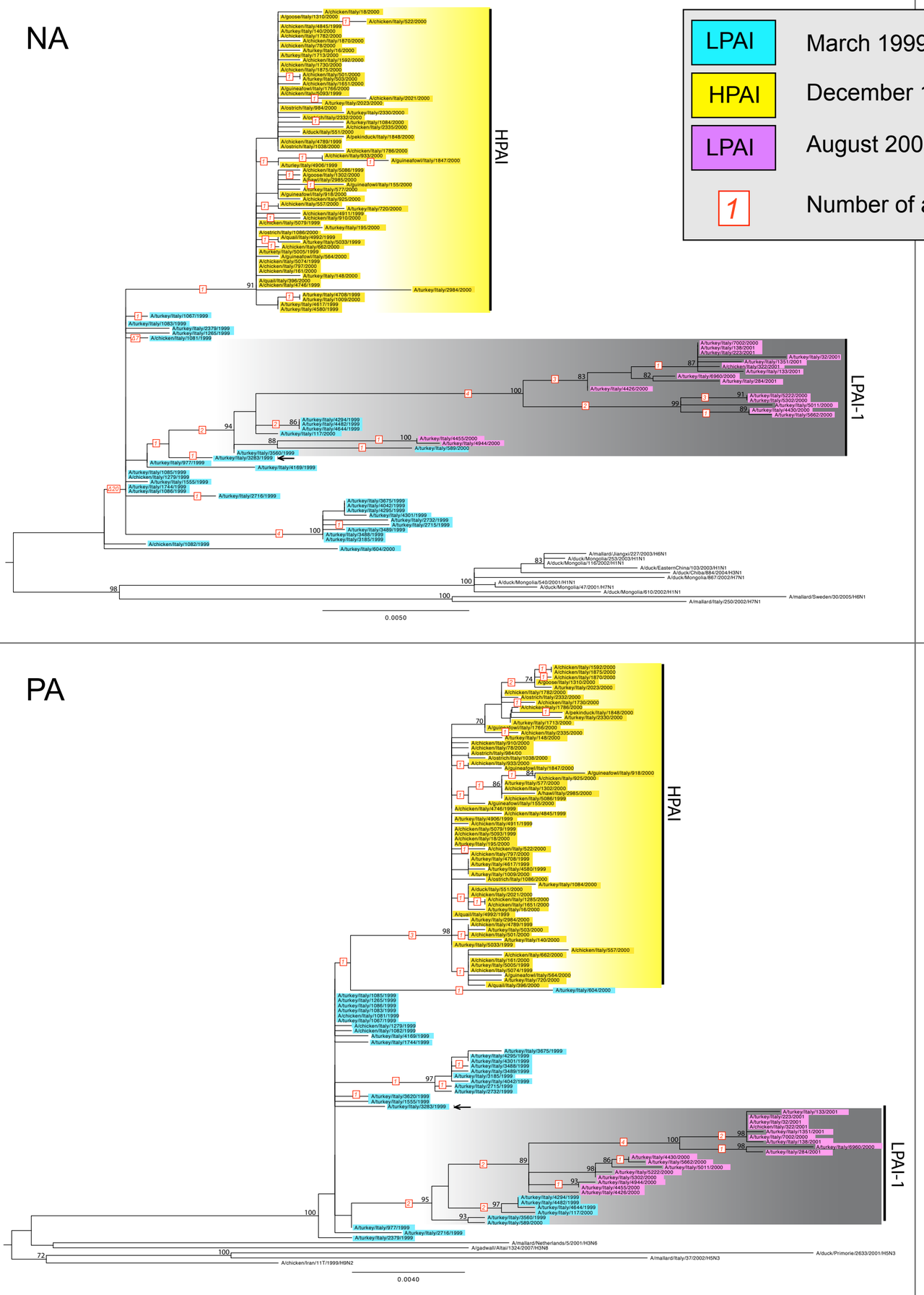
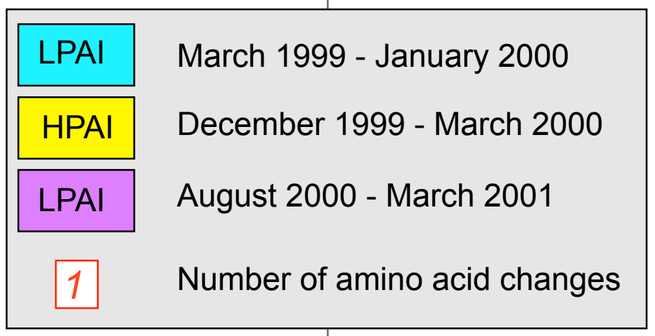

0.05

Fig. S1. HA ML phylogenetic tree. This tree shows phylogenetic relationships among LPAI and HPAI H7N1 viruses (in red) and all the Eurasian H7 sequences available in GenBank (443 sequences). The tree is mid-point rooted for clarity only. Branch lengths are scaled according to the numbers of nucleotide substitutions per site. The tree is mid-point rooted for clarity only.


NA
HPAI
LPAI-1
0.0050
PA
HPAI
LPAI-1
0.0040


LPAI
March 1999 - January 2000
HPAI
December 1999 - March 2000
LPAI
August 2000 - March 2001
1
Number of amino acid changes

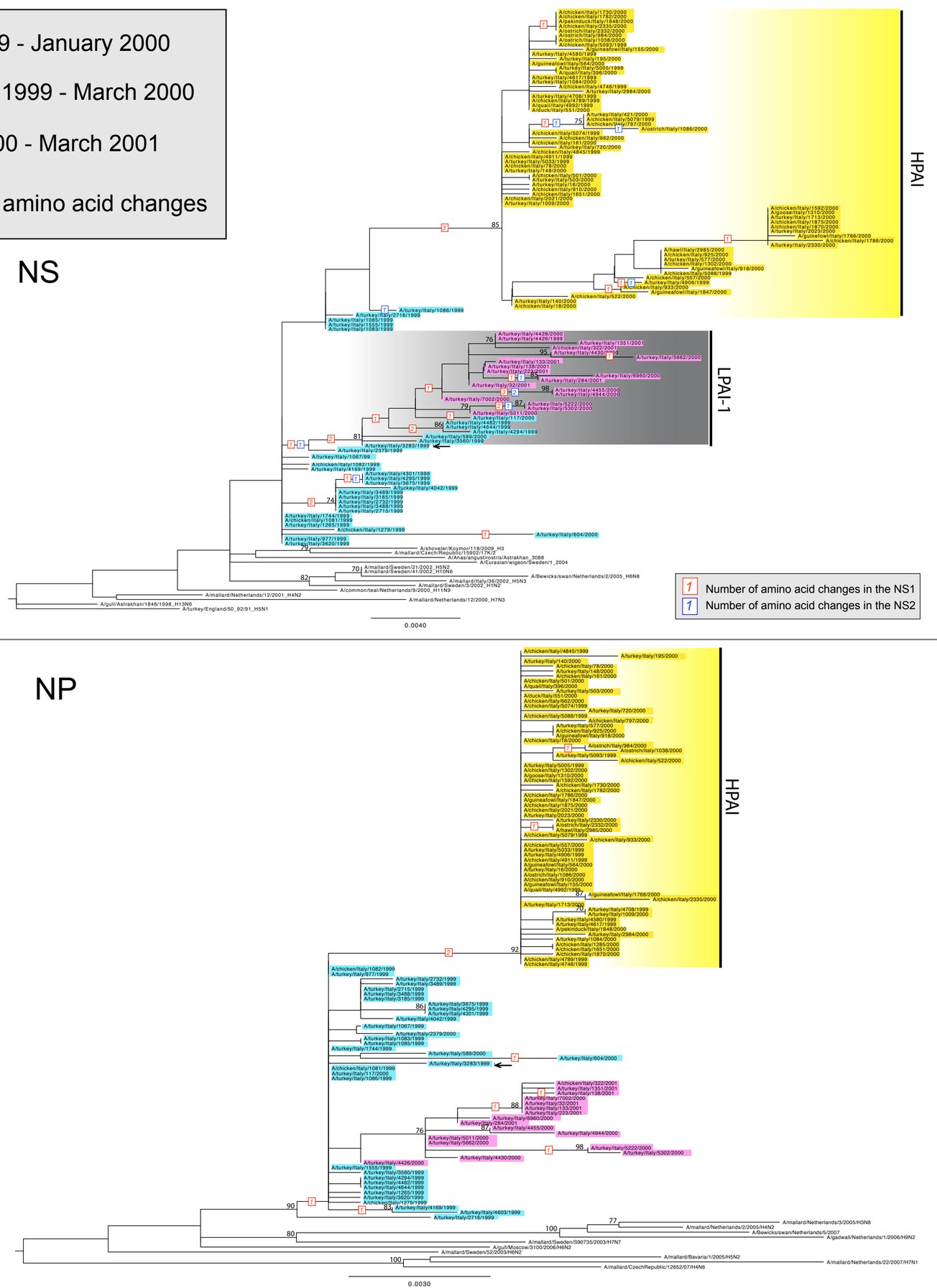
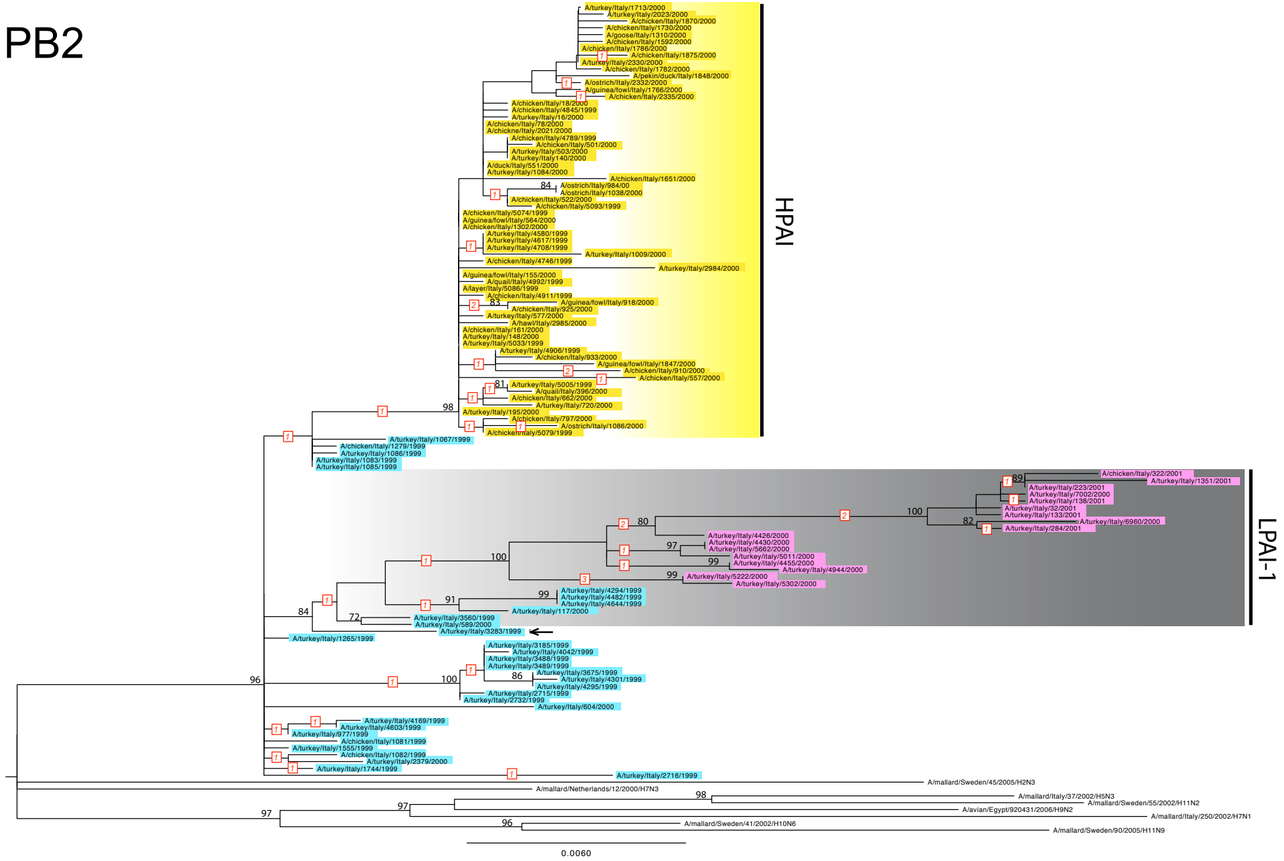

NS
HPAI
LPAI-1
0.0040
NP
HPAI
LPAI-1
0.0030
1 Number of amino acid changes in the NS1
1 Number of amino acid changes in the NS2

Fig. S2. ML phylogenetic tree of the NA, NP, NS and PA gene segments of H7N1 avian influenza subtype. Viruses are coloured as follows: blue for LPAI collected from March 1999 to January 2000, pink for LPAI sampled between August 2000 and February 2001, yellow for HPAI viruses (December 1999-March 2000). The numbers at the nodes represent bootstrap values (>60%), while branch lengths are scaled according to the numbers of nucleotide substitutions per site. The numbers highlighted in red and blue represent the number of amino acid changes identified along the main branches. The closest relative of the LPAI-I group is identified with a black arrow. The trees are mid-point rooted for clarity only.

PB2

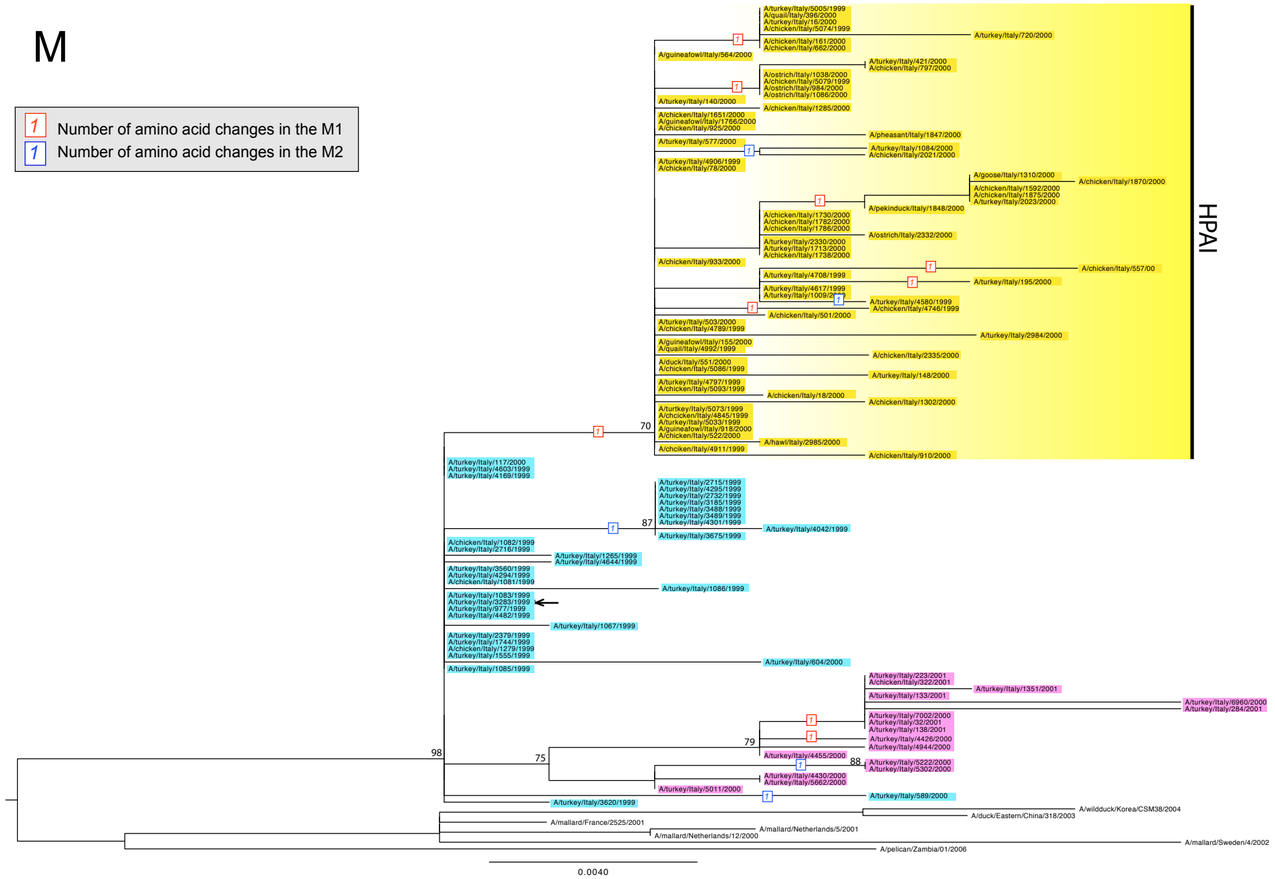


LPAI	March 1999 - January 2000
HPAI	December 1999 - March 2000
LPAI	August 2000 - March 2001
1	Number of amino acid changes

M

1 Number of amino acid changes in the M1

1 Number of amino acid changes in the M2



PB1

1 Number of amino acid changes in the PB1

1 Number of amino acid changes in the PB1-F2

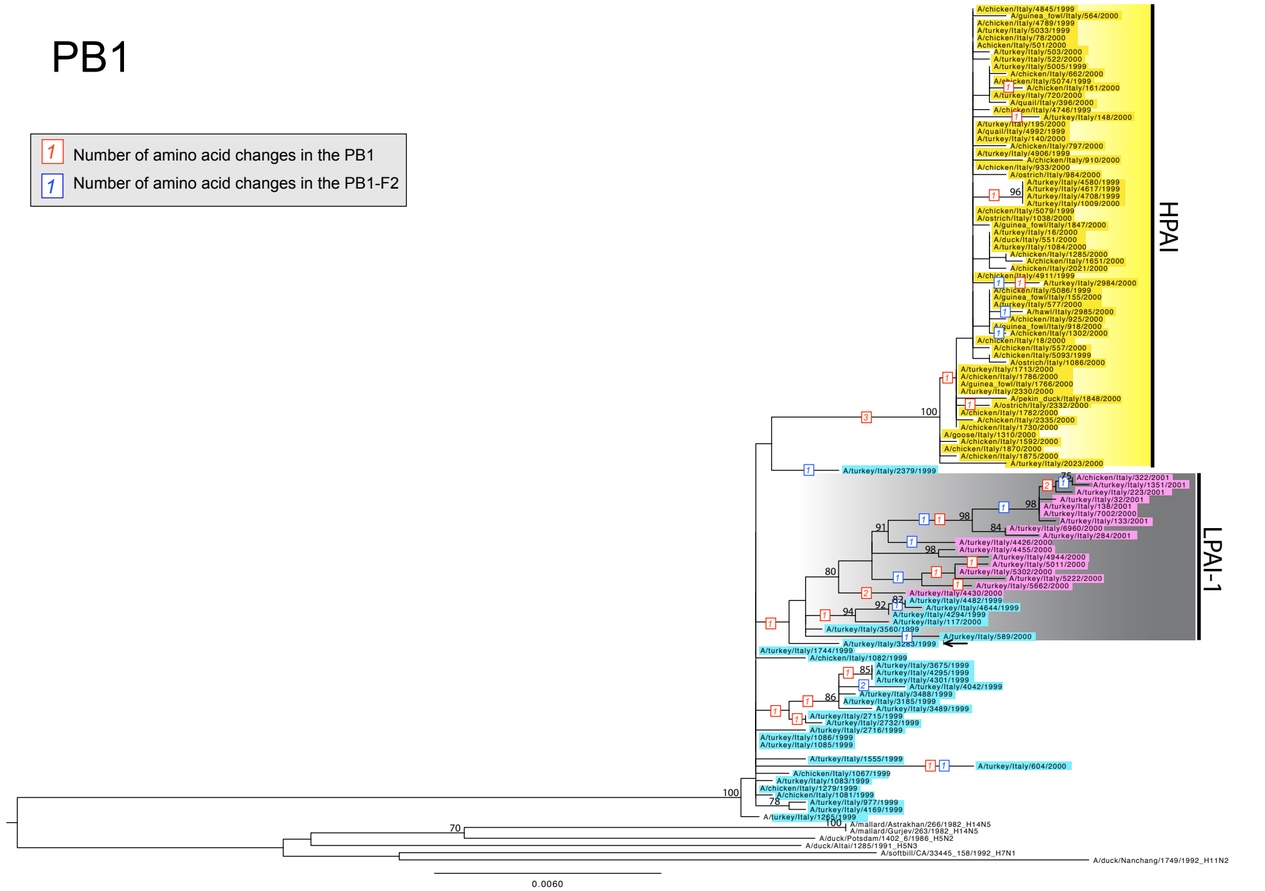


Fig. S3. ML phylogenetic tree of the PB1, PB2, M gene segments of H7N1 avian influenza subtype. The colour scheme is the same as that used in Fig. S2. The number highlighted in red represents the number of amino acid changes identified along the main branches. The numbers at the nodes represent bootstrap values (>60%), while branch lengths are scaled according to the numbers of nucleotide substitutions per site. The numbers highlighted in red and blue represent the number of amino acid changes identified along the main branches. The closest relative of the LPAI-I group is identified with a black arrow. The trees are mid-point rooted for clarity only.

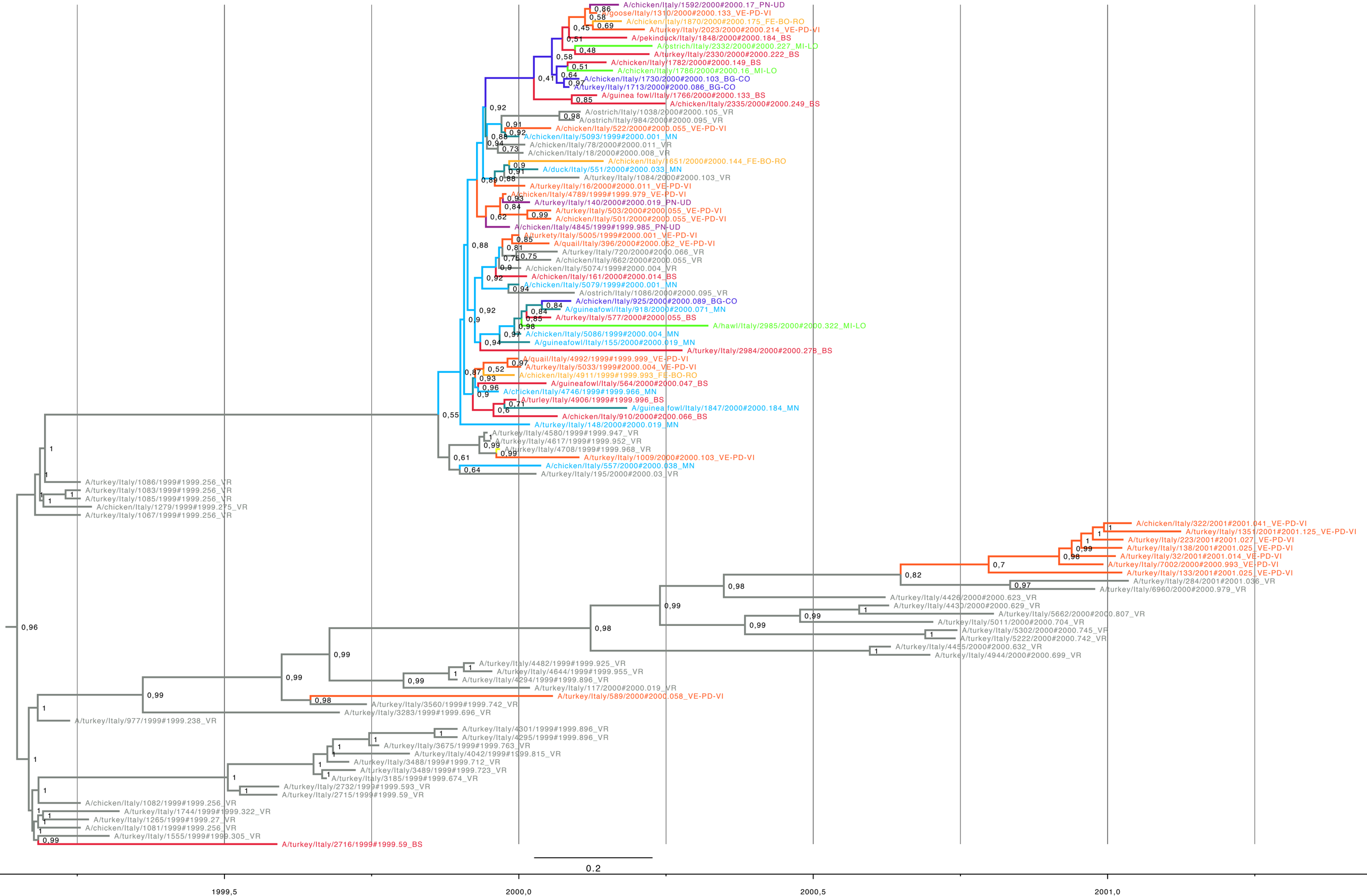
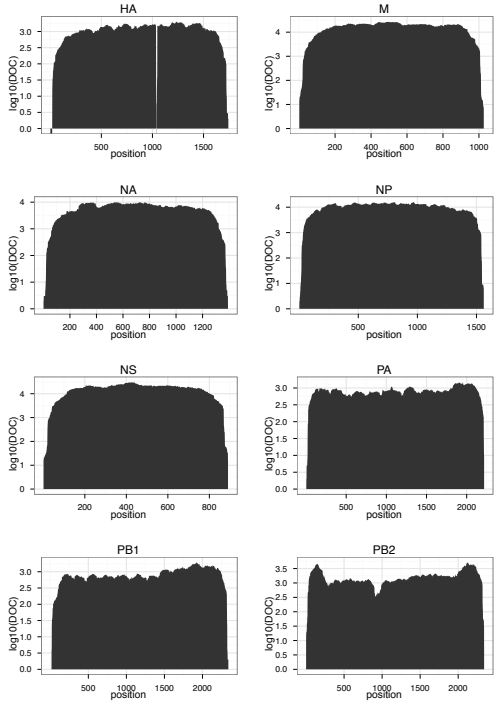
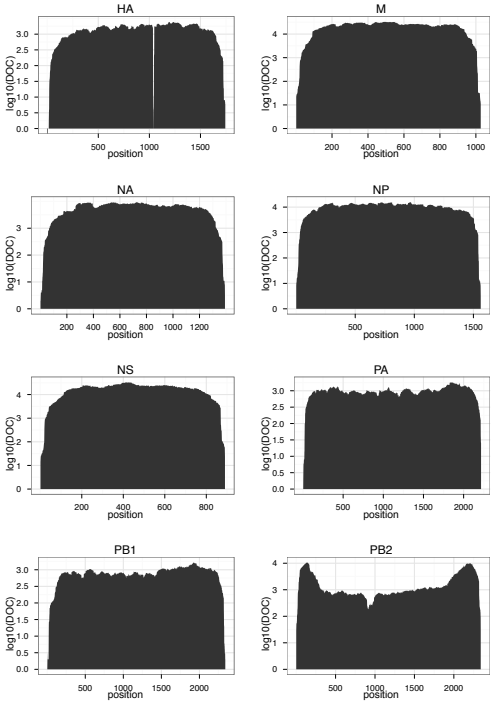

A/chicken/Italy/1592/2000#2000.17_PN-UD
A/goose/Italy/1310/2000#2000.133_VE-PD-VI
A/chicken/Italy/1870/2000#2000.175_FE-BO-RO
A/turkey/Italy/2023/2000#2000.214_VE-PD-VI
A/pekinduck/Italy/1848/2000#2000.184_BS
A/ostrich/Italy/2332/2000#2000.227_MI-LO
A/turkey/Italy/2330/2000#2000.222_BS
A/chicken/Italy/1782/2000#2000.149_BS
A/chicken/Italy/1786/2000#2000.16_MI-LO
A/chicken/Italy/1730/2000#2000.103_BG-CO
A/turkey/Italy/1713/2000#2000.086_BG-CO
A/guinea fowl/Italy/1766/2000#2000.133_BS
A/chicken/Italy/2335/2000#2000.249_BS
A/ostrich/Italy/1038/2000#2000.105_VR
A/ostrich/Italy/984/2000#2000.095_VR
A/chicken/Italy/522/2000#2000.055_VE-PD-VI
A/chicken/Italy/5093/1999#2000.001_MN
A/chicken/Italy/78/2000#2000.011_VR
A/chicken/Italy/18/2000#2000.008_VR
A/chicken/Italy/1651/2000#2000.144_FE-BO-RO
A/duck/Italy/551/2000#2000.033_MN
A/turkey/Italy/1084/2000#2000.103_VR
A/turkey/Italy/16/2000#2000.011_VE-PD-VI
A/chicken/Italy/4789/1999#1999.979_VE-PD-VI
A/turkey/Italy/140/2000#2000.019_PN-UD
A/turkey/Italy/503/2000#2000.055_VE-PD-VI
A/chicken/Italy/501/2000#2000.055_VE-PD-VI
A/chicken/Italy/4845/1999#1999.985_PN-UD
A/turkey/Italy/5005/1999#2000.001_VE-PD-VI
A/quail/Italy/396/2000#2000.052_VE-PD-VI
A/turkey/Italy/720/2000#2000.066_VR
A/chicken/Italy/662/2000#2000.055_VR
A/chicken/Italy/5074/1999#2000.004_VR
A/chicken/Italy/161/2000#2000.014_BS
A/chicken/Italy/5079/1999#2000.001_MN
A/ostrich/Italy/1086/2000#2000.095_VR
A/chicken/Italy/925/2000#2000.089_BG-CO
A/guineafowl/Italy/918/2000#2000.071_MN
A/turkey/Italy/577/2000#2000.055_BS
A/hawl/Italy/2985/2000#2000.322_MI-LO
A/chicken/Italy/5086/1999#2000.004_MN
A/guineafowl/Italy/155/2000#2000.019_MN
A/turkey/Italy/2984/2000#2000.278_BS
A/quail/Italy/4992/1999#1999.999_VE-PD-VI
A/turkey/Italy/5033/1999#2000.004_VE-PD-VI
A/chicken/Italy/4911/1999#1999.993_FE-BO-RO
A/guineafowl/Italy/564/2000#2000.047_BS
A/chicken/Italy/4746/1999#1999.966_MN
A/turley/Italy/4906/1999#1999.996_BS
A/guinea fowl/Italy/1847/2000#2000.184_MN
A/chicken/Italy/910/2000#2000.066_BS
A/turkey/Italy/148/2000#2000.019_MN
A/turkey/Italy/4580/1999#1999.947_VR
A/turkey/Italy/4617/1999#1999.952_VR
A/turkey/Italy/4708/1999#1999.968_VR
A/turkey/Italy/1009/2000#2000.103_VE-PD-VI
A/chicken/Italy/557/2000#2000.038_MN
A/turkey/Italy/195/2000#2000.03_VR
A/turkey/Italy/1086/1999#1999.256_VR
A/turkey/Italy/1083/1999#1999.256_VR
A/turkey/Italy/1085/1999#1999.256_VR
A/chicken/Italy/1279/1999#1999.275_VR
A/turkey/Italy/1067/1999#1999.256_VR
A/chicken/Italy/322/2001#2001.041_VE-PD-VI
A/turkey/Italy/1351/2001#2001.125_VE-PD-VI
A/turkey/Italy/223/2001#2001.027_VE-PD-VI
A/turkey/Italy/138/2001#2001.025_VE-PD-VI
A/turkey/Italy/32/2001#2001.014_VE-PD-VI
A/turkey/Italy/7002/2000#2000.993_VE-PD-VI
A/turkey/Italy/133/2001#2001.025_VE-PD-VI
A/turkey/Italy/284/2001#2001.036_VR
A/turkey/Italy/6960/2000#2000.979_VR
A/turkey/Italy/4426/2000#2000.623_VR
A/turkey/Italy/4430/2000#2000.629_VR
A/turkey/Italy/5662/2000#2000.807_VR
A/turkey/Italy/5011/2000#2000.704_VR
A/turkey/Italy/5302/2000#2000.745_VR
A/turkey/Italy/5222/2000#2000.742_VR
A/turkey/Italy/4455/2000#2000.632_VR
A/turkey/Italy/4944/2000#2000.699_VR
A/turkey/Italy/4482/1999#1999.925_VR
A/turkey/Italy/4644/1999#1999.955_VR
A/turkey/Italy/4294/1999#1999.896_VR
A/turkey/Italy/117/2000#2000.019_VR
A/turkey/Italy/589/2000#2000.058_VE-PD-VI
A/turkey/Italy/3560/1999#1999.742_VR
A/turkey/Italy/3283/1999#1999.696_VR
A/turkey/Italy/977/1999#1999.238_VR
A/turkey/Italy/4301/1999#1999.896_VR
A/turkey/Italy/4295/1999#1999.896_VR
A/turkey/Italy/3675/1999#1999.763_VR
A/turkey/Italy/4042/1999#1999.815_VR
A/turkey/Italy/3488/1999#1999.712_VR
A/turkey/Italy/3489/1999#1999.723_VR
A/turkey/Italy/3185/1999#1999.674_VR
A/turkey/Italy/2732/1999#1999.593_VR
A/turkey/Italy/2715/1999#1999.59_VR
A/chicken/Italy/1082/1999#1999.256_VR
A/turkey/Italy/1744/1999#1999.322_VR
A/turkey/Italy/1265/1999#1999.27_VR
A/chicken/Italy/1081/1999#1999.256_VR
A/turkey/Italy/1555/1999#1999.305_VR
A/turkey/Italy/2716/1999#1999.59_BS
0.2
1999,5
2000,0
2000,5
2001,0

Fig. S4. MCC tree inferred for the concatenated gene sequences of the Italian H7N1 viruses. Sequences are coloured according to the province of origin. Light blue: Mantova; grey: Verona; red: Brescia; yellow: Ferrara-Ravenna-Bologna-Rovigo; orange: Venezia-Padova-Vicenza; blue: Bergamo-Como; green: Milano-Lodi. The numbers at branch points represent the probability of spatial states.

2732/99



3675/99



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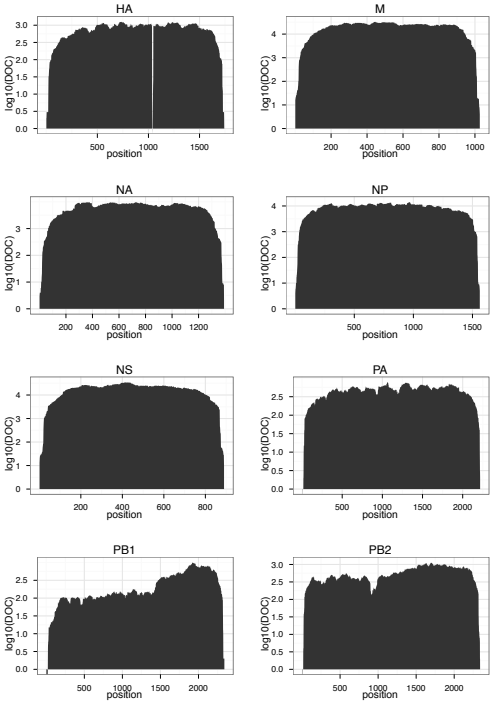

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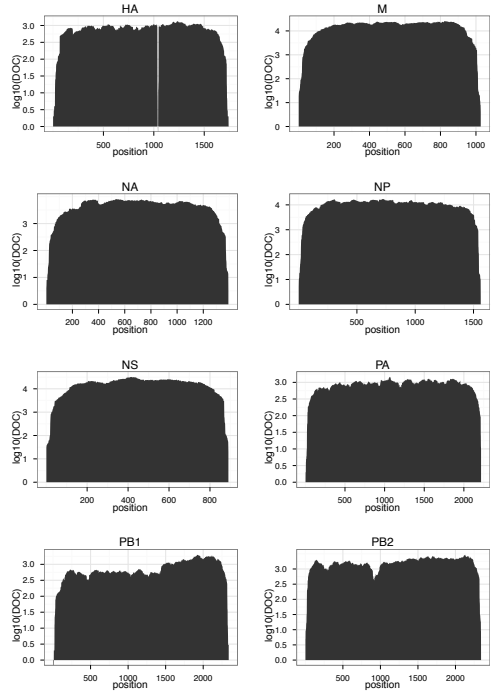
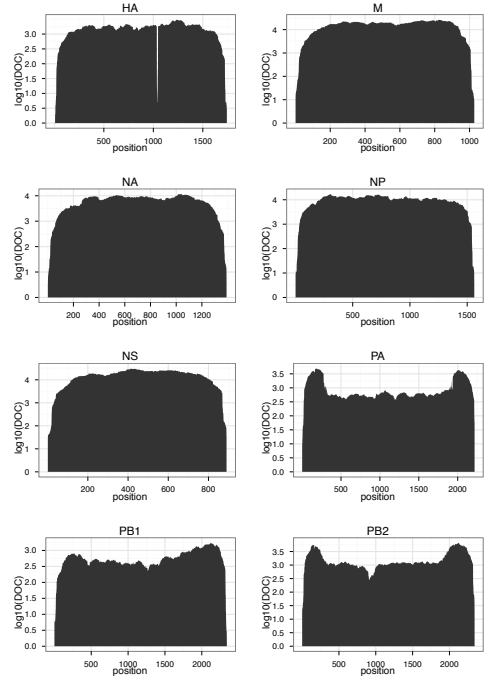


4829/99



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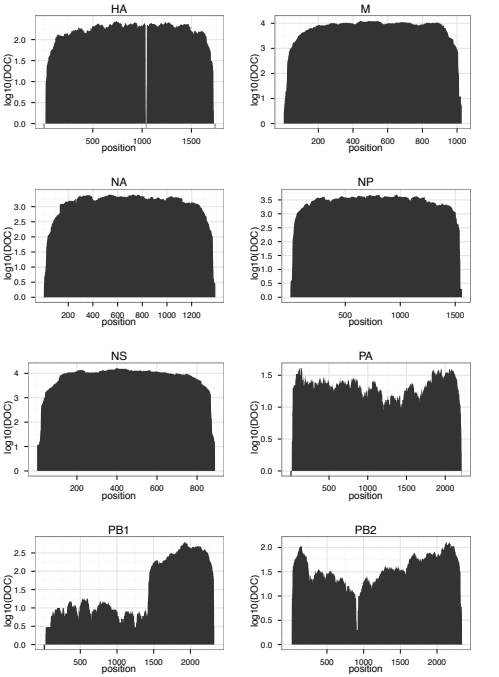
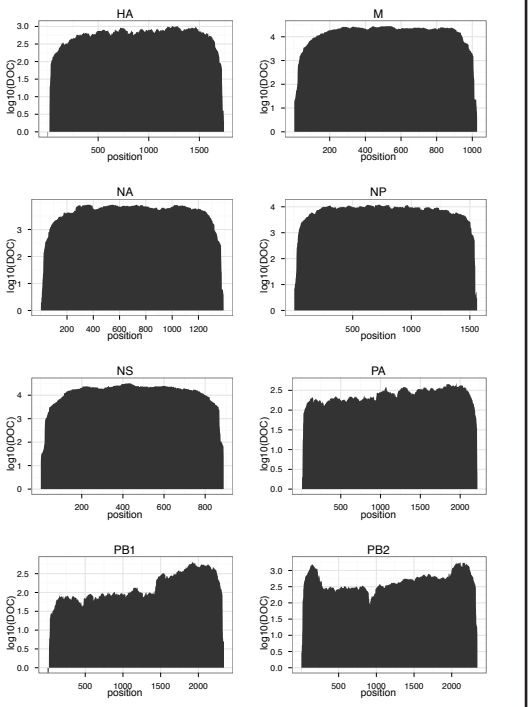
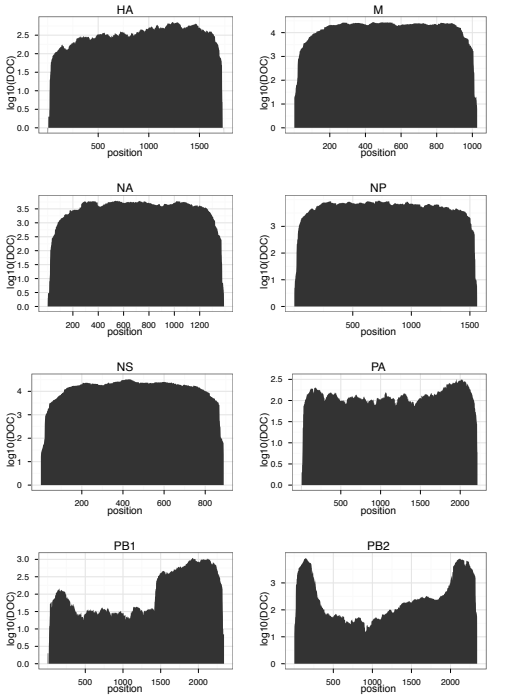


Fig. S5. Depth of coverage plots. The x-axes represent nucleotide position and the y-axes represent depth for LPAI samples. Reads are aligned to the reference A/turkey/Italy/4749/99 (HPAI) virus sequences.

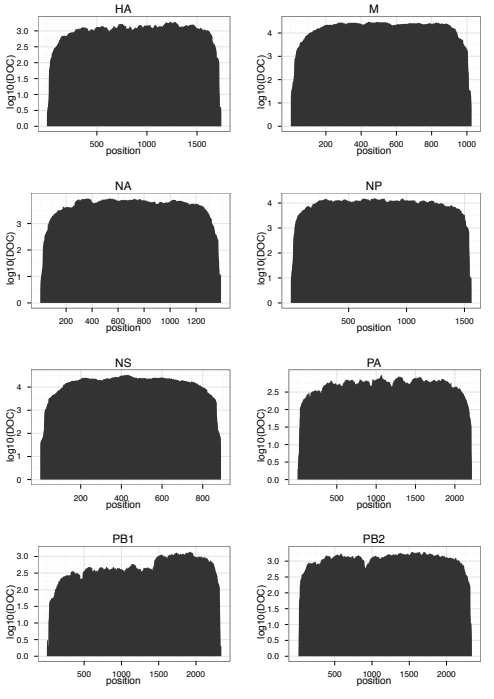
4828/99



4827/99



4749/99



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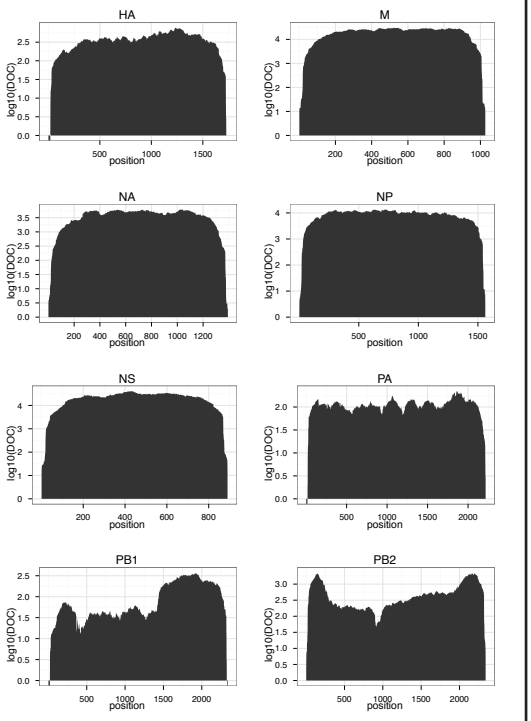
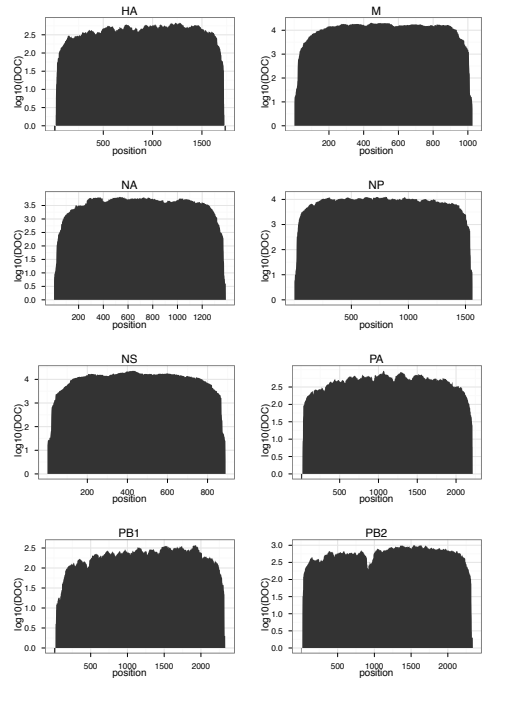
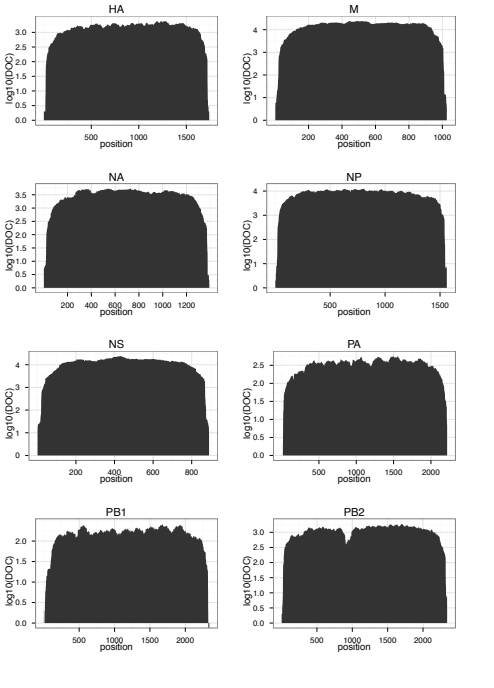


4911/99



4756/99



4618/99

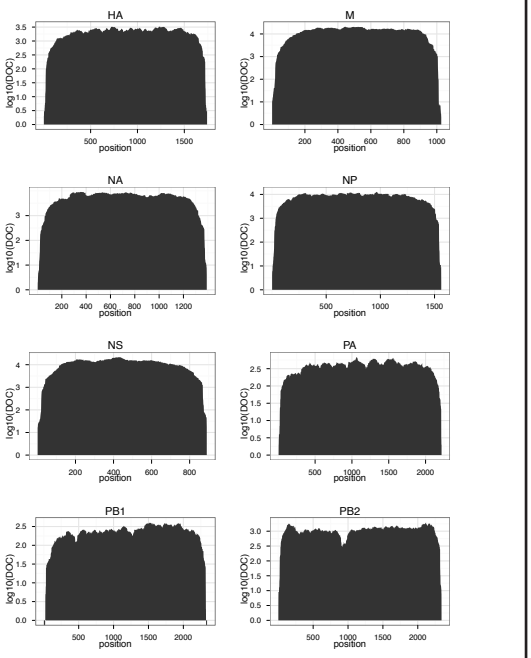


Fig. S6. Depth of coverage plots. The x-axes represent nucleotide position and the y-axes represent depth for HPAI samples. Reads are aligned to the reference A/turkey/Italy/4749/99 (HPAI) virus sequences.

Table S2. Values of Association Index (AI), Parsimony Score (PS) and Monophyletic Clade (MC) obtained using BaTS program from the analysis of all the Italian H7N1 viruses.

Dataset	Statistic	Observed mean	Lower 95% CI	Upper 95% CI	Null mean	Lower 95% CI	Upper 95% CI	P-value
H7N1	AI	6,350	5,325	7,328	10,697	9,739	11,706	0
	PS	52,711	50	56	74,056	70,378	77,059	0
	MC(VR)	11,162	11	12	3,246	2,456	4,994	0,009
	MC(MN)	2,058	2	3	1,416	1,038	2,006	0,110
	MC(BS)	5,103	4	7	1,959	1,402	2,559	0,009
	MC(PN-UD)	1,006	1	1	1,020	1	1,105	1
	MC(FE-RA-RO)	1,011	1	1	1,033	1	1,094	1
	MC(BG-CO)	1,304	1	2	1,014	1	1,089	1
	MC(MI-LO)	1	1	1	1,014	1	1,041	1
	MC(VE-PD-VI)	8	8	8	1,744	1,249	2,252	0,009
LPAI H7N1	AI	1,796	1,154	2,428	4,751	4,078	5,490	0
	PS	16,331	14	18	29,817	27,875	31,352	0
	MC (VR)	11,342	11	13	3,877	2,682	5,653	0,009
	MC (MN)	2,044	2	3	1,100	1	1,586	0,029
	MC (BS)	5,268	4	7	1,689	1,118	2,361	0,009
	MC (VE-PD-VI)	8	8	8	1,309	1,000	2,047	0,009
	MC (FE-RA-BO-RO)	1	1	1	1,007	1	1,025	1
HPAI H7N1	AI	4,949	4,101	5,758	5,749	5,225	6,186	0
	PS	35,480	33	38	40,021	37,550	42,363	0
	MC (PN-UD)	1,011	1	1	1,031	1	1,084	1
	MC (BS)	1,698	1	2	1,654	1,265	2,162	0,180
	MC (FE-RA-BO-RO)	1,018	1	1	1,036	1	1,144	1
	MC (BG-CO)	1,322	1	2	1,054	1	1,260	1
	MC (MI-LO)	1	1	1	1,034	1	1,116	1
	MC (VR)	2,301	2	3	1,645	1,195	2,342	0,139
	MC (MN)	1,207	1	2	1,374	1,048	1,893	1
	MC (VE-PD-VI)	2,161	2	3	1,642	1,208	2,234	0,149

Table S3 - Best-fit phylogeographic model. Log marginal likelihood estimates for four phylogeographic models. The log Bayes factor (BF) comparison between each possible model and a model with rates fixed equally is provided, with the best-fit model highlighted in bold.

	Phylogeographic model	**Log marginal likelihood**	**Log BF**
HA	Rates fixed equally	-4597.83	-
	Rate fixed to population of destination	-4593.2	**2.012**
	Rates fixed to population of origin	-4599.6	-0.769
	Rates fixed to product (destination * origin)	-4598.26	-0.188
	Rates fixed to swine flow	-4594.58	1.414
Complete genome	Rates fixed equally	-26505.95	-
	Rate fixed to population of destination	-26496.86	3.9949
	Rates fixed to population of origin	-26510.41	-1.933
	Rates fixed to product (destination * origin)	-26508-08	-0.924
	Rates fixed to swine flow	-26502.91	1.322