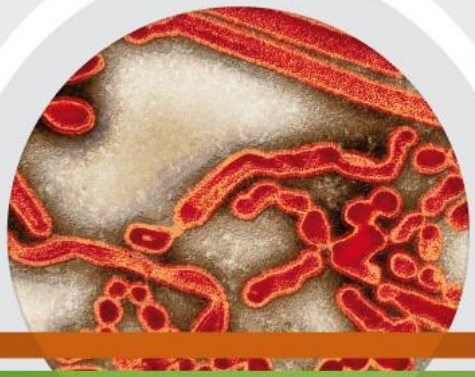


SURVEILLANCE REPORT

# Influenza virus characterisation

Summary Europe, December 2012


## Summary

During the 2012–13 season, A(H1N1)pdm09, A(H3N2) and B/Victoria and B/Yamagata lineage influenza viruses, with collection dates since 1 September 2012, have been detected in relatively low numbers in ECDC-affiliated countries. However, the situation varies from country to country:

- Type A and type B viruses are co-circulating in approximately equal proportions.
- A(H3N2) and A(H1N1)pdm09 viruses have been detected at comparable levels, with a recent rise in the proportion of A(H1N1)pdm09 viruses.
- A(H1N1)pdm09 viruses continued to show genetic drift from the vaccine virus, A/California/07/2009, but the vast majority remained antigenically similar to it.
- The vast majority of A(H3N2) viruses have been antigenically similar to A/Victoria/361/2011, the vaccine virus for the 2012–13 influenza season.
- B/Victoria lineage viruses fell within, and were antigenically similar to, reference cell-propagated viruses of the B/Brisbane/60/2008 genetic clade.
- Recent B/Yamagata-lineage viruses fell into two genetic clades: clade 2 represented by B/Estonia/55669/2012, and clade 3 represented by B/Wisconsin/1/2010 (the vaccine component for the 2012–13 influenza season). Clade 2 viruses have predominated over clade 3.


In December, packages containing viruses and/or clinical samples were received from Denmark, Germany, Norway, Sweden and the United Kingdom (England) by the WHO Collaborating Centre for Reference and Research on Influenza, based at the MRC National Institute for Medical Research in London. A summary of specimens received, with collection dates between 1 September and 31 December 2012, is shown in Table 1.

The proportions of influenza type A (54%) and type B (46%) viruses received were similar. For type A, H3N2 viruses have been received in slightly greater numbers than H1N1pdm09 viruses (ratio 3:2), but there has been a marked difference in levels of propagation: 16/19 (84%) for H3N2 and 2/12 (17%) for H1N1pdm09. Among influenza B receipts, viruses of the B/Yamagata and B/Victoria lineages were received at a ratio of approximately 3:1, and propagation rates were good: 19/21 (90%) and 6/6 (100%), respectively.


This report was prepared by Rod Daniels, Vicki Gregory and John McCauley on behalf of the Community Network of Reference Laboratories for Human Influenza in Europe (CNRL), under contract to the European Centre for Disease Prevention and Control (ECDC).

**Table 1. Summary of clinical samples and isolates received from ECDC-affiliated countries, with collection dates since 1 September 2012**

| MONTH | TOTAL RECEIVED | H1N1pdm09 | | H3N2 | | B Victoria lineage | | B Yamagata lineage | |
|---|---|---|---|---|---|---|---|---|---|
| Country | | Number received | Number propagated[1] | Number received | Number propagated[2] | Number received | Number propagated[1] | Number received | Number propagated[1] |
| **SEPTEMBER** | | | | | | | | | |
| Denmark | 2 | | | 2 | 2 | | | | |
| Norway | 2 | | | | | | | 2 | 2 |
| **OCTOBER** | | | | | | | | | |
| Germany | 2 | | | 1 | 1 | | | 1 | 1 |
| Norway | 12 | 6 | 0 | | | 1 | 1 | 5 | 4 |
| Sweden | 2 | 1 | 1 | 1 | 1 | | | | |
| United Kingdom | 8 | 1 | 0 | 2 | 2 | 2 | 2 | 3 | 3 |
| **NOVEMBER** | | | | | | | | | |
| Denmark | 6 | | | 1 | 1 | 2 | 2 | 3 | 3 |
| Germany | 5 | | | 4 | 1 | | | 1 | 1 |
| Norway | 7 | 3 | 0 | | | | | 4 | 3 |
| Sweden | 7 | 1 | 1 | 5 | 5 | | | 1 | 1 |
| United Kingdom | 3 | | | 2 | 2 | 1 | 1 | | |
| **DECEMBER** | | | | | | | | | |
| Denmark | 2 | | | 1 | 1 | | | 1 | 1 |
| | 58 | 12 | 2 | 19 | 16 | 6 | 6 | 21 | 19 |
| | | | 20.7% | | 32.8% | | 10.3% | | 36.2% |

1. Propagated to sufficient titre to perform HI assay
2. Propagated to sufficient titre to perform HI assay in presence of 20nM oseltamivir

# Influenza A(H1N1)pdm09 virus analyses

The results of an HI assay carried out to examine three A(H1N1)pdm09 viruses received from Sweden since the November report are shown in Table 2. A/Stockholm/32/2012 and A/Stockholm/34/2012 showed good reactivity with post-infection ferret antisera raised against the panel of reference viruses, including antiserum raised against the vaccine virus, A/California/7/2009. A/Stockholm/27/2012 showed at least fourfold reduced reactivity, with eight of nine antisera in the panel, including the antiserum raised against the vaccine strain. Sequence analysis of the HA gene of A/Stockholm/27/2012 revealed an amino acid polymorphism at position G155E=G in the glycoprotein, which has previously been associated with low reactivity in HI assays, and commonly results from propagation of viruses in certain tissue culture cells.

Phylogenetic analysis of the HA gene of representative viruses (Figure 1) shows that the most recently detected H1N1 viruses from EU/EEA countries, including the three recent viruses from Sweden analysed here, cluster within Groups 6 and 7, as previously described, with A/Stockholm/27/2012 falling in Group 7.

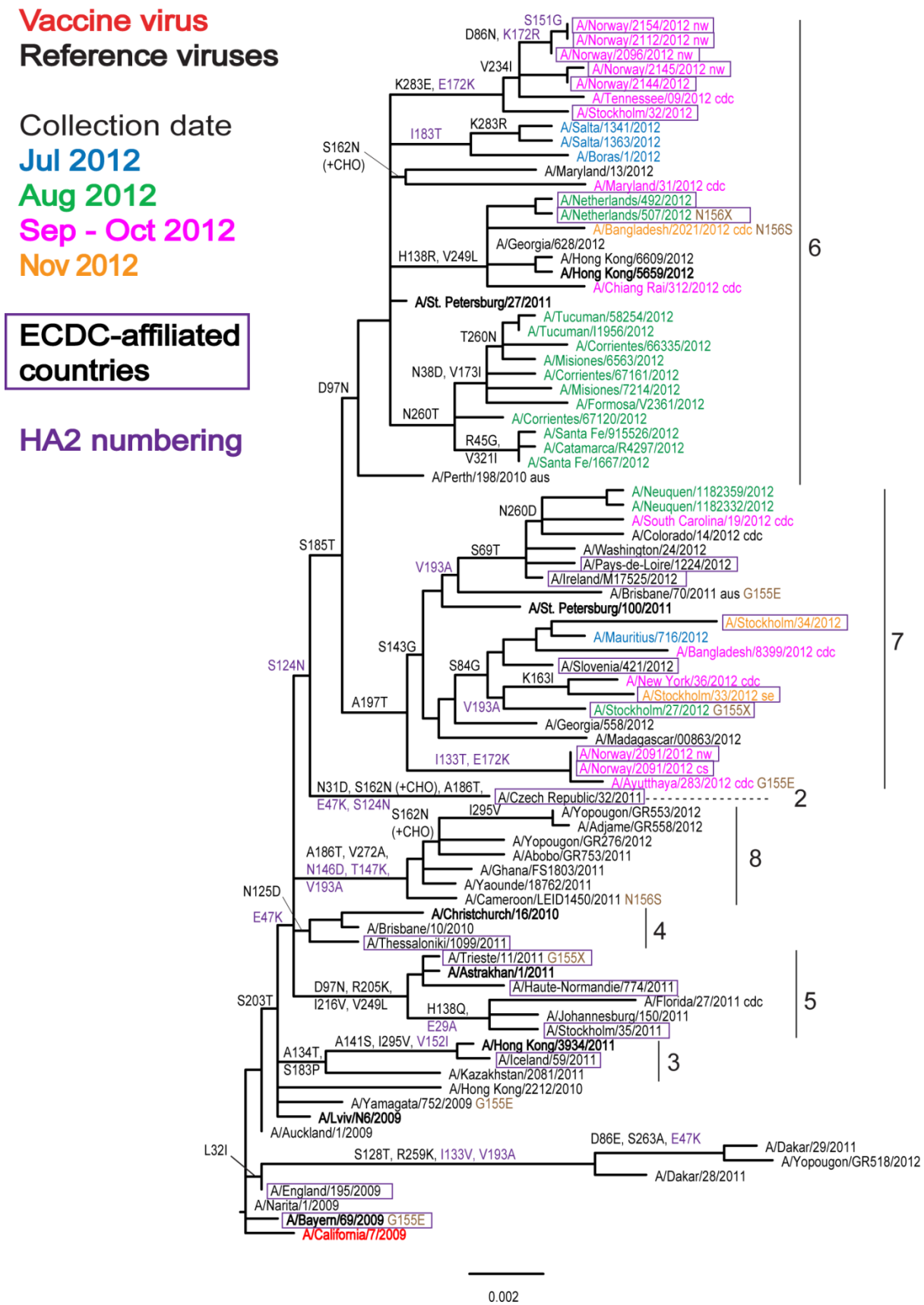
**Table 2. Antigenic analysis of A(H1N1)pdm09 viruses by HI**

| | | | | Haemagglutination inhibition titre[1] | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Post infection ferret sera | | | | | | | | | |
| Viruses | Genetic Group | Collection date | Passage History | A/Cal 7/09 F29/11 | A/Bayern 69/09 F11/11 | A/Lviv N6/09 C4/34/09 | A/C'church 16-Oct F30/10 Group 4 | A/HK 3934/11 F21/11 Group 3 | A/Astrak 1/11 F22/11 Group 5 | A/St. P 27/11 F23/11 Group 6 | A/St. P 100/11 F24/11 Group 7 | A/HK 5659/12 F30/12 Group 6 | |
| *REFERENCE VIRUSES* | | | | | | | | | | | | | |
| A/California/7/2009 | | 2009-04-09 | E1/E2 | 640 | 640 | 640 | 320 | 640 | 320 | 320 | 640 | 640 | |
| A/Bayern/69/2009 | | 2009-07-01 | MDCK5/MDCK1 | 40 | 160 | 80 | 40 | < | 40 | 40 | < | 40 | |
| A/Lviv/N6/2009 | | 2009-10-27 | MDCK4/SIAT1/MDCK2 | 320 | 1280 | 640 | 160 | 80 | 80 | 160 | 160 | 320 | |
| A/Christchurch/16/2010 | 4 | 2010-07-12 | E2/E2 | 640 | 1280 | 1280 | 5120 | 1280 | 1280 | 640 | 2560 | 2560 | |
| A/Hong Kong/3934/2011 | 3 | 2011-03-29 | MDCK2/MDCK3 | 160 | 80 | 160 | 320 | 640 | 640 | 320 | 640 | 640 | |
| A/Astrakhan/1/2011 | 5 | 2011-02-28 | MDCK1/MDCK5 | 640 | 320 | 640 | 640 | 1280 | 640 | 640 | 2560 | 1280 | |
| A/St. Petersburg/27/2011 | 6 | 2011-02-14 | E1/E2 | 640 | 640 | 640 | 640 | 1280 | 1280 | 640 | 2560 | 2560 | |
| A/St. Petersburg/100/2011 | 7 | 2011-03-14 | E1/E2 | 1280 | 640 | 1280 | 640 | 1280 | 1280 | 1280 | 5120 | 2560 | |
| A/Hong Kong/5659/2012 | 6 | 2012-05-21 | MDCK4 | 640 | 320 | 640 | 640 | 1280 | 1280 | 1280 | 2560 | 2560 | |
| *TEST VIRUSES* | | | | | | | | | | | | | |
| A/Stockholm/27/2012 | 7 | 2012-08-15 | MDCK3/MDCK1 | 80 | 320 | 160 | 80 | 80 | 80 | 160 | 160 | 160 | G155X |
| A/Stockholm/32/2012 | 6 | 2012-10-25 | MDCK2/MDCK1 | 1280 | 640 | 1280 | 1280 | 1280 | 1280 | 1280 | 2560 | 5120 | |
| A/Stockholm/34/2012 | 7 | 2012-11-06 | MDCK1/MDCK1 | 320 | 320 | 320 | 320 | 640 | 640 | 640 | 1280 | 1280 | |

Sequences in phylogenetic trees — Vaccine

1. < = <40

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



# Influenza A(H3N2) virus analyses

The majority of H3N2 viruses were successfully propagated but, as described before, A(H3N2) viruses have continued to be difficult to characterise antigenically by HI assay due to variable agglutination of red blood cells from guinea pigs, turkeys and humans. Influenza A(H3N2) viruses were analysed by HI assay using guinea pig red blood cells in the presence of 20nM oseltamivir, added to circumvent the NA-mediated binding of H3N2 viruses to the red blood cells (Lin et al. 2010). HI results are shown in Table 3: all viruses reacted poorly with the post-infection ferret antiserum raised against the currently recommended egg-grown vaccine virus, A/Victoria/361/2011, compared with the homologous titre. Generally, the test viruses also reacted poorly with antisera raised against other reference/previous vaccine viruses propagated in eggs (A/Perth/16/2009, A/Victoria/208/2009, A/Iowa/19/2010 or A/Hawaii/22/2012). However, many reacted somewhat better with antisera raised against egg-propagated A/Texas/50/2012, compared with the homologous titre.

The test viruses reacted well with sera raised against reference viruses exclusively propagated in cells (A/Alabama/5/2010, A/Stockholm/18/2011, A/Berlin/93/2011, A/Athens/112/2012, and an isolate of A/Victoria/361/2011), compared with the titres against the homologous viruses (Table 3).

Phylogenetic analysis of the HA gene sequences of representative viruses has been carried out (Figure 2). Viruses from the EU/EEA collected since 1 September 2012 have HA genes that fall into the A/Victoria/208 clade, genetic groups 5 and 3, subgroup 3C.

The HA1 amino acid substitutions that are associated with each of these groupings are:

- Subgroup 3C: **S45N**, **T48I**, **A198S**, **V223I** and **N312S**, e.g. the prototype vaccine virus A/Victoria/361/2011, with the great majority of viruses also carrying the substitutions **Q33R** and **N278K** (e.g. A/Berlin/93/2011) and a significant number of viruses also carrying **N145S** substitution.
- Group 5: **D53N**, **Y94H**, **I230V** and **E280A**, e.g. A/Alabama/5/2010 and A/Norway/1443/2012.

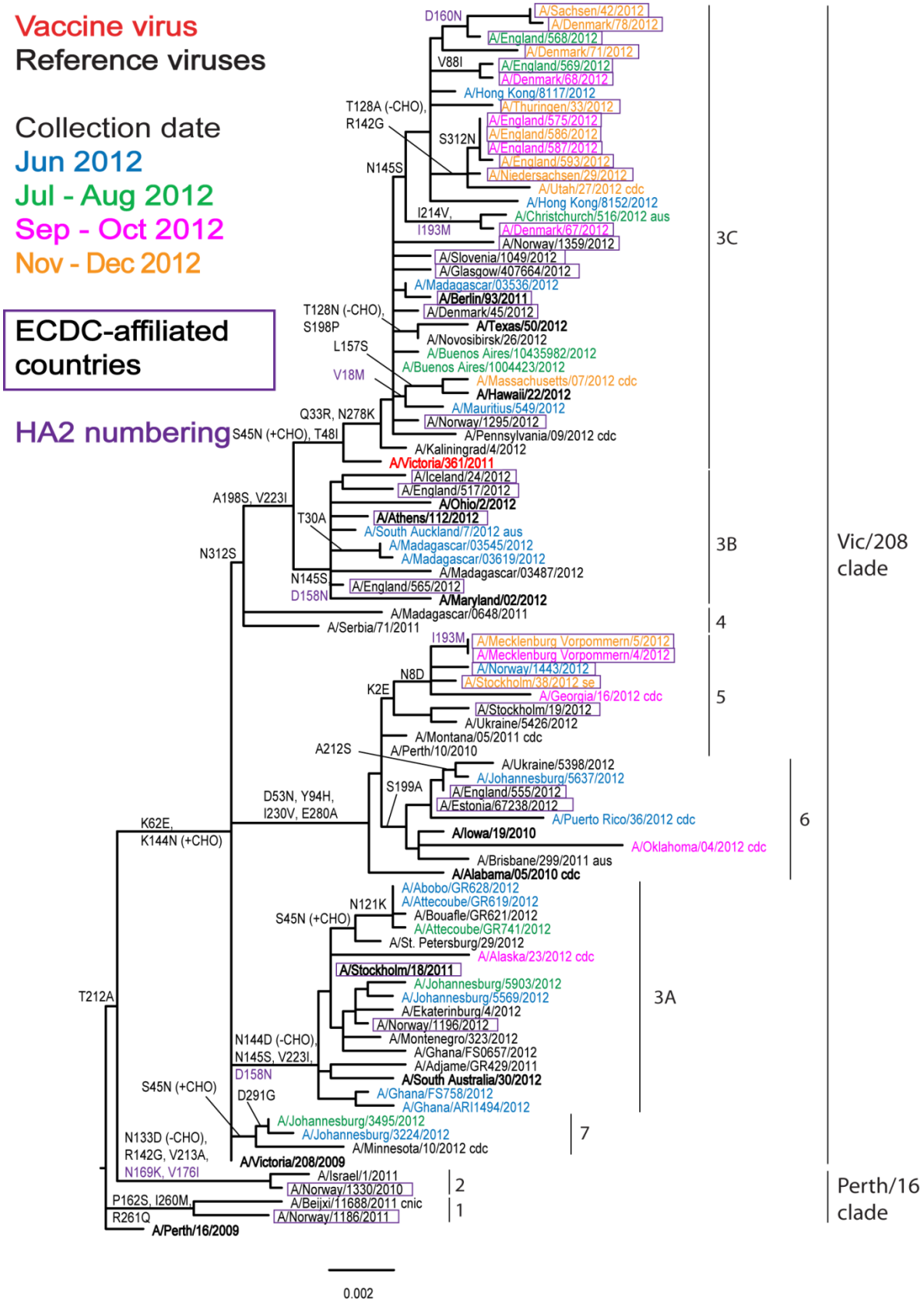
**Table 3. Antigenic analysis of A(H3N2) viruses by HI (guinea pig RBC with 20nM oseltamivir)**

| | | | | Haemagglutination inhibition titre[1] | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Post infection ferret sera | | | | | | | | | | |
| Viruses | Genetic group | Collection Date | Passage History | A/Perth 16/09 F35/11 | A/Vic 208/09 F7/10 | A/Ala 5/10 group 5 | A/Stock 18/11 group 3A | A/Iowa 19/10 group 6 | A/Vic 361/11 Egg F35/12 group 3C | A/Berlin 93/11 T/C F11/12 group 3C | A/Vic 361/11 T/C F14/12 group 3C | A/Athens 112/12 F16/12 group 3B | A/Texas 50/12 F36/12 group 3C | A/Hawaii 22/12 F37/12 group 3C |
| *REFERENCE VIRUSES* | | | | | | | | | | | | | | |
| A/Perth/16/2009 | | 2009-07-04 | E3/E2 | 1280 | 80 | 320 | 160 | 160 | 160 | 320 | 640 | 640 | 320 | 160 |
| A/Victoria/208/2009 | | 2009-06-02 | E3/E1 | 1280 | 5120 | 1280 | 2560 | 5120 | 2560 | 5120 | 2560 | 2560 | 5120 | 5120 |
| A/Alabama/5/2010 | 5 | 2010-07-13 | MK1/C2/SIAT2 | 40 | 40 | 80 | 80 | 80 | 80 | 160 | 320 | 160 | 320 | 80 |
| A/Stockholm/18/2011 | 3A | 2011-03-28 | MDCK2/SIAT5 | 40 | 40 | 40 | 160 | 80 | 80 | 320 | 320 | 160 | 320 | 160 |
| A/Iowa/19/2010 | 6 | 2010-12-30 | E3/E2 | 320 | 640 | 640 | 1280 | 1280 | 640 | 1280 | 1280 | 1280 | 2560 | 640 |
| A/Victoria/361/2011 | 3C | 2011-10-24 | E3/E2 | 320 | 640 | 320 | 160 | 640 | 2560 | 640 | 640 | 160 | 2560 | 1280 |
| A/Berlin/93/2011 | 3C | 2011-12-07 | NVD3/SIAT5 | 160 | 80 | 160 | 320 | 160 | 320 | 640 | 640 | 320 | 640 | 320 |
| A/Victoria/361/2011 | 3C | 2011-10-24 | MDCK2/SIAT2 | 80 | 80 | 160 | 320 | 160 | 160 | 640 | 640 | 320 | 640 | 320 |
| A/Athens/112/2012 | 3B | 2012-02-01 | SIAT8 | 40 | 80 | 80 | 160 | 80 | 160 | 320 | 320 | 640 | 640 | 320 |
| A/Texas/50/2012 | 3C | 2012-04-15 | E5/E1 | 320 | 640 | 640 | 1280 | 640 | 1280 | 1280 | 1280 | 1280 | 2560 | 1280 |
| A/Hawaii/22/2012 | 3C | 2012-07-09 | E4/E1 | 320 | 640 | 640 | 640 | 1280 | 640 | 1280 | 1280 | 1280 | 2560 | 5120 |
| *TEST VIRUSES* | | | | | | | | | | | | | | |
| A/Stockholm/12-16700/2012 | | unknown | MDCK2/SIAT1 | 40 | 80 | 80 | 320 | 160 | 160 | 640 | 640 | 640 | 640 | 320 |
| A/Stockholm/30/2012 | | 2012-07-12 | MDCK1/SIAT1 | 40 | 80 | 160 | 160 | 160 | 160 | 320 | 640 | 320 | 640 | 160 |
| A/Stockholm/29/2012 | | 2012-07-27 | MDCK1/SIAT1 | 40 | 80 | 80 | 160 | 80 | 160 | 320 | 320 | 320 | 640 | 320 |
| A/England/568/2012 | 3C | 2012-08-18 | SIAT1/SIAT3 | < | < | 40 | 80 | 40 | 80 | 160 | 160 | 160 | ND | ND |
| A/England/569/2012 | 3C | 2012-08-18 | SIAT2/SIAT2 | < | 40 | < | 160 | 40 | 80 | 160 | 160 | 320 | ND | ND |
| A/Denmark/68/2012 | 3C | 2012-09-25 | Cx/SIAT1 | 40 | 80 | 80 | 320 | 160 | 160 | 640 | 640 | 320 | 640 | 320 |
| A/Denmark/67/2012 | 3C | 2012-09-27 | MDCK3/SIAT1 | 40 | 80 | 80 | 320 | 160 | 160 | 640 | 640 | 320 | 640 | 320 |
| A/Stockholm/31/2012 | | 2012-10-17 | MDCK2/SIAT1 | 80 | 80 | 320 | 320 | 320 | 320 | 640 | 640 | 640 | 1280 | 320 |
| A/England/575/2012 | 3C | 2012-10-23 | SIAT1/SIAT1 | 40 | 80 | 80 | 320 | 160 | 160 | 320 | 160 | 160 | 640 | 320 |
| A/Mecklenburg Vorpommern/4/2012 | 5 | 2012-10-30 | C2/SIAT2 | 40 | 40 | 80 | 160 | 80 | 80 | 320 | 320 | 320 | 320 | 160 |
| A/England/587/2012 | 3C | 2012-10-31 | SIAT1/SIAT1 | < | 80 | 40 | 160 | 40 | 80 | 320 | 320 | 160 | 320 | 160 |
| A/England/586/2012 | 3C | 2012-11-02 | SIAT1/SIAT1 | < | 80 | 80 | 160 | 80 | 80 | 320 | 320 | 160 | 320 | 160 |
| A/Niedersachsen/29/2012 | 3C | 2012-11-05 | C1/SIAT1 | 40 | 40 | 80 | 160 | 80 | 80 | 640 | 320 | 640 | 640 | 320 |
| A/Stockholm/38/2012 | 5 | 2012-11-05 | MDCK1/SIAT1 | 80 | 80 | 160 | 320 | 320 | 160 | 640 | 640 | 640 | 640 | 320 |
| A/Denmark/71/2012 | 3C | 2012-11-08 | MDCK4/SIAT1 | 40 | 80 | 80 | 320 | 160 | 160 | 640 | 640 | 320 | 640 | 320 |
| A/Stockholm/39/2012 | | 2012-11-08 | MDCK1/SIAT1 | 40 | 80 | 80 | 320 | 160 | 80 | 320 | 320 | 320 | 640 | 160 |
| A/England/593/2012 | 3C | 2012-11-12 | SIAT1/SIAT1 | < | 80 | 40 | 160 | 80 | 80 | 320 | 320 | 160 | 320 | 160 |
| A/Stockholm/40/2012 | | 2012-11-14 | MDCK1/SIAT1 | 80 | 80 | 160 | 160 | 160 | 160 | 320 | 640 | 320 | 640 | 160 |
| A/Stockholm/37/2012 | | 2012-11-15 | MDCK0/SIAT1 | 80 | 160 | 160 | 320 | 320 | 160 | 640 | 640 | 320 | 1280 | 320 |
| A/Stockholm/36/2012 | | 2012-11-16 | MDCK1/SIAT1 | 40 | 80 | 80 | 320 | 160 | 160 | 640 | 640 | 320 | 640 | 320 |
| A/Denmark/78/2012 | 3C | 2012-12-05 | MDCK2/SIAT1 | 40 | 80 | 40 | 160 | 80 | 80 | 320 | 320 | 320 | 320 | 160 |
| | | | | | | | | | Vaccine | | Vaccine | | | |

Sequences in phylogenetic trees

1. < = <40, ND = Not Done

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



# Influenza B virus analyses

## B/Victoria-lineage virus

Table 4 shows the results of HI analyses for viruses of the B/Victoria lineage. All test viruses showed low reactivity with antisera raised against the egg-propagated virus B/Brisbane/60/2008, the influenza vaccine component for the 2010–11 season, compared with the titre against the homologous virus. The test viruses also showed a similar reduction in titre with antiserum raised against the egg-propagated virus B/Malta/636714/2011. The test viruses reacted better with antisera raised against reference viruses genetically closely related to B/Brisbane/60/2008, but propagated in cells; these post-infection ferret antisera were raised against B/Paris/1762/2008, B/Hong Kong/514/2009 and B/Odessa/3886/2010.

Phylogenetic analysis of the HA genes of representative B/Victoria lineage viruses is shown in Figure 3. All recently collected viruses received from EU and EEA laboratories carried HA genes that fell into clade 1, with only a small number of amino acid substitutions, compared with the HA of the previously used vaccine virus, B/Brisbane/60/2008.

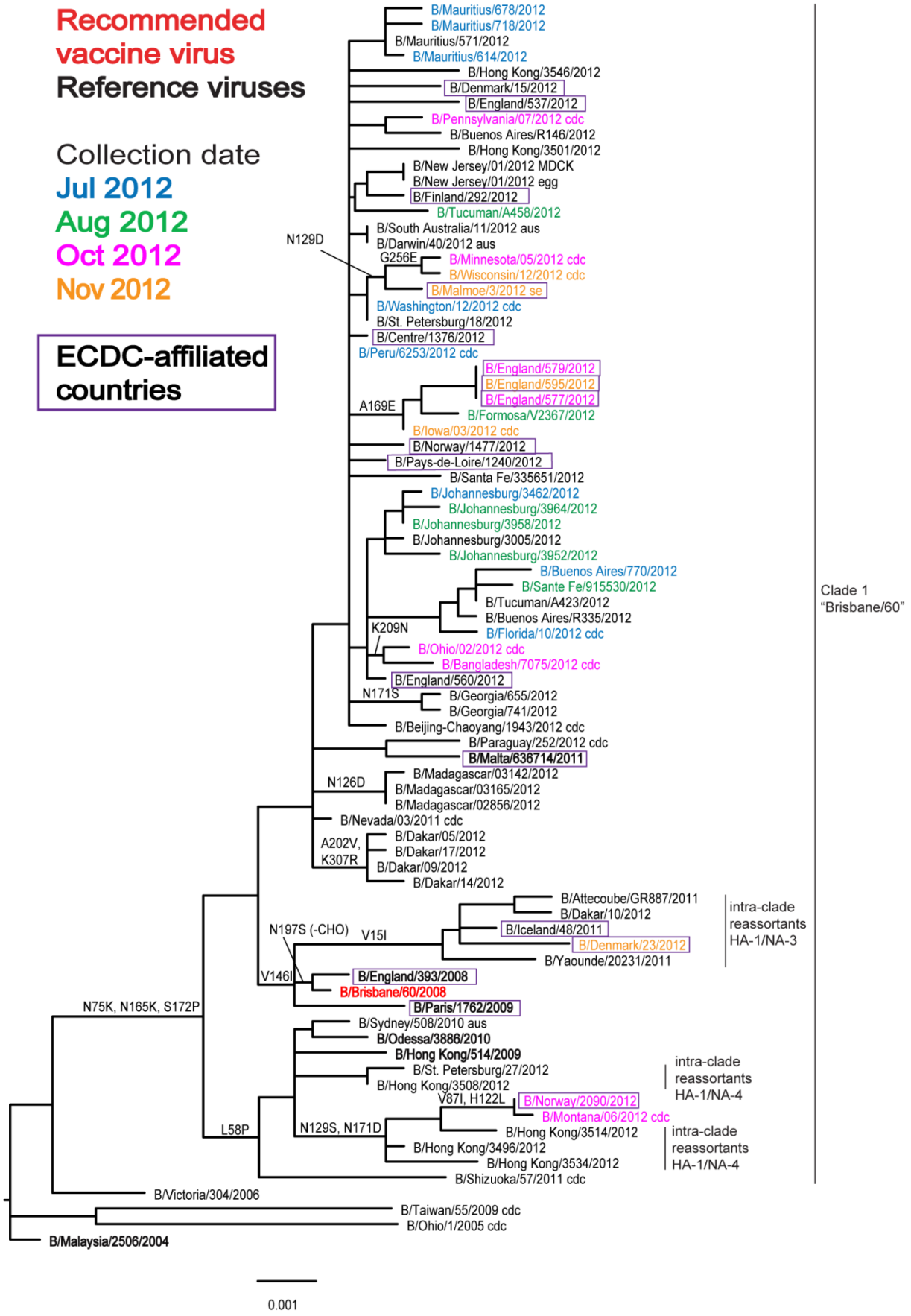
**Table 4. Antigenic analysis of influenza B viruses (Victoria lineage) by HI**

| | | | Haemagglutination inhibition titre[1] | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Post infection ferret sera | | | | | | |
| Viruses | Collection date | Passage History | B/Bris[2] 60/08 Sh 523 | B/Mal 2506/04 F28/05 | B/England 393/08 F05/11 | B/Bris 60/08 F22/12 | B/Paris 1762/08 F17/11 | B/HK 514/09 F13/10 | B/Odessa 3886/10 F19/11 | B/Malta 636714/11 F33/11 |
| *REFERENCE VIRUSES* | | | | | | | | | | |
| B/Malaysia/2506/2004 | 2004-12-06 | E3/E6 | 640 | 320 | 10 | 40 | < | < | < | 40 |
| B/England/393/2008 | 2008-08-29 | E1/E1 | 2560 | 80 | 320 | 640 | 40 | 40 | 40 | 640 |
| B/Brisbane/60/2008 | 2008-08-04 | E4/E3 | 5120 | 160 | 320 | 640 | 40 | 40 | 80 | 640 |
| B/Paris/1762/2008 | 2009-02-09 | C2/MDCK2 | 2560 | 20 | 20 | 40 | 40 | 40 | 40 | 20 |
| B/Hong Kong/514/2009 | 2009-10-11 | MDCK1/MDCK2 | 2560 | < | 10 | 20 | 80 | 80 | 160 | 20 |
| B/Odessa/3886/2010 | 2010-03-19 | MDCK2/MDCK1 | 5120 | < | 10 | 20 | 80 | 80 | 160 | 20 |
| B/Malta/636714/2011 | 2011-03-07 | E4/E1 | 2560 | 160 | 320 | 640 | 40 | 40 | 40 | 640 |
| *TEST VIRUSES* | | | | | | | | | | |
| B/Norway/2090/2012 | 2012-10-02 | LLC-MK2/ MDCK1/MDCK1 | 2560 | < | 10 | 10 | 80 | 80 | 80 | 20 |
| B/England/577/2012 | 2012-10-22 | SIAT1/MDCK1 | 2560 | 10 | 10 | 20 | 40 | 40 | 40 | 10 |
| B/England/579/2012 | 2012-10-30 | SIAT1/MDCK1 | 2560 | < | 10 | 20 | 80 | 80 | 80 | 20 |
| B/Denmark/23/2012 | 2012-11-05 | MDCK3/MDCK1 | 2560 | < | 10 | 20 | 80 | 40 | 80 | 20 |
| B/Denmark/22/2012 | 2012-11-07 | MDCK2/MDCK1 | 2560 | < | 10 | 20 | 80 | 40 | 80 | 40 |
| B/England/595/2012 | 2012-11-09 | SIAT2/MDCK1 | 2560 | < | 10 | 20 | 80 | 40 | 40 | 20 |

Sequences in phylogenetic trees

1. < = <10; 2. hyperimmune sheep serum

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



# B/Yamagata-lineage viruses

Table 5 shows the results of HI analyses of B/Yamagata lineage viruses. The results show heterogeneity with test viruses. There is variable reactivity with the antisera raised against the vaccine virus B/Wisconsin/1/2010 and with antisera raised against viruses in the same genetic group, group 3, (B/Stockholm/12/2011, B/Serbia/1894/2011 and B/Novosibirsk/1/2012). Of the five viruses in group 3, three showed low reactivity with the antisera raised against B/Wisconsin/1/2010. The results for test viruses with antisera raised against viruses in genetic group 2 (B/Estonia/55669/2011 and B/Hong Kong/3577/2012) better correlated with the genetic group of the HA, but not exclusively so. Two of the five viruses from genetic group 3 showed good reactivity with antiserum raised against B/Estonia/55669/2011 and three of the five showed good reactivity with antiserum raised against B/Hong Kong/3577/2012. Antisera raised against the tissue culture propagated viruses in genetic group 3 also showed variable reactivity with genetic group 3 test viruses.

All viruses from genetic group 2 showed good reactivity with post-infection antisera raised against B/Estonia/55669/2011 and B/Hong Kong/3577/2012 and generally low reactivity with sera raised against B/Wisconsin/1/2010, B/Stockholm/12/2011, B/Serbia/1894/2011 and B/Novosibirsk/1/2012, the genetic group 3 reference viruses.

Figure 4 shows a phylogenetic analysis of the HA genes of representative B/Yamagata lineage viruses. The phylogeny clearly shows the two genetic clades: clade 3, represented by the vaccine virus B/Wisconsin/1/2010 and reference viruses B/Bangladesh/3333/2007 and B/Stockholm/12/2011; and clade 2, represented by the reference viruses B/Brisbane/3/2007 and B/Estonia/55669/2011. The two clades are differentiated by substitutions at HA1 residues 48, 108, 150, 165, 181 and 229. The HA genes of viruses of clade 2 encode **K48**, **A108**, **S150**, **N165**, **A181** and **G229**; the HA genes of viruses in clade 3 encode **R48**, **P108**, **I150**, **Y165**, **T181** and **D229**. However, the phylogenetic analysis does not explain the observed heterogeneity of the HI results. There appears to be an increased number of viruses with recent collection dates that have HA genes falling into genetic group 2, but the numbers analysed remain small.

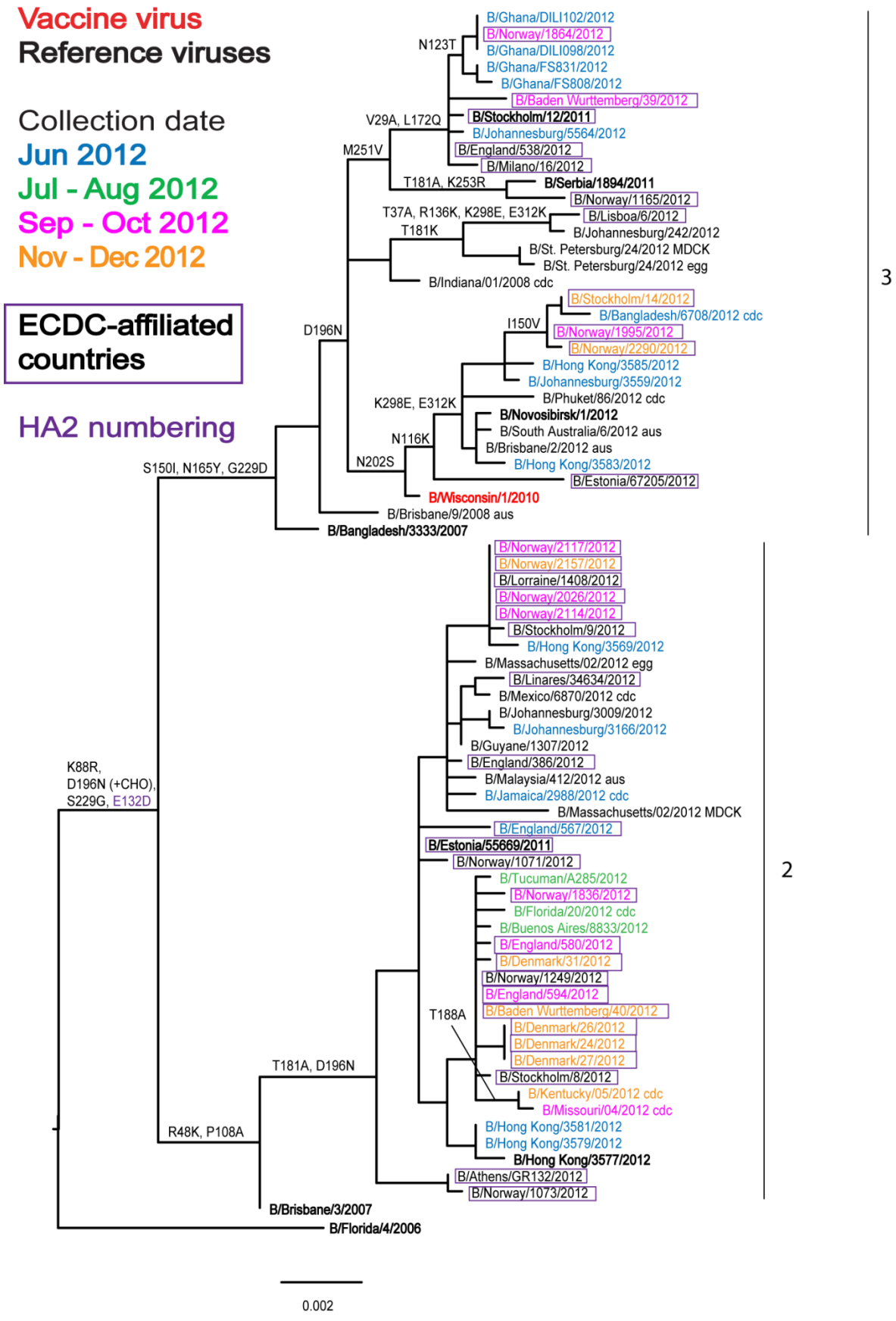
**Table 5. Antigenic analysis of influenza B viruses (Yamagata lineage) by HI**

| | | | | Haemagglution inhibition titre | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Post infection ferret sera | | | | | | | | | |
| Viruses | Genetic group | Collection date | Passage History | B/Fl[3] 4/06 SH479 Group 1 | B/Fl[1] 4/06 F01/10 Group 1 | B/Bris[2] 3/07 F21/12 Group 2 | B/Wis[2] 1/10 F26/10 Group 3 | B/Stock[2] 12/11 F12/12 Group 3 | B/Estonia[2] 55669/11 F32/12 Group 2 | B/Serbia[2] 1894/11 F25/11 Group 3 | B/Stock[2] 12/11 T/C F8/12 Group 3 | B/Novo[2] 1/12 F31/12 Group 3 | B/HK[2] 3577/12 F33/12 Group 2 |
| *REFERENCE VIRUSES* | | | | | | | | | | | | | |
| B/Florida/4/2006 | 1 | 2006-12-15 | E3/E3 | 5120 | 1280 | 1280 | 320 | 1280 | 160 | 10 | 640 | 80 | 640 |
| B/Brisbane/3/2007 | 2 | 2007-09-03 | E2/E1 | 5120 | 2560 | 1280 | 640 | 1280 | 320 | 10 | 1280 | 80 | 640 |
| B/Wisconsin/1/2010 | 3 | 2007-08-07 | E3/E2 | 5120 | 1280 | 1280 | 640 | 1280 | 80 | 40 | 640 | 160 | 160 |
| B/Stockholm/12/2011 | 3 | 2007-08-07 | E4/E1 | 2560 | 640 | 160 | 320 | 640 | 20 | 10 | 320 | 40 | 40 |
| B/Estonia/55669/2011 | 2 | 2011-03-14 | MDCK1/MDCK1 | 2560 | 160 | 80 | 20 | 80 | 320 | 20 | 40 | 80 | 640 |
| B/Serbia/1894/2011 | 3 | 2011-03-08 | MDCK1/MDCK4 | 2560 | 320 | 160 | 80 | 320 | 320 | 160 | 160 | 320 | 320 |
| B/Stockholm/12/2011 | 3 | 2011-03-28 | Cx/MDCK2 | 1280 | 160 | 80 | 40 | 160 | 80 | 80 | 160 | 160 | 320 |
| B/Novosibirsk/1/2012 | 3 | 2012-02-14 | C2/MDCK2 | 1280 | 160 | 80 | 80 | 160 | 80 | 40 | 80 | 160 | 160 |
| B/Hong Kong/3577/2012 | 2 | 2012-06-13 | MDCK2/MDCK1 | 5120 | 160 | 80 | 10 | 80 | 320 | 10 | 20 | 40 | 640 |
| *TEST VIRUSES* | | | | | | | | | | | | | |
| B/Norway/1494/2012 | 2 | 2012-06-12 | MDCK1/MDCK1 | 5120 | 160 | 160 | 20 | 160 | 640 | 40 | 40 | 80 | 1280 |
| B/Norway/1836/2012 | 2 | 2012-09-07 | MDCK2/MDCK1 | 5120 | 160 | 160 | 20 | 160 | 1280 | 80 | 80 | 320 | 1280 |
| B/Norway/1864/2012 | 3 | 2012-09-16 | MDCK2/MDCK1 | 5120 | 160 | 160 | 40 | 320 | 160 | 160 | 160 | 640 | 640 |
| B/Norway/2026/2012 | 2 | 2012-10-09 | MDCK1/MDCK1 | 1280 | 160 | 80 | 20 | 160 | 320 | 20 | 20 | 80 | 640 |
| B/Norway/1995/2012 | 3 | 2012-10-11 | MDCK1/MDCK1 | 1280 | 320 | 160 | 160 | 320 | 320 | 160 | 320 | 320 | 640 |
| B/England/580/2012 | 2 | 2012-10-23 | SIAT1/MDCK1 | 2560 | 160 | 160 | 40 | 160 | 320 | 40 | 40 | 40 | 1280 |
| B/Baden Württemberg/39/2012 | 3 | 2012-10-25 | C1/MDCK1 | 2560 | 320 | 160 | 160 | 320 | 320 | 320 | 320 | 320 | 640 |
| B/Norway/2114/2012 | 2 | 2012-10-29 | MDCK1/MDCK1 | 1280 | 160 | 80 | 20 | 160 | 320 | 40 | 20 | 40 | 640 |
| B/Norway/2117/2012 | 2 | 2012-10-30 | MDCK1/MDCK1 | 1280 | 160 | 80 | 10 | 160 | 160 | 40 | 20 | 40 | 1280 |
| B/England/581/2012 | | 2012-10-30 | SIAT2/MDCK1 | 2560 | 160 | 160 | 20 | 80 | 320 | 20 | 20 | 40 | 640 |
| B/England/594/2012 | 2 | 2012-10-30 | SIAT1/MDCK1 | 640 | 80 | 40 | 10 | 20 | 160 | 10 | 10 | 20 | 640 |
| B/Stockholm/14/2012 | 3 | 2012-11-04 | MDCK1/MDCK1 | 320 | 160 | 80 | 40 | 320 | 20 | 10 | 40 | 40 | 40 |
| B/Norway/2157/2012 | 2 | 2012-11-04 | MDCK1/MDCK1 | 1280 | 160 | 80 | 10 | 80 | 160 | 40 | 20 | 40 | 640 |
| B/Baden Württemberg/40/2012 | 2 | 2012-11-05 | C1/MDCK1 | 1280 | 160 | 160 | 40 | 160 | 320 | 40 | 40 | 40 | 640 |
| B/Norway/2290/2012 | 3 | 2012-11-11 | MDCK2 | 320 | 160 | 80 | 20 | 160 | 10 | < | 40 | 20 | 40 |
| B/Denmark/24/2012 | 2 | 2012-11-15 | MDCK3/MDCK1 | 1280 | 80 | 80 | 10 | 40 | 160 | 10 | 20 | 40 | 640 |
| B/Denmark/26/2012 | 2 | 2012-11-26 | MDCK2/MDCK1 | 640 | 80 | 40 | 10 | 40 | 160 | 10 | 10 | 20 | 640 |
| B/Denmark/27/2012 | 2 | 2012-11-28 | MDCK1/MDCK1 | 1280 | 160 | 80 | 20 | 80 | 320 | 10 | 20 | 40 | 1280 |
| B/Denmark/31/2012 | 2 | 2012-12-06 | MDCK1/MDCK1 | 1280 | 160 | 80 | 10 | 40 | 160 | 10 | 10 | 20 | 640 |

Sequences in phylogenetic trees — Vaccine

1. < = <40; 2. < =<10 ; 3. hyperimmune sheep serum

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



# Influenza A(H3N2)v virus

As we have described previously, on 3 August the United States CDC issued a Health Advisory describing an increase in the number of influenza A(H3N2)v infections in three US states. The US CDC has prepared further background information and has provided updated case counts. As of 1 December 2012, 308 cases have been confirmed. Antigenic and genetic characterisation of H3N2v viruses has been described by Lindstrom et al., 2012. The virus was characterised as being antigenically distinct from currently circulating human seasonal influenza viruses and to be a reassortant virus, with seven genes from swine influenza 'triple reassortant' H3N2 viruses and the M gene from influenza A(H1N1)pdm09 virus.

Risk assessments for these A(H3N2)v viruses, as a risk to public health, have been posted by the United States CDC and ECDC.

A description of results generated by the WHO Collaborating Centre for Reference and Research on Influenza, based at the MRC National Institute for Medical Research in London, and evaluated at the WHO Vaccine Composition Meetings held at WHO Geneva on 20–22 February 2012 and Beijing, China, on 17–19 September 2012, can be found at http://www.nimr.mrc.ac.uk/documents/about/interim-report-feb-2012.pdf and http://www.nimr.mrc.ac.uk/documents/about/Interim_Report_September_2012_2.pdf.

# Note on the figures

The phylogenetic trees were constructed using RAxML and drawn using FigTree. The bars indicate the proportion of nucleotide changes in the sequence. 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 ECDC countries are highlighted within boxes. Sequences for some of the viruses from non-EU/EEA countries were recovered from GISAID, and we acknowledge all laboratories who submitted sequences directly to the London WHO CC.