Influenza virus characterization

Summary report, Europe, October 2023
Contents

Acknowledgements 4

Summary of the latest WHO Influenza Vaccine Composition meetings 5

Influenza by type/subtype 6
  Worldwide ........................................... 6
  European region .................................. 7

Summary of influenza detections in the WHO European Region, week 35/2023 to 42/2023 8

Sentinel surveillance system dynamics, week 35/2023 to 42/2023 9

Genetic diversity by Type/Lineage and group 10

Influenza A H1N1 12
  Genetic analyses: H1N1 .................................. 12
    Maximum likelihood phylogenetic trees: H1N1 .................................. 12

Summary of the antigenic properties of H1N1 viruses circulating in the reporting period 14

A/H1N1: References 14

Influenza A H3N2 15
  Genetic analyses: H3N2 .................................. 15
    Maximum likelihood phylogenetic tree: H3N2 .................................. 15

Summary of the antigenic properties of H3N2 viruses circulating in the reporting period 17

A/H3N2: HI reagents and references 17

Influenza B 18
  Genetic analyses: B/Victoria .................................. 18
    Maximum likelihood phylogenetic tree: B/Victoria .................................. 18

Summary of the antigenic properties of B/Victoria lineage viruses circulating in the reporting period 20

Summaries of data submitted to TESSy 21
  Genetic characterization .................................. 21
    (According to the guidance produced for TESSy reporting at the beginning of the 2022-2023 influenza season) .................................. 21
  Susceptibility to antivirals .................................. 21

Annex 22
  A/H1N1 .................................. 22
  A/H3N2 .................................. 23
  B/Victoria .................................. 24

WHO Collaborating Centre reports 24
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Summary of the latest WHO Influenza Vaccine Composition meetings

Genetic and antigenic characterization data generated at the Worldwide Influenza Centre for viruses with collection dates after 31 January 2023 until 31 August 2023 informed the WHO influenza vaccine composition meeting (VCM) in September 2023 when recommendations were made for the southern hemisphere (SH) 2024 influenza season. At the September 2023 VCM it was recommended to change the A(H1N1)pdm09 and A(H3N2) vaccine components for the 2024 SH season. Previously, at the February 2023 VCM, which focused on data from viruses collected after 31 August 2022 until 31 January 2023, it was also recommended to change the A(H1N1)pdm09 vaccine component for the 2023-2024 northern hemisphere (NH) season.

It is recommended vaccines for use in the 2024 SH influenza season contain the following:

**Trivalent: Egg-based Vaccines**
- an A/Victoria/4897/2022 (H1N1)pdm09-like virus;
- an A/Thailand/8/2022 (H3N2)-like virus; and
- a B/Austria/1359417/2021 (B/Victoria lineage)-like virus.

**Trivalent: Cell- or recombinant-based Vaccines**
- an A/Wisconsin/67/2022 (H1N1)pdm09-like virus;
- an A/Massachusetts/18/2022 (H3N2)-like virus; and
- a B/Austria/1359417/2021 (B/Victoria lineage)-like virus.

**Quadrivalent: egg- or cell culture- or recombinant-based vaccines Above 3 components; and a B/Phuket/3073/2013 (B/Yamagata lineage)-like virus.**

**Influenza B/Yamagata-lineage**

No B/Yamagata-lineage viruses with collection dates after March 2020 have been detected or sequences released in GISAID as of 23 October 2023.

The absence of confirmed detection of naturally occurring B/Yamagata lineage viruses is indicative of very low risk of infection by B/Yamagata lineage viruses. Therefore, it is the opinion of the WHO influenza vaccine composition advisory committee that inclusion of a B/Yamagata lineage antigen in quadrivalent influenza vaccines is no longer warranted, and every effort should be made to exclude this component as soon as possible. A continued effort by all NICs of GISRS is required to identify B/Yamagata-lineage viruses for detailed characterization to determine if there are any in circulation.
Influenza by type/subtype

Worldwide

Geographical distribution of influenza viruses with collection dates from 1st September 2023 through to 23rd October as deposited in GISAID, coloured by Type/subtype. Geographic markers scaled to detection proportions. Full length HA. Map provided by WHO GIS Centre for Health, DNA/DDI (https://www.who.int/data/gis).

Globally, influenza detections declined and remained at inter-seasonal levels through the reporting period. The relative proportions of A/H1N1, A/H3N2 and B/Victoria varied by geographic region with cocirculation of A/H1N1 and A/H3N2 overall and some detections of B/Victoria in the US and Thailand, as indicated by the different colours in the pie charts by country.
European region

Geographical distribution in the European region of influenza viruses with collection dates from 1st September 2023 through to 23rd October as deposited in GISAID, coloured by Type/subtype. Geographic markers scaled to detection proportions. Full length HA.

In the European region, influenza detections remained at inter-seasonal levels through the reporting period. The majority of countries which reported detections showed co-circulation of A/H1N1 and A/H3N2 as indicated by the different colours in the pie charts.
Summary of influenza detections in the WHO European Region, week 35/2023 to 42/2023

Table 1 shows influenza virus detections in the WHO European Region reported to The European Surveillance System (TESSy) database from weeks 35 to 42 of 2023 (end of season 2022-2023 to beginning of season 2023-2024) compared with the same period in the previous season. For type-percentage calculations, the denominator is total detections; for subtype and lineage, it is the total influenza A subtyped and total influenza B lineage determined, respectively. As not all countries have a true non-sentinel testing denominator, no percentage calculations for total tested are shown (Data taken from Flu News Europe reports and from ERVISS (European Respiratory Virus Surveillance Summary)).

<table>
<thead>
<tr>
<th>Virus type/subtype/lineage</th>
<th>Cumulative number of detections for weeks 40/2022-30/2023</th>
<th>Cumulative number of detections for weeks 40/2021-30/2022</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Sentinel sources</td>
<td>Non-sentinel sources</td>
</tr>
<tr>
<td>Influenza A</td>
<td>162</td>
<td>1523</td>
</tr>
<tr>
<td>A(H1N1)pdm09</td>
<td>66</td>
<td>331</td>
</tr>
<tr>
<td>A(H3N2)</td>
<td>66</td>
<td>215</td>
</tr>
<tr>
<td>A not subtyped</td>
<td>30</td>
<td>977</td>
</tr>
<tr>
<td>Influenza B</td>
<td>20</td>
<td>215</td>
</tr>
<tr>
<td>Victoria lineage</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Yamagata lineage</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Lineage not ascribed</td>
<td>20</td>
<td>211</td>
</tr>
<tr>
<td>Total detections</td>
<td>182</td>
<td>1718</td>
</tr>
<tr>
<td>Total tested</td>
<td>16267</td>
<td>254989</td>
</tr>
</tbody>
</table>

Compared with the same period in 2022, for sentinel surveillance the number of tested specimens has nearly doubled, however with a 3-fold decrease in influenza detections. For non-sentinel surveillance, the number of tested specimens has marginally increased from last season to the current, however detections have decreased by 2-fold. The higher number of detections in 2022 was largely driven by the increase in A(H3N2) detections. Relative frequencies of type A vs B influenza viruses have slightly changed, with influenza B detections increasing from 7% in 2022 to nearly 11% in the current period, although the overall frequency of influenza B in season 2022-2023 was of 26%. Relative frequencies of influenza A subtypes were also different from last season when there was 68% predominance of A(H3N2) viruses, compared to the current season with a predominance of 59% for A(H1N1).
During the period from week 35 to week 42 of 2023, influenza activity remained well under the epidemic threshold of 10%.

Across sentinel surveillance, influenza A/H3N2 and A/H1N1 viruses cocirculated during most of this period with overall frequencies of 59% for A(H1N1) and 41% for A(H3N2).
Genetic diversity by Type/Lineage and group

![Genetic diversity of global samples chart]

The chart shows the distribution of genetic diversity among different influenza subtypes and lineages. The x-axis represents the number of samples, ranging from 0 to 80. The y-axis lists the various lineages and subtypes, including:

- 5a.2a (A / H1N1)
- 5a.2a.1 (A / H1N1)
- 2a.1b (A / H3N2)
- 2a.3 (A / H3N2)
- 2a.3a (A / H3N2)
- 2a.3a.1 (A / H3N2)
- 2a.3b (A / H3N2)
- 2b (A / H3N2)
- V1A.3a.2 (B / Victoria)

Each lineage is represented by a different color: red for A / H1N1, green for A / H3N2, and purple for B / Victoria. The bars indicate the number of samples for each lineage.
Gene diversity of samples by region - EURO

<table>
<thead>
<tr>
<th>Clade</th>
<th>Number of Samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>5a.2a (A/H1N1)</td>
<td></td>
</tr>
<tr>
<td>5a.2a.1 (A/H1N1)</td>
<td></td>
</tr>
<tr>
<td>2a.3a (A/H3N2)</td>
<td></td>
</tr>
<tr>
<td>2a.3b (A/H3N2)</td>
<td></td>
</tr>
<tr>
<td>2a.1b (A/H3N2)</td>
<td></td>
</tr>
<tr>
<td>2a.3a.1 (A/H3N2)</td>
<td></td>
</tr>
</tbody>
</table>

Number of samples:

- A/H1N1
- A/H3N2
Influenza A H1N1

Genetic analyses: H1N1

Globally 6B.1A.5a.2a and 6B.1A.5a.2a.1 HA clade viruses both continued to circulate with differing relative proportions depending on region.

In Europe, both 5a.2a and 5a.2a.1 viruses were detected, with 5a.2a viruses being the larger proportion.

Within the 5a.2a viruses, characterised by substitutions K54Q, A186T, E224A, R259K and K308R, two clades were observed: one minor clade with D94N and T216A represented by A/Sydney/5/2021, with viruses detected in Europe, the US and Australia, and a larger clade defined by substitution I418V which was detected in Europe, the US, Japan and Thailand. Other subclades that were reported in previous weeks were not seen during this period, except for one with A48P and additional substitutions A141S, R205K, K239R, L365Q and R450K with only two viruses from Thailand.

Within the 5a.2a.1 viruses, characterised by substitutions P137S, K142R, D260E, T277A, E356D and N451H, there are two main groups of viruses: a major clade with T216A represented by A/Victoria/4897/2022 and a minor clade represented by A/Wisconsin/67/2022. Within the major clade there are 3 distinct subclades: one with R113K and V427I that was detected in Europe and Japan; a second with T120A that was circulating in North America and Spain; and a third subclade with R45K that was seen in the US and the Caribbean region.

Maximum likelihood phylogenetic trees: H1N1

Maximum likelihood time-resolved phylogenetic tree inferred using Iqtree2 from HA sequence data obtained from GISAID from 1st September onwards. Trees were manually curated and, if sequence numbers were > 200, downsampled using Treemer to retain a representative tree topology of 200 sequences with emphasis on viruses from the European region. Annotation of amino acids substitutions was performed with Treetime ancestral reconstruction. References and CVVs are marked as Cell or Egg.
Vaccine viruses
Reference viruses

# recently sequenced at WIC

Influenzavirus characterisation – Summary Europe, October 2023

WHO EUROPE SURVEILLANCE REPORT

Influenza virus characterisation – Summary Europe, October 2023
Summary of the antigenic properties of H1N1 viruses circulating in the reporting period

Very few A/H1N1 viruses collected since 1st September have been phenotypically characterized by haemagglutination inhibition (HI). For an overall picture of the past season, see the Annex.

A/H1N1: References

<table>
<thead>
<tr>
<th>Virus</th>
<th>Genetic group</th>
<th>Virus passage</th>
<th>Ferret ID</th>
</tr>
</thead>
<tbody>
<tr>
<td>A/Sydney/5/2021</td>
<td>6B.1A.5a.2a</td>
<td>MDCK3/MDCK3</td>
<td>F46/22</td>
</tr>
<tr>
<td>A/Sydney/5/2021</td>
<td>6B.1A.5a.2a</td>
<td>E3/E3</td>
<td>F04/22</td>
</tr>
<tr>
<td>A/Victoria/4897/2022</td>
<td>6B.1A.5a.2a.1</td>
<td>SIAT2/MDCK2</td>
<td>F05/23</td>
</tr>
<tr>
<td>IVR-238 (A/Victoria/4897/2022)</td>
<td>6B.1A.5a.2a.1</td>
<td>E3/D6/E1 10-6</td>
<td>F07/23</td>
</tr>
<tr>
<td>A/Wisconsin/67/2022</td>
<td>6B.1A.5a.2a.1</td>
<td>MDCK2</td>
<td>F17/23</td>
</tr>
</tbody>
</table>
Influenza A H3N2

Genetic analyses: H3N2

Clade 3C.2a1b.2a.2 (renamed as 2 since February 2023) predominated since 1st February in all geographic regions where H3N2 circulated.

Within this clade, cocirculation of multiple genetic clades were observed during most of the southern hemisphere influenza season 2022-2023, with clades 2a.2b, the 2a.3a.1 and 2a.1b were the most frequently detected.

During this reporting period, the great majority of H3 viruses detected belong to clade 2a.3a.1, which share substitutions E50K with clade 2a.3a and present additional substitutions I140K and I223V. Within clade 2a.3a.1, viruses with I25V, V347M and I418V were seen in Spain, Russian Federation and Japan, whereas viruses with N122D (potential loss of N-glycosylation) and K276E were detected in Europe, Thailand and the US. A few viruses from Norway with substitutions Q173R and K276E cluster within clade 2a.3a.

Maximum likelihood phylogenetic tree: H3N2

Maximum likelihood time-resolved phylogenetic tree inferred using Iqtree2 from HA sequence data obtained from GISAID from 1st September onwards. Trees were manually curated and, if sequence numbers were > 200, downsampled using Treemer to retain a representative tree topology of 200 sequences with emphasis on viruses from the European region. Annotation of amino acids substitutions was performed with Treetime ancestral reconstruction. References and CVVs are marked as Cell or Egg.
Summary of the antigenic properties of H3N2 viruses circulating in the reporting period

Very few A/H3N2 viruses collected since 1st September have been phenotypically characterized by haemagglutination inhibition (HI). For an overall picture of the past season, see the Annex.

A/H3N2: HI reagents and references

<table>
<thead>
<tr>
<th>Virus</th>
<th>Genetic group</th>
<th>Virus passage</th>
<th>Ferret ID</th>
</tr>
</thead>
<tbody>
<tr>
<td>A/Thuringen/10/2022</td>
<td>2b</td>
<td>P1/SIAT2</td>
<td>F36/22</td>
</tr>
<tr>
<td>A/Stockholm/5/2021</td>
<td>2a</td>
<td>SIAT0/SIAT3</td>
<td>F35/21</td>
</tr>
<tr>
<td>A/Darwin/9/2021</td>
<td>2a</td>
<td>E3/E4</td>
<td>F39/21</td>
</tr>
<tr>
<td>A/Norway/24873/2021</td>
<td>2a.3</td>
<td>SIAT2</td>
<td>F10/22</td>
</tr>
<tr>
<td>A/Norway/24873/2021</td>
<td>2a.3</td>
<td>E3 (Am2Al1)</td>
<td>F11/22</td>
</tr>
<tr>
<td>A/Poland/97/2022</td>
<td>2a.2</td>
<td>S2</td>
<td>F39/22</td>
</tr>
<tr>
<td>A/Slovenia/8720/2022</td>
<td>2a.1</td>
<td>SIAT1/MDCK1/SIAT2</td>
<td>F24/22</td>
</tr>
<tr>
<td>A/Lille/50053/2022</td>
<td>2a.1</td>
<td>MDCK1/SIAT3</td>
<td>F02/23</td>
</tr>
<tr>
<td>A/Catalonia/NSVH161512067/2022</td>
<td>2a.1b</td>
<td>SIAT1/SIAT3</td>
<td>F41/22</td>
</tr>
<tr>
<td>A/Albania/289813/2022</td>
<td>2a.3a.1</td>
<td>MDCK1</td>
<td>F21/23</td>
</tr>
<tr>
<td>A/Albania/289813/2022</td>
<td>2a.3a.1</td>
<td>E3(Am1Al2)</td>
<td>F19/23</td>
</tr>
<tr>
<td>A/Brandenburg/15/2022</td>
<td>2a.3a.1</td>
<td>E5(Am1Al2)</td>
<td>F18/23</td>
</tr>
<tr>
<td>A/Brandenburg/15/2022</td>
<td>2a.3a.1</td>
<td>E3</td>
<td>F18/23</td>
</tr>
<tr>
<td>A/Thailand/08/2022</td>
<td>2a.3a.1</td>
<td>E3/E1</td>
<td>F34/23</td>
</tr>
<tr>
<td>IVR-237(A/Thailand/08/2022)</td>
<td>2a.3a.1</td>
<td>E3/D7/E1</td>
<td>F35/23</td>
</tr>
</tbody>
</table>
Influenza B

Genetic analyses: B/Victoria

Clade V1A.3a.2 viruses are characterised by substitutions A127T, P144L, N150K, G184E, N197D (‑CHO), K203R and R279K predominated since 1st February 2023 in geographic regions where B/Victoria-lineage viruses were detected. Within V1A.3a.2, the most recent viruses are characterised by additional substitution D197E. Some minor subclades within V1A.3a.2 show R80G and E184K (US), D129N (US and Thailand) and a E183K (Europe and US). No Clade V1A.3 viruses were detected since 1st February 2023. No B/Yamagata lineage viruses have been detected since March 2020.

Maximum likelihood phylogenetic tree: B/Victoria

Maximum likelihood time-resolved phylogenetic tree inferred using Iqtree2 from HA sequence data obtained from GISAID from 1st September onwards. Trees were manually curated and, if sequence numbers were > 200, downsampled using Treemer to retain a representative tree topology of 200 sequences with emphasis on viruses from the European region. Annotation of amino acids substitutions was performed with Treetime ancestral reconstruction. References and CVVs are marked as Cell or Egg.
Vaccine viruses
Reference viruses

# recently sequenced at WIC

V1A.3a.2

V1A.3

V1A

V1A.1

V1A.3a.2

19
Summary of the antigenic properties of B/Victoria lineage viruses circulating in the reporting period

Very few B/Victoria viruses collected since 1st September have been phenotypically characterized by haemagglutination inhibition (HI). For an overall picture of the past season, see the Annex.

# B/Victoria: Reagents and references

<table>
<thead>
<tr>
<th>Virus</th>
<th>Genetic group</th>
<th>Virus passage</th>
<th>Ferret ID</th>
</tr>
</thead>
<tbody>
<tr>
<td>B/Brisbane/60/2008</td>
<td>V1A</td>
<td>E4/E4</td>
<td>sheep pool</td>
</tr>
<tr>
<td>B/Washington/02/2019</td>
<td>V1A.3</td>
<td>E3/E3</td>
<td>F20/20</td>
</tr>
<tr>
<td>B/Stockholm/3/2022</td>
<td>V1A.3a.2</td>
<td>SIAT1/MDCK3</td>
<td>F28/22</td>
</tr>
<tr>
<td>B/Austria/1359417/2021</td>
<td>V1A.3a.2</td>
<td>SIAT1/MDCK4</td>
<td>NIB F01/21</td>
</tr>
<tr>
<td>B/Austria/1359417/2021 G141</td>
<td>V1A.3a.2</td>
<td>E3/E5</td>
<td>F15/21</td>
</tr>
<tr>
<td>B/Austria/1359417/2021 G141R</td>
<td>V1A.3a.2</td>
<td>E3/E5</td>
<td>F44/21</td>
</tr>
</tbody>
</table>
Summaries of data submitted to TESSy

Genetic characterization

(According to the guidance produced for TESSy reporting at the beginning of the 2022-2023 influenza season)

Overall, 9,997 viruses detected over the course of the 2022-2023 season (weeks 40/2022-39/2023) were genetically characterized:

- Of 4,066 A/H1N1 viruses, all but five belonged to clade 6B.1A.5a.2 (clade 5a.2) with 714 represented by A/Norway/25089/2022 (5a.2a.1), 2,841 by A/Sydney/5/2021 (5a.2a) and 85 by A/Victoria/2570/2019 (5a.2), while 415 were allocated to the ‘Subgroup Not Listed’ category. Eleven were clade 5a.1 viruses represented by A/Guangdong-Maonan/SWL1536/2019.

- Of 3,319 A/H3N2 viruses, 2,803 belonged to the ‘Bangladesh-like’ clade (3C.2a1b.2a.2, renamed as 2) with 1,947 represented by A/Bangladesh/4005/2020, 221 represented by A/Darwin/9/2021 (clade 2a) and 980 represented by A/Slovenia/8720/2022 (clade 2a.1). Three viruses carried HA genes belonging to clade 3C.2a1b.1a represented by A/Denmark/3264/2019. A total of 160 viruses were allocated to the ‘Subgroup Not Listed’ category.

- Of 2,612 B/Victoria-lineage viruses, 2,612 were clade V1A.3a.2 represented by B/Austria/1359417/2021. The remaining 296 viruses were allocated to the ‘Subgroup Not Listed’ category.

No virus characterisation data was available so far for weeks 40 to 42 of season 2023-2024.

Susceptibility to antivirals

Between weeks 35 and 39/2023, 8 viruses were assessed for susceptibility to neuraminidase inhibitors and 7 were assessed for susceptibility to baloxavir marboxil. Phenotypically and/or genotypically, no markers associated with reduced susceptibility were identified.

No antiviral susceptibility data was available so far for weeks 40 to 42 of season 2023-2024. At the WIC, between weeks 35 and 42/2023 196 influenza viruses detected within the WHO EURO Region since 1st February 2023 were assessed for susceptibility to antivirals. Of these, 63 A/H1N1, 42 A/H3N2 and 91 B/Victoria-lineage were phenotypically assessed against oseltamivir and zanamivir. All viruses showed Normal Inhibition (NI) by both NAIs. Phenotypic testing for susceptibility to baloxavir marboxil was performed for 9 A/H1N1 viruses, with all of them showing Normal Inhibition. Genotypic assessment of 33 H1N1, 14 H3N2 and 120 B/Victoria NA gene sequences from influenza viruses detected within the WHO EURO Region since 1st February 2023 and received at the WIC between weeks 35 and 42/2023 did not find any marker associated with reduced susceptibility to NAI, except for two influenza B viruses B/Kosovo/1306/2023 and B/Kosovo/1561/2023 which exhibit substitution K360E that is associated with highly reduced inhibition by peramivir, an antiviral for which we do not perform susceptibility testing.

For 28 H1N1, 14 H3N2 and 95 B/Victoria PA viruses where PA gene sequencing was successful, no markers associated with reduced inhibition by baloxavir marboxil were identified.
Annex

Heatmapped phylogenetic trees represent the combined genetic and antigenic analyses where the correlation between genetic groups, signature amino acids and their antigenic profile (including microneutralisation for A/H3N2) can be observed.

These outputs were generated by the London WHO Collaborating Centre for the SH 2024 September VCM with influenza viruses with collection dates between 1 February and 31 August 2023.

A/H1N1
**A/H3N2**

[Image of genetic tree and heatmap showing virus strain information]

- Vaccine viruses:
  - A/Albania/290243/2022 Egg
  - A/Albania/289813/2022 Cell
  - A/Pays de Loire/05590/2023
  - A/Albania/290270/2022 Egg

- Potential CVVs:
  - A/Serbia/500898/2023
  - A/Belgium/G0261/2023
  - A/South Africa/R03690/2023
  - A/New York/66/2022 Cell
  - A/Hong Kong/1538/2023

- Feb 2023
  - D493N

- Apr 2023
  - D408N

- May 2023
  - R299K, D408N

- Genetic changes:
  - D7N, D32E, I140M, P227H, N246S
  - D53N, N96S (+CHO), I192F, N378S
  - L42X, N122I, D225N

- HI fold changes:
  - > 8.0
  - < 2.0

- Other markers:
  - Potential CVVs
  - Clock human lineage

[Table showing virus strain information with genetic markers and changes]
B/Victoria

WHO Collaborating Centre reports

A full description of results generated by the London WHO Collaborating Centre at the WIC and used at the September 2023 WHO VCM, and previous ones, can be found at https://www.crick.ac.uk/partnerships/worldwide-influenza-centre/annual-and-interim-reports