# Antigenic and genetic characteristics of H5N1 viruses and candidate H5N1 vaccine viruses developed for potential use as human vaccines

**February 2008**

The development of representative H5N1 candidate vaccine viruses by the WHO Global Influenza Programme is being conducted as one component of the overall global strategy for pandemic preparedness. This summary provides an update on the characterization of H5N1 viruses circulating in birds, those that have caused human infections and the current status of the development of candidate H5N1 vaccine viruses. This information may be used to guide national authorities as regards decisions on procurement of H5N1 vaccines.

H5N1 vaccines are continuing to be developed by manufacturers using clade 1 and clade 2 viruses that have been modified by reverse genetics. Clinical trials have been conducted or are under way in several countries and stockpiles of clade 1 and clade 2 vaccines are being acquired by a number of countries (www.who.int/entity/vaccine_research/diseases/ari/final_report_stockpile_meeting.pdf ). Because it is not known if the next influenza pandemic will be caused by H5N1 viruses or which of the clades or subclades of H5N1 would be responsible, should one occur, clinical trials using both clade 1 and clade 2 viruses should continue as an essential element in pandemic preparedness, to maximize data available on priming, cross-reactivity and cross-protection by vaccine viruses from different clades and subclades.

Companies are recommended to consult individual national authorities on the specific H5N1 viruses to be used for preparing experimental pilot lots and stockpiles of H5N1 vaccines. Decisions should be based on the epidemiology and geographical distribution of the circulating H5N1 viruses that are described below.

Comparisons of the candidate H5N1 vaccine viruses developed from clade 1 and clade 2 viruses with respect to immunogenicity and cross-reactivity and their relationship to newly emerging H5N1 viruses are ongoing, and will be reported periodically by WHO.

## Genetic characteristics of H5N1 viruses

A revised nomenclature for phylogenetic relationships among the haemagglutinin (HA) genes of H5N1viruses was devised by consultation among representatives of OIE, FAO and WHO (http://www.who.int/csr/disease/avian_influenza/guidelines/nomenclature/en/index.html ) (Figure 1). The HA sequences of the majority of H5N1 viruses circulating in avian species separated into ten distinct phylogenetic clades. Clade 1 viruses have caused human infections in Cambodia, China Hong Kong Special Administrative Region , Thailand and Viet Nam and have been recently detected in poultry in Viet Nam and

Cambodia. Clade 2.1 viruses have continued to circulate in poultry and caused human infections in Indonesia. Clade 2.2 viruses have the most geographically diverse distribution and have caused outbreaks in birds in over 60 countries in Africa, Asia and Europe with human infections in Azerbaijan, China, Djibouti, Egypt, Iraq, Nigeria, Pakistan and Turkey. Some recent clade 2.2 viruses have diverged genetically from reference strains. Clade 2.3 viruses are genetically diverse and continue to circulate in birds in a number of Asian countries. Within clade 2.3, clade 2.3.2 and clade 2.3.4 viruses continue to circulate in poultry in Asia; clade 2.3.4 viruses have been responsible for human infections in China, Lao People's Democratic Republic, Myanmar and Viet Nam. Viruses from other clades have been sporadically detected in Asia.

**Antigenic characteristics of H5N1 viruses**

Haemagglutination inhibition tests of available H5N1 viruses demonstrate that current vaccine candidates continue to provide good antigenic coverage of most isolates within corresponding clades. However, some viruses within clades 1, 2.2, and 2.3 show evidence of antigenic heterogeneity (Table 1). Newer clade 1 viruses (e.g. A/duck/Vietnam/NCVD16/2007) are antigenically distinguishable from vaccine candidate viruses A/Vietnam/1194/2004 and A/Vietnam/1203/2004. The majority of clade 2.1 viruses tested, including A/duck/Hunan/795/2002, were antigenically similar to A/Indonesia/5/2005. While the majority of available clade 2.2 viruses were antigenically similar to currently recommended vaccine candidates, some recently characterized clade 2.2 viruses from Egypt show evidence of antigenic heterogeneity. Viruses within clade 2.3.2 are antigenically distinguishable from other 2.3 clades. Clade 2.3.4 viruses remain antigenically similar to existing vaccine candidates.

**Potential H5N1 vaccine viruses**

On the basis of the geographical spread, the epidemiology, and the antigenic and genetic properties of the H5N1 viruses, national authorities may recommend the use of one or more of the H5N1 candidate vaccine viruses listed in table 2 for pilot lot vaccine production and subsequent stockpiling of vaccines, should relevant national policies exist.

Additional H5N1 candidate vaccine viruses are being developed as the viruses continue to evolve, and will be announced as they become available. Institutions, companies and others interested in pandemic vaccine development, who wish to receive these prototype viruses, should contact the WHO Global Influenza Programme at GISN@who.int or the institutions listed in announcements published at WHO web site http://www.who.int/csr/disease/avian_influenza/guidelinestopics/en/index5.html .

**Table 1. Antigenic properties of H5N1 viruses**

| | REFERENCE FERRET ANTISERA | | | | | | |
|---|---|---|---|---|---|---|---|
| **REFERENCE ANTIGENS** | CLADE | VN/1203 | IND/5 | MG/244 | II/33487 | ANH/1 | JP/HK |
| A/VIETNAM/1203/2004 (VN/1203) | 1 | **320** | 20 | <10 | 40 | - | - |
| A/INDONESIA/5/2005 (IND/5) | 2.1 | 10 | **640** | 80 | 40 | - | - |
| A/WHOOPER SWAN/MG/244/05 (MG/244) | 2.2 | 20 | 160 | **320** | 320 | - | - |
| A/CHICKEN/INDIA/NIV-33487/2006 (II/33487) | 2.2 | - | 320 | 320 | **320** | 20 | 80 |
| A/ANHUI/1/05 (ANH/1) | 2.3.4 | 40 | 320 | <10 | 20 | **640** | |
| A/JAPAN. WHITE EYE/HK/1038/06 (JP/HK) | 2.3.4 | 20 | 640 | 40 | 640 | 1280 | **1280** |
| **TEST ANTIGENS** | | | | | | | |
| A/THAILAND/676/05 | 1 | 160 | 20 | <10 | - | 40 | - |
| A/DUCK/VN/NCVD16/2007 | 1 | 40 | <10 | <10 | - | <10 | - |
| A/MUSCKOVY DUCK/VIETNAM/33/07 | 1 | 40 | 40 | - | - | <40 | - |
| A/DUCK/HUNAN/795/02 | 2.1 | 80 | 2560 | - | - | 40 | - |
| A/INDONESIA/6/05 | 2.1 | <10 | 640 | 80 | - | - | - |
| A/INDONESIA/CDC1031/07 | 2.1 | <10 | 640 | 160 | - | 160 | 10 |
| A/INDONESIA/CDC625L/06 (Medan) | 2.1 | 40 | 80 | 20 | 40 | <10 | 20 |
| A/TURKEY/65-596/06 | 2.2 | 160 | 2560 | 5120 | 2560 | 320 | 320 |
| A/EGRET/EGYPT/NAMRU-3-1162/06 | 2.2 | <10 | 80 | 640 | - | 10 | 40 |
| A/CHICKEN/EGYPT/9402NAMRU3/07 | 2.2 | <10 | 320 | 160 | 80 | 80 | - |
| A/CHICKEN/EGYPT/9403NAMRU3/07 | 2.2 | <10 | 160 | 40 | 80 | 40 | - |
| A/CHINESE POND HERON/HONG KONG/18/05 | 2.3.2 | 80 | 40 | - | - | <40 | - |
| A/COMMON MAGPIE/HONG KONG/5052/07 | 2.3.2 | 80 | 320 | - | - | <40 | - |
| A/ANHUI/2/05 | 2.3.4 | 10 | 160 | <10 | - | 320 | |
| A/DUCK/LAOS/3295/06 | 2.3.4 | 10 | 160 | <10 | - | 320 | 80 |
| A/HOUSE CROW/HK/719/2007 | 2.3.4 | <10 | 80 | <10 | 10 | 320 | 80 |
| A/LAOS/JP085/2007 | 2.3.4 | <10 | 80 | - | - | 160 | 40 |
| A/CHICKEN/VN/NCVD74/2007 | 2.3.4 | 20 | <10 | <10 | - | 40 | - |
| A/DUCK/VN/NCVD81/2007 | 2.3.4 | <10 | <10 | <10 | - | 80 | - |
| A/VIETNAM/HN31203I/2007 | 2.3.4 | <10 | <10 | <10 | - | 80 | - |
| A/CHICKEN/MALAYSIA/935/06 | 2.3.4 | <10 | 80 | <10 | 10 | 160 | 40 |

- not done

**Table 2. Status of H5N1 vaccine virus development as of 13 February 2008**

**Reassortants with completed regulatory approval**

| Virus | Clade | Institution* | Availability |
|---|---|---|---|
| A/Vietnam/1203/2004 | 1 | CDC and SJ/NIAID | Yes |
| A/Vietnam/1194/2004 | 1 | NIBSC | Yes |
| A/Indonesia/5/2005 | 2.1 | CDC | Requires Indonesia Government permission |
| A/Bar-headed goose/Qinghai/1A/2005 | 2.2 | SJ/NIAID | Yes |
| A/Whooper swan/Mongolia/244/2005 | 2.2 | SJ/NIAID | Yes |
| A/turkey/Turkey/1/2005 | 2.2 | NIBSC | Yes |
| A/Anhui/1/2005 | 2.3.4 | CDC | Yes |
| A/Japanese white-eye/Hong Kong/1038/2006 | 2.3.4 | SJ/NIAID | Yes |

**Reassortants prepared and awaiting regulatory approval**

| Virus | Clade | Institution* | Availability |
|---|---|---|---|
| A/Chicken/India/NIV33487/2006 | 2.2 | CDC/NIV | Pending |
| A/goose/Guiyang/337/2006 | 4 | SJ/NIAID | May 2008 |
| A/duck/Laos/3295/2006 | 2.3.4 | FDA | May 2008 |
| A/Cambodia/R0405050/2007 | 1 | NIBSC | May 2008 |

**Viruses proposed by WHO for candidate vaccine preparation**

| Virus | Clade | Institution* |
|---|---|---|
| A/duck/Hunan/795/2002-like | 2.1 | SJ/NIAID |
| A/egret/Egypt/1162/2007-like or A/Egypt/2321/2007-like | 2.2 | CDC |
| A/Common Magpie/Hong Kong/5052/2007 | 2.3.2 | SJ/NIAID |

* CDC- Centers for Disease Control and Prevention, USA
FDA- Food and Drug Administration, USA
NIAID- National Institute of Allergy and Infectious Disease, NIH, USA
NIBSC- National Institute of Biological Standards and Control, UK
NIV- National Institute of Virology, India
SJ- St Jude Children's Research Hospital, USA

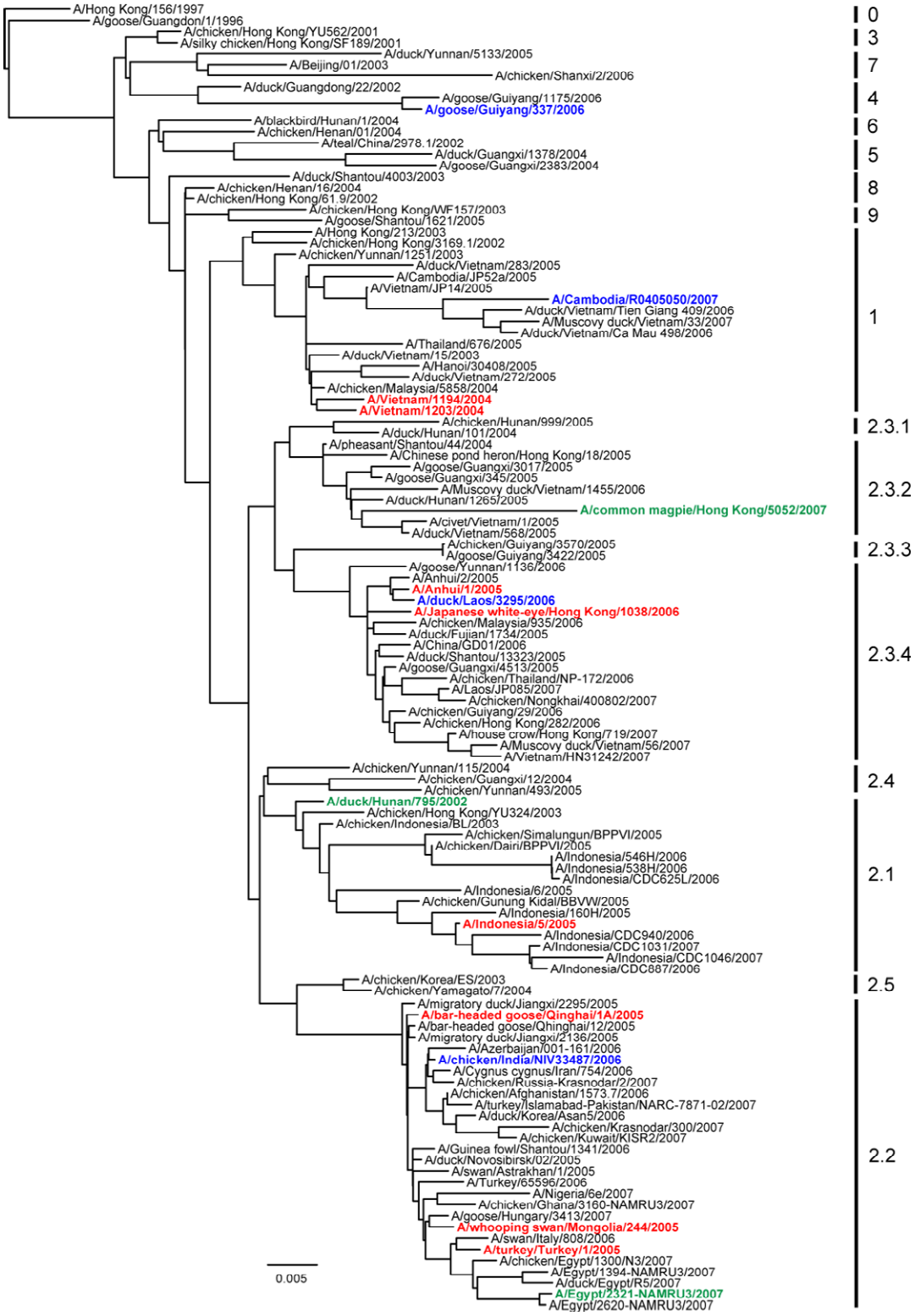


Figure 1. Phylogenetic relationships of H5N1 viruses