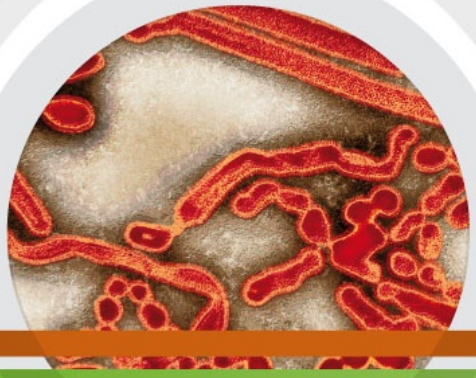

SURVEILLANCE REPORT

Influenza virus characterisation

Summary Europe, March 2019

Summary

This is the fifth report for the 2018–19 influenza season. As of week 14 in 2019, 197 027 influenza detections across the WHO European Region had been reported. Detections were 99.1% type A viruses, with A(H1N1)pdm09 prevailing over A(H3N2), and 0.9% type B viruses, with 72 (67%) of 108 ascribed to a B/Yamagata-lineage.

Since the February 2019 characterisation report[1], a further five shipments of influenza-positive specimens from EU/EEA countries have been received at the London WHO CC, the Francis Crick Worldwide Influenza Centre (WIC). A total of 1 037 virus specimens, with collection dates after 31 August 2018, have been received.

A total of 103/105 (98.1%) A(H1N1)pdm09 test viruses characterised antigenically since the February 2019 characterisation report showed good reactivity with antiserum raised against the 2018–19 vaccine virus, A/Michigan/45/2015 (clade 6B.1). The 304 test viruses with collection dates from week 40 of 2018 genetically characterised at the WIC, including an H1N2 reassortant, have all fallen in a 6B.1 subclade, designated 6B.1A, defined by HA1 amino acid substitutions of S74R, S164T and I295V. Of these recently circulating viruses, 273 also have HA1 S183P substitution, often with additional substitutions in HA1 and/or HA2.

Since the last report, only 46 A(H3N2) viruses successfully recovered had sufficient HA titre to allow antigenic characterisation by HI assay in the presence of oseltamivir. These viruses were poorly recognised by antisera raised against the currently used vaccine virus, egg-propagated A/Singapore/INFIMH-16-0019/2016, in HI assays. Of the 247 viruses with collection dates from week 40 of 2018 genetically characterised at the WIC, 224 were clade 3C.2a (with 29 3C.2a2, nine 3C.2a3, five 3C.2a4 and 181 3C.2a1b) and 23 were clade 3C.3a.

Recent clade 1A B/Victoria-lineage viruses carry HA genes that encode HA1 amino acid substitutions of I117V, N129D and V146I were compared to a previous vaccine virus, B/Brisbane/60/2008. Groups of viruses defined by deletions of two [Δ162–163, 1A(Δ2)] or three [Δ162–164, 1A(Δ3)] amino acids in HA1 have emerged, with the triple deletion group having subgroups of Asian and African origin. HI analyses with panels of post-infection ferret antisera have shown these virus groups to be antigenically

[1] European Centre for Disease Prevention and Control. Influenza virus characterisation – Summary Europe, February 2019. Stockholm: ECDC; 2019. Available from: http://ecdc.europa.eu/publications-data/influenza-virus-characterisation-february-2019

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distinguishable. One virus that was characterised since the last report is of the Asian [Δ162–164, 1A(Δ3)] subgroup. Of the five viruses characterised from EU/EEA countries, one was Δ162–163 and four Δ162–164 (three African and one Asian subgroup).

Including the three B/Yamagata-lineage viruses reported here, nine from the 2018–19 season have been characterised. All have HA genes that fall in clade 3 and encode HA1 amino acid substitutions of L172Q and M251V compared to the vaccine virus B/Phuket/3073/2013, but remain antigenically similar to the vaccine virus that is recommended for use in quadrivalent vaccines for current and subsequent northern hemisphere influenza seasons.

Table 1 shows a summary of influenza virus detections in the WHO European Region reported to ECDC's TESSy database since the start of the 2018–19 season (weeks 40/2018-14/2019). Since week 1 of 2019, the cumulative number of detections has increased from 18 049 to 197 027, with type A (99.1%) predominating over type B (0.9%) viruses which is a common pattern, unlike the 2017-18 season when type B predominated over type A at the start of the season and throughout most of it. Of the type A viruses subtyped (n=74 057) and the type B viruses ascribed to a lineage (n=108), A(H1N1)pdm09 (n=43 254) have continued to prevail over A(H3N2) (n=30 803) viruses and 72 of 108 type B viruses have been B/Yamagata-lineage; these relative proportions have increased in favour of A(H3N2) and decreased slightly for B/Yamagata-lineage viruses compared to the summary in the February 2019 characterisation report. Overall, the ratio of type A to type B detections is dramatically increased compared with the 2017–18 season (0.8:1 to 113:1) and as the 2018-19 influenza season has progressed the early prevalence of A(H1N1)pdm09 over A(H3N2) viruses has decreased such that levels observed in the two seasons have become comparable (58.4% in 2018–19 compared with 50.6% in 2017–18).

Table 1. Influenza virus detections in WHO European Region from start of reporting for 2018–19 season (weeks 40/2018–14/2019)

	Cumulative number of detections			Totals*		Totals for 2017-18 season*		
Virus type/subtype/lineage	Sentinel sources	Non-sentinel sources	Totals	%	Ratios	Number	%	Ratios
Influenza A	**20727**	**174569**	**195296**	**99.1**	**113:1**	**106003**	**44.1**	**0.8:1**
A(H1N1)pdm09	8671	34583	43254	58.4		23121	50.6	
A(H3N2)	7044	23759	30803	41.6	0.7:1	22568	49.4	1:1
A not subtyped	5012	116227	121239			60314		
Influenza B	**203**	**1528**	**1731**	**0.9**		**134618**	**55.9**	
Victoria lineage	10	26	36	33.3		301	1.9	
Yamagata lineage	50	22	72	66.7	2:1	15701	98.1	52.2:1
Lineage not ascribed	143	1480	1623			118616		
Total detections (total tested)	**20930 (50953)**	**176097 (718996)**	**197027 (769949)**			**240621 (903182)**		

*** Percentages are shown for total detections (types A & B [in bold type], and for viruses ascribed to influenza A subtype and influenza B lineage).**

Since week 40 of 2018, 47 shipments of specimens (virus isolates and/or clinical specimens) from 34 centres across 29 EU/EEA countries have been received at the Crick Worldwide Influenza Centre (WIC) containing 1 037 individual virus-related samples with collection dates after 31 August 2018 (Table 2). The proportions of received samples are similar to those reported to TESSy (Table 1) in terms of virus type and virus subtype or lineage. The genetic and antigenic characterisation data generated at the WIC for many of these viruses was presented at the WHO influenza vaccine composition meeting for the northern hemisphere 2019–20 season. Recommendations emerging from the meeting held on 18–21 February 2019 and the subsequent update on 21 March 2019 have been published [1,2].

Table 2. Summary of clinical samples and virus isolates contained in packages received from EU/EEA Member States since week 40, 2018

MONTH / Country	TOTAL RECEIVED Seasonal viruses	A		H1N1pdm09		H3N2			B		B Victoria lineage		B Yamagata lineage	
		Number received	Number propagated[1]	Number received	Number propagated[1]	Number received	Number propagated[2]		Number received	Number propagated[1]	Number received	Number propagated[1]	Number received	Number propagated[1]
2018 SEPTEMBER														
France	7					6	3	3					1	1
Spain	1			1	1									
Sweden	1			1	1									
OCTOBER														
Czech Republic	2			2	2									
Denmark	2					2	0	2						
Estonia	3	1	0	1	0	1	0	1						
Finland	2			1	1	1	0	1						
France	11			3	3	7	5	2					1	in process
Germany	1					1	0	1						
Iceland	2					1	0	1					1	1
Ireland	3			2	1	1	0	1						
Latvia	1			1	1									
Netherlands	1					1	0	1						
Norway	29			12	8	14	0	8					3	1
Portugal	2			2	2									
Slovenia	1			1	1									
United Kingdom	3			1	1	2	0	2						
NOVEMBER														
Austria	4			1	1	3	1	2						
Belgium	5			3	2	2	0	2						
Bulgaria	1			1	0									
Croatia	1			1	1									
Czech Republic	1			1	1									
Denmark	12			8	8	3	0	3			1	1		
Estonia	3			3	1									
Finland	4			2	2	2	0	2						
France	17			10	10	7	4	2						
Germany	8			4	4	4	0	4						
Iceland	15			4	3	11	7	3						
Ireland	17			12	10	4	0	3					1	1
Italy	10			2	2	8	5	3						
Latvia	2					2	1	1						
Lithuania	5					5	0	4						
Netherlands	3			2	2	1	0	1						
Norway	26			14	13	12	1	10						
Portugal	1												1	0
Spain	8			2	in process	6	in process							
Sweden	1			1	1									
United Kingdom	14			6	6	6	2	1			1	0	1	1
DECEMBER														
Austria	1					1	0	1						
Belgium	6			2	1	4	0	2						
Bulgaria	9			5	4	4	0	4						
Croatia	8			6	6	2	0	1						
Cyprus	3			3	1									
Denmark	7			5	5	2	0	2						
Estonia	18	1	0	16	11	1	0							
France	33			17	17	14	10	4			1	1	1	in process
Germany	11			5	5	6	0	6						
Greece	11			8	5	3	0	1						
Hungary	6			4	4	2	1	1						
Iceland	3			3	3									
Ireland	3			3	3									
Italy	1			1	1									
Latvia	6			5	5	1	1	0						
Lithuania	14	1	0	5	3	8	0	3						
Netherlands	5			4	4	1	0	1						
Norway	15			6	4	7	1	4			2	1		
Poland	1			1	0									
Portugal	18			8	8	9	0	9					1	1
Romania	12			2	2	10	1	9						
Slovenia	3			1	1						2	2		
Spain	28			15	in process	13	in process							
Sweden	11			7	7	4	3	1						
United Kingdom	8			5	5	3	1							
2019 JANUARY														
Belgium	47			8	3	39	2	18						
Bulgaria	13			12	in process	1	0	1						
Croatia	2												2	1
Cyprus	17			17	7									
France	26			11	11	15	13	2						
Germany	34			15	15	19	5	14						
Greece	30			19	in process	8	in process		3	in process				
Hungary	2					2	2							
Italy	4			3	3	1	0	1						
Latvia	6			6	6									
Lithuania	1			1	1									
Luxembourg	25			10	8	14	3	6					1	1
Malta	42			23	in process	19	in process							
Netherlands	12			8	8	4	2	2						
Norway	19			10	9	7	0	6					2	1
Poland	6			6	3									
Portugal	5			1	1	4	3	1						
Romania	13			11	6	2	1	1						
Slovenia	11			8	8	3	0	3						
Spain	73			32	in process	41	in process							
United Kingdom	38	3	0	31	2	4	1	0						
FEBRUARY														
Bulgaria	42			22	in process	20	in process							
Germany	26			9	9	17	in process							
Greece	17			11	in process	5	in process		1	in process				
Latvia	7			1	1	6	in process							
Malta	8			5	in process	3	0	1						
MARCH														
Bulgaria						1	in process							
Germany	23			7	in process	16	in process							
Greece	15			8	in process	6	in process		1	in process				
Latvia	2			1	1	1	in process							
	1037	6	0	538	296	466	79	169	5	0	7	5	16	9
29 Countries					51.9%		44.9%					0.7%		1.5%
					97.4%							2.7%		

1. Propagated to sufficient titre to perform HI assay (the totalled number does not include any from batches that are in process)
2. Propagated to sufficient titre to perform HI assay in the presence of 20nM oseltamivir (the totalled number does not include any from batches that are in process)

Numbers in red indicate viruses recovered but with insufficient HA titre to permit HI assay

Numbers highlighted in blue show the number of viruses subjected to HI assay for 'completed' sample sets. Under a 'sequence first' virus characterisation scheme: (i) sequencing only was possible for some clinical specimens that had been collected in lysis buffer; (ii) where sequencing failed, despite samples having good Ct values, virus propagation was attempted for only a few samples; and (iii) where multiple viruses shared the same HA sequence only a selection were propagated to allow assay by HI

one virus is H1N2

As of 2019-04-10

Influenza A(H1N1)pdm09 virus analyses

Tables 3-1 to 3-6 show the results of haemagglutination inhibition (HI) assays of A(H1N1)pdm09 viruses performed with a panel of post-infection ferret antisera. Tables 3-1 and 3-2 are repeated from the February 2019 characterisation report, but with genetic group data now included, while Tables 3-3 to 3-6 were generated in March 2019. Test viruses in each table are sorted by genetic group (where known at the time of preparing this report) and then by date of collection. A summary of the HI results for all test viruses in Tables 3-1 to 3-6 is shown in Table 3-7 and a summary for viruses sorted by genetic group is shown in Table 3-8.

The vast majority of A(H1N1)pdm09 test viruses, 146 of 149 (98%), were antigenically indistinguishable from the egg-propagated vaccine virus for the northern hemisphere 2018–19 influenza season, A/Michigan/45/2015 [3], as assessed with post-infection ferret antisera, being recognised at titres within twofold of the titre of the antiserum with the homologous virus (Table 3-7). The three viruses showing greater than twofold titre reductions are undergoing genetic characterisation.

Antisera raised against eight reference viruses (A/California/7/2009, A/Bayern/69/2009, A/Hong Kong/5659/2012, A/Slovenia/2903/2015, A/Paris/1447/2017, A/Switzerland/3330/2017, A/Norway/3433/2018 and A/Ireland/84630/2018) recognised ≥90% of the test viruses at titres within twofold of the titres of the antisera with their homologous viruses and over 97% at titres within fourfold of the respective homologous titres (Table 3-7). Similarly good reactivities were seen with antiserum raised against egg-propagated A/Brisbane/02/2018, the vaccine virus recommended for the 2019-20 northern hemisphere influenza season [1,2].The antiserum raised against A/Switzerland/2656/2017 recognised 76% of test viruses at titres within twofold of the titre of the antiserum with the homologous virus and 94% within fourfold. The antisera raised against cell culture-propagated A/Astrakhan/1/2011 and A/Lviv/N6/2009 recognised 77% and 3% respectively of test viruses at titres within twofold of the homologous titres and 100% and 26% respectively within fourfold. The antiserum raised against A/Lviv/N6/2009 is an unusual virus/antiserum combination with A/Lviv/N6/2009 encoding HA1 amino acid substitutions of G155G/E, with E predominating, and D222G.

All test viruses for which HA gene sequencing had been completed fell into clade 6B.1, which is defined by the amino acid substitutions S84N, S162N (introducing a potential N-linked glycosylation site) and I216T in HA1, with all recently circulating viruses clustering in a genetic subclade designated as 6B.1A and defined by the HA1 amino acid substitutions S74R, S164T (which alters the glycosylation motif at residues 162–164) and I295V. A number of genetic subgroups defined by specific amino acid substitutions have emerged but the great majority of viruses in the various subgroups have remained antigenically similar to A/Michigan/45/2015 as shown in the February 2019 and earlier characterisation reports, as assessed with post-infection ferret antisera.

Figure 1 shows a phylogenetic tree for the HA genes of A(H1N1)pdm09 viruses sequenced at the Francis Crick Institute, with collection dates since the start of the 2018-19 influenza season, and other representative viruses. Within subclade 6B.1A, clusters of viruses (genetic groups) encoding a range of HA1 amino acid substitutions have emerged, e.g. T120A, or N260D in combination with N129D, many with T185I, or N260D with E235D and V193A in HA2 or N129D with A141E or K302T and N169S and E179D in HA2 or L161I and I77M in HA2. The HA of most recently circulating viruses carry the substitution S183P in HA1, although this is not retained in all genetic groups, and the phylogenetic tree is annotated with HA1 S183P substitution groups assigned for the February 2019 WHO Vaccine Consultation Meeting [1,2]: 6B.1A/183P-1 to -7, abbreviated to 6B.1A1 to 6B.1A7 in Figure 1. The location of vaccine viruses A/Michigan/45/2015 and the recently recommended A/Brisbane/02/2018 for the northern hemisphere 2019-20 influenza season [1,2] are indicated on the phylogeny (Figure 1).

Table 3-9 shows that test viruses from EU/EEA countries in subclade 6B.1A and viruses in each of the genetic groups 183P-1, -2, -3, -4, -5, -6 and -7 show similar patterns of recognition by the panel of post-infection ferret antisera. Generally, test viruses showed good reactivity, with ≥90% reacting within twofold of respective homologous titres and all antisera but for that raised against A/Lviv/N6/2009. However, of the two groups with the highest number of test viruses, 6B.1A5 and 6B.1A7, the group 6B.1A5 viruses (defined by HA1 S183P and N260D amino acid substitutions, with the great majority also having N129D and T185I substitutions) showed lower proportions reacting within twofold of homologous titres with antisera raised against A/California/7/2009 (86%), A/Astrakhan/1/2011 (71%), A/Switzerland/2656/2017 (68%) and A/Ireland/84630/2018 (80%), although all group 6B.1A5 test viruses reacted within fourfold of the respective homologous titres. While such HI studies conducted with post-infection ferret antisera indicated low levels of antigenic drift in A(H1N1)pdm09 viruses, panels of post-vaccination human antisera recognised viruses containing the HA1 substitution S183P less well. Based on these results, A/Brisbane/02/2018 was recommended as the A(H1N1)pdm09 vaccine component for the northern hemisphere 2019–20 influenza season [1,2].

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

Viruses	Other information	Passage history / Ferret number / Genetic group	Collection date	Passage history	Haemagglutination inhibition titre – Post-infection ferret antisera: A/Mich 45/15 Egg NIB 6B.1	A/Cal 7/09 Egg F07/16*1	A/Bayern 69/09 MDCK F09/15*1	A/Lviv N6/09 MDCK F13/18*1	A/Astrak 1/11 MDCK F22/13*1 5	A/HK 5659/12 MDCK F17/15*1 IIB 6A	A/Slov 2903/15 Egg F48/16*1 6B.1	A/Paris 1447/17 MDCK F03/18*2 6B.1A	A/Swit 2656/17 Egg F20/18*1 6B.1A	A/Swit 3330/17 Egg F23/18*1 6B.1A5	A/Norway 3433/18 MDCK F04/19*1 6B.1A5
REFERENCE VIRUSES															
A/Michigan/45/2015		6B.1	2015-09-07	E3/E3	640	640	320	320	640	320	1280	1280	640	320	1280
A/California/7/2009	clone 38-32		2009-04-09	E3/E3	640	320	320	320	320	320	640	1280	640	160	1280
A/Bayern/69/2009	G155E		2009-07-01	MDCK5/MDCK1	<	<	320	320	<	<	<	320	40	40	320
A/Lviv/N6/2009	G155E, D222G		2009-10-27	MDCK4/SIAT1/MDCK3	80	80	1280	1280	80	80	160	640	160	160	1280
A/Astrakhan/1/2011		5	2011-02-28	MDCK1/MDCK7	640	320	640	640	640	320	1280	2560	640	320	2560
A/Hong Kong/5659/2012		6A	2012-05-21	MDCK4/MDCK2	640	320	160	160	320	320	640	1280	320	320	1280
A/Slovenia/2903/2015	clone 37	6B.1	2015-10-26	E4/E2	640	320	320	320	320	320	640	1280	640	320	1280
A/Paris/1447/2017		6B.1A	2017-10-20	MDCK1/MDCK3	640	320	320	160	320	320	640	2560	640	320	2560
A/Switzerland/2656/2017		6B.1A	2017-12-21	E5/E3	640	320	640	640	320	320	1280	1280	1280	320	2560
A/Switzerland/3330/2017	clone 35	6B.1A5	2017-12-20	E6/E2	320	160	160	160	160	160	320	1280	320	320	1280
A/Norway/3433/2018		6B.1A5	2018-10-30	MDCK3	640	160	160	80	160	160	640	1280	320	160	1280
TEST VIRUSES															
A/Saint-Etienne/1883/2018			2018-08-31	MDCK3/MDCK1	640	320	320	80	320	320	1280	1280	640	320	2560
A/Ireland/80216/2018		6B.1A	2018-11-12	MDCK1/MDCK1	640	320	320	160	320	320	640	1280	640	320	1280
A/Marseille/1989/2018		6B.1A5	2018-10-05	MDCK2/MDCK1	1280	320	320	160	320	320	1280	2560	640	320	2560
A/Ireland/70370/2018		6B.1A5	2018-10-07	MDCK2/MDCK1	640	160	160	160	160	160	640	1280	320	160	1280
A/Ireland/78268/2018		6B.1A5	2018-11-06	MDCK1/MDCK1	640	160	160	320	160	320	640	1280	320	320	2560
A/Lyon/2065/2018		6B.1A5	2018-11-08	MDCK3/MDCK1	320	80	320	320	160	160	640	1280	320	320	2560
A/Lyon/2088/2018		6B.1A5	2018-11-12	MDCK2/MDCK1	1280	320	640	320	320	320	1280	2560	640	640	2560
A/Ireland/81752/2018		6B.1A5	2018-11-20	MDCK2/MDCK1	640	320	320	320	320	640	1280	2560	640	640	2560
A/Netherlands/10000/2019		6B.1A5	2019-01-02	MDCK-MIX2/MDCK1	640	160	160	320	160	160	640	1280	320	160	2560
A/Netherlands/10003/2019		6B.1A5	2019-01-10	MDCK-MIX2/MDCK1	640	320	320	320	320	320	640	2560	640	320	2560
A/Netherlands/10004/2019		6B.1A5	2019-01-11	MDCK-MIX2/MDCK1	640	640	640	640	640	640	1280	2560	640	640	2560
A/Ireland/79897/2018		6B.1A7	2018-11-12	MDCK1/MDCK1	640	160	160	80	160	160	640	2560	640	160	2560
A/Netherlands/10615/2018		6B.1A7	2018-12-28	MDCK-MIX2/MDCK1	640	320	320	320	320	320	1280	2560	640	320	2560
A/Netherlands/10614/2018		6B.1A7	2018-12-28	MDCK-MIX2/MDCK1	640	320	320	160	320	320	640	2560	640	320	2560
A/Ireland/78012/2018		6B.1A1	2018-11-05	MDCK1/MDCK1	320	160	160	<	160	160	640	1280	320	160	1280
A/Netherlands/10001/2019		6B.1A6	2019-01-07	MDCK-MIX2/MDCK1	640	320	320	160	320	160	1280	2560	640	320	2560

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

Vaccine

Sequences in phylogenetic trees

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

Viruses	Other information	Passage history / Ferret number / Genetic group	Collection date	Passage history	Haemagglutination inhibition titre – Post-infection ferret antisera: A/Mich 45/15 Egg NIB F42/16*1 6B.1	A/Cal 7/09 Egg F07/16*1	A/Bayern 69/09 MDCK F09/15*1	A/Lviv N6/09 MDCK F13/18*1	A/Astrak 1/11 MDCK F22/13*1 5	A/HK 5659/12 MDCK F17/15*1 6A	A/Slov 2903/15 Egg NIB F48/16*1 6B.1	A/Paris 1447/17 MDCK F03/18*2 6B.1A	A/Swit 2656/17 Egg F20/18*1 6B.1A	A/Swit 3330/17 Egg F23/18*1 6B.1A5	A/Norway 3433/18 MDCK F04/19*1 6B.1A5	NEW A/Ireland 84630/18 MDCK F08/19*1 6B.1A6
REFERENCE VIRUSES																
A/Michigan/45/2015		6B.1	2015-09-07	E3/E3	640	640	640	320	640	640	1280	2560	1280	320	2560	1280
A/California/7/2009	clone 38-32		2009-04-09	E3/E3	640	640	640	320	640	640	1280	2560	1280	320	2560	1280
A/Bayern/69/2009	G155E		2009-07-01	MDCK5/MDCK1	40	<	320	320	<	<	40	320	40	40	320	40
A/Lviv/N6/2009	G155E, D222G		2009-10-27	MDCK4/SIAT1/MDCK3	80	80	1280	1280	80	160	160	1280	320	160	1280	320
A/Astrakhan/1/2011		5	2011-02-28	MDCK1/MDCK7	640	640	640	640	640	640	640	2560	640	320	2560	1280
A/Hong Kong/5659/2012		6A	2012-05-21	MDCK4/MDCK2	640	320	320	160	320	640	1280	2560	640	320	2560	1280
A/Slovenia/2903/2015	clone 37	6B.1	2015-10-26	E4/E2	640	640	320	320	640	640	1280	2560	1280	320	2560	1280
A/Paris/1447/2017		6B.1A	2017-10-20	MDCK1/MDCK3	640	320	320	160	320	320	1280	2560	1280	640	2560	1280
A/Switzerland/2656/2017		6B.1A	2017-12-21	E5/E3	640	640	320	320	640	640	1280	2560	1280	640	2560	1280
A/Switzerland/3330/2017	clone 35	6B.1A5	2017-12-20	E6/E2	640	320	320	160	320	320	640	1280	640	640	1280	1280
A/Norway/3433/2018		6B.1A5	2018-10-30	MDCK3	320	160	160	80	160	320	640	1280	320	320	2560	640
A/Ireland/84630/2018		6B.1A6	2018-11-28	MDCK1/MDCK2	640	320	320	160	320	320	640	2560	640	320	2560	1280
A/Brisbane/02/2018		6B.1A1	2018-01-04	E3/E1	1280	1280	640	640	640	1280	2560	5120	2560	640	5120	2560
TEST VIRUSES																
A/Slovenia/2615/2018		6B.1A	2018-10-15	MDCKx/MDCK1	1280	1280	640	320	640	1280	2560	5120	2560	1280	5120	2560
A/Lisboa/25/2018		6B.1A5	2018-10-25	MDCK1/MDCK1	640	160	320	160	320	160	640	2560	640	320	2560	1280
A/Slovenia/82/2019		6B.1A5	2019-01-03	MDCKx/MDCK1	640	320	320	160	320	320	640	2560	640	320	2560	1280
A/Slovenia/80/2019		6B.1A5	2019-01-03	SIATx/MDCK1	640	320	320	160	320	640	640	1280	640	320	2560	1280
A/Avila/33/2019		6B.1A5	2019-01-15	MDCK1/MDCK1	640	320	320	320	640	320	1280	2560	640	320	2560	1280
A/Valladolid/28/2019		6B.1A5	2019-01-15	MDCK1/MDCK1	1280	640	640	320	640	320	1280	2560	1280	640	2560	2560
A/Lisboa/26/2018		6B.1A7	2018-10-25	MDCK3/MDCK1	1280	640	640	160	640	320	1280	5120	1280	640	5120	2560
A/Ireland/83112/2018		6B.1A7	2018-11-22	MDCK2	1280	640	640	320	640	640	2560	5120	1280	640	2560	2560
A/Ireland/82546/2018		6B.1A7	2018-11-22	MDCK2	1280	640	640	320	640	640	2560	5120	1280	640	5120	2560
A/Lisboa/29/2018		6B.1A7	2018-12-06	MDCK1/MDCK1	1280	320	640	160	640	320	1280	2560	1280	640	2560	1280
A/Lisboa/27/2018		6B.1A7	2018-12-08	MDCK1/MDCK1	640	320	320	160	320	160	1280	5120	640	320	2560	1280
A/Lisboa/28/2018		6B.1A7	2018-12-10	MDCK2/MDCK1	640	320	160	80	320	160	640	2560	640	320	2560	1280
A/Lisboa/30/2018		6B.1A7	2018-12-14	MDCK2/MDCK1	640	320	320	160	320	320	1280	2560	640	320	2560	1280
A/Slovenia/3088/2018		6B.1A7	2018-12-26	SIATx/MDCK1	1280	640	640	320	640	640	2560	5120	2560	640	2560	2560
A/Burgos/561/2018		6B.1A7	2018-12-28	MDCK1/MDCK1	2560	1280	1280	640	640	1280	2560	5120	2560	1280	5120	5120
A/Portugal/119/2018		6B.1A7	2018-12-29	MDCK1	640	320	320	160	320	320	1280	2560	640	320	2560	1280
A/Valladolid/1/2019		6B.1A7	2019-01-02	MDCK1/MDCK1	1280	640	640	320	640	1280	2560	5120	2560	1280	5120	5120
A/Valladolid/8/2019		6B.1A7	2019-01-08	MDCK1/MDCK1	1280	640	640	320	640	640	1280	5120	1280	640	5120	2560
A/Burgos/6/2019		6B.1A7	2019-01-08	MDCK1/MDCK1	1280	640	640	160	320	640	1280	5120	1280	640	5120	2560
A/Valladolid/10/2019		6B.1A7	2019-01-09	MDCK1/MDCK1	640	320	320	160	320	320	1280	2560	1280	640	2560	1280
A/Valladolid/16/2019		6B.1A7	2019-01-11	MDCK1/MDCK1	1280	640	640	320	640	640	2560	5120	2560	1280	5120	5120
A/Palencia/19/2019		6B.1A7	2019-01-12	MDCK1/MDCK1	640	320	320	160	320	320	1280	2560	1280	640	2560	2560
A/Valladolid/22/2019		6B.1A7	2019-01-13	MDCK1/MDCK1	1280	640	320	160	640	320	1280	5120	1280	640	5120	2560
A/Burgos/35/2019		6B.1A7	2019-01-15	MDCK1/MDCK1	640	320	320	160	640	320	1280	2560	1280	640	2560	2560
A/Valladolid/29/2019		6B.1A7	2019-01-15	MDCK1/MDCK1	1280	640	640	320	640	640	2560	5120	1280	640	5120	2560
A/Baden-Wurttemberg/161/2018		6B.1A2	2018-08-16	C1/MDCK1	1280	640	640	320	640	1280	2560	5120	1280	640	5120	2560
A/Valladolid/559/2018		6B.1A6	2018-09-24	MDCK1/MDCK1	640	320	160	80	160	320	640	1280	640	320	2560	1280
A/Schleswig-Holstein/3/2018		6B.1A4		C2/MDCK1	640	320	320	160	320	320	1280	2560	640	320	2560	1280

* Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used)
1 < = <40; 2 < = <80
Sequences in phylogenetic trees

Vaccine

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

Viruses	Other information	Passage history / Ferret number / Genetic group	Collection date	Passage history	Haemagglutination inhibition titre – Post-infection ferret antisera: A/Mich 45/15 Egg NIB F42/16*1 6B.1	A/Bayern 69/09 MDCK F09/15*1	A/Lviv N6/09 MDCK F13/18*1	A/Slov 2903/2015 Egg NIB F48/16*1 6B.1	A/Paris 1447/17 MDCK F03/18*2 6B.1A	A/Swit 2656/17 Egg F20/18*1 6B.1A	A/Swit 3330/17 Egg F23/18*1 6B.1A5	A/Norway 3433/18 MDCK F04/19*1 6B.1A5	A/Ire 84630/18 MDCK F08/19*1 6B.1A6
REFERENCE VIRUSES													
A/Michigan/45/2015		6B.1	2015-09-07	E3/E3	640	640	320	2560	2560	1280	640	2560	1280
A/Bayern/69/2009	G155E		2009-07-01	MDCK5/MDCK1	40	320	320	40	320	80	80	320	40
A/Lviv/N6/2009	G155E, D222G		2009-10-27	MDCK4/SIAT1/MDCK3	80	1280	1280	160	1280	320	160	1280	160
A/Slovenia/2903/2015	clone 37	6B.1	2015-10-26	E4/E2	320	320	160	1280	1280	640	320	1280	640
A/Paris/1447/2017		6B.1A	2017-10-20	MDCK1/MDCK3	640	320	80	1280	2560	640	320	2560	1280
A/Switzerland/2656/2017		6B.1A	2017-12-21	E5/E3	640	320	320	1280	2560	1280	640	2560	1280
A/Switzerland/3330/2017	clone 35	6B.1A5	2017-12-20	E6/E2	320	320	160	640	1280	640	320	1280	1280
A/Norway/3433/2018		6B.1A5	2018-10-30	MDCK3	320	160	80	640	1280	320	320	2560	640
A/Ireland/84630/2018		6B.1A6	2018-11-28	MDCK1/MDCK2	640	320	160	1280	5120	640	320	5120	2560
TEST VIRUSES													
A/Stockholm/21/2018		6B.1A5	2018-09-30	MDCK0/MDCK1	640	160	160	320	1280	320	320	2560	640
A/Luxembourg/1621/2019			2019-01-07	MDCK1	1280	320	160	1280	2560	640	320	5120	1280
A/Bulgaria/1464/2018		6B.1A	2018-12-11	MDCK1	640	320	160	1280	2560	640	640	2560	2560
A/Estonia/118253/2018		6B.1A	2018-12-17	MDCK2	640	320	160	640	1280	640	320	2560	1280
A/Bulgaria/1526/2018		6B.1A	2018-12-27	MDCK1	640	320	80	1280	2560	640	320	2560	1280
A/Bulgaria/1533/2018		6B.1A	2018-12-28	MDCK1	640	320	80	1280	2560	640	320	2560	1280
A/Norway/3379/2018		6B.1A5	2018-10-23	MDCK1/MDCK1	320	160	80	640	1280	320	320	2560	640
A/Norway/3488/2018		6B.1A5	2018-11-07	MDCK1/MDCK1	320	80	40	320	640	320	160	1280	640
A/Norway/3473/2018		6B.1A5	2018-11-07	MDCK1	640	320	320	640	2560	640	320	2560	1280
A/Norway/3524/2018		6B.1A5	2018-11-10	MDCK1/MDCK1	640	640	320	1280	2560	640	640	5120	1280
A/Norway/3585/2018		6B.1A5	2018-11-14	MDCK1	1280	640	640	1280	2560	1280	640	5120	2560
A/Norway/3513/2018		6B.1A5	2018-11-15	MDCK1/MDCK1	640	320	320	1280	2560	1280	320	2560	1280
A/Norway/3637/2018		6B.1A5	2018-11-26	MDCK1	640	320	160	640	2560	640	320	2560	1280
A/Norway/3672/2018		6B.1A5	2018-11-28	MDCK1	640	160	160	640	1280	640	320	2560	1280
A/Norway/3679/2018		6B.1A5	2018-11-29	MDCK1	640	320	160	640	1280	640	320	2560	640
A/Norway/3652/2018		6B.1A5	2018-11-29	MDCK1/MDCK1	640	160	160	640	1280	640	320	2560	640
A/Estonia/117956/2018		6B.1A5	2018-11-29	MDCK1	640	320	160	640	2560	640	320	2560	1280
A/Lulea/1/2018		6B.1A5	2018-12-04	MDCK0/MDCK1	320	160	160	640	1280	320	320	1280	640
A/Eskilstuna/2/2018		6B.1A5	2018-12-06	MDCK0/MDCK1	320	320	160	640	1280	320	320	2560	1280
A/Bulgaria/1441/2018		6B.1A5	2018-12-11	MDCK1	1280	640	640	1280	2560	640	640	5120	1280
A/Norway/4138-2/2018		6B.1A5	2018-12-24	MDCK1	640	320	160	640	2560	320	320	2560	1280
A/Norway/90/2019		6B.1A5	2019-01-06	MDCK1	640	320	160	640	2560	640	320	2560	1280
A/Norway/95/2019		6B.1A5	2019-01-07	MDCK1	640	320	160	1280	2560	320	320	2560	1280
A/Norway/140/2019		6B.1A5	2019-01-07	MDCK1	640	320	160	1280	2560	640	320	2560	1280
A/Norway/223/2019		6B.1A5	2019-01-14	MDCK1	640	320	160	640	2560	640	320	2560	1280
A/Halmstad/3/2018		6B.1A7	2018-11-12	MDCK1/MDCK1	1280	640	160	1280	5120	1280	640	5120	2560
A/Norway/3478/2018		6B.1A7	2018-11-14	MDCK1	320	160	80	640	1280	320	160	1280	640
A/Norway/82/2019		6B.1A7	2019-01-05	MDCK1	1280	320	160	2560	5120	640	640	5120	2560
A/Norway/103/2019		6B.1A7	2019-01-07	MDCK1	640	320	160	1280	2560	640	320	5120	1280
A/Luxembourg/1616/2019		6B.1A7	2019-01-09	MDCK1	640	320	160	640	1280	640	320	2560	1280
A/Luxembourg/1423/2019		6B.1A7	2019-01-10	MDCK1	640	160	80	640	1280	320	320	1280	1280
A/Norway/3737/2018		6B.1A3	2018-11-27	MDCK1	1280	640	640	1280	2560	1280	640	2560	2560

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1 < = <40; 2 < = <80
Sequences in phylogenetic trees

Vaccine
NH 2018-19
SH 2019

Table 3-4. Antigenic analysis of A(H1N1)pdm09 viruses by HI

Viruses	Other information	Passage history / Ferret number / Genetic group	Collection date	Passage history	Haemagglutination inhibition titre – Post-infection ferret antisera: A/Mich 45/15 Egg NIB F42/16*1 6B.1	A/Bayern 69/09 MDCK F09/15*1	A/Lviv N6/09 MDCK F13/18*2	A/Slov 2903/2015 Egg NIB F48/16*1 6B.1	A/Paris 1447/17 MDCK F03/18*2 6B.1A	A/Swit 2656/17 Egg F20/18*1 6B.1A	A/Swit 3330/17 Egg F23/18*1 6B.1A5	A/Norway 3433/18 MDCK F04/19*1 6B.1A5	A/Ire 84630/18 MDCK F08/19*1 6B.1A6	A/Bris 02/18 Egg F09/19*1 6B.1A1
REFERENCE VIRUSES														
A/Michigan/45/2015		6B.1	2015-09-07	E3/E2	640	320	320	1280	2560	640	320	1280	1280	1280
A/Bayern/69/2009	G155E		2009-07-01	MDCK5/MDCK1	<	320	160	<	160	40	40	320	40	80
A/Lviv/N6/2009	G155E, D222G		2009-10-27	MDCK4/SIAT1/MDCK3	160	640	1280	160	1280	320	320	1280	320	640
A/Slovenia/2903/2015	clone 37	6B.1	2015-10-26	E4/E2	640	320	320	1280	2560	640	320	2560	1280	1280
A/Paris/1447/2017		6B.1A	2017-10-20	MDCK1/MDCK3	320	160	40	640	1280	320	160	1280	640	640
A/Switzerland/2656/2017		6B.1A	2017-12-21	E5/E3	640	320	320	1280	2560	640	320	2560	1280	640
A/Switzerland/3330/2017	clone 35	6B.1A5	2017-12-20	E6/E2	160	160	80	320	640	320	160	640	640	320
A/Norway/3433/2018		6B.1A5	2018-10-30	MDCK3	320	160	<	320	1280	160	160	1280	320	320
A/Ireland/84630/2018		6B.1A6	2018-11-28	MDCK1/MDCK2	320	160	80	640	1280	320	160	1280	640	640
A/Brisbane/02/2018		6B.1A1	2018-01-04	E3/E1	640	320	320	1280	2560	640	320	1280	1280	640
IVR-190(A/Brisbane/02/2018)		6B.1A1	2018-01-04	E3/D8/E1	1280	640	320	2560	5120	1280	640	2560	2560	1280
TEST VIRUSES														
A/Belgium/G0024/2019		6B.1A5	2019-01-07	MDCK2	320	160	80	320	1280	160	160	2560	640	640
A/Luxembourg/2575/2019			2019-01-13	MDCK2	640	160	80	640	1280	320	320	2560	640	1280
A/Avila/32/2019			2019-01-15	SIAT1/SIAT1	320	160	80	320	1280	160	160	2560	1280	1280
A/Belgium/S0270/2019		6B.1A7	2019-01-15	MDCK2	320	160	80	640	1280	320	160	1280	640	640
					Vaccine NH 2018-19									Vaccine NH 2019-20
					Sequences in phylogenetic trees SH 2019									

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

Table 3-5. Antigenic analysis of A(H1N1)pdm09 viruses by HI

Viruses	Other information	Passage history / Ferret number / Genetic group	Collection date	Passage history	Haemagglutination inhibition titre – Post-infection ferret antisera									
					A/Mich 45/15	A/Bayern 69/09	A/Lviv N6/09	A/Slov 2903/2015	A/Paris 1447/17	A/Swit 2656/17	A/Swit 3330/17	A/Norway 3433/18	A/Ire 84630/18	A/Bris 02/18
		Passage history			Egg	MDCK	MDCK	Egg	MDCK	Egg	Egg	MDCK	MDCK	Egg
		Ferret number			F31/16*1	F09/15*1	F13/18*1	NIB F48/16*1	F03/18*2	F20/18*1	F23/18*1	F04/19*1	F08/19*1	F09/19*1
		Genetic group			6B.1			6B.1	6B.1	6B.1	6B.1	6B.1	6B.1	6B.1A1
REFERENCE VIRUSES														
A/Michigan/45/2015		6B.1	2015-09-07	E3/E3	640	320	160	640	1280	640	320	1280	1280	640
A/Bayern/69/2009	G155E		2009-07-01	MDCK5/MDCK1	80	320	320	40	320	40	40	320	40	80
A/Lviv/N6/2009	G155E, D222G		2009-10-27	MDCK4/SIAT1/MDCK3	320	1280	1280	160	640	160	160	1280	160	320
A/Slovenia/2903/2015	clone 37	6B.1	2015-10-26	E4/E2	640	320	160	1280	1280	640	320	1280	1280	640
A/Paris/1447/2017		6B.1A	2017-10-20	MDCK1/MDCK3	640	160	80	1280	1280	640	320	2560	1280	640
A/Switzerland/2656/2017		6B.1A	2017-12-21	E5/E2	1280	640	320	1280	2560	1280	640	2560	1280	1280
A/Switzerland/3330/2017	clone 35	6B.1A5	2017-12-20	E6/E2	320	160	80	640	1280	640	320	1280	640	320
A/Norway/3433/2018		6B.1A5	2018-10-30	MDCK3	320	80	40	320	640	160	160	1280	320	320
A/Ireland/84630/2018		6B.1A6	2018-11-28	MDCK1/MDCK3	320	160	80	640	1280	320	160	1280	640	640
A/Brisbane/02/2018		6B.1A1	2018-01-04	E3/E1	1280	640	320	1280	5120	1280	640	2560	2560	640
TEST VIRUSES														
A/England/620/2018			2018-11-12	SIAT2/MDCK1	640	160	80	640	1280	640	320	1280	640	640
A/Hamburg/5/2018			2018-11-28	P2/MDCK1	320	80	80	320	640	160	80	1280	640	320
A/Bayern/50/2018			2018-12-06	P1/MDCK1	1280	320	160	1280	2560	640	320	2560	1280	1280
A/Berlin/58/2018			2018-12-07	P1/MDCK1	640	320	160	640	1280	320	320	2560	1280	640
A/Hamburg/6/2018			2018-12-10	P1/MDCK1	320	160	160	320	640	160	160	1280	640	320
A/Hamburg/7/2018			2018-12-17	P1/MDCK1	640	320	160	640	1280	320	320	2560	1280	640
A/England/733/2018			2018-12-17	SIAT2/MDCK1	160	80	80	80	320	40	80	640	80	160
A/Toulon/2307/2018			2018-12-18	MDCK2/MDCK1	640	160	80	640	1280	320	160	1280	640	640
A/England/700/2018			2018-12-21	SIAT1/MDCK1	320	160	160	320	640	160	160	1280	640	320
A/Saint-Etienne/2322/2018			2018-12-23	MDCK2/MDCK1	320	80	80	320	640	160	160	1280	320	320
A/England/731/2018			2018-12-24	SIAT1/MDCK1	80	160	80	40	320	80	<	320	40	160
A/England/732/2018			2018-12-26	SIAT1/MDCK1	80	160	40	40	160	40	40	1280	40	160
A/Mecklenburg-Vorpommern/5/2018			2018-12-27	P1/MDCK1	1280	320	160	2560	2560	1280	640	2560	2560	1280
A/Lyon/4/2019			2018-12-27	MDCK2/MDCK1	640	160	160	640	1280	320	160	2560	640	640
A/England/708/2018			2018-12-27	SIAT1/MDCK1	640	320	160	640	1280	320	320	1280	1280	640
A/Grenoble/43/2019			2018-12-28	MDCK2/MDCK1	1280	320	160	1280	2560	640	320	2560	1280	640
A/Lyon/CHU/R18.134.99/2018			2018-12-29	MDCK2/MDCK1	640	160	160	640	1280	320	160	1280	640	640
A/Lyon/6/2019			2018-12-29	MDCK2/MDCK1	640	160	160	1280	2560	640	320	2560	1280	640
A/Lyon/10/2019			2018-12-31	MDCK2/MDCK1	1280	320	160	1280	2560	640	640	2560	2560	1280
A/Lyon/CHU/R19.01.51/2019			2019-01-01	MDCK2/MDCK1	1280	320	320	2560	2560	1280	640	2560	2560	2560
A/Niedersachsen/1/2019			2019-01-02	P1/MDCK1	1280	320	160	1280	2560	1280	320	2560	2560	1280
A/Lyon/CHU/R19.03.40/2019			2019-01-03	MDCK2/MDCK1	640	320	160	1280	1280	640	320	2560	640	640
A/England/1/2019			2019-01-03	SIAT1/MDCK1	640	320	160	640	1280	320	320	2560	640	640
A/Lyon/CHU/R19.04.49/2019			2019-01-05	MDCK2/MDCK1	640	160	40	320	1280	320	160	1280	640	320
A/Annecy/35/2019			2019-01-06	MDCK2/MDCK1	640	320	160	1280	1280	640	320	2560	1280	1280
A/Gueret/64/2019			2019-01-07	MDCK2/MDCK1	640	160	80	640	1280	320	160	2560	640	640
A/Lyon/HFME/47/2019			2019-01-07	MDCK2/MDCK1	640	160	80	640	1280	320	320	2560	640	640
A/England/2/2019			2019-01-07	SIAT1/MDCK1	640	320	320	640	1280	640	320	2560	1280	1280
A/Lyon/96/2019			2019-01-08	MDCK2/MDCK1	640	320	80	1280	1280	640	320	2560	1280	1280
A/Lyon/128/2019			2019-01-09	MDCK2/MDCK1	1280	320	160	1280	2560	640	320	2560	1280	1280
A/Saint-Etienne/137/2019			2019-01-11	MDCK2/MDCK1	2560	640	160	1280	5120	1280	640	5120	2560	2560
					Vaccine NH 2018-19									Vaccine NH 2019-20
					SH 2019									

*Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used)
1 < = <40; 2 < = <80
Sequences in phylogenetic trees

Table 3-6. Antigenic analysis of A(H1N1)pdm09 viruses by HI

Viruses	Other information	Passage history	Collection date	Passage history	Haemagglutination inhibition titre: Post-infection ferret antisera									
					A/Mich 45/15	A/Bayern 69/09	A/Lviv N6/09	A/Slov 2903/2015	A/Paris 1447/17	A/Swit 2656/17	A/Swit 3330/17	A/Norway 3433/18	A/Ire 84630/18	A/Bris 02/18
		Passage history			Egg	MDCK	MDCK	Egg	MDCK	Egg	Egg	MDCK	MDCK	Egg
		Ferret number			F31/16*1	F09/15*1	F13/18*2	NIB F48/16*1	F03/18*2	F20/18*1	F23/18*1	F04/19*1	F08/19*1	F09/19*1
		Genetic group			6B.1			6B.1	6B.1A	6B.1A	6B.1A5	6B.1A5	6B.1A6	6B.1A1
REFERENCE VIRUSES														
A/Michigan/45/2015		6B.1	2015-09-07	E3/E3	640	320	320	1280	2560	640	320	1280	1280	1280
A/Bayern/69/2009	G155E		2009-07-01	MDCK5/MDCK1	<	320	160	<	160	40	40	320	40	80
A/Lviv/N6/2009	G155E, D222G		2009-10-27	MDCK4/SIAT1/MDCK3	160	640	1280	160	1280	320	320	1280	320	640
A/Slovenia/2903/2015	clone 37	6B.1	2015-10-26	E4/E2	640	320	320	1280	2560	640	320	2560	1280	1280
A/Paris/1447/2017		6B.1A	2017-10-20	MDCK1/MDCK3	320	160	40	640	1280	320	160	1280	640	640
A/Switzerland/2656/2017		6B.1A	2017-12-21	E5/E2	640	320	320	1280	2560	640	320	2560	1280	640
A/Switzerland/3330/2017	clone 35	6B.1A5	2017-12-20	E6/E2	160	160	80	320	640	320	320	640	640	320
A/Norway/3433/2018		6B.1A5	2018-10-30	MDCK3	320	160	<	320	1280	160	160	1280	320	320
A/Ireland/84630/2018		6B.1A6	2018-11-28	MDCK1/MDCK3	320	160	80	640	1280	320	160	1280	640	640
A/Brisbane/02/2018		6B.1A1	2018-01-04	E3/E1	640	320	320	1280	2560	640	320	1280	1280	640
TEST VIRUSES														
A/Greece/1519/2018				MDCK1/MDCK1	640	160	80	640	1280	320	320	2560	640	320
A/Greece/1589/2018				MDCK1/MDCK1	1280	640	320	1280	2560	640	640	2560	2560	1280
A/Croatia/5299/2018			2018-11-28	MDCKx/MDCK1	640	320	80	640	2560	640	640	1280	1280	640
A/Croatia/5331/2018			2018-12-03	MDCKx/MDCK1	2560	640	320	2560	5120	1280	1280	2560	2560	2560
A/Croatia/5327/2018			2018-12-03	MDCKx/MDCK1	640	160	80	640	2560	320	640	1280	1280	640
A/Bucuresti/238999/2018			2018-12-07	SIAT2/MDCK1	640	160	160	640	1280	320	640	1280	640	640
A/Bucuresti/239045/2018			2018-12-10	SIAT1/MDCK1	320	160	80	320	640	160	320	1280	640	320
A/Greece/1585/2018			2018-12-18	MDCK1/MDCK1	2560	640	320	1280	2560	1280	640	2560	2560	1280
A/Greece/1607/2018			2018-12-27	MDCK1/MDCK1	640	320	320	640	1280	640	640	2560	640	640
A/Greece/1619/2018			2018-12-28	MDCK1/MDCK1	1280	640	320	1280	2560	640	640	2560	2560	1280
A/Greece/1617/2018			2018-12-28	MDCK1/MDCK1	1280	320	320	640	1280	320	640	2560	1280	640
A/Greece/1616/2018			2018-12-31	MDCK1/MDCK1	1280	320	320	1280	1280	640	640	2560	1280	640
A/Bucuresti/239594/2019			2019-01-07	MDCK1	1280	320	160	1280	2560	640	640	2560	1280	1280
A/Saarland/1/2019			2019-01-07	C1/MDCK1	640	160	80	640	640	320	320	2560	1280	640
A/Rheinland-Pfalz/1/2019			2019-01-10	C1/MDCK1	640	320	320	640	640	320	640	2560	1280	640
A/Bucuresti/239839/2019			2019-01-11	MDCK2	640	320	160	1280	2560	640	640	2560	1280	640
A/Thuringen/1/2019			2019-01-11	C1/MDCK1	1280	320	320	640	640	320	640	2560	1280	640
A/Greece/144/2019			2019-01-14	MDCK1	640	160	40	640	1280	320	320	2560	640	640
A/Berlin/7/2019			2019-01-14	C1/MDCK1	640	320	320	1280	640	320	640	2560	1280	640
A/Brandenburg/2/2019			2019-01-15	C1/MDCK1	1280	320	80	1280	2560	640	1280	2560	1280	1280
A/Greece/168/2019			2019-01-17	MDCK1	1280	320	160	640	1280	640	640	1280	1280	640
A/Nordrhein-Westfalen/9/2019			2019-01-17	C1/MDCK1	1280	320	160	1280	1280	640	640	2560	1280	640
A/Rheinland-Pfalz/3/2019			2019-01-17	C1/MDCK1	640	320	160	640	1280	320	640	2560	640	640
A/Greece/199/2019			2019-01-18	MDCK1	1280	320	160	1280	2560	640	640	2560	1280	640
A/Greece/232/2019			2019-01-21	MDCK1	640	320	160	1280	1280	640	320	2560	1280	640
A/Baden-Wurttemberg/7/2019			2019-01-21	C1/MDCK1	640	160	80	640	1280	320	640	1280	640	640
A/Sachsen/5/2019			2019-01-21	C1/MDCK1	640	320	160	640	1280	320	640	2560	640	640
A/Thuringen/8/2019			2019-01-28	C1/MDCK1	640	160	160	640	1280	320	320	1280	640	640
A/Bremen/7/2019			2019-01-31	C1/MDCK1	1280	320	160	640	1280	640	640	2560	1280	640
A/Berlin/26/2019			2019-02-04	C1/MDCK1	2560	640	160	1280	2560	1280	1280	2560	2560	2560
A/Niedersachsen/31/2019			2019-02-05	C1/MDCK1	1280	320	160	640	1280	320	640	2560	640	640
A/Hamburg/1/2019			2019-02-11	C1/MDCK1	640	320	320	640	640	320	640	2560	1280	640
A/Sachsen/28/2019			2019-02-11	C1/MDCK1	640	160	160	640	640	320	640	2560	1280	640
A/Thuringen/16/2019			2019-02-11	C1/MDCK1	640	160	80	640	640	320	320	1280	1280	640
A/Nordrhein-Westfalen/63/2019			2019-02-18	C1/MDCK1	640	320	160	640	640	320	640	1280	640	640
A/Bayern/68/2019			2019-02-19	C1/MDCK1	640	160	80	320	320	160	160	1280	320	320
A/Rheinland-Pfalz/27/2019			2019-02-21	C1/MDCK1	640	160	160	1280	640	640	640	2560	1280	640
A/Brandenburg/18/2019			2019-02-28	C1/MDCK1	640	80	80	640	640	320	320	1280	640	320

*Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used)
1 < = <40; 2 < = <80
Sequences in phylogenetic trees

Vaccine NH 2018-19
SH 2019

Vaccine NH 2019-20

Table 3-7. Antigenic analysis of A(H1N1)pdm09 viruses by HI – Summary all test viruses

Viruses	Other information		Haemagglutination inhibition titre												
			Post-infection ferret antisera												
		Passage history	A/Mich 45/15 Egg	A/Cal 7/09 Egg	A/Bayern 69/09 MDCK	A/Lviv N6/09 MDCK	A/Astrak 1/11 MDCK	A/HK 5659/12 MDCK	A/Slov 2903/15 Egg	A/Paris 1447/17 MDCK	A/Swit 2656/17 Egg	A/Swit 3330/17 Egg	A/Norway 3433/18 MDCK	A/Ire 84630/18 MDCK	A/Bris 02/18 Egg
		Ferret number	NIB F42/16[*1]	F07/16[*1]	F09/15[*1]	F13/18[*1]	F22/13[*1]	F17/15[*1]	NIB F48/16[*1]	F03/18[*2]	F20/18[*1]	F23/18[*1]	F04/19*1	F08/19[*1]	F09/19[*1]
		Genetic group	6B.1				5	6A	6B.1	6B.1A	6B.1A	6B.1A5	6B.1A5	6B.1A6	6B.1A1
REFERENCE VIRUSES															
A/Michigan/45/2015		6B.1	640	640	320	320	640	320	1280	1280	640	320	1280	1280	1280
A/California/7/2009	clone 38-32		640	320	320	320	320	320	640	1280	640	160	1280	ND	ND
A/Bayern/69/2009	G155E		<	<	320	320	<	<	<	320	40	40	320	40	80
A/Lviv/N6/2009	G155E, D222G		80	80	1280	1280	80	80	160	640	160	160	1280	320	640
A/Astrakhan/1/2011		5	640	320	640	640	640	320	1280	2560	640	320	2560	ND	ND
A/Hong Kong/5659/2012		6A	640	320	160	160	320	320	640	1280	320	320	1280	ND	ND
A/Slovenia/2903/2015	clone 37	6B.1	640	320	320	320	320	320	640	1280	640	320	1280	1280	1280
A/Paris/1447/2017		6B.1A	640	320	320	160	320	320	640	2560	640	320	2560	640	640
A/Switzerland/2656/2017		6B.1A	640	320	640	640	320	320	1280	1280	1280	320	2560	1280	640
A/Switzerland/3330/2017	clone 35	6B.1A5	320	160	160	160	160	160	320	1280	320	320	1280	640	320
A/Norway/3433/2018		6B.1A5	640	160	160	80	160	160	640	1280	320	160	1280	320	320
A/Ireland/84630/2018		6B.1A6	320	ND	160	80	ND	ND	640	1280	320	160	1280	640	640
A/Brisbane/02/2018		6B.1A1	640	ND	320	320	ND	ND	1280	2560	640	320	1280	1280	640
TEST VIRUSES															
Number of viruses tested*			149	44	149	149	44	44	149	149	149	149	149	133	73
No with titre reduction ≤2-fold			146	42	144	5	37	41	135	144	114	145	148	123	70
%			*98.0*	*95.5*	*96.6*	*3.4*	*77.3*	*93.2*	*90.6*	*96.6*	*76.5*	*97.4*	*99.3*	*92.5*	*95.9*
No with titre reduction =4-fold			1	2	5	34	7	3	11	4	28	2	1	7	3
%			*0.7*	*4.5*	*3.4*	*22.8*	*22.7*	*6.8*	*7.4*	*2.7*	*18.8*	*1.3*	*0.7*	*5.3*	*4.1*
No with titre reduction ≥8-fold			2			110			3	1	7	2		3	
%			*1.3*			*73.8*			*2.0*	*0.7*	*4.7*	*1.3*		*2.2*	
			Vaccine NH 2018-19 SH 2019												Vaccine NH 2019-20

* Of those with available HA sequence, all were clade 6B.1A

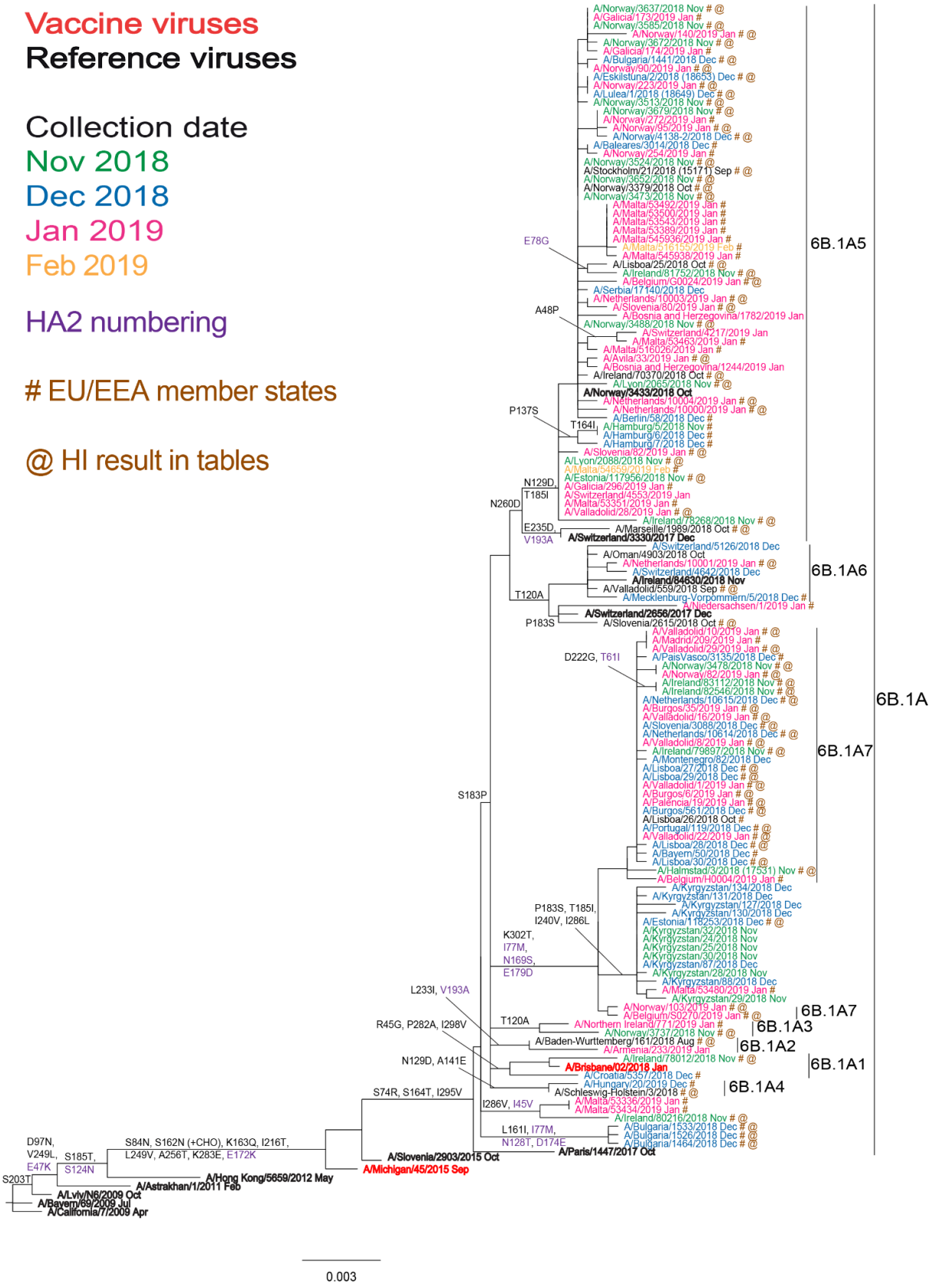
ND = Not Done

Reference virus results are taken from an individual table as an example. Summaries for each antiserum are based on fold-reductions observed on the days that HI assays were performed.

Table 3-8. Antigenic analysis of A(H1N1)pdm09 viruses by HI – Summary by test virus genetic group

Viruses		Haemagglutination inhibition titre												
		Post-infection ferret antisera												
	Passage history	A/Mich 45/15 Egg	A/Cal 7/09 Egg	A/Bayern 69/09 MDCK	A/Lviv N6/09 MDCK	A/Astrak 1/11 MDCK	A/HK 5659/12 MDCK	A/Slov 2903/15 Egg	A/Paris 1447/17 MDCK	A/Swit 2656/17 Egg	A/Swit 3330/17 Egg	A/Norway 3433/18 MDCK	A/Ire 84630/18 MDCK	A/Bris 02/18 Egg
	Ferret number	NIB F42/16[*1]	F07/16[*1]	F09/15[*1]	F13/18[*1]	F22/13[*1]	F17/15[*1]	NIB F48/16[*1]	F03/18[*2]	F20/18[*1]	F23/18[*1]	F04/19*1	F08/19[*1]	F09/19[*1]
	Genetic group	6B.1				5	6A	6B.1	6B.1A	6B.1A	6B.1A5	6B.1A5	6B.1A6	6B.1A1
TEST VIRUSES														
Total number tested		75	43	75	75	43	43	75	75	75	75	75	60	1
Number tested	6B.1A	6	2	6	6	2	2	6	6	6	6	6	5	
No with titre reduction ≤2-fold		6	2	6		2	2	6	6	6	6	6	5	
No with titre reduction =4-fold					1									
No with titre reduction ≥8-fold					5									
Number tested	6B.1A1	1	1	1	1	1	1	1	1	1	1	1		
No with titre reduction ≤2-fold		1	1	1			1	1	1		1	1		
No with titre reduction =4-fold						1				1				
No with titre reduction ≥8-fold					1									
Number tested	6B.1A2	1	1	1	1	1	1	1	1	1	1	1	1	
No with titre reduction ≤2-fold		1	1	1		1	1	1	1	1	1	1	1	
No with titre reduction =4-fold					1									
Number tested	6B.1A3	1		1	1			1	1	1	1	1	1	
No with titre reduction ≤2-fold		1		1	1			1	1	1	1	1	1	
Number tested	6B.1A4	1	1	1	1	1	1	1	1	1	1	1	1	
No with titre reduction ≤2-fold		1	1	1		1	1	1	1	1	1	1	1	
No with titre reduction =4-fold														
No with titre reduction ≥8-fold					1									
Number tested	6B.1A5	34	14	34	34	14	14	34	34	34	34	34	25	1
No with titre reduction ≤2-fold		34	12	33	1	10	13	32	33	23	34	34	20	1
%		*100*	*85.7*	*97.1*	*2.9*	*71.4*	*92.9*	*94.1*	*97.1*	*67.6*	*100*	*100*	*80.0*	
No with titre reduction =4-fold			2	1	11	4	1	2	1	11			5	
%			*14.3*	*2.9*	*32.4*	*28.6*	*7.1*	*5.9*	*2.9*	*32.4*			*20.0*	
No with titre reduction ≥8-fold					22									
%					*64.7*									
Number tested	6B.1A6	2	2	2	2	2	2	2	2	2	2	2	1	
No with titre reduction ≤2-fold		2	2	2		1	2	2	2	2	2	2	1	
No with titre reduction =4-fold						1								
No with titre reduction ≥8-fold					2									
Number tested	6B.1A7	29	22	29	29	22	22	29	29	29	29	29	26	1
No with titre reduction ≤2-fold		29	22	29	1	21	20	29	29	27	29	29	25	1
%		*100*	*100*	*100*	*3.4*	*95.5*	*90.9*	*100*	*100*	*93.1*	*100*	*100*	*96.2*	
No with titre reduction =4-fold					8	1	2			2			1	
%					*27.6*	*4.5*	*9.1*			*6.9*			*3.8*	
No with titre reduction ≥8-fold					20									
%					*69.0*									
		Vaccine NH 2018-19 SH 2019												Vaccine NH 2019-20

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



Influenza A(H3N2) virus analyses

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

Since the February 2019 characterisation report of the viruses recovered, only 46 retained sufficient HA activity to allow antigenic analysis by HI based on positive neuraminidase activity (Tables 4-2 to 4-6). Table 4-1 is repeated from the February 2019 characterisation report, but with genetic group data now included. Of the 46 test viruses, only four were recognised at titres within fourfold of the homologous titre by the antiserum raised against the currently used vaccine virus, egg-propagated A/Singapore/INFIMH-16-0019/2016 (subclade 3C.2a1). Test viruses were analysed with antisera raised against three cell culture-propagated subgroup 3C.2a1b viruses, for which no homologous titres are given due to the inability of these cell culture-propagated reference viruses to agglutinate RBCs. Those raised against A/La Rioja/2202/2018, A/Netherlands/10260/2018 and A/Norway/3275/2018 recognised 9/46 (20%), 5/20 (25%) and 9/42 (21%) test viruses respectively at titres of ≥160. An antiserum raised against egg-propagated A/Netherlands/10260/2018 recognised test viruses poorly: all 20 yielded titres at least 32-fold reduced compared to the homologous titre.

Antisera raised against subclade 3C.2a2 viruses generally recognised the test viruses poorly. Those raised against cell culture-propagated A/Bretagne/1413/2017 recognised none of the 46 test viruses at titres within fourfold of the homologous titre. In addition, those raised against egg-propagated A/Switzerland/8060/2017, the vaccine virus recommended for use in the 2019 southern hemisphere season, recognised only 4/46 (9%) test viruses at a titre within fourfold of the homologous titre.

Antiserum raised against a cell culture-propagated clade 3C.2a virus, A/Hong Kong/5738/2014, recognised all 43/46 (93%) test viruses at titres within fourfold of the homologous titre and 19 (41%) within twofold. Antisera raised against cell culture-propagated cultivars of A/Stockholm/6/2014 and A/England/538/2018, clade 3C.3a viruses, recognised 37/46 (80%) and 36/46 (78%) test viruses respectively at titres within fourfold of the titres of the antisera with their homologous viruses and 21/46 (46%) and 35/46 (76%) within twofold.

A summary of the HI data presented in Tables 4-1 to 4-6 is presented in Table 4-7. For test viruses with known HA sequences at the time this report was prepared, these results are broken down by virus clade/subclade in Table 4-8. It shows the poor recognition of test viruses by post-infection ferret antisera raised against egg-propagated vaccine/reference viruses, poor cross-reactivity of antisera raised against subclade 3C.2a2 viruses, antigenic drift in the clade 3C.3a viruses from 2014–2018 with the response to A/England/538/2018 being more clade 3C.3a-specific and that the antisera raised against cell culture-propagated viruses that raised against A/Hong Kong/5738/2014 gives the broadest cross-clade/subclade reactivity.

HA gene sequences of the test viruses characterised antigenically in the February 2019 report are now available. The genetic clades are shown in Table 4-1 and most are included in the HA phylogenetic analysis (Figure 2). Viruses in clades 3C.2a and 3C.3a have circulated since the 2013–14 northern hemisphere influenza season, with clade 3C.2a viruses having dominated since the 2014–15 influenza season, notably subclade 3C.2a2 viruses, though subgroup 3C.2a1b viruses have predominated in recent months (Figure 2). The HA gene sequences of viruses in both clades continue to diverge. Notably, clade 3C.3a viruses have evolved to carry HA1 amino acid substitutions of L3I, S91N, N144K (loss of a N-linked glycosylation motif at residues 144-146), F193S and K326R compared to A/Stockholm/6/2014 and the number of detections in January 2019 has increased in certain WHO European region countries (Belgium, France, Germany, Israel, Netherlands and Spain; Figure 2) and North America. New genetic groups have also emerged among the clade 3C.2a viruses, designated as subclades/subgroups. Amino acid substitutions that define these subclades/subgroups are:

- Clade 3C.2a – 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) and Q311H in HA1 and D160N in HA2, e.g. A/Hong Kong/7295/2014 a cell culture-propagated surrogate for A/Hong Kong/4801/2014 (a former vaccine virus).
- Subclade 3C.2a1 – those in clade 3C.2a plus N171K in HA1 and I77V and G155E in HA2; most also carry N121K in HA1, e.g. A/Singapore/INFIMH-16-0019/2016 (2018–19 northern hemisphere vaccine virus).
- Subgroup 3C.2a1a – those in subclade 3C.2a1 plus T135K in HA1, resulting in the loss of a potential glycosylation site, and also G150E in HA2, e.g. A/Greece/4/2017.
- Subgroup 3C.2a1b – those in subclade 3C.2a1 plus K92R and H311Q in HA1, e.g. A/La Rioja/2202/2018, with many viruses in this subgroup carrying additional HA1 amino acid substitutions.
- Subclade 3C.2a2 – those in clade 3C.2a plus T131K, R142K and R261Q in HA1, e.g.

[2] For example, the September 2013 report: European Centre for Disease Prevention and Control. Influenza virus characterisation – Summary Europe, September 2013. Stockholm: ECDC; 2014. Available from: http://ecdc.europa.eu/publications-data/influenza-virus-characterisation-september-2013

[3] European Centre for Disease Prevention and Control. Influenza virus characterisation – Summary Europe, November 2014. Stockholm: ECDC; 2014. Available from: http://ecdc.europa.eu/publications-data/influenza-virus-characterisation-november-2014

A/Switzerland/8060/2017 (2019 southern hemisphere vaccine virus).

- Subclade 3C.2a3 – those in clade 3C.2a plus N121K and S144K in HA1, e.g. A/Cote d'Ivoire/544/2016
- Subclade 3C.2a4 – those in clade 3C.2a plus N31S, D53N, R142G, S144R, N171K, I192T, Q197H and A304T in HA1 and S113A in HA2, e.g. A/Valladolid/182/2017.
- Clade 3C.3a – T128A (resulting in the loss of a potential glycosylation site), R142G and N145S in HA1 which defined clade 3C.3 plus A138S, F159S and N225D in HA1, many with K326R, e.g. A/England/538/2018.

Globally, the majority of viruses with collection dates from 1 September 2018 have HA genes that continue to fall into genetic groups within clade 3C.2a, with those in subgroup 3C.2a1b having been more numerous than those in subclade 3C.2a2 from September 2018–February 2019 (Figure 2). Notably, a significant number of the subgroup 3C.2a1b viruses have fallen in two recently emerged clusters: one defined by amino acid substitutions T131K and K135T (a reversion resulting in re-establishment of the 133-135 glycosylation sequon) in HA1 with V200I in HA2 and the other by T128A substitution in HA1 (resulting in loss of a potential glycosylation sequon). Furthermore, as indicated above, the number of clade 3C.3a virus detections has increased in recent weeks in a number of countries/regions.

The locations of A/Singapore/INFIMH-16-0019/2016 (3C.2a1), the A(H3N2) virus recommended for inclusion in vaccines for the northern hemisphere 2018–19 influenza season [3], A/Switzerland/8060/2017 (3C.2a2), the A(H3N2) virus recommended for inclusion in vaccines for the southern hemisphere 2019 influenza season [4], and A/Kansas/14/2017, the A(H3N2) virus recommended for inclusion in vaccines for the northern hemisphere 2019–20 influenza season [1,2], are indicated in Figure 2.

Table 4-1. Antigenic analysis of A(H3N2) viruses by HI

Viruses	Other information	Passage history / Ferret number / Genetic group	Collection date	Passage history	Haemagglutination inhibition titre – Post-infection ferret antisera									
					A/Stock 6/14 SIAT F14/14*1 3C.3a	A/HK 5738/14 MDCK F30/14*1 3C.2a	A/Bretagne 1413/17 SIAT F01/18*1 3C.2a2	A/Singapore 0019/16 Egg 10-4 F46/17*1 3C.2a1	A/La Rioja 2202/18 SIAT F26/18*1 3C.2a1b	A/Swit 8060/17 Egg F27/18*1 3C.2a2	A/Eng 538/18 SIAT F31/18*1 3C.3a	A/Neth 10260/18 Egg F02/19*1 3C.2a1b	A/Neth 10260/18 SIAT F07/19*1 3C.2a1b	A/Norway 3275/18 SIAT F03/19*1 3C.2a1b
REFERENCE VIRUSES														
A/Stockholm/6/2014		3C.3a	2014-02-06	SIAT1/SIAT3	160	40	80	160	80	160	160	<	40	80
A/Hong Kong/5738/2014		3C.2a	2014-04-30	MDCK1/MDCK2/SIAT2	160	80	160	160	80	160	160	<	80	160
A/Bretagne/1413/2017		3C.2a2	2017-10-09	MDCK1/SIAT4	80	<	640	160	80	640	80	<	80	160
A/Singapore/INFIMH-16-0019/2016		3C.2a1	2016-04-14	E5/E2	<	<	40	640	80	80	40	<	40	<
A/Switzerland/8060/2017	clone 57	3C.2a2	2017-12-12	E7/E1	<	40	1280	320	80	1280	80	40	80	40
A/England/538/2018		3C.3a	2018-02-26	MDCK1/SIAT3	40	<	40	80	<	40	640	<	80	<
A/Netherlands/10260/2018		3C.2a1b	2018-02-15	E5/E1	40	<	80	80	320	80	80	1280	320	160
TEST VIRUSES														
A/Parma/177/2018		3C.2a1b	2018-11-20	SIAT2/SIAT2	160	80	40	160	320	160	80	<	320	160
A/Parma/180/2018		3C.2a1b	2018-11-21	SIAT1/SIAT2	80	40	<	80	80	40	40	<	160	80
A/Parma/179/2018		3C.2a1b	2018-11-22	SIAT1/SIAT1	160	40	<	80	160	40	40	<	160	80
A/Parma/174/2018		3C.2a1b	2018-11-22	SIAT1/SIAT1	160	40	<	40	160	40	40	<	160	80
A/England/630/2018		3C.2a1b	2018-11-23	SIAT1/SIAT1	80	40	160	80	80	160	40	<	80	160
A/Palermo/327/2018		3C.2a	2018-11-28	SIAT3/SIAT1	160	40	640	160	160	640	160	<	80	160
A/England/646/2018		3C.2a1b	2018-11-28	SIAT2/SIAT1	<	<	<	<	80	<	<	<	80	80
A/Constanta/239165/2018		3C.2a1b	2018-12-07	SIAT1/SIAT2	<	<	<	<	80	<	<	<	40	40
A/England/754/2018		3C.2a1b	2018-12-13	MDCK1/SIAT1	<	<	<	<	<	40	80	<	<	<
A/Nord Pas de Calais/2726/2018		3C.2a3	2018-12-26	SIAT1	80	<	<	40	80	80	40	<	40	40
A/Bourgogne/074/2019		3C.3a	2019-01-02	SIAT1	40	<	40	<	<	<	320	<	40	<
A/Centre/013/2019		3C.2a1b	2019-01-02	SIAT1	160	<	<	80	160	<	<	<	160	40
A/Pays de Loire/040/2019		3C.2a1b	2019-01-04	SIAT1	80	<	80	40	160	80	40	<	80	160
A/Haute Normandie/085/2019		3C.3a	2019-01-07	SIAT1	<	<	<	<	<	<	320	<	40	<
A/Nord Pas de Calais/054/2019		3C.2a1b	2019-01-07	SIAT1	40	<	<	80	160	40	40	<	80	80
A/England/3/2019		3C.2a1b	2019-01-07	MDCK1/SIAT1	<	<	<	40	<	40	80	<	<	<
A/Paris/105/2019		3C.2a1b	2019-01-08	SIAT1	<	<	<	<	80	<	<	<	80	40
A/Iasi/239836/2019		3C.2a1b	2019-01-09	SIAT1	80	40	80	80	160	80	40	<	80	160
								Vaccine SH 2018 NH 2018-19		Vaccine SH 2019				

* Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used) [1] < = <40; ND = Not Done

Sequences in phylogenetic trees

Table 4-2. Antigenic analysis of A(H3N2) viruses by HI

Viruses	Other information		Collection date	Passage history	Haemagglutination inhibition titre								
					Post-infection ferret antisera								
					A/Stock 6/14	A/HK 5738/14	A/Bretagne 1413/17	A/Singapore 0019/16	A/La Rioja 2202/18	A/Swit 8060/17	A/Eng 538/18	A/Neth 10260/18	A/Neth 10260/18
		Passage history			SIAT	MDCK	SIAT	Egg 10^{-4}	SIAT	Egg	SIAT	Egg	SIAT
		Ferret number			F14/14[*1]	St J F60/17[*1]	F01/18*[1]	F46/17*[1]	F26/18[*1]	F27/18[*1]	F31/18[*1]	F02/19[*1]	F07/19[*1]
		Genetic group			3C.3a	3C.2a	3C.2a2	3C.2a1	3C.2a1b	3C.2a2	3C.3a	3C.2a1b	3C.2a1b
REFERENCE VIRUSES													
A/Stockholm/6/2014		3C.3a	2014-02-06	SIAT1/SIAT2	160	40	80	160	80	160	160	<	80
A/Hong Kong/5738/2014		3C.2a	2014-04-30	MDCK1/MDCK2/SIAT2	160	80	80	160	80	160	160	<	40
A/Bretagne/1413/2017		3C.2a2	2017-10-09	MDCK1/SIAT4	160	80	640	160	80	640	160	<	80
A/Singapore/INFIMH-16-0019/2016		3C.2a1	2016-04-14	E5/E2	<	40	40	640	160	160	80	<	40
A/Switzerland/8060/2017	clone 57	3C.2a2	2017-12-12	E7/E1	40	80	1280	640	80	1280	80	40	80
A/England/538/2018		3C.3a	2018-02-26	MDCK1/SIAT3	80	<	40	80	40	80	640	<	80
A/Netherlands/10260/2018		3C.2a1b	2018-02-15	E5/E1	<	40	80	80	320	80	80	1280	320
TEST VIRUSES													
A/Nordrhein-Westfalen/41/2019		3C.2a1b	2019-01-04	C1/SIAT1	80	<	<	80	160	80	80	<	160
A/Nordrhein-Westfalen/11/2019		3C.3a	2019-01-21	C1/SIAT1	80	<	40	80	<	80	640	<	80
A/Nordrhein-Westfalen/22/2019		3C.3a	2019-01-28	C1/SIAT1	80	<	40	80	<	80	640	<	80
A/Berlin/23/2019		3C.3a	2019-01-31	C1/SIAT1	80	<	40	80	<	80	640	<	80
								Vaccine SH 2018 NH 2018-19		Vaccine SH 2019			

* Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used) [1] < = <40; ND = Not Done

Sequences in phylogenetic trees

Table 4-3. Antigenic analysis of A(H3N2) viruses by HI

Viruses	Other information		Collection date	Passage history	Haemagglutination inhibition titre									
					Post-infection ferret antisera									
		Passage history			A/Stock 6/14	A/HK 5738/14	A/Bretagne 1413/17	A/Singapore 0019/16	A/La Rioja 2202/18	A/Swit 8060/17	A/Eng 538/18	A/Neth 10260/18	A/Neth 10260/18	A/Norway 3275/18
		Ferret number			SIAT F14/14*1	MDCK St J F60/17*1	SIAT F01/18*1	Egg 10-4 F46/17*1	SIAT F26/18*1	Egg F27/18*1	SIAT F31/18*1	Egg F02/19*1	SIAT F07/19*1	SIAT F03/19*1
		Genetic group			3C.3a	3C.2a	3C.2a2	3C.2a1	3C.2a1b	3C.2a2	3C.3a	3C.2a1b	3C.2a1b	3C.2a1b
REFERENCE VIRUSES														
A/Stockholm/6/2014		3C.3a	2014-02-06	SIAT1/SIAT3	160	160	80	320	80	320	320	<	80	80
A/Hong Kong/5738/2014		3C.2a	2014-04-30	MDCK1/MDCK2/SIAT2	160	160	160	320	160	320	320	40	80	320
A/Bretagne/1413/2017		3C.2a2	2017-10-09	MDCK1/SIAT4	160	160	640	320	80	640	320	40	80	160
A/Singapore/INFIMH-16-0019/2016		3C.2a1	2016-04-14	E5/E2	40	160	80	640	160	160	80	40	40	40
A/Switzerland/8060/2017	clone 57	3C.2a2	2017-12-12	E7/E1	40	160	1280	640	160	1280	160	40	80	80
A/England/538/2018		3C.3a	2018-02-26	MDCK1/SIAT3	40	40	40	40	<	40	640	10	80	<
A/Netherlands/10260/2018		3C.2a1b	2018-02-15	E5/E1	40	160	160	160	640	160	80	1280	640	160
TEST VIRUSES														
A/Luxembourg/341/2019		3C.2a1b	2019-01-03	SIAT1	80	80	<	80	160	40	40	<	80	160
A/Luxembourg/1177/2019		3C.2a1b	2019-01-06	SIAT1	<	40	<	<	80	<	80	<	160	160
A/Luxembourg/2293/2019		3C.2a1b	2019-01-14	SIAT1	160	80	40	80	160	80	80	<	160	320

*Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used) [1] < = <40; ND = Not Done

Sequences in phylogenetic trees

Vaccine SH 2018 NH 2018-19

Vaccine SH 2019

Table 4-4. Antigenic analysis of A(H3N2) viruses by HI

Viruses	Other information	Passage history / Ferret number / Genetic group	Collection date	Passage history	Haemagglutination inhibition titre – Post-infection ferret antisera									
					A/Stock 6/14 SIAT F14/14*1 3C.3a	A/HK 5738/14 MDCK St J F60/17*1 3C.2a	A/Bretagne 1413/17 SIAT F01/18*1 3C.2a2	A/Singapore 0019/16 Egg 10-4 F46/17*1 3C.2a1	A/La Rioja 2202/18 SIAT F26/18*1 3C.2a1b	A/Swit 8060/17 Egg F27/18*1 3C.2a2	A/Eng 538/18 SIAT F31/18*1 3C.3a	A/Neth 10260/18 Egg F02/19*1 3C.2a1b	A/Neth 10260/18 SIAT F07/19*1 3C.2a1b	A/Norway 3275/18 SIAT F03/19*1 3C.2a1b
REFERENCE VIRUSES														
A/Stockholm/6/2014		3C.3a	2014-02-06	SIAT1/SIAT3	160	160	80	160	80	160	160	<	40	80
A/Hong Kong/5738/2014		3C.2a	2014-04-30	MDCK1/MDCK2/SIAT2	160	160	160	160	80	160	160	40	80	160
A/Bretagne/1413/2017		3C.2a2	2017-10-09	MDCK1/SIAT4	160	320	1280	320	160	1280	320	40	80	160
A/Singapore/INFIMH-16-0019/2016		3C.2a1	2016-04-14	E5/E2	<	80	40	640	160	160	80	40	40	40
A/Switzerland/8060/2017	clone 57	3C.2a2	2017-12-12	E7/E1	<	160	640	320	80	640	80	40	80	40
A/England/538/2018		3C.3a	2018-02-26	MDCK1/SIAT3	40	<	40	40	40	40	320	<	40	<
A/Netherlands/10260/2018		3C.2a1b	2018-02-15	E5/E1	<	160	80	80	320	80	80	1280	320	160
TEST VIRUSES														
A/Greece/1430/2018				SIAT1/SIAT1	<	40	80	<	<	40	<	<	40	160
A/Saint-Etienne/1998/2018			2018-10-15	MDCK3/SIAT1	40	40	40	40	160	40	40	40	80	80
A/Lyon/CHU/R18.116.67/2018			2018-11-26	MDCK4/SIAT1	40	40	<	40	160	40	40	40	160	160
A/Lyon/CHU/R18.128.2/2018			2018-12-17	MDCK3/SIAT1	80	80	80	80	80	160	40	<	80	320
A/Lyon/2296/2018			2018-12-20	MDCK3/SIAT1	160	160	80	160	160	160	160	<	80	80
A/EHPAD/Montpellier/2320/2018			2018-12-21	MDCK2/SIAT1	160	160	80	160	160	160	160	<	160	80
A/Lyon/2335/2018			2018-12-27	MDCK2/SIAT1	40	40	40	40	<	40	320	<	40	<
A/Lyon/CHU/R18.133.93/2018			2018-12-28	MDCK2/SIAT1	80	40	40	80	<	40	320	40	40	<
A/Lyon/CHU/R19.02.59/2019			2019-01-02	MDCK2/SIAT1	160	80	80	80	160	160	80	<	80	160
A/Lyon/CHU/R19.03.77/2019			2019-01-04	MDCK2/SIAT1	40	40	40	40	<	40	320	<	40	<
A/La Rochelle/85/2019			2019-01-05	MDCK2/SIAT1	40	40	40	40	<	80	320	<	40	<
A/Lyon/95/2019			2019-01-09	MDCK2/SIAT1	40	40	40	40	<	80	320	<	40	<
A/Lyon/EHPAD/108/2019			2019-01-09	MDCK2/SIAT1	40	40	80	40	80	80	<	<	80	160

*Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used) [1] < = <40; ND = Not Done

Sequences in phylogenetic trees

Vaccine SH 2018 NH 2018-19

Vaccine SH 2019

Table 4-5. Antigenic analysis of A(H3N2) viruses by HI

Viruses	Other information		Collection date	Passage history	Haemagglutination inhibition titre									
					Post-infection ferret antisera								NEW	NEW
					A/Stock 6/14	A/HK 5738/14	A/Bretagne 1413/17	A/Singapore 0019/16	A/La Rioja 2202/18	A/Swit 8060/17	A/Eng 538/18	A/Norway 3275/18	A/Kansas 14/17	A/Kansas 14/17
		Passage history			SIAT	MDCK	SIAT	Egg 10^{-4}	SIAT	Egg	SIAT	SIAT	Egg	Egg
		Ferret number			F14/14*1	St J F60/17*1	F01/18*1	F46/17*1	F26/18*1	F27/18*1	F31/18*1	F03/19*1	F11/19*1	F12/19*1
		Genetic group			3C.3a	3C.2a	3C.2a2	3C.2a1	3C.2a1b	3C.2a2	3C.3a	3C.2a1b	3C.3a	3C.3a
REFERENCE VIRUSES														
A/Stockholm/6/2014		3C.3a	2014-02-06	SIAT1/SIAT3	320	160	160	160	160	320	160	160	160	160
A/Hong Kong/5738/2014		3C.2a	2014-04-30	MDCK1/MDCK2/SIAT2	160	160	160	160	80	320	160	320	160	160
A/Bretagne/1413/2017		3C.2a2	2017-10-09	MDCK1/SIAT4	160	160	640	160	80	640	160	320	160	160
A/Singapore/INFIMH-16-0019/2016		3C.2a1	2016-04-14	E5/E2	80	160	80	640	320	160	80	80	40	40
A/Switzerland/8060/2017	clone 57	3C.2a2	2017-12-12	E7/E1	80	160	1280	640	160	1280	80	80	40	80
A/England/538/2018		3C.3a	2018-02-26	MDCK1/SIAT3	80	80	80	80	40	80	640	40	320	320
A/Kansas/14/2017		3C.3a	2017-12-14	E7/E2	40	<	40	40	<	<	320	<	1280	1280
TEST VIRUSES														
A/Belgium/G0023/2019			2019-01-03	SIAT1	160	80	80	80	40	80	640	40	320	320
A/Belgium/S0275/2019			2019-01-15	SIAT1	160	80	80	80	40	80	640	40	320	320
A/Niedersachsen/81/2019			2019-01-25	C1/MDCK1	160	80	80	80	40	80	640	40	320	320
A/Nordrhein-Westfalen/53/2019			2019-02-11	C1/MDCK1	160	80	80	80	40	80	640	40	160	160
A/Bayern/53/2019			2019-02-11	C1/MDCK1	160	80	80	80	40	80	640	40	320	320
A/Berlin/28/2019			2019-02-11	C1/MDCK1	160	80	80	80	40	80	640	40	320	320
A/Nordrhein-Westfalen/60/2019			2019-02-18	C1/MDCK1	160	80	80	80	40	80	640	40	320	320
A/Bremen/12/2019			2019-02-18	C1/MDCK1	160	80	80	160	80	80	640	80	320	320
A/Hessen/34/2019			2019-02-18	C1/MDCK1	160	80	80	160	80	160	640	80	320	320
A/Berlin/37/2019			2019-02-21	C1/MDCK1	80	80	80	80	40	80	640	40	320	320
A/Baden-Wurttemberg/87/2019			2019-02-25	C1/MDCK1	160	80	80	80	40	80	640	40	320	320
								Vaccine SH 2018 NH 2018-19		Vaccine SH 2019			Vaccine NH 2019-20	

* Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used) [1] < = <40; ND = Not Done

Sequences in phylogenetic trees

Table 4-6. Antigenic analysis of A(H3N2) viruses by HI

Viruses	Other information		Collection date	Passage history	Haemagglutination inhibition titre: Post-infection ferret antisera								
					A/Stock 6/14	A/HK 5738/14	A/Bretagne 1413/17	A/Singapore 0019/16	A/La Rioja 2202/18	A/Swit 8060/17	A/Eng 538/18	A/Norway 3275/18	A/Kansas 14/17
		Passage history			SIAT	MDCK	SIAT	Egg 10^{-4}	SIAT	Egg	SIAT	SIAT	Egg
		Ferret number			F14/14[*1]	St J F60/17[*1]	F01/18[*1]	F46/17[*1]	F26/18[*1]	F27/18[*1]	F31/18[*1]	F03/19[*1]	F11/19[*1]
		Genetic group			3C.3a	3C.2a	3C.2a2	3C.2a1	3C.2a1b	3C.2a2	3C.3a	3C.2a1b	3C.3a
REFERENCE VIRUSES													
A/Stockholm/6/2014		3C.3a	2014-02-06	SIAT1/SIAT3	320	160	80	160	80	160	160	80	160
A/Hong Kong/5738/2014		3C.2a	2014-04-30	MDCK1/MDCK2/SIAT1	160	160	160	160	160	320	160	160	160
A/Bretagne/1413/2017		3C.2a2	2017-10-09	MDCK1/SIAT4	160	160	640	320	80	640	160	160	160
A/Singapore/INFIMH-16-0019/2016		3C.2a1	2016-04-14	E5/E2	40	80	40	640	320	160	80	40	40
A/Switzerland/8060/2017	clone 57	3C.2a2	2017-12-12	E7/E1	40	160	1280	640	160	1280	80	80	80
A/England/538/2018		3C.3a	2018-02-26	MDCK1/SIAT3	80	40	40	80	40	80	640	<	320
A/Kansas/14/2017		3C.3a	2017-12-14	E7/E2	<	<	<	80	<	<	320	<	1280
TEST VIRUSES													
A/Valladolid/560/2018			2018-12-27	SIAT1/SIAT1	40	40	40	80	40	80	640	<	160
A/Valladolid/2/2019			2019-01-02	SIAT1/SIAT1	40	40	40	80	40	80	640	<	320
A/Avila/3/2019			2019-01-03	SIAT1/SIAT1	40	40	40	80	40	80	640	<	320
A/Valladolid/5/2019			2019-01-04	SIAT1/SIAT1	40	40	40	80	40	80	640	<	320
A/Valladolid/9/2019			2019-01-08	SIAT1/SIAT1	80	40	80	80	40	80	640	40	320
A/Valladolid/13/2019			2019-01-09	SIAT1/SIAT1	40	<	40	80	40	40	640	<	160
A/Soria/11/2019			2019-01-09	SIAT1/SIAT1	80	40	40	80	320	80	40	160	80
A/Avila/15/2019			2019-01-10	SIAT1/SIAT1	80	40	40	80	40	80	640	<	160
A/Palencia/20/2019			2019-01-12	SIAT1/SIAT1	80	40	40	80	40	80	640	40	160
A/Valladolid/18/2019			2019-01-12	SIAT1/SIAT1	80	40	40	80	80	80	640	<	320
A/Valladolid/17/2019			2019-01-12	SIAT1/SIAT1	40	<	40	80	40	40	320	<	160
A/Valladolid/27/2019			2019-01-14	SIAT1/SIAT1	80	40	40	80	40	40	640	<	160
A/Valladolid/26/2019			2019-01-14	SIAT1/SIAT1	80	80	40	80	80	80	640	<	320
A/Valladolid/25/2019			2019-01-14	SIAT1/SIAT1	80	80	40	80	80	80	640	<	320
A/Valladolid/24/2019			2019-01-14	SIAT1/SIAT1	40	<	40	80	<	40	320	<	160
								Vaccine SH 2018 NH 2018-19		Vaccine SH 2019			Vaccine NH 2019-20

[*] Superscripts refer to antiserum properties (< relates to the lowest dilution of antiserum used) [1] < = <40; ND = Not Done

Sequences in phylogenetic trees

Table 4-7. Antigenic analysis of A(H3N2) viruses by HI – Summary all test viruses

Viruses	Other information		Haemagglutination inhibition titre: Post-infection ferret antisera										
		Passage history	SIAT	MDCK	SIAT	Egg 10^{-4}	SIAT	Egg	SIAT	Egg	SIAT	SIAT	Egg
		Ferret number	F14/14*1	F30/14*1	F01/18*1	F46/17*1	F26/18*1	F27/18*1	F31/18*1	F02/19*1	F07/19*1	F03/19*1	F11/19*1
		Genetic group	3C.3a	3C.2a	3C.2a2	3C.2a1	3C.2a1b	3C.2a2	3C.3a	3C.2a1b	3C.2a1b	3C.2a1b	3C.3a
REFERENCE VIRUSES													
A/Stockholm/6/2014		3C.3a	160	40	80	160	80	160	160	<	40	80	160
A/Hong Kong/5738/2014		3C.2a	160	80	160	160	80	160	160	<	80	160	160
A/Bretagne/1413/2017		3C.2a2	80	<	640	160	80	640	80	<	80	160	160
A/Singapore/INFIMH-16-0019/2016		3C.2a1	<	<	40	640	80	80	40	<	40	<	40
A/Switzerland/8060/2017	clone 57	3C.2a2	<	40	1280	320	80	1280	80	40	80	40	80
A/England/538/2018		3C.3a	40	<	40	80	<	40	640	<	80	<	320
A/Netherlands/10260/2018		3C.2a1b	40	<	80	80	320	80	80	1280	320	160	ND
A/Kansas/14/2017		3C.3a	<	<	<	80	<	<	320	ND	ND	<	1280
TEST VIRUSES													
Number of viruses tested*			64	64	64	64	64*	64	64	38	38*	60*	26
No with titre reduction ≤2-fold			31	26	1		17	1	37		10	14	
%			*48.4*	*40.6*	*1.6*		*26.6*	*1.6*	*57.8*		*26.3*	*23.3*	
No with titre reduction =4-fold			18	35	1	6		4	2				18
%			*28.1*	*54.7*	*1.6*	*9.4*		*6.2*	*3.1*				*69.2*
No with titre reduction ≥8-fold			15	3	62	58		59	25	38			8
%			*23.5*	*4.7*	*96.8*	*90.6*		*92.2*	*39.1*	*100*			*30.8*

* Homologous HI titres not available - only results for viruses yielding HI titres of ≥160 with the respective antisera are shown

Vaccine SH 2018 NH 2018-19 | Vaccine SH 2019 | Vaccine NH 2019-20

Reference virus results are taken from individual tables as examples. Summaries for each antiserum are based on fold-reductions observed on the days that HI assays were performed.

Table 4-8. Antigenic analysis of A(H3N2) viruses by HI – Summary by test virus genetic group

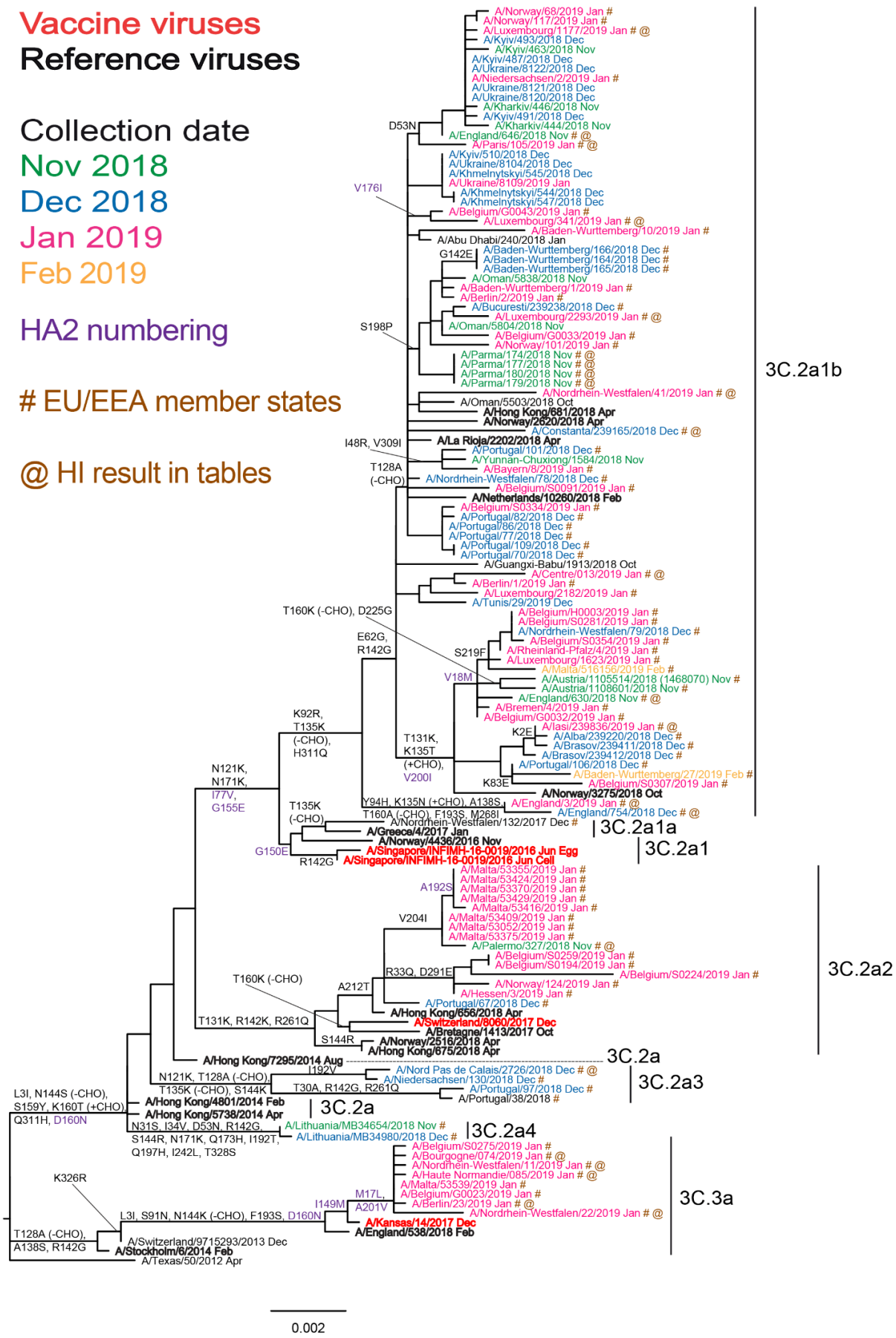
Viruses	Other information		Haemagglutination inhibition titre									
			Post-infection ferret antisera									
			A/Stock 6/14	A/HK 5738/14	A/Bretagne 1413/17	A/Singapore 0019/16	A/La Rioja 2202/18	A/Swit 8060/17	A/Eng 538/18	A/Neth 10260/18	A/Neth 10260/18	A/Norway 3275/18
		Passage history	SIAT	MDCK	SIAT	Egg 10^{-4}	SIAT	Egg	SIAT	Egg	SIAT	SIAT
		Ferret number	F14/14*1	F30/14*1	F01/18*1	F46/17*1	F26/18*1	F27/18*1	F31/18*1	F02/19*1	F07/19*1	F03/19*1
		Genetic group	3C.3a	3C.2a	3C.2a2	3C.2a1	3C.2a1b	3C.2a2	3C.3a	3C.2a1b	3C.2a1b	3C.2a1b
TEST VIRUSES												
Total number tested			25	25	25	25	25	25	25	25	25	21
Number tested		3C.2a1b	18	18	18	18	18*	18	18	18	18*	17*
No with titre reduction ≤2-fold			11	8			10				8	7
%			*61.1*	*44.4*			*55.6*				*44.4*	*41.2*
No with titre reduction =4-fold			1	10	1	1						
%			*5.6*	*55.6*	*5.6*	*5.6*						
No with titre reduction ≥8-fold			6		17	17		18	18	18		
%			*33.3*		*94.4*	*94.4*		*100*	*100*	*100*		
Number tested		3C.3a	5	5	5	5	5*	5	5	5	5*	2*
No with titre reduction ≤2-fold			3				0		5		0	0
%			*60.0*						*100*			
No with titre reduction =4-fold			1	5								
%			*20.0*	*100*								
No with titre reduction ≥8-fold			1		5	5		5		5		
%			*20.0*		*100*	*100*		*100*		*100*		
Number tested		3C.2a	1	1	1	1	1*	1	1	1	1*	1*
No with titre reduction ≤2-fold			1	1	1		1	1			0	1
No with titre reduction =4-fold						1			1			
No with titre reduction ≥8-fold										1		
Number tested		3C.2a3	1	1	1	1	1*	1	1	1	1*	1*
No with titre reduction ≤2-fold			1				0				0	0
No with titre reduction =4-fold				1								
No with titre reduction ≥8-fold					1	1		1	1	1		

* Homologous HI titres not available - only results for viruses yielding HI titres of ≥160 with the respective antisera are shown

Vaccine SH 2018 NH 2018-19

Vaccine SH 2019

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



Influenza B virus analyses

Influenza B viruses represented only 2.7% of the samples received with collection dates after 31 Augist 2018 and were received from national influenza centres (NICs) in 11 countries: Croatia, Denmark, France, Greece, Iceland, Ireland, Luxembourg, Norway, Portugal, Slovenia and the United Kingdom (Table 1). Of the small number received, 16 were B/Yamagata-lineage and seven were B/Victoria-lineage.

Influenza B/Victoria-lineage

A single B/Victoria lineage virus from an EU/EEA country has been tested by HI since the February 2019 characterisation report (Table 5). B/Norway/4183/2018 reacted poorly with sheep hyperimmune serum raised against egg-propagated B/Brisbane/60/2008 (clade 1A) and 8 of 10 components of the post-infection ferret antisera panel. However, it showed certain reactivity with post-infection ferret antisera raised against cell culture-propagated B/Colorado/06/2017 and B/Cote d'Ivoire/1661/2018, viruses with deletions of two [group 1A(Δ2)] and three [group 1A(Δ3)] amino acids in HA1 respectively. HA gene sequencing showed B/Norway/4183/2018 to be a subclade 1A(Δ3) virus.

A relatively small number (478 as of 12 April 2019) of HA sequences for viruses collected since 1 September 2018 have been deposited in the EpiFlu database of GISAID and the great majority of these have been from China and the US, with only 28 from Europe. All recently collected viruses continue to have HA genes that fall in the B/Brisbane/60/2008 clade (clade 1A; Figure 3), with virtually all falling in a subclade defined by HA1 amino acid substitutions I117V, N129D and V146I within clade 1A. Two groups within this subclade have deletions in the HA gene. A geographically dispersed group seen in Europe, the Americas, Asia, and Oceania have HA genes encoding an HA with deletion of residues K162 and N163 of HA1 [1A(Δ2) in Figure 3]. These viruses have additional substitutions of D129G and I180V in HA1, and R151K in HA2. This group of viruses is more prevalent than the subclade viruses that show no deletions. Of the low numbers of B/Victoria-lineage viruses detected recently, those with HA genes encoding a deletion of three HA1 amino acids, K162, N163 and D164 [1A(Δ3) in Figure 3], are predominant. This group splits into an Asian subgroup with viruses carrying additional substitutions of I180T and K209N in HA1 and a West African subgroup with viruses carrying the HA1 substitution K136E, often with additional HA1 substitutions of K52N and E198G or E198K (within the 197–199 glycosylation site) or G133R. Of the viruses detected in Europe, the majority fall in the West African subgroup, but B/Norway/4183/2018 falls in the Asian subgroup. It was noted in the September 2018 characterisation report[4] and earlier ones that the clade 1A viruses without deletions, the 1A(Δ2) group and the 1A(Δ3) subgroups are antigenically distinct from one another. Following the emergence and spread of viruses in the 1A(Δ2) group, a representative, B/Colorado/06/2017, was been recommended for use in trivalent influenza vaccines for the 2018–19 and 2019–20 northern hemisphere [1–3] and 2019 southern hemisphere seasons [4].

Influenza B/Yamagata-lineage

HI results for the three B/Yamagata-lineage viruses characterised since the February 2019 report are shown in Table 6, sorted by date of collection. The antiserum raised against egg-propagated B/Phuket/3073/2013, recommended for inclusion in quadrivalent vaccines for the 2018–2019 and 2019–20 northern hemisphere [1–3] and 2019 southern hemisphere seasons [4], recognised all three test viruses at a titre within twofold of the titre of the antiserum with the homologous virus. An antiserum raised against the cell culture-propagated cultivar of B/Phuket/3073/2013 recognised all test viruses poorly at titres reduced at least 16-fold compared to the homologous titre. Antisera raised against two other egg-propagated clade 3 viruses, B/Wisconsin/1/2010 (a former vaccine virus) and B/Stockholm/12/2011, recognised all three test viruses at titres within fourfold of the homologous titres. Antisera raised against two recently circulating clade 3 cell culture-propagated viruses yielded different reactivity patterns with the test viruses. Those raised against B/Mauritius/1791/2017 yielded a homologous titre of 40 and recognised all three test viruses well, while those raised against B/Mauritius/I-762/2018 gave a homologous titre of 640 and reacted well with only two test viruses, one each within twofold and fourfold of the homologous titre.

Antisera raised against cell culture-propagated clade 2 viruses, B/Estonia/55669/2011 and B/Massachusetts/02/2012, each recognised 1/3 (33%) test virus at titres within fourfold of the homologous titres, while that raised against egg-propagated B/Massachusetts/02/2012 recognised all three test viruses at titres within twofold of the homologous titre.

All test viruses carried an HA gene in genetic clade 3 (Table 6). Worldwide, all HA genes from viruses collected in the 2017–2018 season and since have fallen in clade 3, the B/Wisconsin/1/2010–B/Phuket/3073/2013 clade. Figure 4 shows a phylogenetic analysis of the HA genes of representative B/Yamagata-lineage viruses, including recently circulating ones. HA sequences of all viruses with collection dates after 31 August 2018 deposited in the EpiFlu database of GISAID (n=459 as of 12 April 2019), including those from Europe (n=26), fall in a subgroup

[4] European Centre for Disease Prevention and Control. Influenza virus characterisation Summary Europe, September 2018. Stockholm: ECDC; 2018. Available from: http://ecdc.europa.eu/publications-data/influenza-virus-characterisation-summary-europe-september-2018

defined by HA1 L172Q and M251V amino acid substitutions compared to B/Phuket/3073/2013. Some subclustering of sequences, defined by specific amino acid substitutions (e.g. HA1 S120T or D229N or D232N [introducing a potential N-linked glycosylation site]), is occurring. It has been noted in previous characterisation reports for 2018 that none of these amino acid substitutions have any obvious antigenic effects based on HI assays using post-infection ferret antisera raised against egg-propagated B/Phuket/3073/2013 which has been recommended for inclusion in quadrivalent vaccines for the 2018–2019 and 2019–20 northern hemisphere [1–3] and 2019 southern hemisphere seasons [4].

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

Viruses	Other information	Collection date	Passage history	Haemagglutination inhibition titre										
				Post-infection ferret antisera										
	Passage history			B/Bris 60/08 Egg	B/Bris 60/08 Egg	B/Malta 636714/11 Egg	B/Sth Aus 81/12 Egg	B/HK 514/09 MDCK	B/Ireland 3154/16 MDCK	B/Nord-West 1/16 MDCK	B/Norway 2409/17 MDCK	B/Colorado 06/17 MDCK	B/Colorado 06/17 Egg	B/CIV 1662/18 MDCK
	Ferret number			Sh 539, 540, 543, 544, 570, 571, 574[*1,3]	F44/17[*4]	F29/13[*2]	F25/16[*2]	NIB F47/16[*2]	F15/16[*2]	F16/16[*2]	F40/17[*2]	F09/18[*1]	F10/18[*2]	F37/18[*2]
	Genetic group			1A	1A	1A	1A	1B	1A	1A	1A(Δ2)	1A(Δ2)	1A(Δ2)	1A(Δ3)
REFERENCE VIRUSES														
B/Brisbane/60/2008	1A	2008-08-04	E4/E4	2560	640	320	640	320	40	40	<	40	80	<
B/Malta/636714/2011	1A	2011-03-07	E4/E1	2560	320	320	320	320	40	40	<	40	80	<
B/South Australia/81/2012	1A	2012-11-28	E4/E2	2560	320	160	640	160	40	40	<	40	80	10
B/Hong Kong/514/2009	1B	2009-10-11	MDCK1/MDCK2	2560	40	40	80	320	80	80	<	20	<	<
B/Ireland/3154/2016	1A	2016-01-14	MDCK1/MDCK4	2560	20	<	20	80	160	80	<	<	<	<
B/Nordrhein-Westfalen/1/2016	1A	2016-01-04	C2/MDCK2	1280	20	<	20	40	80	80	<	<	<	<
B/Norway/2409/2017	1A(Δ2)	2017-04-27	MDCK1/MDCK3	80	<	<	<	<	<	<	40	160	40	<
B/Colorado/06/2017	1A(Δ2)	2017-02-05	MDCK1/MDCK2	160	<	<	<	<	<	<	40	80	40	<
B/Colorado/06/2017	1A(Δ2)	2017-02-05	E5/E2	640	160	40	80	20	<	<	40	160	160	<
B/Cote D'Ivoire/1662/2018	1A(Δ3)	2018-07-25	P0/MDCK3	320	10	<	20	<	<	<	<	20	<	40
TEST VIRUSES														
B/Norway/4183/2018	1A(Δ3)	2018-12-27	MDCK1/MDCK1	320	<	<	<	<	<	<	<	40	<	10
					Vaccine[#]								Vaccine[$]	

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

[1] < = <40; [2] < = <10; [3] hyperimmune sheep serum; [4] < = <20; ND = Not Done

[#] B/Victoria-lineage virus recommended for use in trivalent vaccines NH 2017-18 and quadravalent vaccines SH 2018

[$] B/Victoria-lineage virus recommended for use in trivalent vaccines NH 2018-19, SH 2019 and NH 2019-20

Sequences in phylogenetic trees

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

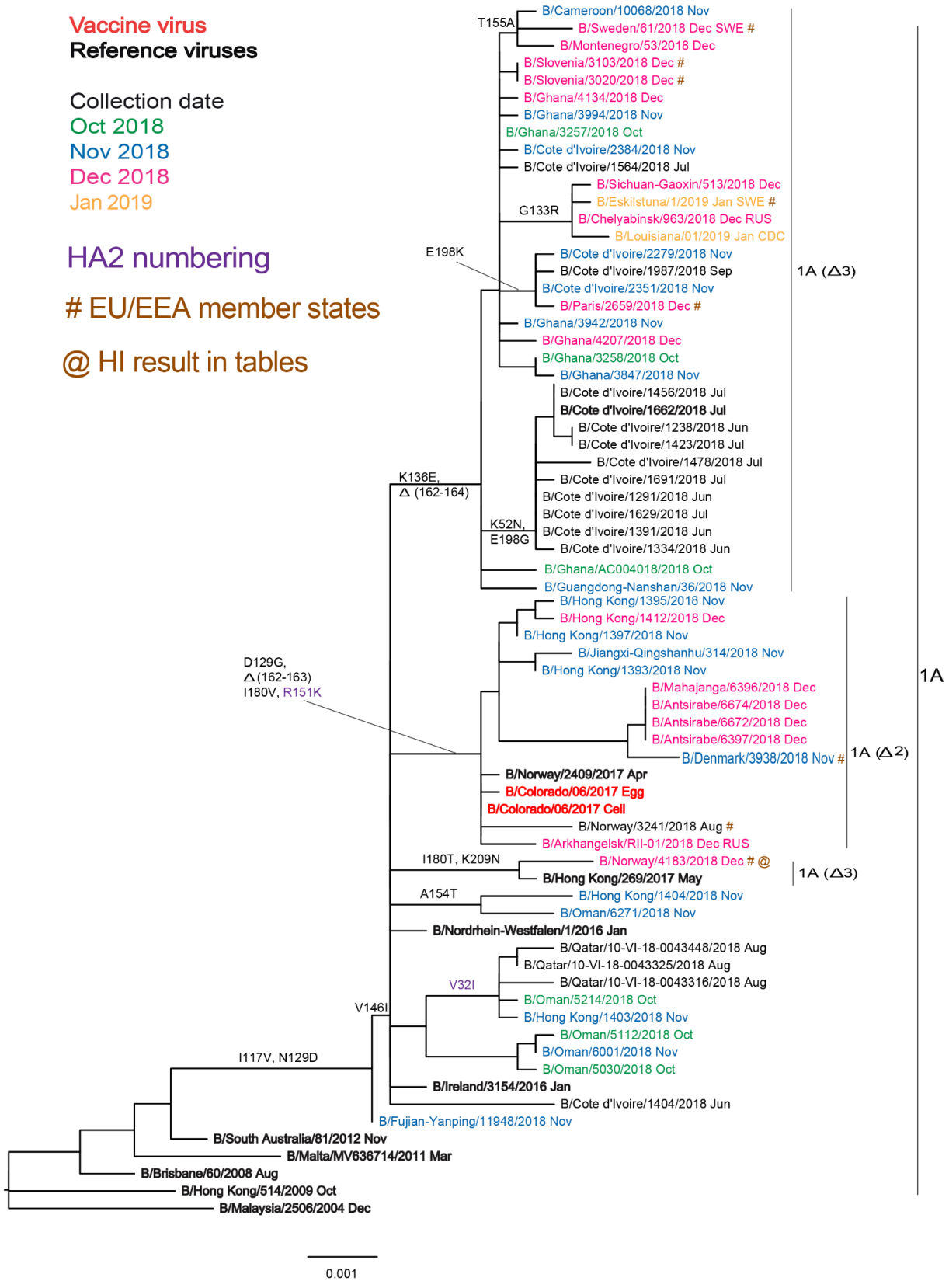


Table 6. Antigenic analysis of influenza B/Yamagata-lineage viruses by HI

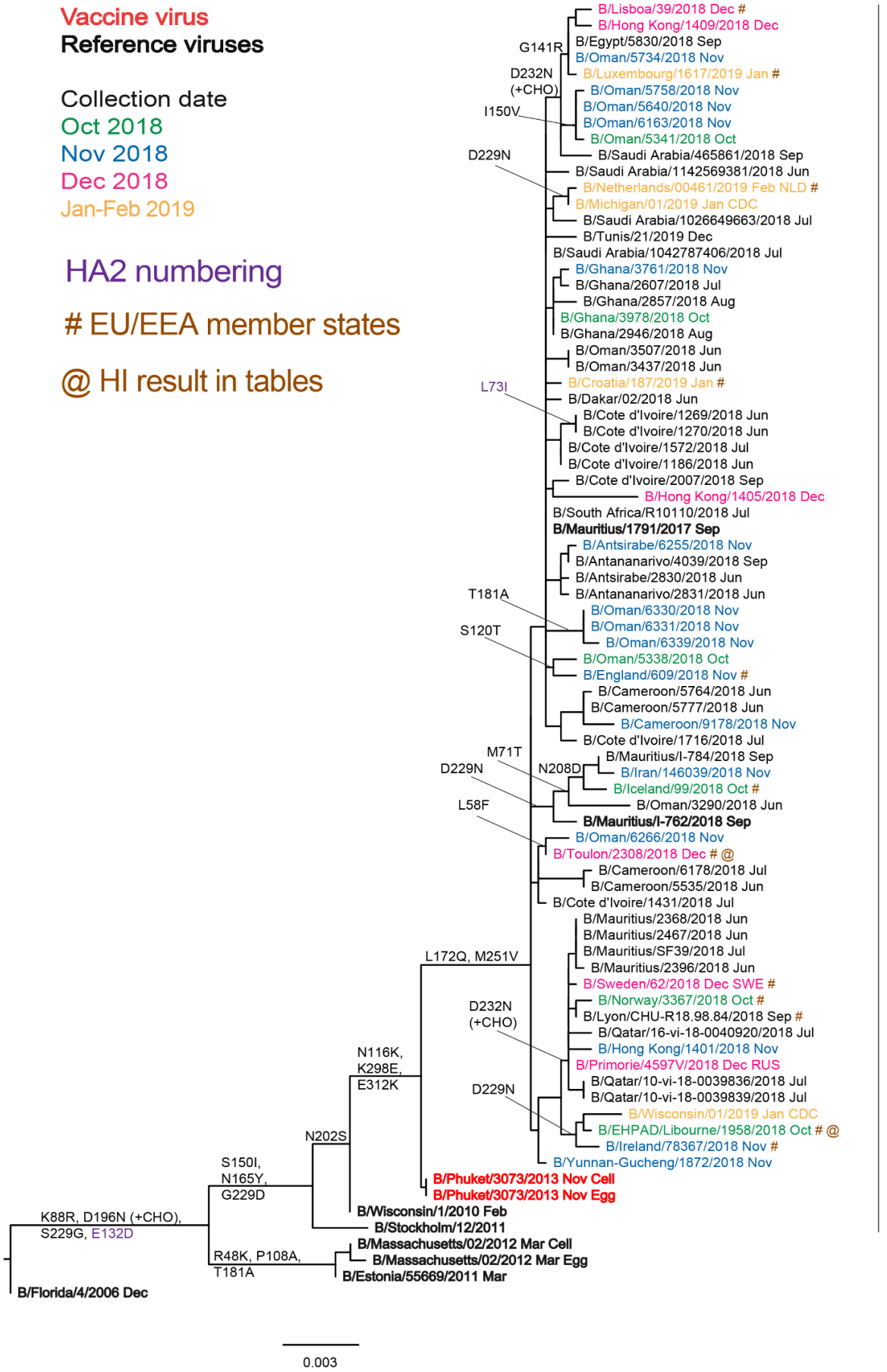
Viruses	Other information	Passage history / Ferret number / Genetic Group	Collection date	Passage history	Haemagglutination inhibition titre									
						Post-infection ferret antisera								
					B/Phuket 3073/13 Egg SH614[*1,4] 3	B/Estonia 55669/11 MDCK F39/17*[3] 2	B/Mass 02/12 MDCK F10/16[*2] 2	B/Mass 02/12 Egg F06/17[*2] 2	B/Wis 1/10 Egg F36/15[*2] 3	B/Stock 12/11 Egg F05/17[*2] 3	B/Phuket 3073/13 MDCK F27/15[*2] 3	B/Phuket 3073/13 Egg F25/17[*2] 3	B/Maur 1791/17 MDCK F04/18[*2] 3	B/Maur I-762/18 MDCK F05/19*[2] 3
REFERENCE VIRUSES														
B/Estonia/55669/2011		2	2011-03-14	MDCK2/MDCK3	1280	160	80	160	20	40	<	80	10	160
B/Massachusetts/02/2012		2	2012-03-13	MDCK1/C2/MDCK3	1280	320	160	640	80	160	10	320	20	160
B/Massachusetts/02/2012		2	2012-03-13	E3/E4	640	<	10	320	20	80	<	80	<	20
B/Wisconsin/1/2010		3	2010-02-20	E3/E2	2560	<	20	320	80	160	10	320	20	160
B/Stockholm/12/2011		3	2011-03-28	E4/E1	2560	<	20	320	40	160	10	160	20	160
B/Phuket/3073/2013		3	2013-11-21	MDCK2/MDCK3	5120	320	320	640	160	320	160	320	160	>1280
B/Phuket/3073/2013		3	2013-11-21	E4/E3	1280	<	20	160	40	160	<	160	20	80
B/Mauritius/1791/2017		3	2017-09-20	MDCK1/MDCK4	2560	<	40	160	20	80	20	160	40	160
B/Mauritius/I-762/2018		3	2018-09-02	MDCK1/MDCK2	2560	40	80	160	40	80	40	160	80	640
TEST VIRUSES														
B/EHPAD/Libourne/1958/2018		3	2018-10-12	MDCK2/MDCK1	1280	20	20	160	20	40	<	80	40	160
B/Toulon/2308/2018		3	2018-12-18	MDCK2/MDCK1	2560	40	40	320	40	80	10	160	40	320
B/Norway/252/2019		3	2019-01-11	MDCK2	1280	<	20	160	20	40	<	80	20	80
												Vaccine[#]		

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

[#] B/Yamagata-lineage virus recommended for use in quadravalent vaccines NH 2018-19, SH 2019 and NH 2019-20

Sequences in phylogenetic trees

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



Summaries of data submitted to TESSy

Genetic characterisation

For the 2018–19 season, as of week 14/2019, 3244 viruses had been characterised genetically and ascribed to a genetic clade:

- A total of 1 606 A(H1N1)pdm09 were subclade 6B.1, represented by the vaccine virus A/Michigan/45/2015, with a further 3 attributed to a subgroup not listed.
- A total of 1 591 were A(H3N2) viruses, with 1046 being subgroup 3C.2a1b represented by A/Alsace/1746/2018, 65 being subclade 3C.2a2 represented by A/Switzerland/8060/2017, 30 being subclade 3C.2a3 represented by A/Cote d'Ivoire/544/2016, 373 being clade 3C.3a represented by A/England/538/2018 (this represents a 15-fold increase in the proportion of 3C.3a viruses compared to the same period in the 2017-18 season), 57 being subclade 3C.2a1 represented by A/Singapore/16-0019/2016, 4 being clade 3C.2a represented by A/Hong Kong/4801/2014, 9 being subgroup 3C.2a1a represented by A/Greece/4/2017 and 7 were attributed to a subgroup not listed in current TESSy reporting categories.
- Twenty-two were B/Yamagata-lineage clade 3 represented by the vaccine virus B/Phuket/3073/2013.
- Twenty-two were B/Victoria-lineage viruses, with 5 being clade 1A represented by B/Brisbane/60/2008, 5 being subclade 1A(Δ2) with a two amino acid deletion in HA represented by the vaccine virus B/Colorado/06/2017 and 12 being subclade 1A(Δ3) with a three amino acid deletion in HA represented by B/Hong Kong/269/2017.

Antiviral susceptibility

For viruses collected during the course of the 2018–19 season, as of week 14 of 2019, 1 262 A(H1N1)pdm09, 773 A(H3N2) and 29 type B viruses have been tested for susceptibility to neuraminidase inhibitors. Eight A(H1N1)pdm09 viruses carried NA H275Y amino acid substitution indicative of highly reduced inhibition (confirmed phenotypically for 3) and 1 type B virus showed evidence of reduced inhibition (RI) by oseltamivir and zanamivir.

At the WIC, 565 viruses from EU/EEA countries have been assessed phenotypically against oseltamivir and zanamivir for this season: 301 A(H1N1)pdm09, 247 A(H3N2), 7 B/Victoria-lineage and 10 B/Yamagata-lineage. All but one virus showed normal inhibition by the two neuraminidase inhibitors. B/Norway/3241/2018 (Victoria-lineage) showed RI by the inhibitors and the NA gene encoded D197N amino acid substitution.

Influenza A(H7N9) virus

On 1 April 2013, WHO Global Alert and Response [5] reported that the China Health and Family Planning Commission notified WHO of three cases of human infection with influenza A(H7N9). A description of the characteristics of H7N9 viruses can be found on the WHO website [6]. An increased number of cases were reported over the course of the following seasons and cases were reported in 2017, including the fifth (2016–17) and largest wave to date, which included the emergence of highly pathogenic avian influenza strains that caused certain zoonoses, though few human cases were reported during the 2017–18 season [7]. WHO posted an analysis of information on A(H7N9) viruses on 10 February 2017 [8]. A summary and assessment of influenza viruses at the human-animal interface on 12 February 2019 indicates that there have been no publicly available reports from animal health authorities in China of influenza A(H7N9) virus detections in animals in recent months [9], with the latest human case occurring early in February 2018 [10]. The latest overview of avian influenza by ECDC in collaboration with the European Food Safety Authority and the EU Reference Laboratory for Avian Influenza was published on 27 September 2018 [11].

Influenza A(H5) virus

The most recent monthly risk assessment of influenza at the human–animal interface published by WHO on 12 February 2019 indicated that various A(H5Nx) subtypes continue to be detected in birds in Africa, Europe and Asia, notably A(H5N6) viruses. No new human cases have been detected since the last update published on 21 January 2019 [9]. As of 12 February 2019, no cases of human infection by A(H5N1) viruses have been reported to WHO in 2018–19 [12]. On 18 November 2016, ECDC published a rapid risk assessment related to outbreaks of highly pathogenic avian influenza H5N8 viruses in Europe [13]. The EU Reference Laboratory for Avian Influenza, in collaboration with ECDC and the European Food Standards Agency, published the latest overview of avian influenza on 27 September 2018 [11].

WHO CC reports

A description of results generated by the London WHO CC at the WIC and used at the most recent WHO vaccine composition meeting (held in Beijing, China 18–20 February 2019) and previous ones, can be found at: http://www.crick.ac.uk/partnerships/worldwide-influenza-centre/annual-and-interim-reports

Note on figures

The phylogenetic trees were constructed using RAxML (http://cme.h-its.org/exelixis/web/software/raxml/index.html), drawn using FigTree (http://tree.bio.ed.ac.uk/software/figtree) and annotated using Adobe Illustrator. 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 most viruses from non-EU/EEA countries were recovered from the EpiFlu database of GISAID. ECDC gratefully acknowledges the authors, originating and submitting laboratories of the sequences from the EpiFlu database of GISAID that were downloaded for use in the preparation of this report (all submitters of data may be contacted directly via the GISAID website at http://www.gisaid.org), along with all laboratories who submitted sequences directly to WHO CC London.

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