

TECHNICAL DOCUMENT

Community Network of Reference Laboratories (CNRL) for Human Influenza in Europe

Influenza virus characterisation

Summary Europe, April 2010

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Viruses collected in Europe between January and March and characterised by the London-based WHO Collaborating Centre (WHO CC) for Reference and Research on Influenza have all been pandemic A(H1N1) 2009.

Table 1 shows the number of pandemic influenza A(H1N1) clinical samples and virus isolates from Europe received at the WHO CC and collected between January and April 2010. Note: only representative clinical samples were taken for virus recovery in tissue culture, the choice being based in part on qRT/PCR Ct values.

Table 1. Summary of H1N1 pandemic analyses conducted on samples collected January to April 2010.

Country	Clinical samples received		Isolates received	
	Number received	Number grown	Number received	Number grown
JANUARY				
Belgium	11	10		
Cyprus	4	3		
Czech Republic			1	1
Georgia	3	2		
Greece	2	1		
Latvia			6	6
Luxembourg			15	15
Macedonia	9			
Moldova	98	8		
Portugal			5	5
Tajikistan	20			
FEBRUARY				
Belgium	7	6		
Georgia	1	1		
Greece	20	6	4	4
Luxembourg			2	2
Moldova	33	1		
Tajikistan	13	1	1	1
MARCH				
Moldova	2			
TOTAL	223	39	34	34

All propagated viruses have been antigenically characterised by HI assay.

Antigenic analysis of a representative set of European viruses, including viruses recovered from specimens collected in January and February 2010, is shown in Table 2. The table shows haemagglutination inhibition (HI) results using a panel of reference post-infection ferret antisera. As described in the previous ECDC Virus Characterisation reports ([January 2010](#), [February 2010](#), [March 2010](#)) the viruses continue to react well with ferret antisera raised against the reference pandemic A(H1N1) viruses. A minority of the viruses show a 4-fold reduction in titre with the antiserum raised against the vaccine virus A/California/7/2009 but these are not considered epidemiologically or antigenically significant. A similar pattern continues to be observed with viruses from countries outside of Europe that have been assayed at the WHO CC in London.

Table 2. Antigenic analyses of pandemic A(H1N1) influenza viruses conducted at WHOCC.

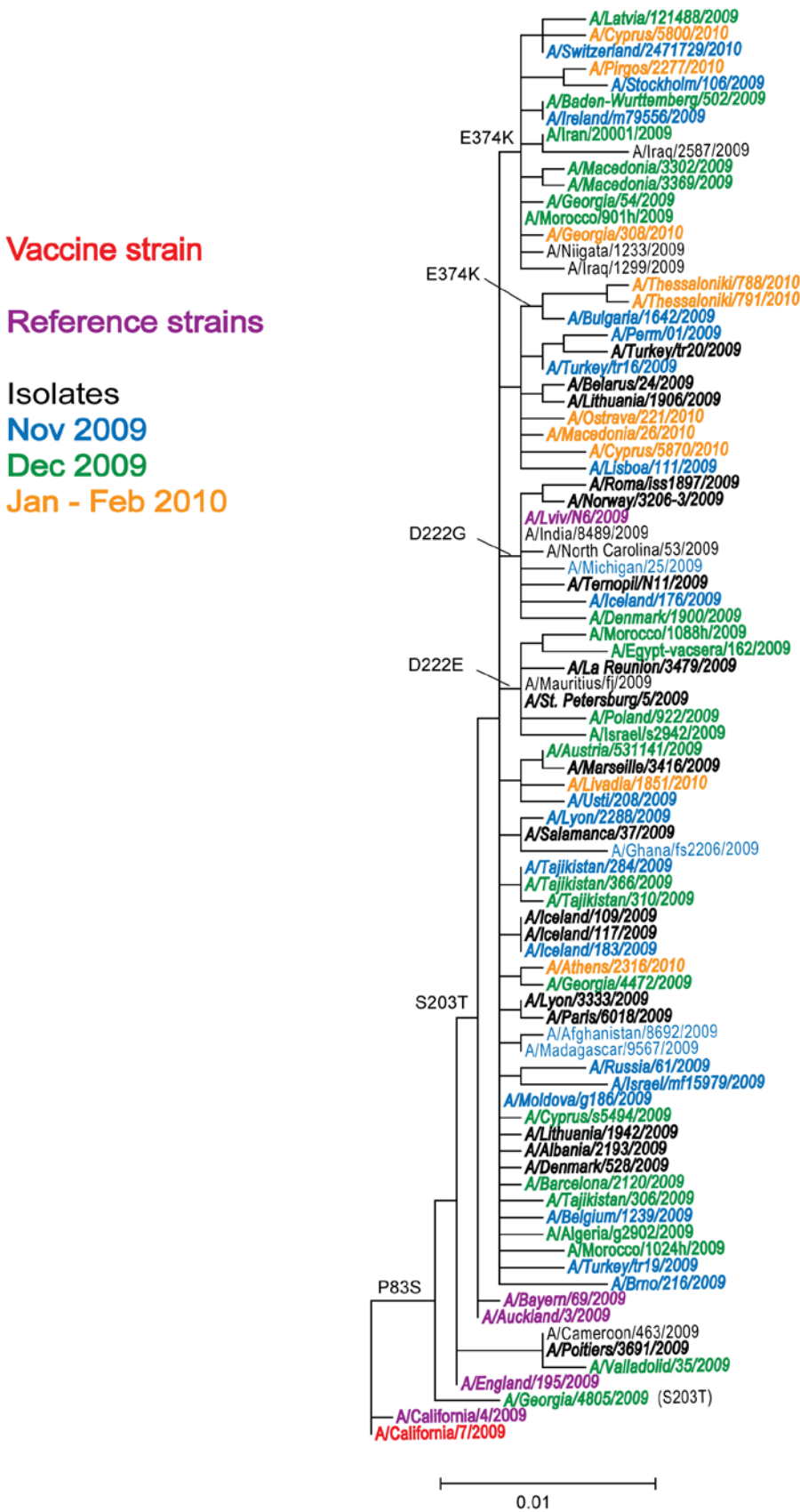
Viruses	Collection date	Passage History	Haemagglutination inhibition titre ¹					
			Post infection ferret sera					
			A/Cal 4/09 C4/F14/09	A/Cal 7/09 C4/31/09 NIBSC	A/Eng 195/09 F17/09	A/Auck 3/09 C4/17/09	A/Bayern 69/09 C4/33/09	A/Lviv N6/2009 C4/34/09
REFERENCE VIRUSES								
A/California/4/2009		C1,E2	2560	2560	2560	5120	2560	5120
A/California/7/2009		E6	5120	2560	2560	5120	1280	5120
A/England/195/2009		MDCK3/SIAT1	1280	1280	1280	2560	640	1280
A/Auckland/3/2009		Ex+3	5120	5120	2560	5120	2560	5120
A/Bayern/69/2009		MDCK4/SIAT1	80	320	40	80	640	320
A/Lviv/N6/2009		MDCK4/SIAT1	640	1280	160	320	2560	2560
TEST VIRUSES								
A/Athens/2243/2010	04/02/2010	SIAT2	2560	2560	1280	2560	1280	2560
A/Belgium/G132/2010	11/01/2010	SIAT4	2560	2560	2560	2560	1280	2560
A/Belgium/G161/2010	04/02/2010	SIAT4	1280	1280	1280	2560	1280	1280
A/Belgium/G82/2010	21/01/2010	SIAT3	1280	1280	1280	1280	1280	1280
A/Bulgaria/1325/2009	03/11/2009	SIAT2	2560	5120	1280	5120	2560	2560
A/Georgia/4032/2009	09/12/2009	SIAT3	1280	1280	640	2560	640	1280
A/Georgia/4802/2009	27/12/2009	SIAT2	640	1280	640	2560	640	1280
A/Georgia/670/2010	04/02/2010	SIAT3	320	640	160	320	640	1280
A/Latvia/1-1346/2010	27/01/2010	MDCKx / SIAT1	1280	2560	1280	2560	1280	1280
A/Latvia/1-1403/2010	28/01/2010	MDCKx / SIAT1	1280	1280	1280	2560	1280	2560
A/Latvia/2-37/2010	29/01/2010	MDCKx / SIAT1	2560	1280	1280	2560	1280	2560
A/Lisboa/2/2010	04/01/2010	MDCK1 / SIAT1	1280	1280	1280	1280	640	1280
A/Lisboa/3/2010	18/01/2010	MDCK1 / SIAT1	640	640	1280	1280	640	640
A/Lisboa/4/2010	06/01/2010	MDCK2 / SIAT1	1280	1280	1280	1280	640	1280
A/Lisboa/5/2010	09/01/2010	MDCK2 / SIAT1	1280	1280	1280	1280	640	1280
A/Livadia/1851/2010	26/01/2010	SIAT3	1280	2560	1280	2560	1280	1280
A/Luxembourg/184/2010	25/01/2010	MDCKx / SIAT1	640	1280	640	1280	640	1280
A/Luxembourg/185/2010	25/01/2010	MDCKx / SIAT1	640	1280	640	2560	640	1280
A/Luxembourg/251/2010	02/02/2010	MDCKx / SIAT1	2560	2560	2560	5120	1280	2560
A/Luxembourg/8/2010	04/01/2010	MDCKx / SIAT1	1280	640	320	1280	640	640
A/Moldova/112/2010	05/01/2010	SIAT2	2560	2560	2560	2560	1280	2560
A/Moldova/63/2010	04/01/2010	SIAT2	2560	5120	2560	2560	2560	2560
A/Moldova/66/2010	03/01/2010	SIAT3	2560	2560	2560	2560	1280	2560
A/Moldova/8/2010	02/01/2010	SIAT3	2560	2560	2560	2560	1280	2560
A/Samos Island/2161/2010	02/02/2010	SIAT3	1280	2560	1280	5120	1280	2560

Vaccine strain

Gene sequence analysis of a subset of recent viruses and clinical specimens shows that circulating viruses continue to remain genetically similar to the prototype and vaccine viruses. Figure 1 shows a phylogenetic tree of the haemagglutinin (HA) gene; viruses from Europe (highlighted in bold-type italic script) are shown compared with viruses from other parts of the world. As described in the previous reports (*ibid*), most viruses carry the amino acid substitution S203T in the HA glycoprotein. Additional distinct subsets can also be seen; two carrying a substitution at amino acid 222, and another at position 374 (corresponding to residue 47 of HA2). These substitutions do not significantly affect the antigenicity of the viruses.

No seasonal influenza viruses (H1N1, H3N2, influenza B) of European origin have been received at the WHO CC in London since the [February 2010](#) report.

Figure 1. Phylogenetic comparison of pandemic A(H1N1) HA genes



The phylogenetic tree was constructed using maximum parsimony in PAUP (Sinauer Associates). The bar indicates the proportion of nucleotide changes in the sequence. Reference strains are viruses to which post-infection antisera have been developed. The colours indicate the date of sample collection. Isolates from Europe are in italics.