Influenza virus characterisation

Summary Europe, November 2021

Summary

This is the second influenza virus characterisation report for the 2021-2022 influenza season. As of week 47/2021, 2,707 influenza detections across the World Health Organization (WHO) European Region were reported to the European Surveillance System (TESSy); 93% type A viruses, with A(H3N2) (96%) dominating over A(H1N1)pdm09 (4%), and 7% type B, with only two having been ascribed to a lineage (B/Victoria). This represents a large increase (2,627, 3384%) in detections compared to the 2020-2021 season, following a large increase (354 256, 519%) in the number of samples tested, and is closer to the more usual number of detections seen at this time in earlier seasons. The increased testing is probably related to the emergence of SARS-CoV-2 and measures introduced to combat it.

Since the October 2021 characterisation report¹, no shipments from EU/EEA countries were received at the London WHO Collaborating Centre, the Francis Crick Worldwide Influenza Centre (WIC). This report therefore focuses on genetic characterisation of HA genes of representative seasonal influenza viruses submitted and/or released in GISAID during November 2021, with a focus on those viruses with collection dates after 31 August 2021, together with sequences recently determined at the WIC. The genetic clade nomenclature system adopted during the September 2021 vaccine composition meeting (VCM) is used throughout the document. The data continued to show low levels of influenza detections globally but with a predominance of A(H3N2) viruses.

Very low numbers of A(H1N1)pdm09 detections have been reported. The three with the most recent collection dates fell in the 6B.1A.5a.1 subgroup, represented by the vaccine virus for the northern hemisphere 2020-2021 season, A/Guangdong-Maonan/SWL1536/201960. A batch of viruses from Bangladesh with collection dates in September 2021 all fell in the 6B.1A.5a.2 subgroup, represented by A/Victoria/2570-like and A/Wisconsin/588/2019-like viruses which have been recommended respectively for egg- and cell-based vaccines in the 2021-2022 northern and 2022 southern hemisphere influenza seasons, but with additional HA amino acid substitutions seen previously in viruses from India.

All recently detected A(H3N2) viruses have fallen in subgroup 3C.2a1b.2a and been ‘Bangladesh-like’ (3C.2a1b.2a.2), with a number of EU/EEA countries reporting detections. A/Cambodia/e0826360/2020-like (3C.2a1b.2a.1) viruses were recommended for use in the 2021-2022 northern hemisphere season, while A/Darwin/9/2021-like and


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A/Darwin/6/2021-like (3C.2a1b.2a.2) viruses were recommended for egg- and cell-based vaccines in the 2022 southern hemisphere season. Recently detected viruses in Croatia were Bangladesh-like genetically and antigenically.

While the vast majority of B/Victoria-lineage HA sequences derived from viruses collected after 31 January 2021 were subclade V1A.3, represented by B/Washington/02/2019, the vaccine virus recommended for inclusion in influenza vaccines for the 2021-2022 northern hemisphere season, such viruses have only been detected in the US recently. The vast majority of sequences from recently detected viruses, in geographically dispersed countries, have fallen in the V1A.3.2 subgroup with defining HA1 A127T; P144L and K203R amino acid substitutions. B/Austria/1359417/2021-like (V1A.3.2) viruses have been recommended for use in the southern hemisphere 2022 influenza season. Antigenically, viruses in subgroups of the V.1A.3a group differ and show some loss of reactivity with post-infection ferret antisera raised against B/Washington/02/2019.

No B/Yamagata-lineage HA sequences from clinical specimens collected in 2021, and none with collection dates after March 2020, were available. All of the 77 sequences from viruses detected in 2020, inclusive of 12 from EU/EEA countries, belong to genetic clade Y3 and carry three HA1 amino acid substitutions (L172Q, D229N and M251V) compared to B/Phuket/3073/2013-like viruses which have been recommended for use in quadrivalent influenza vaccines for the 2021-2022 northern hemisphere and 2022 southern hemisphere seasons. The antigenic effects of these amino acid substitutions have been minimal as assessed in earlier reports.

Table 1 shows a summary of influenza virus detections in the WHO European Region reported to the European Surveillance System (TESSy) database during the 2021-2022 season (weeks 40-47/2021), compared to the 2019-2020 season. There has been a vast increase in the number of samples from patients fulfilling Influenza-Like Illness (ILI) and/or Acute Respiratory Infection (ARI) criteria being tested (354 256, 519%), even when compared with a more ‘normal’ season, 2019-2020 (326 453, 391%: results not shown), which preceded the COVID-19 pandemic. With this increased testing there has been a rise in the number of influenza-positive samples (2 627, 3384%), although there was a reduction compared to the same period in 2019-2020 (1 649, 37.9%: results not shown). These data probably relate to a number of factors: (i) significant numbers of samples taken from patients fulfilling ILI and/or ARI criteria being infected with other agents, possibly SARS-CoV-2, the virus responsible for the COVID-19 pandemic; (ii) restrictions on travel and social/work place gatherings, imposed to help curtail the spread of SARS-CoV-2, also impeding the spread of influenza viruses, and; (iii) viral interference, with SARS-CoV-2 infection impeding infection by influenza viruses.

With these caveats, and being mindful of the low number of detections during the 2020-2021 season, the ratio of type A to type B detections has increased compared to the 2020-2021 season (1.2:1 to 12.3:1), with a greater dominance of A(H3N2) over A(H1N1)pdm09 viruses. While the number of influenza B virus detections has increased from 37 to 203 (549%), only two viruses in each period were ascribed to a lineage (Table 1), though B/Yamagata lineage viruses were not detected after March 2020. Currently, it appears that measures introduced relating to the COVID-19 pandemic are still having an effect but that A(H3N2) viruses will dominate in 2021-2022.

Table 1. Influenza virus detections in the WHO European Region from the start of reporting for the 2021-22 season (weeks 40-47/2021)*

<table>
<thead>
<tr>
<th>Virus type/subtype/lineage</th>
<th>Cumulative number of detections for weeks 40-47/2021</th>
<th>Totals*</th>
<th>Ratios</th>
<th>Cumulative number of detections for weeks 40-47/2020</th>
<th>Totals*</th>
<th>Ratios</th>
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<tbody>
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<td>2371</td>
<td>1</td>
<td>2504</td>
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<td>1501</td>
<td>1</td>
<td>1599</td>
<td>66</td>
<td>45</td>
</tr>
<tr>
<td>A not subtyped</td>
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<td>1</td>
<td>841</td>
<td>26</td>
<td>16</td>
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<tr>
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<tr>
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<td>6</td>
<td>195</td>
<td>201</td>
<td>0</td>
<td>35</td>
<td>35</td>
</tr>
<tr>
<td>Total detections (total tested)</td>
<td>139 (10 461)</td>
<td>2 586 (&gt;426 255)</td>
<td>2 707 (&gt;438 716)</td>
<td>1 (4 433)</td>
<td>79 (&gt;80 027)</td>
<td>60 (&gt;84 460)</td>
</tr>
</tbody>
</table>

* Numbers taken from Flu News Europe to week 47/2021 and week 47/2020 reports

Since week 40/2021, two shipments of specimens (virus isolates and/or clinical specimens) were received at the Crick Worldwide Influenza Centre (WIC) from Croatia and the Netherlands containing five and 14 samples, respectively (Table 2). Of the 18 samples, 17 were type A viruses and one was type B/Victoria-lineage. No shipments were received in November 2021.

Genetic and antigenic characterisation data generated at the WIC for viruses with collection dates after 31 August 2020 until 1 January 2021, up to a report deadline of 15 February 2021, contributed to the WIC virus characterisation report that was presented at the WHO influenza vaccine composition meeting (VCM) in February 2021, when recommendations were made for the northern hemisphere 2021-2022 season. Data generated on viruses with collection dates after 31 January 2021 informed the recent VCM where recommendations were made for the 2022 southern hemisphere season. Recommendations for the 2020-2021 northern hemisphere, the 2021 southern hemisphere and upcoming 2022-2023 northern hemisphere and 2022 southern hemisphere seasons have been published [1, 2, 3, 4].

Due to the low number of influenza-positive specimens detected and thereby available for sharing with WIC, recent influenza characterisation reports, and this one, have been based mainly on phylogenetic analyses of complete HA gene sequences submitted to the Epiflu™ database of the Global Initiative on Sharing All Influenza Data (GISAID), inclusive of sequences generated at the WIC, with those from EU/EEA countries highlighted. Four A(H3N2) viruses from Croatia were characterised antigenically since the October 2021 report (Table 3).
Influenza A(H1N1)pdm09 virus analyses

All recently circulating viruses have fallen into clade 6B.1A, defined by the amino acid substitutions S74R, S84N, S162N (introducing a potential N-linked glycosylation site), S164T (which alters the glycosylation motif at residues 162 to 164), I216T and I229S in HA1. Within clade 6B.1A, clusters of viruses (genetic groups) encoding a range of HA amino acid substitutions have emerged, with most recently circulating viruses carrying the substitution S183P in HA1, although this is not retained in all genetic groups. Figures 1a and 1b are annotated with HA1 S183P substitution groups assigned for the February 2019 WHO VCM, with new nomenclature introduced at the time of the September 2021 WHO VCM (6B.1A.1 to 6B.1A.7). The recommended vaccine viruses for the northern hemisphere 2020–2021 (egg-based A/Guangdong-Maonan/SWL1536-like and cell-based A/Hawaii/70/2019-like) and southern hemisphere 2021, 2022 and northern hemisphere 2021–2022 (egg-based A/Victoria/5270/2019-like and cell-based A/Wisconsin/588/2019-like) influenza seasons are shown in red [1, 2, 3, 4]. The seven subclades are defined by the following HA amino acid substitutions:

1. Subclade 6B.1A.1 viruses, represented by the 2019-2020 vaccine virus A/Brisbane/02/2018, carry an HA gene mutation encoding HA1 S183P amino acid substitution.
2. Subclade 6B.1A.2 viruses, represented by A/Denmark/2728/2019, carry HA gene mutations encoding HA1 S183P and L233I with HA2 V193A amino acid substitutions – a group within this subclade has emerged with additional HA1 amino acid substitutions of N129D, K130N, P137S, N156K and K211R (e.g. A/Hong Kong/110/2019).
5. Subclade 6B.1A.5 viruses carry HA gene mutations encoding HA1 S183P and N260D amino acid substitutions and splits into two groups designated 6B.1A.5a represented by A/Norway/3433/2018 with additional HA1 amino acid substitutions of N129D and T185A, and 6B.1A.5b represented by A/Switzerland/3330/2017 with additional amino acid substitutions of HA1 E235D and HA2 V193A. Two subgroups within the 6B.1A.5a group have been defined based on HA1 amino acid substitutions of D187V, K211R and Q189E (6B.1A.5a.1) or K130N, N156K, L161I and V250K (6B.1A.5a.2).
6. Subclade 6B.1A.6 viruses, represented by A/Ireland/84630/2018, carry HA gene mutations encoding HA1 T120A and S183P amino acid substitutions, like subclade 6B.1A.3 viruses, but fall within a separate phylogenetic branch which is closer to subclade 6B.1A.5 viruses.
7. Subclade 6B.1A.7, represented by A/Slovenia/1489/2019, carry HA gene mutations encoding HA1 K302T and HA2 I77M, N169S and E179D amino acid substitutions sometimes with additional HA1 substitutions of E68D, S121N and L161I (e.g. A/Moscow/193/2019). Note: a group within this subclade has emerged with P183S (reversion), T185I, I240V and S286S substitutions in HA1 (e.g. A/Estonia/120012/2019).

The two A(H1N1)pdm09 HA phylogenies show similar profiles. The first is repeated from the October 2021 report and included 35 recently submitted/released sequences available in GISAID, derived from 16 6B.1A.5a.1 and 19 6B.1A.5a.2 viruses (Figure 1a). The 16 6B.1A.5a.1 viruses were largely detected in West Africa but with one each detected in France (in March), Qatar (in August) and USA (in August), while the 6B.1A.5a.2 viruses were largely detected in India with one each in Bangladesh (in August) and USA (in September). The recently emerged 6B.1A.5a.2 viruses were mostly detected in India and carried additional HA1 amino acid substitutions of K54Q, K130N, A188T, Q189E and E224A, often with R259K and K308R. A zoonotic A(H1N1)v H1N1pm09-like virus, A/North Dakota/12226/2021, belonging to the swine 1A.3.3.2 clade was detected in September (Figure 1a). The second phylogeny is based on sequences submitted/released in November (n = 39), only 23 of which had collection dates after 31 August (Figure 1b).
Three of the latter viruses, one each from England, Spain and the US, fell in subgroup **6B.1A.5a.1**, with the remainder falling in subgroup **6B.1A.5a.2**, 19 from Bangladesh and one from the US. The 20 **6B.1A.5a.2** viruses were like those detected in India, carrying additional **HA1** amino acid substitutions of **K54Q, K130N, A186T, Q189E, E224A, R259K** and **K308R**.

The great majority of **A(H1N1)pdm09** viruses characterised antigenically by the WIC in the course of the 2019-2020 influenza season, with the exception of those in subgroup **6B.1A.5a.2**, were antigenically similar to A/Guangdong-Maonan/SWL1536/2019 (H1N1)pdm09-like viruses (**6B.1A.5a.1** with **HA1 D187A** and **Q189E** amino acid substitutions), recommended for use in the northern hemisphere 2020-2021 influenza season [1], as assessed by haemagglutination inhibition (HI) assays with a panel of post-infection ferret antisera raised against vaccine and reference viruses. Results of HI assays for viruses detected in EU/EEA countries can be seen in previous influenza characterisation reports: [https://www.ecdc.europa.eu/en/seasonal-influenza/surveillance-and-disease-data/influenza-virus-characterisation](https://www.ecdc.europa.eu/en/seasonal-influenza/surveillance-and-disease-data/influenza-virus-characterisation) [accessed 1 December 2021].
Figure 1a. Phylogenetic comparison of influenza A(H1N1)pdm09 HA genes (GISAID, October 2021)
Figure 1b. Phylogenetic comparison of influenza A(H1N1)pdm09 HA genes (GISAID, November 2021)
Influenza A(H3N2) virus analyses

The first A(H3N2) HA phylogeny is repeated from the October 2021 report and was based on H3 HA sequences deposited/released in GISAID in October (n = 453), including only those with collection dates in October included together with sequences that had been generated recently at the WIC (Figure 2a). The second phylogeny is based on H3 HA sequences deposited/released in GISAID in November (n = 511), including only those with collection dates from 21 October included (n = 147) together with sequences generated recently at the WIC (Figure 2b).

Viruses in clade 3C.2a have been dominant since the 2014-15 influenza season, with group 3C.2a1b viruses predominating over the course of the 2019-2020 season in most WHO-defined regions of the world but for the European Region, there was equivalence of clade 3C.3a viruses. The HA gene sequences of viruses in both clades 3C.2a and 3C.3a continue to diverge. Notably, clade 3C.3a1 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, and D160N in HA2, compared with cell culture-propagated A/Stockholm/6/2014. Greater variation has been observed among clade 3C.2a viruses, resulting in the designation of new subclades/groups/subgroups. Amino acid substitutions that define these subclades/groups/subgroups are:

- 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 (a former vaccine virus).
- Group 3C.2a1a: Those in subclade 3C.2a1 plus T135K in HA1, resulting in the loss of a potential glycosylation site, and G150E in HA2, e.g. A/Greece/4/2017.
- Group 3C.2a1b: Those in subclade 3C.2a1 plus E62G, R142G and H311Q in HA1, often with additional amino acid substitutions – notably HA1 T135K (resulting in the loss of a potential glycosylation site) commonly with T128A (resulting in the loss of a potential glycosylation site), the 3C.2a1b.1 subgroup (e.g. A/La Rioja/2202/2018) or HA1 T131K and HA2 V200I, the 3C.2a1b.2 subgroup (e.g. A/South Australia/34/2019). Distinct clusters of viruses within both these subgroups have emerged defined by specific HA1 and/or HA2 amino acid substitutions: 3C.2a1b.1a with additional amino acid substitutions of HA1 A138S, F193S and S198P, many also with G186D and D190N (e.g. A/Denmark/3284/2019); 3C.2a1b.1b with additional amino acid substitutions of HA1 S137F, A138S and F193S (e.g. A/Hong Kong/2671/2019); 3C.2a1b.2a with additional amino acid substitutions of HA1 K83E and Y94N with HA2 I193M (e.g. A/Slovenia/1637/2020); 3C.2a1b.2b with HA2 V18M substitution, often with additional HA1 substitutions (e.g. A/Bretagne/1323/2020).
- Clade 3C.3a: represented by a former vaccine virus, A/Switzerland/9715293/2013, with recently circulating clade 3C.3a1 viruses carrying additional substitutions of S91N, N144K (resulting in the loss of a potential glycosylation site), and F193S in HA1 and D160N in HA2, e.g. A/England/538/2018 and A/Kansas/14/2017, the A(H3N2) vaccine virus for the 2019-2020 northern hemisphere influenza season.

The significant geographic spread of viruses in the antigenically distinct 3C.2a1b.1b cluster influenced the selection of an A/Hong Kong/2671/2019-like or an A/Hong Kong/45/2019-like virus as the A(H3N2) component of vaccines for the 2020-2021 northern hemisphere and 2021 southern hemisphere influenza seasons [1, 2].

The HA phylogeny generated for the October report showed that all viruses detected in October were 'Bangladesh-like' (3C.2a1b.2a with HA1 substitutions of Y159N, T160I (loss of a glycosylation site), L164Q, G186D, D190N and Y195F) (Figure 2a). Within this group, subgroups showing limited geographic spread had emerged defined by specific HA1 amino acid substitutions. For example, s subgroup defined by S205F and A212T substitutions detected in Qatar in August and recently detected in the Russian Federation and Spain, and a more geographically dispersed subgroup defined by H156S substitution. The latter subgroup showed more diversification with viruses carrying D53N, N96S and I192F substitutions detected in Qatar, Spain and the Netherlands; viruses with D53G, R201K and S219Y substitutions detected in the Russian Federation; viruses with D53G, D104G and K276R substitutions detected in Lebanon with a branch of this subgroup carrying additional L157I and S262N substitutions as identified in an outbreak in Maryland USA.

The second phylogeny incorporated 147 sequences submitted/released in GISAID in November with collection dates from 21 October 2021, together with sequences recently determined at the WIC (Figure 2b). All viruses with collection dates after 31 August 2021 (n = 176) were 'Bangladesh-like' (3C.2a1b.2a with HA1 substitutions of Y159N, T160I (loss of a glycosylation site), L164Q, G186D, D190N and Y195F). The latter viruses were split into three subgroups defined by specific HA1 amino acid substitutions: (i) S205F and A212T (n = 5) with viruses from the Netherlands, Spain and the Russian Federation; (ii) D53G (n = 58), often with additional amino acid substitutions, in viruses from Lebanon, Spain, the Russian Federation and the US; and (iii) D53N, N96S and I192F (n = 113) with viruses from Croatia, Italy, the Netherlands (n = 86), Spain (n = 23) and the US.

'Bangladesh-like' 3C.2a1b.2a viruses, A/Darwin/9/2021 and A/Darwin/6/2021 for egg- and cell-based vaccines respectively, were recently recommended for use in the southern hemisphere 2022 influenza season [4].

The locations of HA sequences for egg- and cell culture-propagated cultivars of A/Cambodia/e0826360/2020 (3C.2a1b.2a.1) are recommended for use in northern hemisphere 2021-2022 vaccines [3], are indicated on the phylogenies, as are egg- and cell-culture based vaccines to be used in the 2022 southern hemisphere season, A/Darwin/9/2021 and A/Darwin/6/2021 (3C.2a1b.2a.2) respectively [4] (Figures 2a and 2b).
As described in many previous reports, 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 highlighted first in the November 2014 report, this has been a significant problem for most viruses that fall in genetic clade 3C.2a, although there was some alleviation of this during 2019-2020 with continuation into the 2020-2021 influenza season. This issue is now much alleviated for 'Bangladesh-like' 3C.2a1b.2a.2 viruses which agglutinate guinea pig RBCs well, allowing HI assays to be performed.

While the number of detections of seasonal influenza viruses was low from April 2020 to July 2021, compared to previous years, the WHO Collaborating Centres for Influenza have shown viruses in these emerged virus clusters to be antigenically distinguishable from one another and other A(H3N2) virus subgroups.

Antigenic characterisation of four A(H3N2) viruses from Croatia, all of which are 3C.2a1b.2a.2 viruses with HA1 H156S substitution, is presented here (Table 3). All test viruses were inhibited well, by antisera raised against 3C.2a1b.2a.2 viruses, notably so for antisera raised against cell culture-propagated A/Stockholm/5/2021 and slightly less well by antisera raised against cell culture-propagated A/Bangladesh/4005/2020 and egg-propagated A/Darwin/9/2021. Antisera raised against single 3C.2a1b.1a, 3C.2a1b.1b and 3C.3a1 viruses, and two 3C.2a1b.2a.1 did not recognise any of the test viruses.

Results of HI assays with panels of post-infection ferret antisera raised against A(H3N2) vaccine and reference viruses for viruses detected in EU/EEA countries can be seen in previous influenza characterisation reports on ECDC's website. Overall, these data show strong clade/subclade-specific recognition of test viruses by post-infection ferret antisera raised against cell culture-propagated reference viruses, with limited cross-clade/subclade recognition and further reductions in recognition of cell culture-propagated recently circulating viruses by antisera raised against A(H3N2) egg-propagated vaccine viruses.

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2 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:

**Figure 2a.** Phylogenetic comparison of influenza A(H3N2) HA genes (GISAID, October 2021)
Figure 2b. Phylogenetic comparison of influenza A(H3N2) HA genes (GISAID, November 2021)
### Table 3. Antigenic Analysis of A(H3N2) Viruses by HI

<table>
<thead>
<tr>
<th>Country</th>
<th>Year of Isolation</th>
<th>Genetic Group</th>
<th>Antigenic Analysis by HI</th>
</tr>
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<tr>
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<tr>
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<td>A/Stockholm/5/2021</td>
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<td>A/Kansas/14/2017</td>
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</tr>
</tbody>
</table>

| **TEST VIRUSES** | | | |
| A/Croatia/97333/2021 | 2021-09-17 | 3C.2a1b.2a.2 | <, <80, 80, <160, 640, 320, 160, < |
| A/Croatia/99546/2021 | 2021-09-25 | 3C.2a1b.2a.2 | <, <80, 80, <160, 640, 320, 160, < |
| A/Croatia/103473/2021 | 2021-09-07 | 3C.2a1b.2a.2 | <, <80, 80, <160, 640, 320, 160, < |
| A/Croatia/109108/2021 | 2021-09-20 | 3C.2a1b.2a.2 | <, <80, 80, <160, 320, 320, 160, < |

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

\[ 1 < = <40, \text{ND} = \text{Not Done} \]
Influenza B virus analyses

Influenza B/Victoria-lineage

All recently circulating B/Victoria-lineage viruses have fallen in genetic clade V1A, represented by B/Brisbane/60/2008, a former vaccine virus, but with additional HA1 amino acid substitutions of I117V and N129D (e.g. B/Ireland/3154/2016). Viruses retaining full-length HAs had remained similar antigenically to B/Brisbane/60/2008. However, three genetic groups (described below with amino acid substitutions/deletions relative to B/Brisbane/60/2008 indicated) containing deletions of HA gene codons have emerged. Viruses in these groups are antigenically distinct from B/Brisbane/60/2008 and each other (as noted in the September 2018 characterisation report⁴ and earlier ones), such that four antigenically distinguishable groups had been circulating:

- A group with double deletion of HA1 residues 162 and 163 (subclade V1A.1) with amino acid substitutions of D129G and N180V, and HA2 R151K that spread worldwide and is represented by a previous vaccine virus, B/Colorado/06/2017.
- A group with triple deletion of HA1 residues 162 to 164 (subclade V1A.2) first detected in Asia, with amino acid substitutions of I180T and K209N that showed limited geographic spread (with no detections having been made recently), represented by B/Hong Kong/269/2017.
- A group with triple deletion of HA1 residues 162 to 164 (subclade V1A.3) first detected in Africa, with amino acid substitution K136E often with G133N that showed geographic spread and became dominant, represented by B/Washington/02/2019 the vaccine virus recommended after WHO VCMs in February and September 2020, and February 2021 [1, 2, 3].

The phylogeny generated for the October report included 58 new full-length HA gene sequences from viruses with collection dates in 2021 available in GISAID, or generated by the WIC, in October (Figure 3a). One of these viruses, B/La Reunion/438/2021 collected in July, was B/Washington/02/2019-like (V1A.3). All other viruses fell in subgroup V1A.3a.2 (with HA1 A127T, P144L and K203R substitutions).

The second phylogeny incorporated 52 sequences submitted/released in GISAID in November with collection dates from 1 July 2021 (Figure 3b). Of these, just 20 had collection dates after 31 August 2021, seven viruses from the US fell in subclade V1A.3 being B/Washington/02/2019-like, and 13 (three from England, one from the Netherlands and nine from India) fell in subgroup V1A.3a.2 being B/Austria/1359417/2021-like.

The WHO Collaborating Centres for Influenza have shown the V1A.3a group viruses with additional HA1 substitutions to be antigenically distinct from one another and, despite the low number of B/Victoria-lineage viruses detected, there is indication of geographic spread of viruses in these recently emerged virus subgroups, notably those in the subgroup B/Austria/1359417/2021-like (V1A.3a.2) viruses were recently recommended for southern hemisphere 2022 vaccines [4].

Influenza B/Yamagata-lineage

It is assumed that no B/Yamagata-lineage viruses have been detected after March 2020 as no sequences for such viruses with collection dates after this had been released in GISAID as of 30 November 2021. Figure 4 is repeated from the September report with recently designated nomenclature indicated in bold/red type and was generated based on the 77 HA sequences from viruses with collection after 31 December 2019 to 31 March 2020 available in GISAID. All sequences fell in genetic clade 3 (Y3), the B/Wisconsin/1/2010–B/Phuket/3073/2013 clade, within a subgroup defined by HA1 L172Q and M251V amino acid substitutions compared to B/Phuket/3073/2013 which is recommended for inclusion in quadrivalent vaccines for the 2020-2021 and 2021-2022 northern hemisphere, 2021 and 2022 southern hemisphere seasons [1, 2, 3, 4]. Some sub-clustering of sequences, defined by specific amino acid substitutions (e.g. HA1 N164K, K211R, D229N or D232N [introducing a potential N-linked glycosylation site] sometimes with R48K), has occurred. As noted in previous characterisation reports, 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.

Figure 3a. Phylogenetic comparison of influenza B/Victoria-lineage HA genes (GISAID, October 2021)
Figure 3b. Phylogenetic comparison of influenza B/Victoria-lineage HA genes (GISAID, November 2021)
Figure 4. Phylogenetic comparison of influenza B/Yamagata-lineage HA genes (GISAID, September 2021)
Summaries of data submitted to the European Surveillance System

Genetic characterisation

165 viruses detected over the course of the 2021-2022 season (weeks 40-47/2021) were genetically characterised:

- Two A(H1N1)pdm09 viruses, one of which was not assigned to a genetic group, while the other belonged to clade 6B.1A.5a.1 (represented by A/Guangdong-Moan/SWL1536/2019).
- 161 A(H3N2) viruses, all of which belonged to the 'Bangladesh-like' clade (3C.2a1b.2a.2) represented by A/Bangladesh/4005/2020.
- Two B/Victoria-lineage viruses, one each belonging to clades V1A.3 (represented by B/Washington/02/2019) and V1A.3a.2 (represented by B/Austria/1359417/2021).

Antiviral susceptibility

Up to week 47/2021, 122 and 69 A(H3) viruses, respectively, were assessed for susceptibility to neuraminidase inhibitors and the PA inhibitor baloxavir marboxil. No amino acid substitutions previously associated with reduced susceptibility to any of the drugs were identified.

At the WIC, fifteen A(H3N2) influenza viruses detected within EU/EEA countries during the 2021-2022 season have been assessed phenotypically against oseltamivir and zanamivir. All showed NI by both NA enzymes.

Influenza A(H7N9) virus

On 1 April 2013, the World Health Organization (WHO) Global Alert and Response System [5] reported that the China Health and Family Planning Commission had notified WHO of three cases of human infection with influenza A(H7N9). Increased numbers 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 (HPAI) strains that have caused some zoonoses, although few human cases were reported during the 2017-18 season [6]. WHO posted an analysis of information on A(H7N9) viruses on 10 February 2017 [7], and ECDC published a rapid risk assessment on the implications of A(H7N9) for public health on 3 July 2017 [8]. Current risk assessments can be found on WHO's website https://www.who.int/teams/global-influenza-programme/avian-influenza/monthly-risk-assessment-summary (accessed 1 December 2021). The assessment published on 1 October 2021 indicated that there had been no publicly available reports from animal health authorities in China or other countries on influenza A(H7N9) virus detections in animals in recent months [9]. The H7N9 situation update published by the Food and Agricultural Organization of the United Nations on 3 February 2021 indicated there had been 14 detections in chickens in Shandong province, China during October 2020, but the report published on 1 December 2021 indicated that there have been no additional detections since then [10]. The most recent human case was detected in mid-March 2019 [11]. 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 29 September 2021 and can be found on ECDC's website [12].

Influenza A(H5) virus

The most recent monthly risk assessment of influenza at the human-animal interface was published by WHO on 1 October 2021. Since the previous risk assessment on 8 August 2021, 11 human cases of infection with avian influenza A(H5N6) viruses were reported by China with disease onset dates in July through September [9]. All cases reported exposure to poultry and, at the time of report publication, two cases were fatal, seven were severe/critical, one was mild and information was not available for the 11th case. The last human case of known A(H5N1) infection was reported by India [13].

On 30 September 2020, ECDC published an alert related to outbreaks of avian influenza viruses in Europe [14]. The latest collaborative report from ECDC and the European Food Safety Authority (EFSA), reported 162 highly pathogenic avian influenza (HPAI) A(H5) detections between 25 May and 15 September 2021, 51 in poultry, 91 in wild birds and 20 in domestic birds [12]. Detections occurred in 17 EU/EEA countries and the UK. Of the poultry detections, 20 were reported by Kosovo, 17 by Poland and six by Albania, and the wild bird detections were reported in resident populations mainly in northern Europe. Nineteen different virus genotypes have been detected in Europe and Central Asia since July 2020, confirming a high propensity for A(H5) viruses to undergo reassortment events, with most recent detections being subtype A(H5N8). According to reports compiled by the Food and Agricultural Organization of the United Nations (FAO) as of 24 November 2021, various influenza A(H5Nx) subtypes continued to be detected in wild and/or domestic birds in Africa, Asia and Europe, and since 27 October 2021 a total of 272 HPAI (14 H5Nx, 254 H5N1, one H5N5 and three H5N8) and 14 LPAI outbreaks had been reported, with one A(H5N6) human infection in China (symptom onset 29 August 2021) [15].
Influenza A(H9N2) virus

Since the previous WHO update on 8 August 2021, three laboratory-confirmed human cases of influenza A(H9N2) virus infection in children were reported by China with onset dates in June, August and September [9]. For two cases, poultry exposure was reported and disease symptoms were mild, this information was not available for the third case. The most recent FAO report mentions a further case in China with disease onset date of 29 October 2021 [15]. Public Health England recently published an updated risk assessment of avian influenza A(H9N2) [16]. Avian influenza A(H9N2) viruses are enzootic in poultry in Asia and increasingly reported in poultry in Africa.

Other influenza zoonotic events

Since the previous WHO update on 8 August 2021, five A(H1N1)v zoonotic events with swine-related variant influenza A viruses were reported by China with disease onset dates in late 2020/early 2021 [9]. The swine exposure history of these five cases is unknown and while four of the cases had mild disease, the fifth developed pneumonia. In addition, two adult cases were detected in Wisconsin USA, both reported exposure to swine, one was hospitalised but both recovered.

Four zoonotic cases of A(H1N2)v infection were also reported, one each by Austria and France, and two by the US (one each in Iowa and Ohio). The European cases were in adults and the US cases in children, with all reporting exposure to swine. All cases recovered and there was no evidence of human-to-human transmission.

One case of A(H3N2)v infection was reported from Iowa, USA. The infected child was not hospitalised, made a full recovery and no human-to-human transmission was identified.

WHO Collaborating Centre reports

A description of results generated by the London WHO Collaborating Centre at the WIC and used at the September 2021 WHO vaccine composition meeting (held online: 13-23 September 2021 for seasonal influenza viruses), and previous ones, can be found at https://www.crick.ac.uk/partnerships/worldwide-influenza-centre/annual-and-interim-reports (accessed 1 December 2021).

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(s) of sample collection. Isolates from WHO NICs in EU/EEA countries are highlighted in yellow. Sequences for most viruses from non-EU/EEA countries were recovered from the GISAID EpiFlu database. We gratefully acknowledge the authors, originating and submitting laboratories of the sequences from the GISAID 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 WHO CC London.
References


