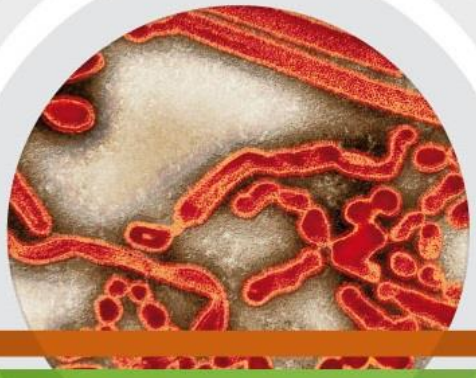


SURVEILLANCE REPORT

Influenza virus characterisation

Summary Europe, May 2016

Summary

From week 40/2015, the start of weekly reporting on influenza activity in the WHO European Region, to week 20/2016 over 138 000 influenza detections across the Region have been reported. Influenza type A viruses have prevailed over type B but, unlike the situation in the 2014–15 season, A(H1N1)pdm09 viruses greatly outnumbered A(H3N2) and the proportion of B/Victoria-lineage detections has risen substantially, representing ~92% of those assigned to a lineage.

To date, 26 EU/EEA countries have shared 631 influenza-positive specimens with the Francis Crick Institute, London, for detailed characterisation: one additional country and 31 specimens since the April 2016 report. Since the latter report, 75 viruses have been characterised antigenically, and genetic analyses are ongoing.

All of 49 A(H1N1)pdm09 viruses characterised antigenically were similar to the vaccine virus A/California/7/2009. Worldwide new genetic subclusters of viruses within the 6B clade have emerged, with two being designated as subclades: 6B.1 defined by HA1 amino acid substitutions S162N and I216T and 6B.2 defined by HA1 amino acid substitutions V152T and V173I. Of the 246 viruses characterised genetically for the 2015–16 season, 25 (10%) were clade 6B, 214 (87%) were subclade 6B.1 and seven (3%) were subclade 6B.2.

The three A(H3N2) test viruses characterised by haemagglutination inhibition (HI) assay were poorly recognised by reference antiserum raised against egg-propagated A/Switzerland/9715293/2013, the vaccine virus recommended for use in the 2015–2016 northern hemisphere influenza season. The test viruses were recognised somewhat better by antisera raised against egg-propagated A/Hong Kong/4801/2014, the virus recommended for use in 2016 southern hemisphere and 2016–2017 northern hemisphere influenza vaccines. Of 65 A(H3N2) viruses characterised genetically for the 2015–2016 season, one (1%) was clade 3C.3, 40 (62%) were subclade 3C.2a and 24 (37%) were subclade 3C.3a.

The 22 B/Victoria-lineage viruses were antigenically similar to tissue culture-propagated surrogates of B/Brisbane/60/2008. All 78 viruses characterised genetically for the 2015–2016 season fell in genetic clade 1A, as do recently collected viruses worldwide.

One B/Yamagata virus have been characterised since the previous report; it reacted well with post-infection ferret antiserum raised against egg-propagated B/Phuket/3073/2013, the recommended vaccine virus for the northern hemisphere 2015–16 influenza season and for quadrivalent vaccines in the 2016 southern hemisphere and 2016–17 northern hemisphere seasons. All 16 viruses characterised genetically for the 2015–2016 season fell in genetic clade 3.

This report was prepared by Rod Daniels, Vicki Gregory, Burcu Ermetal, Aine Rattigan and John McCauley on behalf of the European Reference Laboratory Network for Human Influenza (ERLI-Net), under contract to the European Centre for Disease Prevention and Control (ECDC).

Suggested citation: European Centre for Disease Prevention and Control. Influenza virus characterisation, summary Europe, May 2016. Stockholm: ECDC; 2016.

Table 1 shows a summary of influenza virus detections in the WHO European Region reported to The European Surveillance System (TESSy) for the first 34 weeks (weeks 40/2015–20/2016) of reporting for the 2015–16 season. A total of over 138 000 detections had been made with type A viruses prevailing over type B at a ratio of 2.3:1; this compares to a ratio of 5.8:1 to week 7/2016 indicating a surge in influenza type B circulation over the last 13 weeks. So far, of the type A viruses subtyped (n = 66 707) and the type B viruses ascribed to lineage (n = 7 834), A(H1N1)pdm09 have prevailed over A(H3N2) and B/Victoria over B/Yamagata by ratios of 10.2:1 and 11.1:1, respectively.

Since the start of weekly reporting for the 2015–16 influenza season (week 40/2015), 47 shipments of specimens have been received at the Crick Worldwide Influenza Centre (WIC), from 26 countries in the EU/EEA (Table 2). Of the 631 specimens received – a mix of clinical samples and virus isolates – the majority (79%) were type A viruses, and A(H1N1)pdm09 outnumbered A(H3N2) at a ratio of 5.2:1. Of the 134 type B specimens received (21% of the specimens), 114 were B/Victoria-lineage and 16 B/Yamagata-lineage. A number of specimens are still in the process of being characterised. The antigenic and genetic properties of influenza virus isolates characterised since the April 2016 report[1] are presented and discussed in this report.

Table 1. Influenza virus detections in the WHO European Region since the start of reporting for the 2015–16 season (weeks 40/2015–20/2016)[a]

	Cumulative number of detections			Totals*	
Virus type/subtype	Sentinel sources	Non-sentinel sources	Totals	%	Ratios
Influenza A	**10496**	**85919**	**96415**	**69.7**	**2.3:1**
A(H1N1)pdm09	8665	52083	60748	91.1	10.2:1
A(H3N2)	1365	4594	5959	8.9	
A not subtyped	466	29242	29708		
Influenza B	**8144**	**33791**	**41935**	**30.3**	
Victoria lineage	3974	3210	7184	91.7	11.1:1
Yamagata lineage	145	505	650	8.3	
Lineage not ascribed	4025	30076	34101		
Total detections (total tested)	**18 640 (50 861)**	**119 710 (536 625)**	**138 350 (587 486)**		

*** Percentages are shown for total detections (types A & B, and for viruses ascribed to subtype/lineage). Ratios are given for type A:B, A(H1N1)pdm09: A(H3N2) and Victoria:Yamagata lineages.**

[a] There was a data problem in TESSy relating to weeks 16-19/2016 with double entry of data from some countries. It affected sentinel, non/sentinel detections and SARI data. This problem was identified in week 19 and corrected. As a consequence the data included in the April report (to week 17/2016) was incorrect - the numbers given above (to week 20/2016) are correct, but reduced compared to the same table in the April report.

[1] European Centre for Disease Prevention and Control. Influenza virus characterisation, summary Europe, April 2016. Stockholm: ECDC; 2016. Available from: http://ecdc.europa.eu/en/publications/Publications/ERLI-Net%20report%20April%202016.pdf

Table 2. Summary of clinical samples and virus isolates received from EU/EEA Member States: packages received since the start of the 2015–16 reporting period (week 40/2015)

MONTH* / Country	TOTAL RECEIVED	A		H1N1pdm09		H3N2			B		B Victoria lineage		B Yamagata lineage	
		Number received	Number propagated	Number received	Number propagated[1]	Number received	Number propagated[2]		Number received	Number propagated	Number received	Number propagated[1]	Number received	Number propagated[1]
2015														
SEPTEMBER														
Iceland	1			1	1									
Poland	1					1	0	0						
Spain	1			1	1									
Sweden	1			1	1									
United Kingdom	3					3	3	0						
OCTOBER														
Austria	1			1	1									
Belgium	1			1	1									
France	2			2	2									
Germany	2			1	1								1	1
Italy	2			1	1								1	1
Latvia	1			1	1									
Netherlands	1			1	1									
Norway	11			8	6	2	2	0			1	1		
Portugal	1			1	1									
Romania	1			1	1									
Slovenia	2	1	0	1	1									
Spain	2			2	1									
Sweden	1					1	0	1						
United Kingdom	8			7	7						1	1		
NOVEMBER														
Austria	9			5	5	4	4	0						
Belgium	9			2	1	1	0	1			6	6		
Denmark	1			1	1									
Estonia	3			3	0									
Finland	4			2	2	2	1	1						
France	5			1	1	1	1	0			3	3		
Germany	14			11	11	2	0	2			1	1		
Italy	2					1	1	0			1	1		
Netherlands	7			5	5						2	2		
Norway	9			7	6	1	1	0			1	1		
Poland	1	1	0											
Portugal	8			8	7									
Slovenia	3					2	0	2	1	0				
Spain	5	2	0	2	1	1	1	0						
Sweden	11			7	7	4	4	0						
United Kingdom	7			7	5									
DECEMBER														
Austria	3					3	2	1						
Czech Republic	2			2	2									
Denmark	7			7	7									
Estonia	6			1	0				1	0	4	3		
Finland	8			7	6	1	1	0						
France	24			7	7	2	2	0			15	15		
Germany	21			17	17	3	3	0			1	1		
Greece	6			5	3	1	0	1						
Iceland	3			1	1	1	0	1			1	1		
Ireland	4			2	2	1	0	1			1	1		
Italy	8			3	3	5	2	3						
Latvia	4			3	3						1	1		
Netherlands	5			5	5									
Norway	5			3	3	2	1	1						
Poland	12	10	0	1	1						1	0		
Portugal	14			10	7	3	2	1					1	1
Romania	1					1	0	1						
Slovenia	5					5	1	3						
Spain	14			8	7	2	0	1			4	4		
United Kingdom	3			3	3									
2016														
JANUARY														
Bulgaria	18			18	in process									
Cyprus	15			9	5	1	1	0			5	3		
Czech Republic	3			3	3									
Estonia	3			2	0						1	1		
Germany	26			11	11	5	5	0			8	8	2	2
Greece	27			27	17									
Hungary	7			4	4						3	3		
Iceland	6			5	5								1	1
Ireland	10			9	9						1	1		
Italy	1			1	1									
Latvia	8			6	6						2	2		
Netherlands	2			2	2									
Portugal	6			6	6									
Romania	8			7	7	1	0	1						
Slovakia	6			4	in process	1	in process				1	in process		
Slovenia	8			3	3	3	0	3	2	0				
Spain	19			16	15	1	0	1			2	2		
FEBRUARY														
Bulgaria	47			34	33	1	0	1			12	12		
Cyprus	9			8	8						1	1		
Germany	13			6	6	2	2	0					5	5
Greece	4			4	2									
Latvia	2			2	2									
Netherlands	1			1	1									
Romania	6			3	3	2	2	0			1	1		
Slovakia	9			3	in process	1	in process				5	in process		
Spain	17			15	14						2	2		
MARCH														
Bulgaria	16			6	6	2	0	2			8	8		
Germany	9					1	0	1			7	7	1	1
Romania	9			6	6	1	1	0			1	1	1	1
Slovakia	10			3	in process						5	in process	2	in process
APRIL														
Romania	8			3	3						5	5		
Slovakia	2			1	in process								1	in process
	631	14	0	405	335	78	43	30	4	0	114	99	16	13
					64.2%		12.4%					18.1%		2.5%
26 Countries					78.8%							21.2%		

*** Month indicates the months in which the clinical specimens were collected**

1. Propagated to sufficient titre to perform HI assay

2. Propagated to sufficient titre to perform HI assay in presence of 20nM oseltamivir; numbers in red indicate viruses recovered but with insufficient HA titre to permit HI assay

Influenza A(H1N1)pdm09 virus analyses

Haemagglutination inhibition (HI) analyses of viruses that have been performed since the April 2016 report are shown in Tables 3-1 to 3-3. All of the 49 A(H1N1)pdm09 viruses from EU/EEA countries characterised antigenically were similar to the vaccine virus, A/California/7/2009. Generally, the test viruses were recognised by the panel of antisera at titres within fourfold of the titres for the homologous viruses, with the exception of the antiserum raised against A/Christchurch/16/2010. This antiserum recognised only 3/49 (6%) test viruses at a titre within fourfold of the titre for the homologous virus. In addition, antiserum raised against A/Lviv/N6/2009 showed eightfold reduced titres with five of the test viruses, compared with the homologous titres. Reference viruses carrying HA1 G155E amino acid substitutions, A/Bayern/69/2009 and A/Lviv/N6/2009, showed reduced recognition by the antisera raised against A/California/7/2009 and reference viruses in genetic clades 4, 5, 6, 7 and subclades 6A, 6B, 6B.1 and 6B.2.

While sequencing is ongoing for many of the test viruses indicated in Tables 3-1 to 3-3, the 27 viruses characterised genetically all fell in subclade 6B.1 (Tables 3-1 & 3-2). Since 2009, the HA genes have evolved, and nine clades have been designated. For well over a year, viruses in clade 6, represented by A/St Petersburg/27/2011 and carrying amino acid substitutions of **D97N**, **S185T** and **S203T** in **HA1** and **E47K** and **S124N** in **HA2** compared with A/California/7/2009, have predominated worldwide with a number of subclades emerging. All EU/EEA viruses characterised since the September 2014 report[2] carry HA genes in subclade 6B, which is characterised by additional amino acid substitutions of **K163Q, A256T** and **K283E** in **HA1** and **E172K** in **HA2** compared with A/California/7/2009, e.g. A/South Africa/3626/2013. A number of virus clusters have emerged within clade 6B, and two of these have been designated as subclades: viruses in subclade 6B.1 are defined by **HA1** amino acid substitutions **S84N, S162N** (which results in the formation of a new potential glycosylation motif at residues 162-164 of HA1) and **I216T**, while those in subclade 6B.2 are defined by **HA1** amino acid substitutions **V152T** and **V173I** (Figure 1).

[2] European Centre for Disease Prevention and Control. Influenza virus characterisation, summary Europe, September 2014. Stockholm: ECDC; 2014. Available from: http://ecdc.europa.eu/en/publications/Publications/Influenza-ERLI-Net-report-Sept-2014.pdf

Table 3-1. Antigenic analysis of A(H1N1)pdm09 viruses by HI

Viruses	Other information	Collection date	Passage history	Haemagglutination inhibition titre												
				Post-infection ferret antisera												
				A/Cal	A/Bayern	A/Lviv	A/Chch	A/Astrak	A/St. P	A/St. P	A/HK	A/Sth Afr	A/Slov	A/Israel		
				7/09	69/09	N6/09	16/10	1/11	27/11	100/11	5659/12	3626/13	2903/2015	Q-504/15		
				Egg	MDCK	MDCK	Egg	MDCK	Egg	Egg	MDCK	Egg	Egg	MDCK		
	Passage history			F06/16[-1]	F09/15[-1]	F14/13[-1]	F15/14[-1]	F22/13[-1]	F26/14[+1]	F24/11[+1]	F30/12[-1]	F03/14[+1]	F02/16[-2]	F08/16[-2]		
	Ferret number						4	5	6	7	6A	6B	6B.1	6B.2		
	Genetic group														G155E	G155E>G, D222G
REFERENCE VIRUSES																
A/California/7/2009	clone 38-32	2009-04-09	E3/E2	2560	1280	640	640	2560	640	2560	2560	1280	2560	2560		
A/Bayern/69/2009		2009-07-01	MDCK5/MDCK1	80	320	320	80	40	40	80	40	80	40	40		
A/Lviv/N6/2009		2009-10-27	MDCK4/SIAT1/MDCK3	160	1280	640	320	160	80	160	320	160	320	160		
A/Christchurch/16/2010	4	2010-07-12	E1/E3	1280	1280	1280	5120	2560	640	2560	1280	1280	2560	1280		
A/Astrakhan/1/2011	5	2011-02-28	MDCK1/MDCK5	1280	640	320	640	1280	640	2560	1280	1280	1280	1280		
A/St. Petersburg/27/2011	6	2011-02-14	E1/E4	1280	640	320	640	1280	640	2560	1280	1280	2560	1280		
A/St. Petersburg/100/2011	7	2011-03-14	E1/E4	1280	1280	640	640	1280	640	2560	1280	640	1280	1280		
A/Hong Kong/5659/2012	6A	2012-05-21	MDCK4/MDCK2	640	320	160	160	320	160	1280	640	320	640	320		
A/South Africa/3626/2013	6B	2013-06-06	E1/E3	640	320	320	640	640	320	1280	640	640	640	640		
A/Slovenia/2903/2015	clone 37 6B.1	2015-10-26	E4/E1	2560	640	640	1280	1280	640	5120	2560	1280	2560	1280		
A/Israel/Q-504/2015	6B.2	2015-12-15	C1/MDCK2	1280	640	320	640	1280	640	2560	1280	1280	2560	2560		
TEST VIRUSES																
A/Bulgaria/404/2016	6B.1	2016-02-09	SIAT2/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	2560		
A/Bulgaria/359/2016	6B.1	2016-02-09	SIAT1/MDCK1	1280	640	320	640	640	640	1280	1280	1280	2560	1280		
A/Bulgaria/333/2016	6B.1	2016-02-09	SIAT2/MDCK1	1280	640	320	640	640	640	1280	1280	1280	2560	2560		
A/Bulgaria/332/2016	6B.1	2016-02-09	SIAT2/MDCK1	1280	640	640	1280	1280	640	2560	2560	1280	2560	2560		
A/Bulgaria/381/2016	6B.1	2016-02-10	SIAT1/MDCK1	1280	640	320	640	640	320	1280	1280	1280	1280	1280		
A/Bulgaria/507/2016	6B.1	2016-02-13	SIAT1/MDCK1	1280	640	320	640	1280	320	1280	1280	1280	2560	2560		
A/Bulgaria/505/2016	6B.1	2016-02-13	SIAT1/MDCK1	1280	640	320	640	1280	640	1280	1280	1280	2560	2560		
A/Bulgaria/531/2016	6B.1	2016-02-16	SIAT1/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	1280	1280		
A/Bulgaria/559/2016	6B.1	2016-02-17	SIAT2/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	1280	1280		
A/Bulgaria/552/2016	6B.1	2016-02-17	SIAT2/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	1280	1280		
A/Bulgaria/671/2016	6B.1	2016-02-19	SIAT2/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	1280		
A/Bulgaria/670/2016	6B.1	2016-02-19	SIAT2/MDCK1	1280	640	320	640	640	640	2560	1280	1280	2560	1280		
A/Bulgaria/665/2016	6B.1	2016-02-19	SIAT2/MDCK1	1280	640	320	640	640	640	2560	1280	1280	1280	1280		
A/Bulgaria/694/2016	6B.1	2016-02-22	SIAT1/MDCK1	1280	640	320	640	1280	640	1280	2560	1280	2560	2560		
A/Bulgaria/691/2016	6B.1	2016-02-22	SIAT1/MDCK1	640	640	320	320	640	320	1280	1280	640	1280	1280		
A/Bulgaria/811/2016	6B.1	2016-03-04	SIAT2/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	2560		

Vaccine

* Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used)
[1] < = <40; [2] < = <80

Table 3-2. Antigenic analysis of A(H1N1)pdm09 viruses by HI

Viruses	Other information	Collection date	Passage history	Haemagglutination inhibition titre												
				Post-infection ferret antisera												
				A/Cal	A/Bayern	A/Lviv	A/Chch	A/Astrak	A/St. P	A/St. P	A/HK	A/Sth Afr	A/Slov	A/Israel		
				7/09	69/09	N6/09	16/10	1/11	27/11	100/11	5659/12	3626/13	2903/2015	Q-504/15		
				Egg	MDCK	MDCK	Egg	MDCK	Egg	Egg	MDCK	Egg	Egg	MDCK		
	Passage history			F06/16[-1]	F09/15[-1]	F14/13[-1]	F15/14[-1]	F22/13[-1]	F26/14[-1]	F24/11[-1]	F30/12[-1]	F03/14[-1]	F02/16[-2]	F08/16[-2]		
	Ferret number						4	5	6	7	6A	6B	6B.1	6B.2		
	Genetic group															
REFERENCE VIRUSES																
A/California/7/2009	clone 38-32	2009-04-09	E3/E2	1280	1280	640	640	1280	640	2560	2560	1280	2560	2560	G155E	
A/Bayern/69/2009		2009-07-01	MDCK5/MDCK1	40	320	160	80	40	40	40	40	40	40	40		
A/Lviv/N6/2009		2009-10-27	MDCK4/SIAT1/MDCK3	160	1280	1280	320	160	160	160	320	160	320	160	G155E>G, D222G	
A/Christchurch/16/2010	4	2010-07-12	E1/E3	1280	2560	2560	5120	2560	1280	5120	2560	2560	2560	2560		
A/Astrakhan/1/2011	5	2011-02-28	MDCK1/MDCK5	1280	640	640	640	1280	640	2560	2560	1280	2560	1280		
A/St. Petersburg/27/2011	6	2011-02-14	E1/E4	1280	640	640	640	1280	640	2560	1280	1280	2560	1280		
A/St. Petersburg/100/2011	7	2011-03-14	E1/E4	1280	640	640	640	1280	640	2560	1280	640	2560	1280		
A/Hong Kong/5659/2012	6A	2012-05-21	MDCK4/MDCK2	640	320	160	320	640	320	1280	1280	640	1280	640		
A/South Africa/3626/2013	6B	2013-06-06	E1/E3	640	640	640	640	640	640	1280	640	1280	1280	1280		
A/Slovenia/2903/2015	clone 37 6B.1	2015-10-26	E4/E1	1280	1280	640	1280	2560	1280	5120	2560	1280	2560	2560		
A/Israel/Q-504/2015	6B.2	2015-12-15	C1/MDCK2	1280	640	320	640	1280	640	2560	1280	1280	2560	2560		
TEST VIRUSES																
A/Bulgaria/265/2016		2016-02-03	SIAT2/MDCK1	1280	640	320	640	640	320	2560	1280	1280	1280	1280		
A/Bulgaria/301/2016		2016-02-04	SIAT2/MDCK1	1280	640	640	640	1280	640	2560	1280	1280	2560	2560		
A/Bulgaria/276/2016		2016-02-04	SIAT2/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	1280		
A/Bulgaria/314/2016	6B.1	2016-02-05	SIAT2/MDCK1	640	640	320	640	640	640	2560	1280	1280	1280	1280		
A/Bulgaria/312/2016	6B.1	2016-02-05	SIAT1/MDCK1	1280	640	640	640	1280	640	2560	1280	1280	2560	1280		
A/Bulgaria/311/2016	6B.1	2016-02-05	SIAT1/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	1280		
A/Bulgaria/306/2016	6B.1	2016-02-05	SIAT2/MDCK1	640	320	320	640	640	320	1280	1280	640	1280	1280		
A/Bulgaria/349/2016	6B.1	2016-02-07	SIAT2/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	2560		
A/Bulgaria/345/2016	6B.1	2016-02-07	SIAT2/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	1280		
A/Bulgaria/378/2016	6B.1	2016-02-08	SIAT2/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	1280		
A/Bulgaria/363/2016	6B.1	2016-02-08	SIAT1/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	1280		
A/Bulgaria/353/2016	6B.1	2016-02-08	SIAT2/MDCK1	640	640	320	640	640	640	2560	1280	1280	1280	1280		
A/Bulgaria/331/2016	6B.1	2016-02-08	SIAT2/MDCK1	1280	640	320	640	640	640	2560	1280	1280	1280	1280		
A/Bulgaria/329/2016	6B.1	2016-02-08	SIAT1/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	1280		
				Vaccine												

* Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used)
[1] < = <40; [2] < = <80

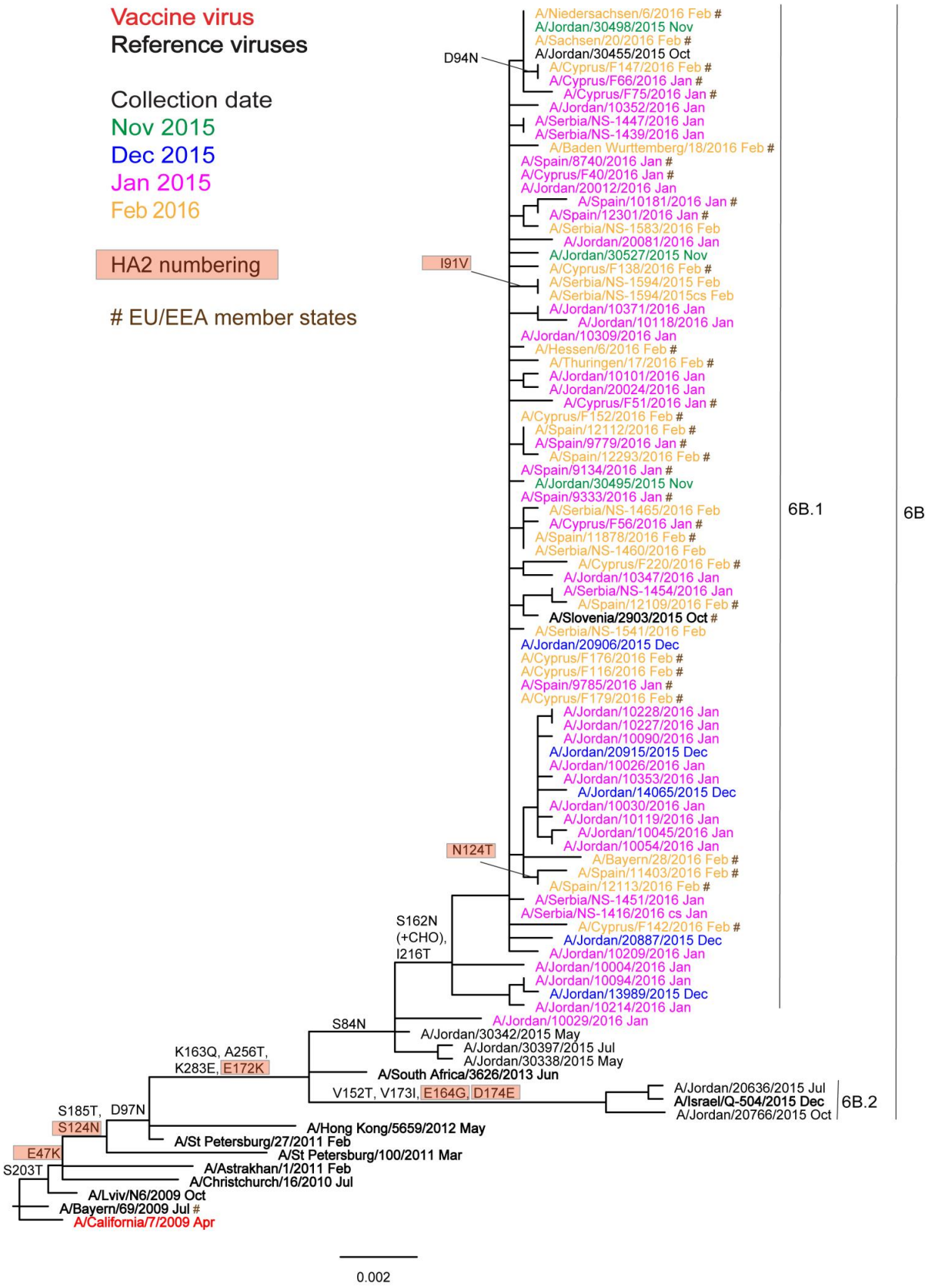
Table 3-3. Antigenic analysis of A(H1N1)pdm09 viruses by HI

Viruses	Other information	Collection date	Passage history	Haemagglutination inhibition titre														
				Post-infection ferret antisera														
				A/Cal	A/Bayern	A/Lviv	A/Chch	A/Astrak	A/St. P	A/St. P	A/HK	A/Sth Afr	A/Slov	A/Israel	A/Ukraine		
				7/09	69/09	N6/09	16/10	1/11	27/11	100/11	5659/12	3626/13	2903/2015	Q-504/15	6907/15		
				Egg	MDCK	MDCK	Egg	MDCK	Egg	Egg	MDCK	Egg	Egg	MDCK	MDCK		
	Passage history			F06/16[1]	F09/15[1]	F14/13[1]	F15/14[1]	F22/13[1]	F26/14[1]	F24/11[1]	F30/12[1]	F03/14[1]	F02/16[2]	F08/16[2]	F-17/16[2]		
	Ferret number						4	5	6	7	6A	6B	6B.1	6B.2	6B.2		
	Genetic group														New		
REFERENCE VIRUSES																	
A/California/7/2009 Clone38-32		2009-04-09	E3/E3	1280	640	640	640	1280	640	2560	1280	1280	2560	2560	640	G155E	
A/Bayern/69/2009		2009-07-01	MDCK5/MDCK1	40	640	320	80	40	40	40	40	80	80	40	<		
A/Lviv/N6/2009		2009-10-27	MDCK4/SIAT1/MDCK3	160	1280	1280	320	160	160	160	320	160	320	160	80	G155E>G, D222G	
A/Christchurch/16/2010	4	2010-07-12	E1/E3	1280	2560	2560	5120	2560	1280	5120	2560	2560	5120	2560	640		
A/Astrakhan/1/2011	5	2011-02-28	MDCK1/MDCK5	1280	1280	640	1280	2560	1280	5120	2560	2560	2560	2560	640		
A/St. Petersburg/27/2011	6	2011-02-14	E1/E4	1280	640	640	640	1280	640	2560	1280	1280	2560	1280	640		
A/St. Petersburg/100/2011	7	2011-03-14	E1/E4	1280	1280	1280	640	2560	1280	5120	2560	1280	2560	2560	640		
A/Hong Kong/5659/2012	6A	2012-05-21	MDCK4/MDCK2	640	320	160	320	640	320	1280	1280	640	640	640	320		
A/South Africa/3626/2013	6B	2013-06-06	E1/E3	640	640	640	320	640	640	1280	640	1280	640	640	320		
A/Slovenia/2903/2015	clone 37 6B.1	2015-10-26	E4/E1	2560	1280	640	1280	2560	1280	5120	2560	2560	5120	2560	640		
A/Israel/Q-504/2015	6B.2	2015-12-15	C1/MDCK2	1280	640	640	640	1280	1280	5120	2560	1280	2560	2560	640		
A/Ukraine/6907/2015	6B.2	2015-12-16	C2/MDCK2	320	640	320	320	640	640	1280	640	1280	1280	2560	640		
TEST VIRUSES																	
A/Bulgaria/133/2016		2016-01-27	SIAT1/MDCK1	2560	1280	640	1280	2560	1280	5120	2560	2560	5120	2560	1280		
A/Bulgaria/178/2016		2016-01-29	SIAT2/MDCK1	640	640	320	320	640	320	2560	1280	1280	1280	1280	320		
A/Bulgaria/175/2016		2016-01-29	SIAT2/MDCK1	640	320	160	320	640	320	1280	640	640	1280	640	320		
A/Bulgaria/171/2016		2016-01-29	SIAT2/MDCK1	1280	320	160	320	640	320	1280	1280	640	1280	640	320		
A/Bulgaria/213/2016		2016-02-01	SIAT2/MDCK1	640	320	160	320	640	320	2560	1280	640	1280	1280	320		
A/Bulgaria/205/2016		2016-02-01	SIAT2/MDCK1	1280	1280	640	640	1280	640	2560	2560	1280	2560	2560	640		
A/Bulgaria/242/2016		2016-02-02	SIAT2/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	1280	640		
A/Sibiu/190931/2016		2016-02-02	MDCK1/MDCK1	1280	640	320	640	640	320	1280	640	640	1280	1280	640		
A/Bucuresti/191223/2016		2016-02-06	MDCK1/MDCK1	1280	640	320	640	640	320	1280	640	640	1280	2560	640		
A/Dolj/193185/2016		2016-02-29	MDCK1/MDCK1	2560	1280	640	1280	2560	1280	5120	2560	2560	5120	2560	1280		
A/Iasi/193746/2016		2016-03-09	MDCK1/MDCK1	320	320	160	160	320	320	640	320	640	640	1280	640		
A/Iasi/193859/2016		2016-03-10	MDCK1/MDCK1	640	320	160	320	640	320	1280	1280	640	1280	1280	320		
A/Iasi/193967/2016		2016-03-11	MDCK1/MDCK1	640	320	320	320	640	640	1280	1280	1280	1280	1280	640		
A/Bacau/194305/2016		2016-03-15	MDCK1/MDCK1	1280	1280	640	640	1280	640	2560	2560	1280	2560	2560	640		
A/Suceava/194312/2016		2016-03-16	MDCK1/MDCK1	1280	640	640	640	1280	640	2560	1280	1280	2560	1280	640		
A/Botosani/195345/2016		2016-03-29	MDCK1/MDCK1	640	320	320	320	640	320	1280	1280	640	1280	640	ND		
A/Iasi/195762/2016		2016-04-06	MDCK1/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	1280	ND		
A/Iasi/196079/2016		2016-04-11	MDCK1/MDCK1	640	640	320	640	640	320	2560	1280	640	1280	640	ND		
A/Dolj/196287/2016		2016-04-11	MDCK1/MDCK1	1280	640	320	640	1280	640	2560	1280	1280	2560	1280	ND		
																Vaccine	

* Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used)

[1] < = <40; [2] < = <80; ND = Not Done

Figure 1. Phylogenetic comparison of influenza A(H1N1)pdm09 HA genes



Influenza A(H3N2) virus analyses

As described in many previous reports[3], influenza A(H3N2) viruses continue to be difficult to characterise antigenically by HI assay due to variable agglutination of red blood cells (RBCs) from guinea pigs, turkeys and humans or the loss of the ability of viruses to agglutinate any of the RBCs. This is a particular problem for most viruses that fall in genetic subgroup 3C.2a, as was highlighted first in the November 2014 report[4].

Results of HI tests performed with guinea pig RBCs in the presence of 20nM oseltamivir, added to circumvent NA-mediated binding of A(H3N2) viruses to the RBCs, are shown in Table 4. Three test viruses, from Romania, retained sufficient HA titre to be analysed by HI assay; genetic characterisation of these is ongoing.

The test viruses, propagated in MDCK-SIAT1 cells, reacted poorly in HI assays with the panel of post-infection ferret antisera relative to the titres of the antisera with their respective homologous viruses (shown in red: Table 4). However, the antisera raised against A/Stockholm/6/2014 (3C.3a: tissue culture- and egg-propagated), tissue culture-propagated A/Hong Kong/5738/2014 (3C.2a), tissue culture-propagated A/Georgia/532/2015 and A/Hong Kong/4801/2014 (3C.2a: tissue culture- and egg-propagated), gave reactivity with all test viruses. The antiserum raised against egg-propagated A/Switzerland/9715293/2013 (3C.3a), the northern hemisphere 2015–16 vaccine component, reacted with test viruses at titres reduced at least eightfold compared to the homologous titre, while antiserum raised against egg-propagated A/Hong Kong/4801/2014, the virus recommended for use in vaccines for the southern hemisphere 2016 and northern hemisphere 2016–17 influenza seasons, yielded titres reduced by two-to-fourfold compared to the homologous titre and gave absolute titres of 80 or greater.

Since 2009, seven genetic groups based on the HA gene have been defined for A(H3N2) viruses. Phylogenetic analysis of the HA genes of representative A(H3N2) viruses with recent collection dates is shown in Figure 2. The HA genes fall within clade 3C. This clade has three subdivisions: 3C.1 (represented by A/Texas/50/2012, the vaccine virus recommended for use in the 2014–15 northern hemisphere season), 3C.2 and 3C.3. Viruses in these three subdivisions had been antigenically similar. In 2014 three new subclades emerged, one in subdivision 3C.2, 3C.2a, and two in 3C.3, 3C.3a and 3C.3b, with subclade 3C.2a viruses dominating in recent months (Figure 2). While viruses in subclades 3C.2a and 3C.3a are antigenic drift variants, those in 3C.3b have remained antigenically similar to previously circulating viruses in the 3C.3 subdivision. Amino acid substitutions that define these subdivisions and subclades are:

- (3C.2) **N145S** in **HA1**, and **D160N** in **HA2**, e.g. A/Hong Kong/146/2013
- (3C.2a) Those in 3C.2 plus **L3I**, **N144S** (resulting in the loss of a potential glycosylation site), **F159Y**, **K160T** (in the majority of viruses, resulting in the gain of a potential glycosylation site), **N225D** and **Q311H** in **HA1**, e.g. A/Hong Kong/5738/2014
- (3C.3) **T128A** (resulting in the loss of a potential glycosylation site), **R142G** and **N145S** in **HA1**, e.g. A/Samara/73/2013
- (3C.3a) those in 3C.3 plus **A138S**, **F159S** and **N225D** in **HA1**, many with **K326R**, e.g. A/Switzerland/9715293/2013
- (3C.3b) those in 3C.3 plus **E62K, K83R, N122D** (resulting in the loss of a potential glycosylation site), **L157S** and **R261Q** in **HA1** with **M18K** in **HA2**, e.g. A/Netherlands/525/2014

Based on results available at the time of the February 2015 vaccine composition meeting showing cross-reactivity of antisera raised against subclade 3C.3a and 3C.2a viruses, but with changes acquired on egg-adaptation of genetic subgroup 3C.2a viruses and, at that time, the lack of a suitable 3C.2a vaccine candidate, the World Health Organization recommendation was to use an A/Switzerland/9715293/2013-like virus as the A(H3N2) component of vaccines for the northern hemisphere 2015–16 influenza season [1]. After February 2015, a new subclade designated 3C.3b emerged, these three subclades being antigenically distinguishable, but subclade 3C.2a viruses became prevalent and have remained so. While ferret antisera raised against 3C.3a and 3C.2a subclade viruses showed some cross-reactivity with viruses in all three subclades, antisera raised against 3C.3b viruses were subclade specific. With the availability of new subclade 3C.2a vaccine candidates and the continued cross-reactivity of antisera raised against viruses in subclades 3C.3a and 3C.2a viruses, the World Health Organization recommendation for the A(H3N2) component of influenza vaccines for the southern hemisphere 2016 [2] and northern hemisphere 2016–17 [3] influenza seasons was for an A/Hong Kong/4801/2014-like (3C.2a) virus.

[3] For example, the September 2013 report: European Centre for Disease Prevention and Control. Influenza virus characterisation, summary Europe, September 2013. Stockholm: ECDC; 2013. Available from: http://www.ecdc.europa.eu/en/publications/Publications/influenza-virus-characterisation-sep-2013.pdf

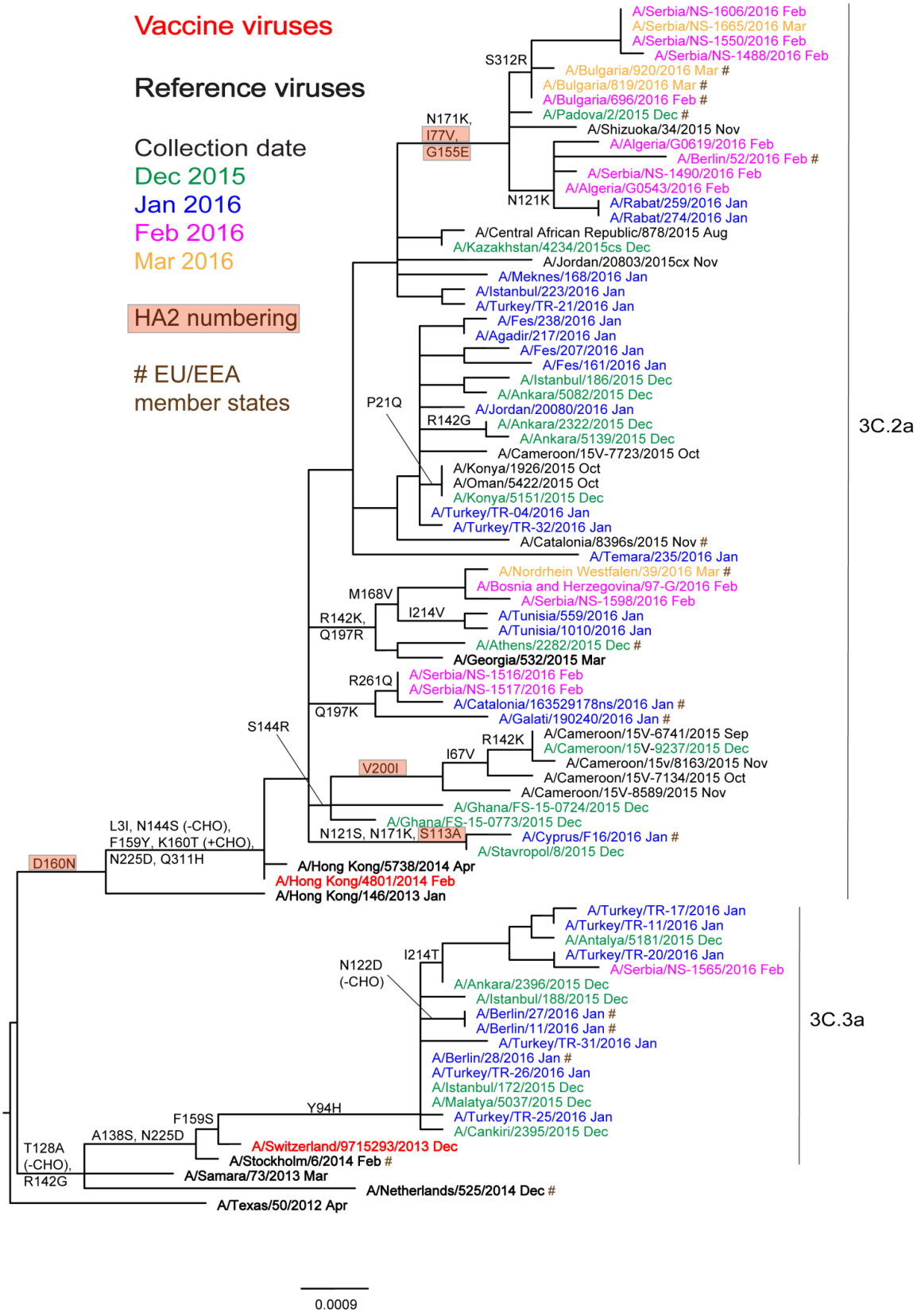
[4] European Centre for Disease Prevention and Control. Influenza virus characterisation, summary Europe, November 2014. Stockholm: ECDC; 2014. Available from: http://www.ecdc.europa.eu/en/publications/Publications/ERLI-Net report November 2014.pdf

Table 4. Antigenic analysis of A(H3N2) viruses by HI (guinea pig RBC with 20nM oseltamivir)

Viruses	Other information	Collection date	Passage history	Haemagglutination inhibition titre													
				Post-infection ferret antisera													
				A/Texas	A/Samara	A/Stock	A/Stock	A/Switz	A/Switz	A/Neth	A/HK	A/HK	A/HK	A/HK	A/HK	A/HK	A/Georgia
				50/12	73/13	6/14	6/14	9715293/13	9715293/13	525/14	146/13	5738/14	4801/14	4801/14	4801/14	4801/14	532/15
				Egg	SIAT	SIAT	Egg	SIAT	Egg	SIAT	Egg	MDCK	MDCK	Egg	Egg	Egg	SIAT
	Passage history			F36/12[-1]	F35/15[-1]	F14/14[-1]	F20/14[-1]	F18/15[-1]	F32/14[-1]	F23/15[-1]	F10/15[-1]	F30/14[-1]	F43/15[-1]	F12/15[-1]	F12/15[-1]	F12/15[-1]	F33/15[-1]
	Ferret number			3C.1	3C.3	3C.3a	3C.3a	3C.3a	3C.3a	3C.3b	3C.2	3C.2a	3C.2a	3C.2a	3C.2a	3C.2a	3C.2a
	Genetic group																
REFERENCE VIRUSES																	
A/Texas/50/2012	3C.1	2012-04-15	E5/E2	1280	640	80	640	40	320	320	320	160	320	80	80	160	160
A/Samara/73/2013	3C.3	2013-03-12	C1/SIAT3	640	640	640	320	80	320	320	320	320	640	160	160	160	320
A/Stockholm/6/2014	3C.3a	2014-02-06	SIAT1/SIAT2	80	80	320	320	160	160	80	160	160	320	160	160	160	160
A/Stockholm/6/2014 isolate 2	3C.3a	2014-02-06	E4/E1	320	80	160	320	80	640	80	40	160	320	40	40	40	80
A/Switzerland/9715293/2013	3C.3a	2013-12-06	SIAT1/SIAT2	<	40	320	160	80	80	40	40	80	80	80	80	80	80
A/Switzerland/9715293/2013 clone 123	3C.3a	2013-12-06	E4/E1	160	160	160	320	80	640	40	80	160	160	320	320	80	160
A/Netherlands/525/2014	3C.3b	2014-12-17	SIAT2/SIAT4	320	160	160	160	40	80	640	160	80	160	80	80	80	160
A/Hong Kong/146/2013	3C.2	2013-01-11	E6	1280	640	80	320	40	640	320	640	320	640	160	160	160	320
A/Hong Kong/5738/2014	3C.2a	2014-04-30	MDCK1/MDCK2/SIAT2	40	80	160	80	40	80	40	80	160	320	160	160	160	160
A/Hong Kong/4801/2014	3C.2a	2014-02-26	MDCK4/MDCK2	40	40	160	80	40	40	<	80	160	320	160	160	160	160
A/Hong Kong/4801/2014 isolate 1	3C.2a	2014-02-26	E6/E2	<	80	80	40	40	40	40	<	160	320	320	320	320	320
A/Georgia/532/2015 plaq 20	3C.2a	2015-03-09	SIAT1/SIAT3	80	80	160	80	40	80	<	80	160	320	160	160	160	640
TEST VIRUSES																	
A/Bucuresti/191366/2016		2016-02-10	SIAT1/SIAT1	<	<	160	160	40	40	40	40	80	80	80	80	80	80
A/Arges/1480-3059/2016		2016-02-10	SIAT1/SIAT1	40	40	320	160	80	80	40	80	160	160	160	160	160	160
A/Bucuresti/193605/2016		2016-03-08	SIAT1/SIAT1	<	40	320	160	80	80	40	80	160	80	80	80	80	80
								Vaccine NH 2015-16						Vaccine SH 2016 NH 2016-17			

* Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used) [1] < = <40

Figure 2. Phylogenetic comparison of influenza A(H3N2) HA genes



Influenza B virus analyses

EU/EEA countries have provided 134 influenza type B viruses, of which 130 were ascribed to a lineage: 114 B/Victoria-lineage and 16 B/Yamagata-lineage (Table 2).

Influenza B – Victoria lineage

Since the April 2016 report 22 viruses of this lineage from Bulgaria and Romania have been characterised antigenically. HI results are shown in Table 5; genetic characterisation is ongoing.

The test viruses showed similar HI reactivity patterns to those from the 2014–15 influenza season: greater than eightfold reductions in HI titres compared with the titre for the homologous virus with post-infection ferret antisera raised against the recommended vaccine virus for quadrivalent live and inactivated vaccines for the northern hemisphere 2015–2016 influenza season, B/Brisbane/60/2008. Similarly, they were poorly recognised by post-infection ferret antisera raised against the reference viruses propagated in eggs B/Malta/636714/2011, B/Johannesburg/3964/2012 and B/South Australia/81/2012. In contrast, all test viruses showed reactivity within fourfold, the majority within twofold, of the titres for the corresponding homologous viruses with antisera raised against viruses that are considered to be surrogate tissue culture-propagated antigens representing the egg-propagated B/Brisbane/60/2008 prototype virus; these antisera were raised against tissue-culture-propagated viruses B/Hong Kong/514/2009 (clade 1B), B/Ireland/3154/2015 and B/Nordrhein-Westfalen/1/2016 (both clade 1A).

Phylogenetic analysis of the HA gene of representative B/Victoria lineage viruses is shown in Figure 3. Throughout the previous season and this season to date, viruses from Europe and elsewhere have HA genes that fall into the B/Brisbane/60/2008 clade (clade 1A) and remain antigenically similar to the vaccine virus B/Brisbane/60/2008. The great majority of viruses, with collection dates since October 2015, fall in a major subcluster defined by amino acid substitutions I117V, N129D and V146I within clade 1A.

These results, linked with the rise in the proportion of B/Victoria-lineage viruses seen in the 2015 southern hemisphere and 2015–2016 northern hemisphere influenza seasons, support the recommendations made to include B/Brisbane/60/2008 in trivalent influenza vaccines for the southern hemisphere 2016 [2] and northern hemisphere 2016–2017 [3] influenza seasons and in the quadrivalent vaccines.

Influenza B – Yamagata lineage

HI results for one B/Yamagata-lineage test virus analysed since the April 2016 report are shown in Table 6, and genetic characterisation is ongoing.

The homologous titres of the 10 post-infection ferret antisera, shown in red, ranged from 160–640, and the test virus showed reactivity with all 10 (Table 6).

Antisera raised against egg-propagated clade 3 viruses B/Phuket/3073/2013 (the virus recommended for inclusion in trivalent influenza vaccines for the northern hemisphere 2014–2015 season) and B/Hong Kong/3417/2014 recognised the test virus at titres within twofold of their respective homologous titres. The test virus showed fourfold or greater reductions in HI reactivity, compared to homologous titres, with the other eight antisera, including that raised against egg-propagated B/Massachusetts/02/2012, the clade 2 vaccine virus recommended for use in the 2014–15 northern hemisphere influenza season.

Figure 4 shows a phylogenetic analysis of the HA genes of representative B/Yamagata-lineage viruses. Worldwide, the vast majority of HA genes from recently collected viruses have fallen in the B/Wisconsin/1/2010–B/Phuket/3073/2013 clade (clade 3), with the great majority falling in a subgroup defined by HA1 L172Q amino acid substitution. A few viruses, annotated in the phylogenetic tree (e.g. B/Oman/6341/2015), are reassortants carrying NA genes normally associated with the B/Victoria-lineage.

Based on such results, a B/Phuket/3073/2013-like virus has been recommended for inclusion in quadrivalent vaccines for the 2016 southern hemisphere [2] and 2016–2017 northern hemisphere [3] influenza seasons.

Table 5. Antigenic analysis of influenza B/Victoria-lineage viruses by HI

Viruses	Collection date	Passage history	Haemagglutination inhibition titre											
			Post-infection ferret antisera											
			B/Bris	B/Mal	B/Bris	B/Malta	B/Jhb	B/For	B/Sth Aus	B/HK	B/Ireland	B/Nord-West		
			60/08	2506/04	60/08	636714/11	3964/12	V2367/12	81/12	514/09	3154/16	1/16		
		Passage history	Egg	Egg	Egg	Egg	Egg	MDCK	Egg	MDCK	MDCK	MDCK		
		Ferret number	Sh 630[*1]	F37/11[*2]	F22/12[*2]	F29/13[*2]	F01/13[*4]	F04/13[*4]	F41/13[*2]	F09/13[*2]	F15/16[*2]	F16/16[*2]		
		Genetic group	1A		1A	1A	1A	1A	1A	1B	1A	1A		
REFERENCE VIRUSES														
B/Malaysia/2506/2004	2004-12-06	E3/E7	2560	320	20	80	160	40	160	20	<	<		
B/Brisbane/60/2008	2008-08-04	E4/E3	1280	80	160	320	320	320	640	160	80	40		
B/Malta/636714/2011	2011-03-07	E4/E1	1280	40	80	320	320	160	640	80	40	40		
B/Johannesburg/3964/2012	2012-08-03	E1/E2	5120	320	640	1280	1280	1280	1280	320	320	160		
B/FormosaV2367/2012	2012-08-06	MDCK1/MDCK3	1280	20	40	160	160	160	320	80	40	40		
B/South Australia/81/2012	2012-11-28	E4/E2	1280	40	80	160	320	320	640	80	80	40		
B/Hong Kong/514/2009	2009-10-11	MDCK3	160	<	<	40	20	40	40	80	80	80		
B/Ireland/3154/2016	2016-01-14	MDCK1/MDCK1	160	<	<	20	<	40	40	80	80	80		
B/Nordrhein-Westfalen/1/2016	2016-01-04	C2/MDCK1	320	<	<	40	40	80	80	80	160	80		
TEST VIRUSES														
B/Bulgaria/177/2016	2016-02-01	SIAT1/MDCK1	160	<	<	40	40	80	80	40	80	40		
B/Bulgaria/238/2016	2016-02-03	SIAT1/MDCK1	320	<	<	40	40	80	80	80	80	80		
B/Bulgaria/261/2016	2016-02-03	SIAT1/MDCK1	160	<	<	20	20	40	80	80	80	80		
B/Bulgaria/343/2016	2016-02-06	SIAT1/MDCK1	320	<	<	40	40	80	80	80	80	80		
B/Bulgaria/350/2016	2016-02-09	SIAT2/MDCK1	320	<	<	40	40	80	80	80	160	80		
B/Bucuresti/679-C7895/2016	2016-02-11	MDCK1/MDCK1	160	<	<	40	20	80	80	40	40	40		
B/Bulgaria/489/2016	2016-02-12	SIAT2/MDCK1	160	<	<	20	<	40	40	80	160	80		
B/Bulgaria/669/2016	2016-02-19	SIAT2/MDCK1	320	<	<	40	40	80	80	80	160	80		
B/Bulgaria/692/2016	2016-02-22	SIAT2/MDCK1	320	<	<	40	40	40	80	80	80	80		
B/Bulgaria/757/2016	2016-02-25	SIAT2/MDCK1	320	<	<	40	80	80	80	160	160	80		
B/Bulgaria/790/2016	2016-02-29	SIAT2/MDCK1	160	<	<	20	20	40	80	80	80	40		
B/Bulgaria/774/2016	2016-02-29	SIAT2/MDCK1	160	<	<	20	40	40	80	80	160	80		
B/Bulgaria/792/2016	2016-03-01	SIAT2/MDCK1	160	<	<	20	<	40	40	40	80	40		
B/Bulgaria/830/2016	2016-03-09	SIAT2/MDCK1	160	<	<	10	<	40	40	80	80	40		
B/Bulgaria/831/2016	2016-03-10	SIAT2/MDCK1	320	<	<	40	80	80	80	80	160	80		
B/Bulgaria/841/2016	2016-03-10	SIAT2/MDCK1	320	<	<	10	<	40	40	80	80	40		
B/Bucuresti/1949978/2016	2016-03-24	MDCK1/MDCK1	80	<	<	20	10	40	40	40	40	20		
B/Bucuresti/1956675/2016	2016-04-05	MDCK1/MDCK1	160	<	<	20	20	40	80	40	40	40		
B/Mures/195946/2016	2016-04-07	MDCK1/MDCK1	160	<	<	20	<	20	40	80	40	40		
B/Alba/195948/2016	2016-04-07	MDCK1/MDCK1	160	<	<	20	20	40	40	80	40	40		
B/Bucuresti/1960001/2016	2016-04-08	MDCK1/MDCK1	160	<	<	20	20	40	40	80	40	40		
B/Mures/196205/2016	2016-04-10	MDCK1/MDCK1	160	<	<	20	20	20	40	80	80	40		

Vaccine
NH 2015-16[#]
SH 2016
NH 2016-17

* Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used)
1 < = <40; 2 < = <10; 3 hyperimmune sheep serum; 4 < = <20

B/Victoria-lineage virus recommended for use in quadravalent vaccines

Figure 3. Phylogenetic comparison of influenza B/Victoria-lineage HA genes

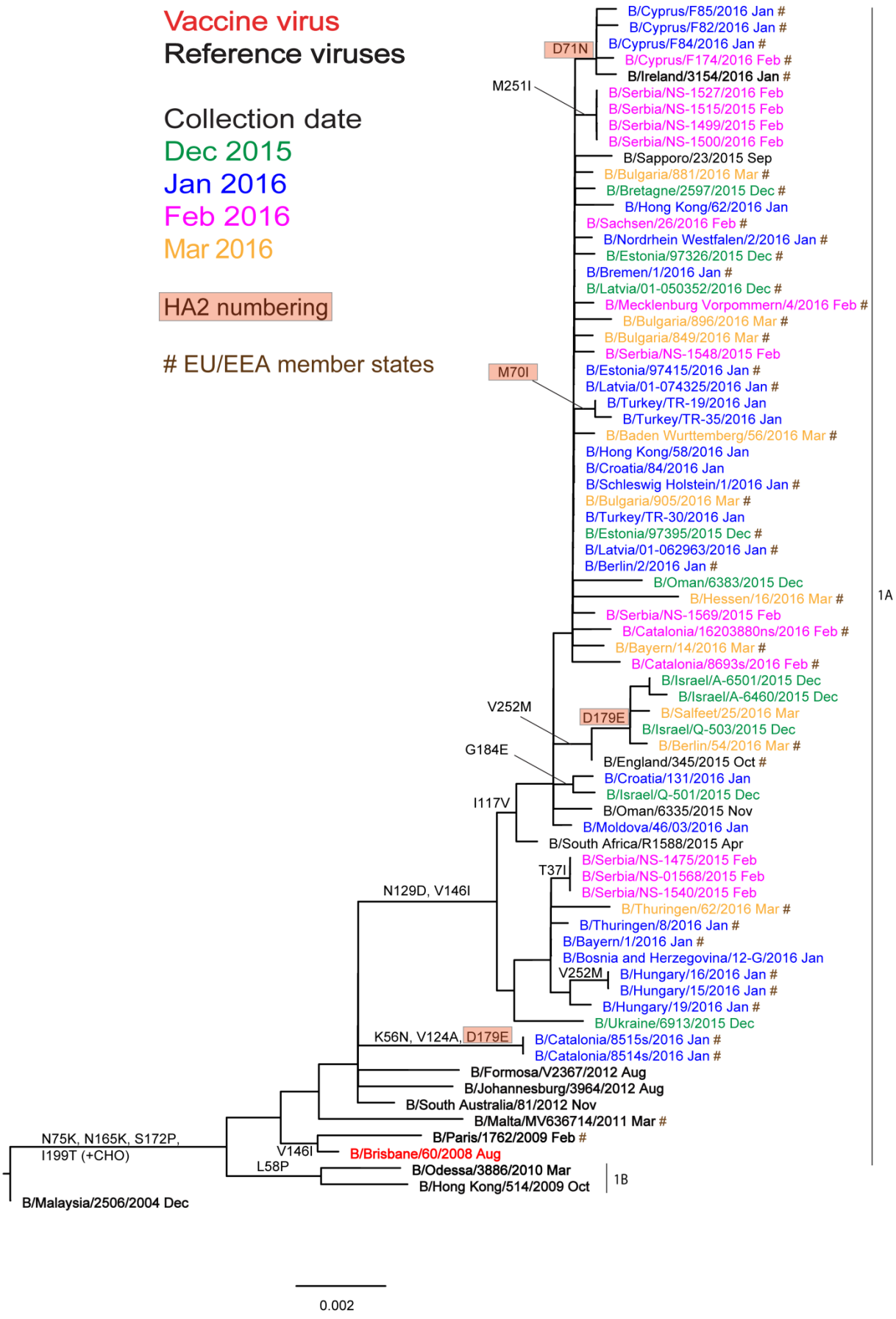


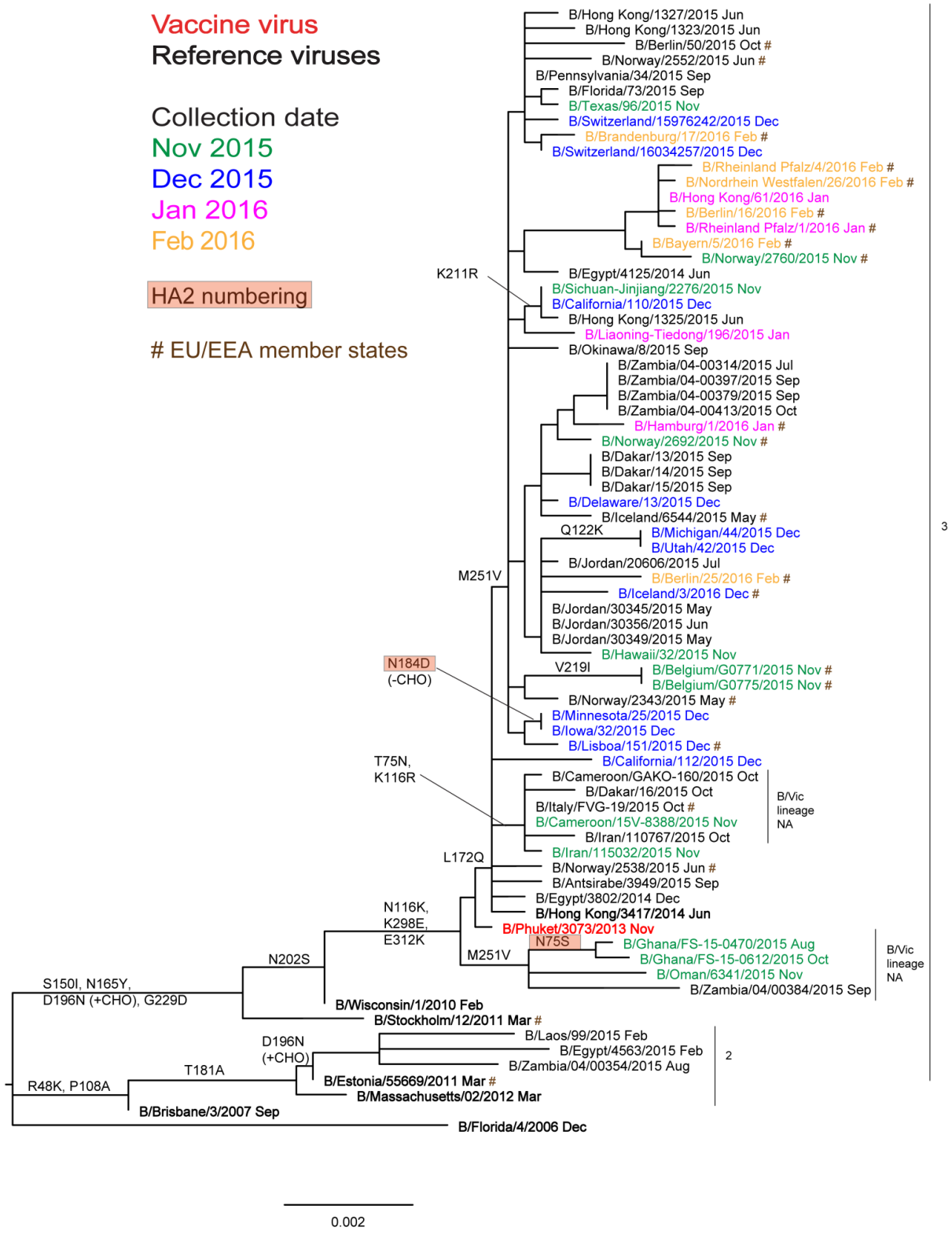
Table 6. Antigenic analysis of influenza B/Yamagata-lineage viruses by HI

Viruses		Collection date	Passage history	Haemagglutination inhibition titre										
				Post-infection ferret antisera										
				B/Phuket 3073/13	B/Fl 4/06	B/Bris 3/07	B/Estonia 55669/11	B/Mass 02/12	B/Mass 02/12	B/Wis 1/10	B/Stock 12/11	B/Phuket 3073/13	B/Phuket 3073/13	B/HK 3417/14
	Passage history			Egg	Egg	Egg	MDCK	MDCK	Egg	Egg	Egg	MDCK	Egg	Egg
	Ferret number			SH614[*1,3]	F1/10[*1]	F38/14[*2]	F32/12[*2]	F05/15[*1]	F42/14[*2]	F10/13[*2]	F06/15[*1]	F35/14[*2]	F36/14[*2]	St Judes F715/14[*2,4]
	Genetic Group			3	1	2	2	2	2	3	3	3	3	3
REFERENCE VIRUSES														
B/Florida/4/2006	1	2006-12-15	E7/E1	1280	320	640	80	160	640	160	160	20	160	160
B/Brisbane/3/2007	2	2007-09-03	E2/E3	1280	320	320	40	80	640	80	80	20	160	160
B/Estonia/55669/2011	2	2011-03-14	MDCK2/MDCK3	1280	80	80	160	160	80	40	<	40	80	160
B/Massachusetts/02/2012	2	2012-03-13	MDCK1/C2/MDCK3	1280	640	640	320	320	640	160	80	80	320	320
B/Massachusetts/02/2012	2	2012-03-13	E3/E3	1280	320	640	80	80	640	160	160	20	160	160
B/Wisconsin/1/2010	3	2010-02-20	E3/E3	2560	160	160	20	10	160	160	80	40	160	160
B/Stockholm/12/2011	3	2011-03-28	E4/E1	1280	160	80	20	<	160	80	160	40	80	160
B/Phuket/3073/2013	3	2013-11-21	MDCK2/MDCK2	5120	160	160	160	320	160	160	80	320	320	160
B/Phuket/3073/2013	3	2013-11-21	E4/E3	2560	320	160	20	20	320	320	160	80	160	160
B/Hong Kong/3417/2014	3	2014-06-04	E4/E3	1280	160	40	10	<	80	80	40	20	80	160
TEST VIRUSES														
B/Bucuresti/194094/2016		2016-03-15	MDCK1/MDCK1	2560	40	40	40	40	40	40	40	40	80	160
													Vaccine NH 2015-16 SH2016[#] NH 2016-17[#]	

***** Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used) 1 < + <40; 2 < + <10; 3 hyperimmune sheep serum; 4 RDE serum pre-absorbed with TRBC

[#] B/Yamagata-lineage virus recommended for use in quadravalent vaccines

Figure 4. Phylogenetic comparison of influenza B/Yamagata-lineage HA genes



Summary of genetic data submitted to TESSy

For the period covering weeks 40/2015–20/2016, 2 601 viruses have been characterised genetically: 1 770 A(H1N1)pdm09 clade 6B represented by A/South Africa/3626/2013 (6B.1 and 6B.2 subclade designations were not available as reporting categories at the start of the 2015–2016 influenza season); 211 A(H3N2) subclade 3C.2a represented by A/Hong Kong/4801/2014, 65 subclade 3C.3a represented by A/Switzerland/9715293/2013, two subclade 3C.3b represented by A/Stockholm/28/2014, and two subclade 3C.3 represented by A/Samara/73/2013; 496 B/Victoria-lineage clade 1A represented by B/Brisbane/60/2008; and 55 B/Yamagata-lineage clade 3 represented by B/Phuket/3073/2013.

Note: the numbers quoted above are reduced compared to the March and April reports as a consequence of correcting a problem encountered with the reporting of virus characterisation data to TESSy.

Antiviral susceptibility

For weeks 40/2015–20/2016 of the 2015–2016 influenza season, countries reported on the antiviral susceptibility of 2 700 A(H1N1)pdm09 viruses, 172 A(H3N2) viruses and 523 influenza type B viruses from sentinel and non-sentinel sources. All but 31 showed no molecular or phenotypic evidence of reduced inhibition (RI) by neuraminidase inhibitors (oseltamivir and zanamivir). Twenty-six A(H1N1)pdm09 viruses carried NA H275Y amino acid substitution associated with highly reduced inhibition (HRI) by oseltamivir, one A(H3N2) virus showed RI by oseltamivir associated with NA-E119V amino acid substitution and four B/Victoria-lineage viruses showed HRI by both drugs due to NA-R374K amino acid substitution.

Phenotypic testing for susceptibility to oseltamivir and zanamivir has been conducted on 501 viruses at the WIC: 316 A(H1N1)pdm09, 69 A(H3N2), 99 B/Victoria-lineage and 17 B/Yamagata-lineage viruses. All but one A(H1N1)pdm09 virus showed normal inhibition (NI) by these neuraminidase inhibitors: A/Bayern/151/2015 showed reduced inhibition (RI) by zanamivir and carried NA I117R amino acid substitution.

Influenza A(H7N9) virus

On 1 April 2013, the World Health Organization (WHO) Global Alert and Response [4] reported that the China Health and Family Planning Commission notified the WHO of three cases of human infection with influenza A(H7N9). The cases were confirmed by laboratory testing on 29 March 2013 by the Chinese CDC. A description of the characteristics of H7N9 viruses can be found on the WHO website [5]. Increased numbers of cases were reported over the course of the 2013–14, 2014–15 and 2015–16 seasons and cases have been reported recently [6]. A revised Rapid Risk Assessment [7] for these A(H7N9) viruses was carried out by ECDC and posted on 2 February 2015. WHO posted a summary of human infection on 31 January 2014 [8], updated on 09 May 2016 [9] with 18 new cases since the report of 04 April 2016, and conducted a risk assessment on 23 February 2015 [10]. In light of the assessment, WHO advised that countries continue to strengthen influenza surveillance. WHO last summarised the numbers of cases of human infection related to their geographic location on 14 July 2014 [11] and has provided subsequent situation updates, with the latest dated 17 May 2016 [6].

Influenza A(H5) virus

The most recent monthly risk assessment of influenza at the human-animal interface was published by WHO on 9 May 2016 [9]. Since the last WHO Influenza update on 4 April 2016, three laboratory-confirmed human cases of avian influenza A(H5N6) virus infection in China have been reported to WHO. While no human cases of A(H5N1) infection in Egypt have been reported for the same period, a single case of human infection with A(H9N2) was reported. ECDC published an updated rapid risk assessment on the situation in Egypt on 13 March 2015 [12] and an epidemiological update 10 April 2015 [13]. On 2 December 2015, ECDC published a rapid risk assessment related to identification highly pathogenic H5 viruses in poultry in France [14].

WHO CC reports

A description of results generated by the WHO Collaborating Centre for Reference and Research on Influenza at the Crick Worldwide Influenza Centre, the Francis Crick Institute, Mill Hill Laboratory and used at the WHO Vaccine Composition Meetings held in Memphis, USA, 21–23 September 2015, and at WHO Geneva, 22–24 February 2016, can be found at:

https://www.crick.ac.uk/media/273950/crick_sep2015_vcm_report_to_post.pdf and
https://www.crick.ac.uk/media/286458/crick_feb2016_vcm_report_to_post.pdf

Note on the Figures

The phylogenetic trees were constructed using RAxML, drawn using FigTree and annotated using Adobe Illustrator. The bars indicate the proportion of nucleotide changes between sequences. Reference strains are viruses to which post-infection ferret antisera have been raised. The colours indicate the month of sample collection. Isolates from WHO NICs in EU/EEA countries are marked (#). Sequences for some viruses from non-EU/EEA countries were recovered from GISAID. We gratefully acknowledge the authors, originating and submitting laboratories of the sequences from GISAID's EpiFlu database which were downloaded for use in the preparation of this report (all submitters of data may be contacted directly via the GISAID website), along with all laboratories who submitted sequences directly to the London WHO Collaborating Centre.

References

1. World Health Organization. Recommended composition of influenza virus vaccines for use in the 2015–2016 northern hemisphere influenza season. Weekly Epidemiological Record 90, 97-108.
2. World Health Organization. Recommended composition of influenza virus vaccines for use in the 2016 southern hemisphere influenza season. Weekly Epidemiological Record 90, 545-558.
3. World Health Organization. Recommended composition of influenza virus vaccines for use in the 2016–2017 northern hemisphere influenza season. Weekly Epidemiological Record 91, 121-132.
4. World Health Organization. Global alert and response: Human infection with influenza A(H7N9) virus in China. 1 April 2013. Available from: http://www.who.int/csr/don/2013_04_01/en/index.html
5. World Health Organization. Avian influenza A(H7N9) virus. Available from: http://www.who.int/influenza/human_animal_interface/influenza_h7n9/en/
6. World Health Organization. Situation updates - avian influenza. Available from: http://www.who.int/csr/don/17-may-2016-ah7n9-china/en/
7. European Centre for Disease Prevention and Control. Updated rapid risk assessment. Human infection with avian influenza A(H7N9) virus. Fourth update. 2 February 2015. Available from: http://ecdc.europa.eu/en/publications/Publications/RRA-Influenza-A-H7N9-update-four.pdf
8. World Health Organization. Background and summary of human infection with avian influenza A(H7N9) virus – as of 31 January 2014. Geneva: WHO; 2014. Available from: http://www.who.int/influenza/human_animal_interface/20140131_background_and_summary_H7N9_v1.pdf
9. World Health Organization. Influenza at the human-animal interface. Summary and assessment as of 25 February 2016. Available from: http://www.who.int/influenza/human_animal_interface/Influenza_Summary_IRA_HA_interface_05_09_2016.pdf
10. World Health Organization. WHO risk assessment: Human infections with avian influenza A(H7N9) virus, 23 February 2015. Available from: http://www.who.int/influenza/human_animal_interface/influenza_h7n9/RiskAssessment_H7N9_23Feb20115.pdf
11. World Health Organization. Map and epidemiological curve of confirmed human cases of avian influenza A(H7N9). Report 18 – data in WHO/HQ as of 14 July 2014. Available from: http://www.who.int/influenza/human_animal_interface/influenza_h7n9/18_reportwebh7n9number_20140714.pdf
12. European Centre for Disease Prevention and Control. Rapid Risk Assessment. Human infection with avian influenza A(H5N1) virus, Egypt. Available from: http://ecdc.europa.eu/en/publications/Publications/Rapid-Risk-Assessment-Influenza-A-H5N1-Egypt-March-2015.pdf
13. European Centre for Disease Prevention and Control. Epidemiological update: increase in reporting of human cases of A(H5N1) influenza, Egypt. Available from: http://ecdc.europa.eu/en/press/news/_layouts/forms/News_DispForm.aspx?List=8db7286c-fe2d-476c-9133-18ff4cb1b568&ID=1199
14. European Centre for Disease Prevention and Control. Rapid Risk Assessment. Situation overview: highly pathogenic avian influenza virus A of H5 type. Available from: http://ecdc.europa.eu/en/publications/Publications/highly-pathogenic-avian-influenza-virus-A-H5-rapid-risk-assessment-2-dec-2015.pdf