World Health Organization

Review of latest available evidence on potential transmission of avian influenza (H5N1) through water and sewage and ways to reduce the risks to human health

Last updated 10/10/2007

Water, Sanitation and Health
Public Health and Environment
Geneva 2006

Review of latest available evidence on potential transmission of avian influenza (H5N1) through water and sewage and ways to reduce the risks to human health

WHO/SDE/WSH/06.1

Printed by the WHO Document Production Services, Geneva, Switzerland

WHO/SDE/WSH/06.1

Review of latest available evidence on potential transmission of avian influenza (H5N1) through water and sewage and ways to reduce the risks to human health

Last updated 10/10/2007

This document is periodically revised as more information becomes available. Questions and answers are being separately developed and will also be periodically updated. To this end, we welcome comments, which can be submitted via email to WSHavianflu@who.int

The most up-to-date version of this document can be accessed at:
http://www.who.int/water_sanitation_health/emerging/ai review.pdf.

Purpose and Scope

The purpose of this document is to summarize the latest available studies and findings on avian influenza pertaining to water resources, water supplies, sanitation (human excreta, sewerage systems and health care waste) and hygiene. Based on this information, we discuss the evidence of avian influenza-related risks to human health associated with these areas of interest. We also lay the groundwork for risk management strategies that will evolve as we improve our understanding of the virus. The document is intended to serve as the scientific basis to inform technical briefing notes, including questions and answers, directed at public health authorities and others involved in pandemic influenza planning. Non-technical question and answer reviews also are planned to provide information and recommendations that are derived from this document and that are of interest to the general public.

Although the "pandemic H5N1" virus does not yet exist, it may behave like avian influenza H5N1 virus in many respects. In this paper, information on avian H5N1 virus is provided where available, but data on other avian influenza viruses are also provided to help fill knowledge gaps until we learn more. As new or revised information is received both this document and the questions and answer reviews will be periodically updated. For information about the current situation or for answers to frequently asked questions about avian influenza and the possible progression to pandemic influenza, please visit WHO's website at http://www.who.int/csr/disease/avian_influenza/en/index.html.

Background

There are three types of influenza viruses: A, B and C. Influenza A viruses can infect humans, birds, pigs, horses, and other animals, but wild birds are the natural hosts for these viruses. Influenza A viruses can cause pandemics. Influenza B viruses are usually found only in humans and generally are associated with less severe epidemics than influenza A viruses. Influenza C viruses cause mild illness in humans and are not a significant concern for human health. Only influenza A viruses are discussed in this document.

Influenza A viruses have different subtypes, defined by the H (haemagglutinin) and N (neuraminidase) proteins on the surface of the virus. The H subtypes are epidemiologically most important, as they govern the ability of the virus to bind to and enter cells, where multiplication of the virus then occurs. The N subtypes govern the release of newly formed virus from the cells. Some subtypes have low pathogenicity (the capacity to cause disease) and others that have high pathogenicity.

Human influenza, the seasonal affliction that causes symptoms such as fever, cough, sore throat and headaches, is caused by human influenza A viruses. Three human influenza A subtypes: H1N1, H1N2 and H3N2 have caused major outbreaks in humans. Avian influenza or bird flu is less well known, although it has captured media attention in recent years. Avian influenza is an infection caused by avian influenza A viruses, with

transmission normally occurring between birds. Less commonly, avian influenza A has infected pigs, and on rare occasions, humans. The subtype of avian influenza A virus known as H5N1 is very contagious among birds and can cause significant mortality in some avian species. In the rare instances that the virus is transmitted from birds to humans, H5N1 can cause pneumonia, multi-organ failure and often death. As of 12 May 2006, there have been 208 cases of transmission to humans, with 115 fatalities reported to WHO (these statistics are frequently updated by WHO and can be found at *http://www.who.int/csr/disease/avian_influenza/country/cases_table_2006_03_24/en/index.html*).

Wild waterfowl are considered the natural reservoir of all avian influenza A viruses. Most infected birds exhibit no symptoms, even when they are excreting large quantities of infectious virus. These asymptomatic birds act as "silent" reservoirs of the virus, perpetuating its transmission to other birds. Domestic waterfowl (e.g., ducks) may also act as a two-way intermediary in the transmission pathway of avian influenza between wild waterfowl and domestic terrestrial poultry (e.g., chickens). Although usually transmitted from wild birds as a virus of low pathogenicity, it may mutate during replication in domestic poultry and highly pathogenic avian influenza (HPAI) strains may arise.

The overriding concern with respect to the HPAI H5N1 virus is that it may change into a form that is highly infectious for humans and that spreads easily from person to person. This could mark the start of a global outbreak or pandemic. No one will have immunity to the virus, as no one will have been exposed to it or developed antibodies. No vaccine with guaranteed efficacy can be prepared in advance of such an outbreak, because the causative virus does not yet exist. Potential vaccines are being prepared and stockpiled in advance, in the hopes that they may match a pandemic strain.

A pandemic strain also may have characteristics of pathogenicity that will not be immediately known. Such changes may have implications for the efficacy of control measures established during pre-pandemic planning. However, we do know that some actions, such as strengthening personal hygiene practices, will reduce human-to-human transmission and help stop or slow the spread of pandemic virus.

Routes of entry into water

Birds infected with avian influenza virus shed large quantities of virus in their faeces as well as in their saliva and nasal secretions. Shedding occurs in the first two weeks of infection (*1*). It has been shown that one infected duck excretes up to 10^{10} EID_{50} (the median egg infective dose) in 24 hours (*2*). As ducks are known to excrete 7.5–10 kg of faeces per year and geese excrete 12.5–15 kg (*3*), infected waterfowl may be able to excrete up to 3×10^9 EID_{50} per gram faeces.

It is likely that infected droppings or other secretions from both symptomatic and asymptomatic migratory waterfowl will enter water environments where the birds gather. Avian influenza virus has been isolated from unconcentrated water from six lakes in

Canada where ducks gathered and deposited large amounts of faeces (*4*); from lakes in the United States (*5,6*); and from concentrated pond water from Hong Kong (*7*). There are no quantitative data available on levels of H5N1 virus in lake water where waterfowl gather, although its detection in unconcentrated water and in small sample volumes suggests that levels are relatively high.

Besides direct deposition of faeces into lake water by migratory waterfowl, it has been suggested that faecal waste from duck and chicken farms may spread to bodies of water via wind, surface runoff or possibly enter groundwater through disposal and composting of waste on poultry farms.

Routes of entry into sewage

In addition to birds, avian influenza virus may be shed in the faeces of mammals, including infected humans, domestic animals and livestock. The H5N1 virus could potentially enter into sewage in urine or faeces excreted by infected humans or in animal waste that is combined with human sewage. Information on the excretion of H5N1 viruses in urine or faeces by mammalian species, including humans, is exceedingly limited and unlikely to be representative of a potential future human pandemic strain. The isolation of the H5N1 virus from the faeces of a child presenting with diarrhoea followed by seizures, coma and death (*8*) suggests that the virus may be excreted by infected humans and can enter sewage in this manner. No virus was detected in the urine of this patient (*8*).

Persistence in water

Avian influenza viruses can persist for extended periods of time in water, although quantitative information on the subtype H5N1 is lacking. Data and findings from studies on other avian influenza subtypes are presented below and summarized in Annex 1.

One study (*2*) showed the avian influenza subtype H3N6 resuspended in Mississippi River (USA) water was detected for up to 32 days at 4°C and was undetectable after 4 days at 22°C. The data showed a decrease of about 4 logarithmic units (LU) in 32 days at 4°C (T_{90}, the time taken to eliminate 90% of the virus in the sample, was estimated to be 8 days) and of more than 8 LU at 22°C (estimated T_{90} = 0.5 days). In a second study (*9*), which used five low-pathogenicity avian influenza viruses (H3N8, H4N6, H6N2, H12N5, and H10N7), infectivity of virus in distilled water (initial concentration 10^6 $TCID_{50}$, or median tissue culture infective dose, per ml) was retained for up to 207 days at 17°C and 102 days at 28°C. The T_{90} ranged between 21 and 32 days at 17°C and between 5 and 17 days at 28°C, depending on the strain. In a study that showed a high level of positive water samples (23%) for a strain of influenza A virus in a lake where ducks were nesting, the proportion of positive samples remained high (14%) in the autumn after the ducks had left for migration, indicating that the virus is able to persist in water (*6*).

Other studies of the persistence of avian influenza viruses in water have shown that these viruses persist for different periods of time depending on temperature, pH and salinity.

For example, one study showed that viruses survived longest (T_{90} = 17 days) at 17°C, with low levels of salinity and a pH of 8.2 (*17*). The shortest viability was observed when virus was exposed to 28°C with 20 ppt salinity and a pH of 8.2 (T_{90} = 1.5 days) (*10*).

Persistence in sewage, excreta and animal wastes

Data and findings from studies on persistence of various avian influenza subtypes in untreated sewage, waste and animal faeces are summarized in Annex 1. No specific information is available on factors affecting the persistence of H5N1 virus in sewage or on the effect of waste treatment processes on H5N1 concentrations. Virus concentrations are reduced at different rates and to various extents in both human and animal waste treatment processes, depending on conditions, but they are not completely eliminated. Furthermore, virus concentrations may be enriched in certain treated or separated waste fractions (such as waste solids) by sedimentation and solid–liquid separation processes (*11*).

Studies on the survival of viruses in human faecal wastes and agricultural animal wastes have indicated that persistence is dependent on several factors, including the virus type, waste type, temperature and other environmental conditions and processes (*11*). The period of avian influenza infectivity in bird faeces and secretions depends on pH and temperature conditions, but generally four weeks after infection, avian influenza virus can no longer be detected (*1*). Ducks infected with the H5N1 virus have been found to shed the virus at high titres from the trachea as well as from the cloacae, with peak levels of virus shedding after three days (*12,13).*

H5N1 virus was isolated from duck faeces on day 3 post-infection (ducks had been infected with 10^6 EID_{50} of virus in 1 ml volume) at 2.25–3.75 $\log_{10}$ EID_{50} per gram fresh faeces, but became undetectable after the faeces were dried overnight at room temperature (20°C). Virus titres declined in wet faeces kept at 25°C but remained detectable for 7 days. When the wet faeces were stored at 4°C, the virus remained viable when testing ceased at day 20. At 37°C, the viruses remained detectable in wet faeces until day 4 (two viruses) and day 6 (two viruses) (*14*). These results suggest that freshly deposited faeces are highly infective (at any temperature), that H5N1 in faecal deposits on land may be more rapidly inactivated than H5N1 in water and that the virus's survival decreases at higher temperatures.

H5N1 typically persists in colder temperatures and produces outbreaks during the colder months of the year (*15*). However, recent (unpublished) studies mentioned above have shown that current H5N1 strains survive longer in faeces at warmer temperatures than previously circulating viruses (*14*), which may explain how the virus has resurfaced in summer months in Asia.

Does the release of antiviral compound into sewers lead to influenza virus resistance in nature?

Potential concern has been raised regarding the possibility that an influenza-specific antiviral used for treatment and prophylaxis (oseltamivir) is not removed or degraded during normal sewage treatment. This may lead to concentrations in natural waters under circumstances such as an influenza pandemic, during which the drug will be administered to large numbers of population, reaching levels to which influenza viruses in nature may develop resistance. This would then increase the risk that influenza viruses being introduced to human beings may become resistant.

To date, there is no credible evidence for such concern because levels of this antiviral compound in wastewater will be low and probably lower than levels needed for biological activity of oseltamivir in waste water (order of uM *in vitro*). Therefore, it should not be concluded that biological events will occur in a virus (an obligate, intracellular biological agent) or its host (cells) that will lead to the emergence of viral resistance to the therapeutic drug as a result of its presence in wastewater.

The activated sludge process of biological wastewater treatment has a mean residence time of 4-8 (conventional) to 24 (extended aeration) hours. The ability of an anti-influenza virus chemical as oseltamivir to survive to such a biologically active exposure and biodegradation and biotransformation process is uncertain and has not been adequately studied.

Exposure of influenza viruses in (waste) water to an antiviral compound may lead to chemical reactions between the virus and the antiviral chemical, such that they would react and somehow result in a complex that would lead to biological selection of resistant mutants. However, in order for biological selection of resistant virus mutants to occur, there must be a living virus host (i.e. a mammal or bird) involved in the process. The influenza virus and the antiviral compound must be present together in a living host, perhaps as a complex, for virus-host interactions to occur that would lead to virus mutations resulting in virus resistance to the compound. These genetic selection reactions or mutations will not occur de novo in the extracellular environment, such as in sewage or water. To assume that such a biological selection event can simply occur because the viruses and the chemicals are both present in the water or wastewater