

Table S3. Summary of regions of significant constraint found in H1N2 (swine host) influenza A genes, using weighted and raw (un-ranked) codon variability values. Reference sequences used are GenBank AB731582.1, AB731583.1, AB731584.1, AB731585.1, AB731586.1, AB731587.1, AB731588.1, AB731589.1, for segments 1–8, respectively. Citation details may be found in S1 Appendix. *Denotes a region only found by excluding a potentially interfering signal. *Z*- and *p*-values in parentheses denote values prior to removal of the next most significant signal. If parenthetical values are absent, then such a signal was removed in an earlier step only.

Gene	Order found	Refseq nt location	<i>Z</i>	<i>p</i>	Comment
PB2	2	7–81	1.19	0.0058	Packaging-associated(21, 22); conserved RNA structure(18)
	1	2212–2277	1.55	<0.0001	Packaging-associated(4–6, 21, 23, 24); conserved RNA structure(3, 25)
PB1	1	2224–2265	1.46	<0.0001	Packaging-associated(5, 6, 21, 22); conserved RNA structure(3, 18)
PB1-F2	Nil found				
PA	3	10–69	1.12	0.0312	Packaging-associated(6, 22)
	2	559–609	1.29	0.0038	Proposed frameshift stimulator (see main text)
	4	640–759	1.12	0.047	Overlap PA-X(26)
	1	2002–2130	1.48	<0.0001	Packaging-associated(5, 6, 21) – but longer than previously described region; conserved cRNA structure(18)
PA-X	2*	10–72	0.99 (0.82)	0.0232 (0.9949)	Packaging-associated(6, 22)
	1	559–572, 574–700	1.12	0.047	Frameshift and overlap PA
HA	1	1612–1692	2.63	<0.0001	Packaging-associated(8, 9, 27)
NP	2	4–57	1.29	0.0056	Packaging-associated(3, 28, 29); conserved cRNA structure(18)
	1	1357–1494	1.62	<0.0001	Packaging-associated(28–31); conserved RNA structure(3, 31)
NA	1	1318–1380	1.40	0.0077	Packaging-associated(32, 33); conserved cRNA structure(18)
M1	2	4–69	0.99	0.0004	Packaging-associated(7, 16); M2 splice donor
	3	115–177	0.89	0.0001	m4 splice junction(17); conserved RNA structure(3, 14, 15)
	1	190–231	1.04	<0.0001	Conserved RNA structure(18)
	4	364–393	0.89	0.0001	Conserved cRNA structure(18)
	5*	682–717	1.01 (0.96)	0.0387 (0.0853)	Structured region 5' to M2 splice acceptor(34–36)
M2	1*	938–970	0.44 (0.38)	0.0004 (0.1164)	Packaging-associated(7, 16); conserved cRNA structure(18)
NS1	1	490–564	1.25	0.002	Splice acceptor; conformationally important region(20); overlapping ORFs
NS2	3*	19–30, 503–508	1.10 (0.71)	0.0152 (0.0639)	Splice donor/acceptor
	1*	515–613	0.68 (0.49)	0.0002 (0.0728)	Conformationally important region(20); overlapping ORFs
	4*	623–718	0.80	0.0417	?Artefact; some overlapping ORFs
	2*	748–835	0.70	<0.0001	Packaging-associated(11)