Preparing for Highly Pathogenic Avian Influenza

A Manual for Countries at Risk

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1. Introduction

Highly pathogenic avian influenza (HPAI) has seriously affected poultry farmers whenever and wherever it has appeared. Historically, outbreaks of HPAI have occurred in all continents (see Table 1). The current avian influenza epidemic, caused principally by the H5N1 strain, has been continuing since it was first recognised in Vietnam in December 2003. Despite concerted attempts at control, Thailand, Vietnam, Indonesia and PR China are still recording outbreaks and there are major control campaigns being implemented in Vietnam and Indonesia. Some outbreaks are still being recorded in Cambodia. Lao PDR, where a few outbreaks occurred, is now apparently free at the time of this writing.

However, two circumstances have increased international concern about the behaviour and spread of this disease. The first is that, to date, more than 130 cases of transmission of the virus to humans have been recorded, with an approximately 50% fatality rate. There is increasing concern that in the future the virus will adapt to enable human-to-human transmission with ease and result in a global human influenza pandemic if not contained in time. Secondly, virus has been introduced through wild birds, along their migratory flyways, to cause disease poultry and avian wildlife\(^1\). Between August and December 2005, the disease has been reported in Russia, Turkey, Croatia, Romania, and Ukraine. In February 2006, the disease has now been reported on the African continent with the first notification of the HPAI H5N1 strain in Nigeria. This first occurrence on the disease in Africa is of major concern, putting at immediate risk the livelihood of millions of people relying on poultry production for income generation and sources of protein. If this situation gets out of control, it will have a devastating impact on the poultry population in the region and will increase the exposure of humans to the virus.

It is difficult to predict the severity of either of these threats. The virus has been present in China since at least 1996 and probably disseminated to South-East Asian countries at least some months before it developed into the epidemic beginning in 2003. There has been enormous opportunity for the virus to infected humans, which has probably occurred much more than has been identified, and yet adaptation for human-to-human transmission has not yet occurred. This however, does not imply that it will not occur and the greater the shedding of virus from infected poultry, the greater the risk of adaptation to cause a human pandemic. Similarly, despite opportunities for the virus to spread in wild birds, to date it has caused minimal disease in poultry outside of South-East and East Asia. Again, whether this occurs in the future is difficult to predict.

Countries may be under threat of introduction of avian influenza through exposure of poultry to wild birds, especially waterfowl. They may also be at risk from introduction of infected or contaminated poultry, poultry products or fomites. This represents a threat to poultry industries around the world, people’s livelihoods, and a source of high quality and inexpensive protein complement to diets. Human populations are also at risk if an influenza pandemic occurs.

This Manual is intended to assist national animal health authorities and other stakeholders consider the needs for preparing for a possible incursion of HPAI, to detect disease at the earliest opportunity and to respond as rapidly as possible to contain the disease.

The international community has a vested interest in minimising the spread of this disease. FAO, together with OIE and WHO, are the key agencies for coordinating an international response to the threat. This Manual also assists countries in determining means of sourcing outside assistance to improve their preparedness for avian influenza.

\(^1\) In this document, poultry is referred as ‘all birds reared or kept in captivity for the production of meat or eggs for consumption, for the production of other commercial products, for restocking supplies of game, or for breeding these categories of birds’. This definition has recently adopted by OIE in the 2005 edition of the *Terrestrial Animal Health Code* Chapter on Avian Influenza. (OIE, 2005a)
2. Avian influenza and the virus that causes it

Avian influenza is caused by influenza viruses which are common in wild birds and occasionally infect poultry. When poultry are infected, they may have no disease, mild disease or very severe disease. Chickens, quail and turkeys are especially susceptible while ducks more commonly show no disease, but act as a reservoir for the virus. Other poultry species, including guinea fowl and pheasants, and also ostriches, can all become affected. While generally wild birds are not affected by the AI viruses that they carry, occasionally they can suffer disease. This has been observed in Asia and parts of Europe as a result of infection with the H5N1 virus and may be because they have become infected with the virus from domestic birds.

Influenza viruses have two main surface antigens, the haemagglutinin (H) and neuraminidase (N). There are many H and N subtypes, but highly pathogenic avian influenza viruses have historically been either H5 or H7, and to a lesser degree H9. The avian influenza virus currently causing the major epidemic in Asia is H5N1, with some occurrences of H5N2 being reported as well. The virus causing disease in Pakistan is past several years is H7N9. AI viruses are also classified as to their pathotype: highly pathogenic (HPAI) and low pathogenic (LPAI) – a biological characteristic of the virus’ virulence in chickens. Currently, the pathotype definition has been expanded to depending on the genetic sequence coding for basic amino acids in the cleavage site of the H protein. All AI viruses that have these sequences at the critical site are considered notifiable and the viruses are denoted as HPNAI (highly pathogenic notifiable avian influenza) and LPNAI.

2.1. Clinical signs

The clinical signs of AI infection are variable and influenced greatly by the virulence of the viruses involved, the species infected, age, concurrent viral or bacterial disease and the environment. The virulence exhibited in chickens can vary during an outbreak.

**Infection with non-pathogenic viruses**
- No clinical signs in infected birds, with seroconversion.
- Some of these viruses have potential to become virulent through genetic mutation.

**Infection with low or mild pathogenic viruses**
- Clinical signs in chickens and turkeys range from inapparent to mild or severe respiratory disease and can be confused with infectious laryngotracheitis and other respiratory tract infections.
- Mortality ranges from 3% in caged hens (layers) to 15% in meat chickens (broilers).
- Egg production in layers can drop sometimes to 45% of the expected egg yield of a large flock, returning to normal levels of production in 2–4 weeks.
- Mutation to virulence has been demonstrated in outbreaks.

**Infection with highly pathogenic viruses**
- In peracute cases involving sudden death, as seen in the 2004-5 outbreak in Viet Nam, clinical signs may not be seen and mortalities occur within hours after onset of depression. Overall mortality rates for peracute/acute cases nearing 100% have been reported.
- In acute cases, mortalities occur as early as 24 hours after the first signs of the disease, and frequently within 48 hours. In other cases, more diverse visible signs are seen and mortalities can be delayed for as long as a week.
• Clinical signs in chickens and turkeys include severe respiratory distress with excessively watery eyes and sinusitis, cyanosis of the combs, wattle and shanks, oedema of the head and eyelids, ruffled feathers, diarrhoea and nervous signs.
• The last eggs laid after the onset of illness frequently have no shells.
• Some severely affected hens may recover, but rarely come back into lay.
The disease in turkeys is similar to that seen in chickens, but is often complicated by secondary infections such as those due to fowl cholera (*Pseudoalteria multocida*), turkey coryza (*Hemophilus gallinarum*), or colibacillosis (*Escherichia coli*).

2.2 Gross pathology

In many cases, poultry dying from the peracute form of the disease lack visible gross pathological lesions. With acute infections in chickens, there is severe lung congestion, haemorrhage and oedema in dead chickens; other organs and tissues appeared normal. More varied visible lesions are seen in chickens surviving 3 to 5 days, including congestion and/or cyanosis of the comb and wattles and swollen heads. The changes in the combs and wattles progress to depressed areas of dark red to blue areas of ischaemic necrosis. Internally, the characteristics of acute infections with viruses causing HPAI are haemorrhagic, necrotic, congestive and transudative changes. The oviducts and intestines often have severe haemorrhagic changes.
As the disease progresses, the pancreas, liver, spleen, kidney and lungs can display yellowish necrotic foci. Haemorrhages (petechial and ecchymotic) cover the abdominal fat, serosal surfaces and peritoneum. The peritoneal cavity is frequently filled with yolk from ruptured ova, associated with severe inflammation of the airsacs and peritoneum in birds that survive 7–10 days. Haemorrhages may be present in the proventriculus, particularly at the junction with the ventriculus (gizzard).
caption: haemorrhage in the muscle and the fat around the gizzard.
Credit: USDA

In cases due to mild pathogenic avian influenza viruses, lesions may be seen in the sinuses characterised by catarrhal, serofibrinous, mucopurulent or caseous inflammation. The tracheal mucosa may be oedematous with exudates varying from serous to caseous. The airsacs may be thickened and have a fibrinous to caseous exudates. Catarrhal to fibrinous peritonitis and egg yolk peritonitis may be seen. Catarrhal to fibrinous enteritis may be seen in the caeca and/or intestine, particularly in turkeys. Exudates may be seen in the oviducts of laying birds (Easterday et al 1997). Histopathological lesions seen in the gross changes described above are not definitive for HPAI, although vasculitis in the brain and other organs may be highly suggestive of the disease.

2.3 Differential diagnosis

The following diseases must be considered in the differential diagnosis of virulent AI:

- Other diseases causing sudden high mortality
  - Newcastle disease
  - Infectious laryngotracheitis
  - Duck plague
  - Acute poisonings

- Other diseases causing swelling of the combs and wattles
  - Acute fowl cholera and other septicaemic diseases
  - Bacterial cellulitis of the comb and wattles

Less severe forms of the disease may be confused with, or complicated by, many other diseases with respiratory or enteric signs. AI should be suspected in any disease outbreak in poultry that persists despite the application of preventive and therapeutic measures for other diseases or when the epidemiological context is highly suggestive of the introduction of the infection.
3. The risk of introduction and dissemination of avian influenza

3.1 Risk of introduction by migrating birds

Migration of water birds present the main risk of carrying AI viruses over long distances, provides a complex network because different bird flyways overlap geographically.

Birds infected with AI virus can shed virus for up to one month. Birds from different regions intermingle with each other in areas where large water bodies attract them and transmission of viruses can occur between them. The outcome is that potentially viruses can be transmitted from infected countries in Southeast and East Asia to Central Asia, Eastern Europe, the Middle East and Africa, North and South America. In the course of the current epidemic, a large number of wild bird species have been found dead, with AI virus type H5N1 being isolated. Recent findings show that the virus can be isolated from other bird species without signs of disease. However, it is not yet fully determined which species are implicated in the long distance introduction of the virus and its transmission to poultry.

If infection occurs in domestic poultry, it is likely to be in areas where wild waterfowl congregate and where poultry are not in bird-proof sheds. Transmission of virus can occur from contaminated water as well as from direct contact of wild birds with poultry. Good biosecurity therefore requires physical barriers erected between poultry and wild birds and also the provision of clean or treated water before being provided to poultry.
An important component of preparedness in the context of the current avian influenza epidemic is to identify wild bird migratory patterns, timing and destination sites and to assess the risk of close contact with domestic poultry providing an entry point for establishment of avian influenza.

3.2. Risk of importation

Many countries currently impose bans on the importation of poultry and products from infected countries with notifiable AI. Given the potential for transboundary spread of the disease, it would be wise to take great care with all poultry products, especially the ones that can carry the virus. Live birds, represent by far the greatest risk but dressed carcasses of infected birds, eggs from infected hens, poultry waste, and fomites contaminated with faeces can all be a source of infection. A detailed risk assessment for each poultry product has been carried out by the European Food Safety Agency (EFSA) and is available on their Web site (see Annex 1).

It should be recognised that illegal movement of live birds also represents a risk that will not be mitigated by imposing bans on legal importation.

3.3 Risk of spread from infected poultry

To prevent further spread of H5N1, surveillance in poultry as well as in wild birds should be strengthened in countries at immediate risk, especially where birds come to rest along their migrating routes. Resources should be focused on the reduction of close contacts between humans, poultry and wildlife through better management practices and improved biosecurity practices in poultry production enterprises, especially those that are small and 'open-air'- where poultry and waterfowl mingle with wild birds. The influenza viruses are easily spread by fomites and generally survive well in water. Furthermore, certain species of ducks are able to carry influenza viruses without exhibiting any clinical signs of disease. Juvenile ducks have the highest rates of infection and shedding. High titres of virus occur in late-summer, when birds leave their northern breeding areas, although these titres decrease as birds continue southwards.

Once a highly pathogenic virus (HPAI) has been recognised in the marketing environment or country, all persons working with poultry should greatly increase the level of hygienic practises to avoid bringing in virus (bio-exclusion) into an operation and to prevent virus exiting (bio-containment) if it has already entered a flock, village or region. The main risks of virus leaving a region to another area are: sale of birds to markets, departure of wild waterfowl which have visited backyard poultry units, people working or selling poultry wearing contaminated footwear or clothes, or moving cages and egg crates to markets or poultry farms. Therefore poultry keepers and communities have to take practical measures to avoid introducing the virus, and to reduce the risk of spread when disease has been detected.

3.4. Virus survival in the environment

Survival of influenza viruses is prolonged by low relative humidity and low temperature in aerosols, whereas low temperature and high moisture levels prolong survival in faeces. Most studies on viral environmental persistence have been undertaken in cool northern climates with following findings:

- AI virus can survive in faeces for at least 35 days at 4°C. AI virus can survive within the poultry house environment for up to 5 weeks (Webster et al 1978).
- Virus may remain infective in lake water for up to 4 days at 22°C and over 30 days at 0°C (Webster et al 1978).
- As an enveloped virus, influenza viruses are susceptible to several disinfectants, including detergents.
- The virus is stable over a pH range of 5.5 – 8.
AI virus can be isolated from lake water where waterfowl are present (Hinshaw et al 1979). Acidification of potentially contaminated drinking water to pH 2.5 or chlorination should minimise spread of infection.

4. Preparing for an Outbreak

Good preparedness planning can be of enormous benefit in successfully managing an outbreak and minimising its impact. This planning should involve a consideration of how best to rapidly detect an outbreak, confirm the diagnosis and implement a rapid and effective control programme. It requires an assessment of veterinary service capabilities and capacity and the legal framework in which these services operate. The structure of national poultry industries should be examined to determine the potential for entry of virus. Industry support should be sought in the planning process. Provision for good public awareness programs should be made as it is critical to have public support for disease control activities and good public knowledge to minimise the risk of human infection.

4.1 Early detection

4.1.1 Wild bird surveillance

Where the risk is from migrating birds, it is essential to identify the migratory habits of different species, their origins, destinations and timing of migration. While disease in wild birds would not be expected, it is of value to alert wildlife personnel to report unusual deaths in wild birds. Active surveillance can be undertaken by catching wildfowl species and sampling (generally by taking cloacal swabs) to test for the presence of AI viruses.

Caption: sampling of wild birds in Mongolia (August 2005)
Credit: B. Karesh, WCS

4.1.2 Domestic poultry surveillance

The identification of poultry at risk should involved poultry flocks located in high risk agro-ecological systems where migrating birds congregate as well as high risk farming systems and practices such as free-grazing ducks associated with rice production systems. At times of particular perceived risk, such as the time of arrival of migrating birds, it might be considered of value to undertake active serological and
virological surveillance of sentinel birds within flocks, especially domestic ducks which are most likely to be exposed. It is therefore advisable that countries perform risk assessment studies for the introduction of AI in order to define the most appropriate surveillance strategy they should adopt.

In any event, each country will have particular priorities and surveillance systems which should be refined to reflect these priorities. For example, HPAI-free countries or those having a lower risk of becoming infected will seek access to detailed, updated information on risks and will focus on the detection of incursions — making early warning and surveillance their priority. For infected countries or those at high risk of infection introduction, surveillance priorities will include the collection of detailed, current information on human health risks, ecological zones and production systems representing the highest risk for introduction and maintenance of HPAI infection.

The FAO guidelines for HPAI surveillance are particularly relevant to countries exposed to the risk of HPAI introduction and should be read in conjunction with this document (see Annex 1).

The objectives of HPAI surveillance and monitoring include:

- To detect clinical disease and infection.
- To understand the epidemiology and ecology of AI, as well as its socioeconomic impact, to help to design effective and implementable control programs for poultry production systems.
- To assess the temporal and spatial patterns and thereby to improve the effectiveness of control efforts.
- To understand the evolution in Asia of AI virus variants.
- To help define and control risks to public health.
- To monitor for antigenic drift in AI viruses through frequent analysis at competent labs.
- To maintain livelihood and assist in ensuring food security through the by implementation of appropriate control measures.
- To demonstrate freedom from clinical disease and absence of infection in a country or compartment and thereby facilitate trade.
- To assess the efficacy of vaccination when used as part of a comprehensive disease control programme.

**MINIMUM REQUIREMENTS FOR EFFECTIVE SURVEILLANCE**
(taken from “FAO Guiding principles for HPAI surveillance”)

The following minimum requirements apply to all countries and compartments:

- HPAI is a notifiable disease (i.e. there should be a legal requirement for suspected cases of disease to be reported to the official veterinary services).
- The official veterinary services must have a formal system for detecting and investigating outbreaks of disease and for reporting confirmed cases internationally, in accordance with OIE guidelines.
- The country and/or region must have the technical capability to diagnose HPNAI and LPNAI (see Annex 2).
- The country and/or region must have a system for recording, managing and analysing diagnostic and surveillance data.
- The country should participate in the regional surveillance and diagnostic network, including the public health sector, to enable sharing of information to characterize risk, prevent disease spread, and enhance control efforts.
- The frequency of surveillance could be a minimum of every six months within a country or could be less than this if selected ‘pilot’ areas are targeted for more frequent surveillance.
In addition to formal surveillance and reporting procedures, raising awareness of the risk of avian influenza is an important means of increasing the likelihood that an outbreak of disease in poultry will be reported rapidly. However, it must also be kept in mind that such public awareness should not be undertaken in such a manner that it causes undue concern within the community. Also, veterinary disease control authorities will need to be prepared for an increased work load, since if formal and informal reporting mechanisms work, there will probably be a significant number of investigations that must be made following increased publicity of the risk of disease.

### 4.2. Rapid response

#### 4.2.1 Disease surveillance

Active surveillance should be initiated as soon as a country is considered at high risk of HPAI incursion. In case the disease is suspected, a sample of all domestic species of bird that die in the restricted area should be investigated and specimens submitted to approved laboratories for virus analysis and characterisation. Field surveillance examinations should seek to detect changes in flock health. Trained personnel should be aware of the potential risk to human health and wear protective gear (goggles or face shield, mask, gloves, and disposable gowns or coveralls, and rubber boots that can be disinfected before leaving the investigation site.

Surveillance needs to include:

- Integrated commercial level poultry producers carrying out their own surveillance and timely reporting;
- Local disease control centre officers carrying out regular telephone surveillance of independent premises; and
- Targeted surveillance of premises in the restricted area and control area, particularly focusing on:
  - infected places, suspect places and dangerous contact places; and
  - premises with unusual sickness and/or mortality.

All reports of a decline in health status of birds or egg production should be investigated and samples taken.

Although surveillance will begin immediately around the infected place or flock, it will have to be extended very quickly to all other sites where birds, products and contaminated materials might have been moved from the Infected Area. Surveillance of wild birds to determine their potential involvement in the dissemination of the disease may also be attempted, but this is likely to have a limited impact on controlling spread of the disease if biosecurity mechanisms are high.

#### 4.2.2 Planning for avian influenza control

It is difficult to put in place a rapid and effective response to an outbreak of a new disease if the process has not been planned and the required resources made available. FAO has guidelines in the publication “Manual on the preparation of national animal emergency preparedness plans”. These guidelines recommend the development of four sets of complementary technical contingency plans:

1. Specific disease contingency plans that document the strategies to be followed in order to detect, contain and eliminate the disease.
2. Standard operating procedures that may be common to several or all emergency disease campaigns.
3. Enterprise manuals that set out zoosanitary guidelines for enterprises that may be involved in and emergency animal disease outbreak.
4. Simple job description cards for all individual officers.

Each national veterinary authority needs to consider their particular needs and be careful not to embark on a programme of emergency manual development that is not sustainable with their resources. A critical element of emergency plans is that they must specifically consider the situation and needs of a particular country (e.g. structure of the poultry sector, organisation of the Veterinary Services). In addition, the
implementation of contingency plans through simulation exercises is key to define gaps or overlap in responsibilities or resources during an outbreak.

The Australian veterinary contingency plan, AUSVETPLAN, is a set of such plans, including a Disease Strategy for Highly Pathogenic Avian Influenza. It can be downloaded from the internet (Annex 1). Some of the most important considerations for the contingency planning process are:

1. Consideration of the disease control strategies that are available, the implications of applying them and which is the most appropriate strategy in different circumstances in a particular country.
2. Financial planning, to determine where the necessary funds will come from and ensure that there is a mechanism and commitment to provide them immediately. A particular issue that needs to be considered is whether adequate compensation will be provided to poultry owners whose birds are destroyed. In the current context of the global avian influenza threat, countries might consider negotiating commitments from international donors to assist in control, should a disease incursion occur.
3. Resource planning, which include the needs for personnel, equipment and other physical resources. Plans will indicate resource requirements at the time of an outbreak. However, they should also provide a means to determine requirements in advance and either obtain them or make provision to rapidly obtain them when they are needed.
4. The need for appropriate legislation must be considered, as in most administrative systems this requires long-term planning. Laws, regulations and proclamations are required to give authorised people the power to:
   - proclaim a notifiable disease
   - enter a poultry enterprise to inspect birds or collect specimens
   - define infected areas and disease control zones
   - institute quarantine of affected or suspect premises
   - place movement controls on poultry, poultry products and potentially contaminated materials
   - destroy and dispose of infected or potentially infected birds and contaminated materials
   - undertake other disease control operations, such as compulsory vaccination
   - place controls on the operation of enterprises, such as poultry processing plants.
5. Obtaining consensus and commitment from all regulatory authorities and industry, as appropriate.
6. Undertaking training of personnel so that the appropriate skills are available.
7. Conducting simulation exercises to identify deficiencies and undertaking periodical review of the contingency plan.

4.3 Avian Influenza control strategies

The primary aim of an early and rapid response to any occurrence of HPAI is to contain the disease before it spreads and eliminate it by stamping out affected premises and proper disposal. Only if this fails should other measures be considered, including vaccination using effective and quality controlled vaccines. This approach is endorsed by OIE, FAO and WHO. Where it proves impossible to eradicate the disease in the short- to medium-term, it may be possible to aim for compartment freedom (e.g., freedom within the commercial sector, in which birds are housed and protected from infection), or zone freedom (freedom in defined geographic areas). In the case of compartmentalisation, the poultry industry must take responsibility for its biosecurity (bioexclusion) with the necessary veterinary regulatory oversight in place; and in the latter, commercial compliance must follow national restrictions to ensure infection does not enter the free zone, and regulators prove to trading partners of the zone’s clean status at any given time.

There is no pre-described strategy to control avian influenza outbreaks. In order to effectively control the disease, countries should have a complete plan of action and the financial and humans resources to implement it under the particular conditions prevailing in their country. A regional approach is also necessary.
4.3.1 Culling

The basis of HPAI eradication by stamping out is:

- immediately impose quarantine of the affected area (premises or village).
- slaughter all infected and potentially infected birds and dispose of the carcases.
- decontaminate sheds and other poultry housing areas.
- carry out rapid surveillance of surrounding areas to determine the extent of possible spread.
- Close and disinfect markets.
- Sick and dead birds should not enter the human food chain, nor should they be sold for feed to other animals (i.e., zoos).

Quarantine and movement controls

As AI is readily transmitted via contaminated objects, strict control of movement of anything that may have become contaminated with virus and immediate imposition of tightly controlled quarantine on all places suspected of being infected is essential to a successful eradication program. Ideally, quarantine should be imposed on all farms/villages on which infection is either known or suspected and should be strictly policed to ensure that no-one, including the residents, owners, staff and other visitors, leaves without changing clothes and footwear.

Particular attention needs to be paid to workers on poultry farms who keep backyard poultry at home.

Strict on-farm biosecurity and hygiene is needed to control spread of the disease from wild birds. Access of wild birds to commercial poultry sheds and flocks should also be considered during depopulation operations. In areas where poultry are raised in a village environment, particular consideration needs to be given as to how effective quarantine disposal and decontamination can be imposed.

Credit: V. Martin
Caption: restriction zone. Outbreak in Anhui province, China (July 2004)

Effective quarantine of an area requires around-the-clock security to ensure that only authorised personnel in protective clothing are allowed to enter. It will be necessary to supervise the movements of residents onto and off the property and to ensure that all pets are confined. It is also strongly recommended to ban cockfighting, pigeon racing and other avian concentrations in the outbreak area.
Slaughter of infected and potentially infected poultry

All susceptible poultry species in infected and dangerous contact premises, or in a large area if this is deemed necessary, whether they are obviously diseased or apparently healthy must be slaughtered.

Although it not possible to provide specific and universally applied standards for controlling the disease through zoning for all potential outbreak occurrences, the following definitions and distances should be considered as a guide to rapidly contain HPAI. It must be noted that distances are indicative and subject to changes according to the epidemiological characteristics, physical and geographical barriers, poultry density and farming systems (more details are provided in Annex 6).

- Infected area

An area classified as an infected area (IA) will be a defined area (village, farm) in which HPAI has been detected. Infected premises (IP’s) will be subject to quarantine and all susceptible animals will be destroyed in this area.

- Restricted area

A restricted area (RA) will be a relatively small declared area (compared to the control area - see below) around infected places which are subject to intense surveillance and movement controls. Movement out of the RA will, in general, be prohibited, while movement into the area would only be through regulatory approval. Multiple RAs may exist within one control areas (CA). The RA does not need to be circular but can have an irregular perimeter depending on known physical and geographical barriers, markets, poultry density and farming systems. This distance will vary with the size and nature of the potential source of virus, but will be approximately 1–5 km around the IP, depending on the density of poultry premises. The boundary could be the perimeter fence of the IP if the IP is in an isolated location. The boundary in a densely populated area will take into account the distribution of susceptible birds and traffic patterns to markets, service areas, abattoirs and areas that constitute natural barriers to movement. If possible hatcheries should be kept out of the RA.

- Control area
The CA will be a larger declared geographical area around one or several RAs (possibly as large as a Province initially) where restrictions will reduce the risk of disease spreading from the RAs. The boundary of the CA will be adjusted as confidence about the extent of the outbreak becomes clearer but must remain consistent with the OIE Code chapters on surveillance and zoning (Chapters 1.3.4 and 1.3.5; see Appendix 3). In general, surveillance and movement controls will be less intense and animals and products may be permitted to move under permit from the area.

The declaration of a CA also helps to control the spread of the outbreak from within the RA. The perimeter of the CA is a buffer zone between the RA and the rest of the country. The boundary does not have to be circular or parallel to that of the RA but should be 2–10 km from the boundary of the RA. In general, the movement of possibly contaminated articles and materials within the CA is allowed but movement out of the CA is prohibited without approval from the Chief Veterinary Office. This type of control area allows reasonable and safe commercial activities to continue.

When declaring RAs and CAs, the areas must not be larger than necessary, thus restricting the number of properties to be quarantined to only those deemed prudent. If flocks in a quarantine area are not depopulated, then the cost of keeping the birds beyond their normal market age could be substantial.
Example of zoning: the case of China

Premises with infected poultry or relevant slaughtering houses and other departments are considered as infected point; areas within the 3km radius are considered as infected zones; areas within 5km around the infected zones are considered as threatened zones.

Stamping out. All poultry within infected zones should be stamped out.

Emergency vaccination. All susceptible poultry in the threatened zones are vaccinated compulsorily with the vaccines approved by Ministry of Agriculture. Only healthy birds should receive vaccine.

Disposal. All poultry carcasses and poultry products in infected points, and excretion material, contaminated feed, litter and sewerage from the infected points shall be subject to bio-treatment or disposal.

Cleaning and disinfection. All contaminated items within the infected zones, transportation vehicles, utensils, poultry counters and ground must be cleaned and disinfected.

Movement control. Warning signs are widely posted around the infected zone, disinfection stations are set up in the transportation entrance of infected zones to disinfect vehicles and items entering and exiting zones; movement of all susceptible live birds and their products was controlled.

Closing the market. All the poultry and their products markets in the infected zones, and the live birds markets with the 10km around the infected zones must be closed.

Tracing. If poultry and their products are sold out during the incubation and clinical manifestation period or moved out, tracing should be conducted on the suspect contaminated items to prevent these items from spreading disease.

Financial support. Financial support systems are established for all poultry destroyed because of HPAI.

Public health. Surveillance of occupational staff of poultry rearing, trade and transportation and process, especially the staff in the infected zones should be intensified, and epidemiological investigation should be conducted. Stringent protective measures must be implemented by staff participating in the destroying infected birds and cleaning of contaminated premises.

Quarantine lift. The conditions for quarantine lift are stipulated as follows: 21 days for infected point and infected zone after strict treatment according to “National Contingency Plan for Highly Pathogenic Avian Influenza” and standard technical requirements for treatment of HPAI; over 14 days for the...
threatened zone where all the susceptible birds are emergency vaccinated with the national approved vaccines and no new cases occur, it is inspected and accepted by related authorities. When quarantine is lifted, live bird trade markets 10 km around the infection zone may be reopened. The infected points are re-stocked 6 months after strict disposal.

Birds should be slaughtered by methods that take account of animal welfare concerns and the safety of operatives, preferably without moving them from the site.

- For *small numbers of birds*, the preferred methods are dislocation of the neck (using burdizzos, bone cutters, secateurs or bare hands). Burdizzos are particularly useful when large numbers of poultry with strong necks (geese, ducks etc) are to be destroyed.

- For *large numbers of birds* in commercial poultry units the preferred method is gassing with carbon dioxide. This method involves lining large garbage waste bins (skips) with plastic sheeting that also forms a canopy over the top of the bin. Birds can be caught using teams of labourers. Experienced catching teams may be available. Chicks are easily caught under heaters and are transferred to skips in plastic garbage bins. Broilers on the ground are driven, using a movable hessian wall, to the catching area where they are caught and placed directly into skips.

- *Caged birds* are more difficult and progress is slower. Each catcher removes 3 or 4 birds from cages and carries them by the legs to skips. Layers on perches are best caught at night or during low light when they are quiet.

Carbon dioxide (CO₂) is transferred to the bottom of the skips through 2.5 cm garden hose fitted to the top of the cylinders. The carbon dioxide should be decanted in 30–45 second bursts. It is essential not to decant too quickly or the bottles will freeze when about half empty. The concentration of CO₂ must be in the range of 60-70% in the skip, with the lid tightly closed for a 1-2 minute period to properly stun and kill the birds. On average, half a 45 kg cylinder of carbon dioxide is needed for the three cubic metre skips and three or more cylinders for the 20 cubic metre skips. Carbon dioxide should be added at a sufficient rate to ensure birds succumb before other birds are placed on top of them. Skips should be three quarters (75%) filled with birds, sealed and transported to the disposal site. Care must be taken to ensure no bird is still alive when dropped into the burial pit. Should this happen birds must be immediately caught and humanely killed.

**Safe disposal of carcasses**

Disposal of dead birds, poultry litter and other contaminated waste is best done by burial. Sometimes this is not practicable or desirable because the required equipment is not available or it would have adverse environmental effects, such as potential contamination of ground water. In these circumstances, the best alternative might be composting.
Burial - Burial is best undertaken at the infected site. It is best to minimise the distance that infected material needs to be transported. A burial place outside infected premises may be the best option in situations where a number of infected foci would have to be depopulated and decontaminated in a given area and where a common burial site would be more efficient. The preferred equipment for digging burial pits is an excavator. This equipment is the most efficient available for the construction of long, deep, vertically sided pits. Other advantages include the ability to easily store topsoil separate to subsoil and the equipment can be used if required to fill the pit with carcasses or other materials and closing the pit without disturbance of the carcasses. Loaders, bulldozers, road graders and backhoes (for small jobs) may be used if excavators are unavailable. With the exception of backhoes, all other equipment requires the continual movement of the machine over the site while digging the pit. Excavators and backhoes essentially remain in a fixed position while digging; hence they move soil faster, with less cost and less damage to the site surrounding the pit. Most excavators have an attachable hammer for rock work if necessary. The dimensions of the burial pit will be dependant on the equipment used, site considerations and the volume of material to be buried. The preferred dimensions are for pits to be as deep as practically possible (reach of machinery, soil type and water-table level being the usual constraints), with vertical sides.

Gas production from decomposition within unopened carcasses may result in considerable expansion in the volume of the buried material to the extent that the surface of the closed pit may rise and carcasses may be expelled from the pit. Lime may be added to pits to prevent earthworms bringing contaminated material to the surface after pit closure. Covering the carcasses with 40 cm of soil is suggested, and add an unbroken layer of slaked lime [Ca(OH)₂] before filling is completed. Lime should not be placed directly on carcasses because it slows, and may prevent, decomposition.

Inspection of the burial site after closure is recommended so that appropriate action can be taken in the event of seepage or other problems.

Composting – Biological decomposition, or composting, is an effective way of dealing with manure and litter waste and can be undertaken within sheds or otherwise on site, thus overcoming the risks of disseminating the virus during transport. Composting should be done in a secure area not accessible to susceptible birds. The procedure requires the piling of carcasses with other bulky contaminated or non-contaminated material, such as wood chips, straw bedding, to allow for proper aeration and covered with a biological filter (i.e., not whole plastic). The pile should not be pressed or otherwise compacted. The site selection is important – away from any residences, with at least 1 m of ground between the pile and any
known water source, with any run off water from the decomposing material collected and treated. Care must be taken to ensure that susceptible animals or pets (dogs) do not have access to the compost pile. Temperatures for proper composition should reach 55-60°C within 10 days and the material kept in place for several weeks; at which time, mixing of the material within the pile should be done, but never pressed. Properly decomposing material at this time should be dark kin colour with minimal foul odour.

Burning/Incineration - A burning area outside an infected place may be the best option in situations where a number of infected foci would have to be depopulated and decontaminated in a given area and where a common burning site would be more practical. The principle is to place carcasses on top of sufficient combustible material, ensuring the arrangement of fuel and carcasses allows adequate air flow to enter the pyre from below, so thus achieving the hottest fire and the most complete combustion in the shortest time. When loading of the carcasses is complete and weather conditions suitable, saturate the fire-bed and carcasses with diesel or heating oil (NOT PETROL) and prepare ignition points about every 10 metres along the length of the fire-bed. These can be made of rags soaked in kerosene. Remove all vehicles, personnel and other equipment well away from the fire-bed. Start the fire by walking into the wind and lighting the ignition points along the way. The fire must be attended at all times and be re-fuelled as necessary, use a tractor with a front mounted blade or a front-loader. Ensure any carcasses or parts thereof that fall off the fire are replaced on the fire. A well constructed fire will burn all carcasses within 48 hours. The ashes should be buried and the site restored as well as possible.

Rendering - Rendering is a closed system for mechanical and thermal treatment of waste, and a good way for carcass disposal if the plant has sufficient capacity and if it is possible to effectively decontaminate the rendering plant afterwards. A medium sized facility could render some 12 tonnes per hour of operation. However, private rendering plants may not be willing to handle infected birds and eggs unless an emergency order is imposed. A disadvantage is that infected material would need to be transported from infected sites to the plant.

Where burial, cremation or rendering are not considered practical or are difficult to carry out on the infected place, permission should be sought to transfer carcasses and/or infectious material to another site for disposal by burial, cremation or rendering. Transport should be in a leak-proof container, such as a large skip, covered with tough polyethylene covers and sealed at the top. It should not be overloaded — half a metre or more (depending on distance to be travelled and temperature) should be left clear for expansion of carcasses. Vehicles should travel slowly to avoid splashing of contaminated material and should be accompanied by a police vehicle to minimise the chances of accidents and to prevent breaches of biosecurity. The escorting officer must carry a supply of an approved disinfectant and basic equipment to deal with minor spills en route. All vehicles must be cleaned and disinfected before leaving the infected place and after unloading.

Decontamination

Soapy water and detergents are first choice for many items. The avian influenza virus is more easily destroyed than many other viruses since it is very sensitive to detergents which destroy the outer lipid envelope of the virus. Therefore washing of contaminated surfaces should always be with detergents (soapy water) or specific disinfectants. The most difficult material to decontaminate are bird droppings as the virus can survive in moist environments with high organic content, so it is essential to thoroughly clean and disinfect items that have been in contact with bird droppings - cages, shoes, clothes before working with poultry/entry to a place where poultry are kept. Simple hygienic measures can reduce risk - but national authorities are encouraged to prepare and communicate specific guidance of each type of poultry enterprise. More guidance for veterinary services on selection and application of decontamination procedures is given below (Ausvetplan Manual). Adaptation to the specific country circumstances will be needed.
### Item to be disinfected | Disinfectant/chemical/procedures
---|---
- Live birds | - Euthanase (carbon dioxide gas; dislocation of neck)
- Carcasses | - Bury or burn
- Animal housing/equipment | - 1, 2, 3 (See key below)
- Humans | - 1
- Electrical equipment | - 5
- Water | - Drain to pasture where possible
- Feed | - Bury
- Effluent, manure | - Bury or burn; 4, 3
- Human housing | - 1, 2
- Machinery, vehicles | - 1, 3
- Clothing | - 1, 2, 3

**Key**

1. Soaps and detergents: leave in contact for 10 minutes
2. Oxidising agents:
   a. sodium hypochlorite: liquid, dilute to final 2-3% available chlorine, not good for organic materials. 10-30 minute contact time.
   b. calcium hypochlorite: Solid or powder, dilute 2-3% available chlorine (20 g/litre powder, 30 g/litre solid), not good for organic materials. 10-30 minute contact time.
   c. Virkon®: 2% (20 g/litre). 10 minutes contact time.
3. Alkalis: (do not use with aluminium and similar alloys)
   - sodium hydroxide (NaOH): 2% (20 g/litre). 10 minute contact time.
   - sodium carbonate anhydrous (Na₂CO₃ .10H₂O): 4% (40 g/litre from powder, 100g/litre from crystals), recommended for use in presence of organic materials as above. 10-30 minute contact time.
4. Acids:
   - hydrochloric acid (HCl): 2% (20 ml/litre), corrosive, use only when other chemicals are not available
   - citric acid: 0.2% (2 g/litre), safe for clothes and body decontamination. 30 minute contact time.
5. Formaldehyde gas: Toxic, only if others cannot be used. 15-24 h exposure time.

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**Destocking period**

After slaughter, disposal and decontamination procedures have been completed; the premises must be left without susceptible species (destocked) for a period of time, determined by the estimating survival time of the pathogen in the particular environment. Restocking should not take place until at least 21 days after satisfactory cleaning and disinfection has been completed and the outbreak has been brought under control in the area. Restocking should be undertaken by introducing a small number of poultry first, and these monitored daily for signs of disease. Should this occur, notification to the authorities must be immediate and sampling of the sick or dead birds done to determine the cause. If the poultry remain healthy, full repopulation can be carried out. Of course, improvement of biosecurity should be instituted at all stages of production to decrease the likelihood of AI or other diseases entering the recovered premises. After repopulation, monitoring should be continuous through the sampling of dead birds to determine whether re-infection has occurred.
4.3.2 Financial Support

The issue of compensation for slaughtered birds, property damaged during decontamination and/or loss of income needs to be carefully considered. In principle, offering compensation encourages owners to report disease. However, it can become very expensive undertaking and usually guidelines are developed that strictly limit the categories for compensation. The important issue is to consider what the cost of compensation might be and how it would be funded should a major outbreak of disease be experienced.

If compensation is paid, it can be controlled as follows:

- Have a registration process
- Only pay for animals slaughtered, not those that have died.
- Pay promptly and at a level that is close to market value (Some countries have used innovative strategies such paying slightly more than market value for healthy in-contact birds, and less for sick birds – which allowed for quick reporting to authorities).
- Do not compensate losses other than livestock (birds).
- Ensure that people with very small flocks are also compensated.

There are alternatives to payment of compensation in cash and experience has shown them to sometimes be acceptable.

- Rather than paying cash, provide replacement birds (can be difficult sometimes, governments have to think in advance about logistical questions of where to get replacements, their transport and the required destocking times).
- Provide credit for owners to re-establish their poultry production, including village birds or facilitate entry into alternative livelihood.
- Provide area assistance to enable market conditions to become re-established without undue delay.
- Provide farmers with free technical and veterinary services in re-establishing production schemes.

4.3.3 Vaccination

Vaccination as a support strategy may be considered when the disease has spread to such an extent that it has overwhelmed the resources of disease control authorities or the economic cost of a widespread slaughter campaign cannot be borne. It can also be considered at an earlier stage when Veterinary Services infrastructures and capacities prove to be very weak and available capacities insufficient to curb the spread of the disease. FAO and OIE have made recommendations for the use of OIE-approved AI vaccines, and several such vaccines are commercially available. If used in accordance with FAO/OIE recommendations (FAO Position Paper, September 2004) and the OIE Manual for Diagnostic Tests and Vaccines for Terrestrial Animals, these vaccines provide excellent protection against clinical disease in chickens by reducing mortality and production losses. Vaccination of poultry also reduces the viral load in the environment and thereby decreasing the risk of transmission to poultry and humans. According to current OIE recommendations, HPAI-vaccinated poultry are not excluded from international trade, although specific technical guidelines must be followed to ensure that the vaccine is being applied properly and monitored effectively.

Vaccination, when it is applied, must be done in combination with other disease control measures, including the slaughter of affected flocks. Efforts to control the disease by vaccination alone, without slaughtering affected birds to reduce the virus load in the environment, will probably not be successful. Depending on the incidence and distribution of outbreaks, vaccination may be undertaken around outbreaks (ring vaccination) or throughout the poultry population (mass vaccination).
### VACCINE type

<table>
<thead>
<tr>
<th>VACCINE type</th>
<th>Advantages</th>
<th>Disadvantages</th>
</tr>
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<tbody>
<tr>
<td><strong>INACTIVATED HOMOLOGOUS VACCINE</strong>&lt;br&gt;The same H and N antigens as the strain isolated in the outbreak</td>
<td>• Readily available&lt;br&gt;• Rapid onset of immunity with adjuvants&lt;br&gt;• Inexpensive&lt;br&gt;• Safe</td>
<td>• Impossibility of differentiating vaccinated from infected birds serologically&lt;br&gt;• Monitoring by using sentinel unvaccinated birds (identification, bleeding and swabbing) is time-consuming, requires planning and monitoring&lt;br&gt;• Requires boosters in long-lived species&lt;br&gt;• Requires percutaneous injection</td>
</tr>
<tr>
<td><strong>INACTIVATED HETEROLOGOUS VACCINE</strong>&lt;br&gt;(DIVA Strategy: Differentiation of Infected from Vaccinated Animals)&lt;br&gt;The same HA subtype and a different NA subtype compared to the virus isolated in the outbreak</td>
<td>NA: marker of field infection. Serology can determine whether birds in a vaccinated flock have also been infected.</td>
<td>• Laboratory capacity to perform the discriminatory test based on the N antigen&lt;br&gt;• Serology is expensive, requires additional reagents and requires a complete knowledge of circulating N antigen sub-types&lt;br&gt;• Requires boosters in long-lived species&lt;br&gt;• Requires percutaneous injection</td>
</tr>
<tr>
<td><strong>RECOMBINANT FOWLPOX VIRUS</strong></td>
<td>• Enables the differentiation between infected and vaccinated birds by serologic tests&lt;br&gt;• Specificity of the immune response directed exclusively against HA components&lt;br&gt;• Vaccination is rapid and only one dose is required.&lt;br&gt;• Inexpensive</td>
<td>• Can only be used to vaccinate chickens without previous fowlpox exposure. Therefore, usually applied only to day-old chicks&lt;br&gt;• Cannot be used in ducks/geese&lt;br&gt;• Requires percutaneous injection</td>
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### Sourcing vaccine

There are a number of different avian influenza vaccines available. Conventional vaccine is prepared from the allantoic fluid of infected eggs, which is inactivated and emulsified with an adjuvant. Attenuated live influenza virus vaccines are not recommended, because of the risk that the vaccine virus could either mutate or reassort with other influenza viruses to become virulent. However, recombinant vaccines have been produced, including fowl-pox virus with the influenza haemagglutinin gene inserted.
Generally, conventional inactivated vaccines are used. The main immunogenic component is the haemagglutinin protein. It must be of the same subtype as the outbreak virus (for the current widespread Asian epidemic, this is H5). The neuraminidase antigen can be the same as the outbreak strain. However, if differential serology is to be undertaken for monitoring vaccine response or virus activity (the DIVA method), then a different neuraminidase should be used in the vaccine (e.g., H5N2 or H5N9).

It is also possible to leave a small number of identified sentinel birds unvaccinated which will aid monitoring for flock infection. If sentinel birds show disease symptoms or die, virus isolation and serological test have to be done to confirm flock infection.

The DIVA strategy requires testing of serum samples for antibody to the neuraminidase, to differentiate that of field strain(s) from that of the vaccine strain(s). It assumes that the heterologous N antigen is not circulating in the field; and thus knowledge of circulating AI viruses (virulent or not) is known. In circumstances, such as currently prevails in Asia, where there may be several strains of avian influenza virus circulating, and where in some countries there are vaccines in use with several different neuraminidase antigens, it may be difficult to apply the strategy. Also, the differentiating assay involves the use of additional reagents, which may be beyond the scope of most national laboratories as it represents an additional expense. A decision to apply the DIVA strategy therefore needs to be carefully considered against these constraints; and may be more appropriate towards the end of a successful control campaign.

For inactivated vaccines, two doses of the vaccine must be given, approximately 30 days apart, to achieve adequate protection. Vaccinated birds are generally not fully protected from infection but have increased resistance to infection, suffer less clinical disease and shed substantially less virus in the event that they become infected. Longer lived species (ducks, geese, yellow chickens) require booster injections of vaccine to maintain protection.

Recombinant fowl pox vaccine can be used for vaccinating day-old chicks. Since it is a live pox virus, it can be applied by stab inoculation into the wing web, which can be performed quickly with minimal training. This vaccine cannot be used in older birds, since they are likely to have already become exposed to fowl pox and will not respond to vaccination. A disadvantage of the fowl pox-vectored AI vaccine is that it is ineffectual in ducks. Specifications for purchasing vaccine and a list of potential suppliers is provided in Annex 2.

4.4 Management of disease control

The FAO Manual on the Preparation of National Animal Disease Emergency Preparedness Plans makes recommendations in regard to management of disease control operations. The following recommendations should be considered:

1. To manage disease control on a national basis, there needs to be a suitable command structure for veterinary services. It is essential that information can flow quickly and efficiently from the field to national headquarters and that conversely, control mechanisms are continuous from headquarters to the field. In recent years, government veterinary services in many countries have been restructured, including regionalisation, rationalisation and downsizing, privatisation, separation of policy and operational functions, and separation of authority for field and laboratory operations. Countries may need to consider whether they need to make structural changes or alternative arrangements to adequately deal with animal health emergencies.

2. It is often advisable to have a consultative committee which can meet during the period of an animal disease emergency, to provide the best technical advice to outbreak management personnel. The committee might comprise of the CVO, national directors of field and laboratory services, head of the epidemiology unit, AI expert, directors of state, regional or provincial veterinary services, privates industry,
representatives of other key groups and other technical experts as required. Gaining the cooperation of police, military, and public works offices would be beneficial for the success of any plan.

3. At the time of an emergency, it can be a great advantage to have made arrangements in advance for a national animal disease control centre and local animal disease control centres. The control of diseases in difficult or marginalised areas of some countries may require special consideration.

4. Arrangements need to be made in advance of an outbreak. It should include negotiation with all government authorities and others who will be involved in assisting in disease control. Because certain strains of avian influenza viruses can infect humans, the respective roles of human health and veterinary services need to be considered in advance.

Lessons learned from the 2004 epidemic in Asia:

As observed in South East Asia during the 2004 epidemic, many countries did not have a plan of action supported by national legislation at the time of HPAI detection.

The following points were particularly lacking in several affected countries:

- A structured surveillance program including surveillance protocols in suspected or at-risk farms.
- Protective equipment not available for workers and animal health personnel.
- Investigations procedures: standardised protocols and sets of epidemiological questions for outbreak investigation and mapping. These protocols should also include information on collection of a standard set of samples for disease investigation from infected farms, neighbouring farms and uninfected farms outside of the control areas to allow for more detailed pathological examination of dead birds.
- Restocking programmes were often incomplete with no detailed method to control the flock after restocking.
- Vaccination plans were often incomplete and did not have a coherent strategy for post-vaccination surveillance activities. Vaccines used were often of unknown quality and of dubious efficacy.
- Declaration of disease freedom from areas or zones were made without proper scientific data supporting such claims and were subsequently proven to be false creating additional uncertainty in the population and loss of consumer confidence.
- Absence of adequate legislation or difficulties of enforcement.

Field services capacity

Veterinary services must have the capacity to undertake disease surveillance, investigate and respond to disease outbreaks and report to various levels of the official veterinary service structure. In designing a proposed strategy to control avian influenza, careful consideration needs to be given to the capacity of the field services to assume disease management activities in accordance with legislation that enables and empowers decision makers. Very few national veterinary administrations have the capacity within their own resources to undertake all of the necessary measures required to combat a major transboundary disease incursion. Assistance is often sought from police, military, national disaster and other emergency organisations late in the process. The immediate and early needs from other public offices and cooperation from private industry must be anticipated to assist field veterinary services from performing their duties. The access to extra budgetary resources should be part of the contingency planning process and appropriate arrangements for collaboration negotiated in advance.

An electronic disease information database can be a great asset in tracking the great amount of detail that can be accumulated in the course of a disease epidemic. There are many systems available, including the FAO-designed and supported system, TADinfo. FAO, OIE and WHO have collaborated in establishing a Global Early Warning and Response System (GLEWS) to enable international animal and human health
authorities to receive early notice of outbreaks, or the likelihood of certain diseases occurring based on disease intelligence and forecasting risks. Data is analysed and information made available to international and national disease control authorities.

4.5 Laboratory diagnosis

A laboratory service is required that is able to:

- undertake testing of samples collected for surveillance activities.
- test samples collected for diagnosis.
- test samples collected for monitoring response to vaccination.

Consideration needs to be given to the capacity of veterinary services to collect and deliver specimens to a competent national laboratory. There are benefits to having regional laboratories that might have better access to the field. However, in many countries expert personnel are lacking and it may be more effective to have just one central laboratory that is well resourced and properly staffed.

4.5.1 Samples

Samples taken from dead birds should include intestinal contents (faeces) or cloacal swabs and oropharyngeal swabs. Samples from trachea, lungs, air sacs, spleen, kidney, brain, liver and heart may also be collected and processed either separately or as a pool. Intestines (loops) should always be collected last and packaged separately to avoid bacterial contamination. Samples from live birds should include both tracheal and cloacal swabs, although swabs of the latter site are the most likely to yield virus. As small delicate birds may be harmed by swabbing, the collection of fresh faeces may serve as an adequate alternative. To optimize the chances of virus isolation, it is recommended that at least one gram of faeces be processed either as faeces or coating of the swab. Should the investigator not be well prepared to perform a necropsy (autopsy), whole birds should be bagged twice (one bag inside another), maintained refrigerated at all times and submitted to a laboratory where a proper necropsy and sample collection can be completed.

The samples should be placed in isotonic phosphate buffered saline (PBS), pH 7.0–7.4, containing antibiotics. The antibiotics can be varied according to local conditions, but could be, for example, penicillin (2000 units/ml), streptomycin (2 mg/ml), gentamycin (50 µg/ml) and mycostatin (1000 units/ml) for tissues and tracheal swabs, but at five-fold higher concentrations for faeces and cloacal swabs. It is important to adjust the pH of the PBS solution to pH 7.0–7.4 following the addition of the antibiotics. Faeces and finely minced tissues should be prepared as 10–20% (w/v) suspensions in the antibiotic solution. Suspensions should be processed as soon as possible after incubation for 1–2 hours at room temperature (22-25°C). When immediate processing is impracticable, samples may be stored at 4°C for up to 4 days. For prolonged storage, diagnostic samples and isolates should be kept at –80°C without PBS. Never use alcohol to preserve samples.

Submission of samples to any laboratory outside the country of origin should always be subject to prior agreement with the recipient laboratory (see Annex 4 *Information for shipping International Diagnostic Specimens*) and be transported in containers meeting IATA (International Air Transport Association) regulations. Infectious substances which cause disease only in animals are categorized as UN 2900. Infectious substances which cause disease in humans (or both in humans and animals) must be assigned to UN 2814. All H5 and H7 samples must be assigned to UN 2814. All materials should be in leak-proof containers.
4.5.2 Laboratory tests

National laboratory services should be able to undertake the following tests.

As a minimum requirement:

1. Virus isolation in eggs (SPF or SAN), identification of isolates as “A” influenza virus, haemagglutinin and neuraminidase typing.
2. Serology - including ELISA (for antibody to matrix protein), haemagglutination-inhibition testing.
3. Antigen detection – ELISA or haemagglutinin testing.

In addition, the capacity to perform the following tests is highly desirable:

5. Pathogenicity testing of virus isolates by chicken inoculation
6. Polymerase chain reaction (PCR) technology for rapid detection of virus genome.

Methods are described in the OIE Manual, Chapter 2.1.14. Consideration needs to be given not just to having the appropriate technology but to having the capacity to handle a large number of specimens in the event of a disease control emergency.

Countries also have access to international expertise to assist in confirming the identity and characteristics of isolates and undertaking further key studies that are of particular importance at the international level. It is important to monitor field viruses for early detection of changes and this can be done by systematic sampling of birds, during an epizootic and in inter-epizootic periods and sending suspect samples to a reference laboratory for confirmation and further characterisation.

A list of the OIE/FAO reference laboratories is in Annex 3.

In April 2005, OIE and FAO launched a joint scientific worldwide network to support the veterinary services in the control Avian Influenza (OFFLU). The objectives of the new network are:

- to collaborate with the WHO human influenza network on issues relating to the animal-human interface, including early preparation of vaccines for possible human use.
- to promote research on avian influenza
- to offer veterinary expertise and new skills to countries to assist in the control and eradication of HPAI.

Through an active and permanent scientific cooperation, the network will develop collaborative research proposals; provide multidisciplinary teams to countries requiring assistance; and act as a link between OIE/FAO reference laboratories, and regional and nation ones. Sharing permanently updated scientific information and expertise on efficient control methods will provide a pro-active approach in helping infected countries to progressively control and eradicate the disease and for disease-free countries to better protect themselves.

For more detailed information, please see the OFFLU website www.offlu.net

Field diagnosis:

Often decisions need to be made immediately in the field during an emergency. Waiting for a laboratory diagnosis before taking action to quarantine an area can lead to critical delays. Clinical and pathological findings in the field can often be sufficient to make a presumptive diagnosis of HPAI and actions taken accordingly. Though some rapid on-site diagnostic assays are available these, to date, have proven to be of
poor sensitivity and are expensive. Several institutes are actively researching the development of improved sensitive, specific and cost effective rapid on-site assays, but these remain in the development stage and would require validation in the field before their recommendation.

4.5.3 Communication and Public Awareness

There are several objectives which should be considered in public awareness campaigns:

- Inform farmers and consumers on the infection channels and risks related to AI.
- Communicate information (e.g. time and venues for vaccination, procedures for obtaining compensation).
- Promote better farming practices and improved hygiene.
- Raising awareness of the risk of poultry infection (see Annex 7: poster example).
- Recognising that in many village situations, it may be unrealistic to expect to be able to improve biosecurity.
- Seeking assistance from the community by poultry owners reporting unusual sickness and mortalities in their flocks.
- Cooperation during disease control activities, should an emergency occur.
- In conjunction with human health authorities, raising awareness of the risk of transmission from poultry to human and about disease in humans.

4.5.4 Human health and safety considerations

Certain avian influenza viruses can infect humans, occasionally causing severe disease and sometimes death. The WHO website (www.who.int) provides updates and recommendations with regard to symptoms, patient care and management, and recommendations to reduce occupational risks (i.e., personal protective equipment). The following should be considered:

**General considerations for the human population**

- vaccination with seasonal human influenza vaccine (which contain H1, H3 and B virus antigens), to minimise the risk of multiple influenza infections causing a new virus to emerge.
- avoid unnecessary contact with infected or exposed poultry, poultry products and poultry waste.
- in particular, people in poor health and children should avoid all contact with affected birds and their environment.
- Do not purchase or consume poultry that are not healthy or wholesome.

**Specific considerations for all personnel involved in disease control**

- selection of workers should avoid those in high risk categories.
- workers should wear protective overalls and where gross contamination is likely, a waterproof apron.
- disposable or rubber work gloves should be worn.
- a disposable P2 or N97 respirator should be worn.
- goggles or a visor should be worn to protect against eye splash.
- disposable footwear or rubber boots should be worn.
- protective clothing should be disinfected after use.
- hands should be washed after protective clothing is removed.
- use of antiviral drugs is recommended, during and for 7 days after exposure.
- workers should monitor their health, watching for signs of fever, respiratory symptoms (e.g., cough) and conjunctivitis (eye inflammation).
- illness in workers or their close contacts should be reported to supervision public health authorities immediately.
5. Prevention and Biosecurity

Areas that have not been affected by the HPAI outbreak or those that have undergone culling, disinfection, and even vaccination should improve their biosecurity. Improved biosecurity at whatever level is cost effective in comparison to the losses from disease, depopulation, and further anguish, be it at the village level or commercial farm.

Likely the most difficult environment to improve biosecurity and disease prevention is in a village, where poultry and other animals are allowed to move without any restrictions and there are no costs to animal care (feeding), but their losses due to disease or scavenging animals (dogs, cats, wildlife) are high. Under these circumstances, the role of rural developing agencies can be beneficial, in educating the advantages of keeping their animals in a fenced enclosure where environmental stresses are minimised, stealing less likely, be safer from scavenging animals, or eliminate loses from valued animals being run over by motorcycles, cars or lorries.

5.1 Restricted Access means keeping the disease out.

Restricting access to the property or farm through the use of fences and enclosures creates a barrier between clean areas where the poultry are kept and the outside environment. Allow only people known to the owner to have access to where the poultry kept and who themselves do not have poultry of their own and do not participate in events where birds congregate, such as cockfights. Particular attention needs to be paid to workers on poultry farms who keep backyard poultry at home - best practices would stipulate that no workers should have poultry of their own, as this is a high risk avenue for disease introduction. Wild birds – resident fowl or migratory birds – should not have contact with the flock through the use of screens, or overlying nets. Visitors that wish to see the poultry should wash their hands and change their shoes and use footwear that could be provided by the owner (e.g., rubber boots that are kept for such visitors). If visitors have birds of their own, they should not be allowed near the birds.

Ducks kept in ponds or paddies shared with other ducks of a different owner represents a high risk, unless all duck owners agree on the measures that collectively can be taken. For instance, erecting poles with nettings that separate one owner’s flock from another; and taking turns in scaring away wild birds from landing or feeding within the production flocks.

5.2 Clean areas means healthy chickens, geese, and ducks

Keep the area of the flock clean from trash and garbage (food waste, plastic bottles, glass bottles, tins or drums). When the owner or care person needs to attend to the chickens or other poultry (e.g., collecting eggs, feeding or watering chores, change the bedding, or the repair of fencing material), a change of clothing and boots should be required. These clothes and boots can be cleaned and disinfected upon exiting the enclosure and be ready for the next use. Dirty clothes should be washed with detergent and hung out to dry in the sun; boots should be washed in chlorinated water, or with soapy water. Washing hands with soap before entering the caged area should be practiced each and every time. Tools (feeding scoops, shovels, brooms) and feeding pans used in the caged areas should be kept clean daily. All manure should be removed and disposed of properly (i.e., compost pile). Keeping a wide pale with chlorinated or soapy water for frequent before entering or exiting the enclosure is a good reminder to follow biosecurity.

Keeping the cages clean prevents pathogens from accumulating and causing health problems. Clean cages keep the birds and eggs clean as well - which translates into better market prices.
Sick or dead chickens must be removed quickly and community animal health workers or the local veterinarian informed of such illness or death.

5.3 Buy healthy: keep healthy

Transportation of birds to the farm can represent considerable a risk – not only should the owner be aware of the “good” price obtained, but also awareness should be made as to that vehicles, (trucks, motorcycles, bicycles), cages, equipment and feed may be contaminated when returning or entering the farmer’s property. Newly purchased equipment should be thoroughly washed with soapy water or otherwise disinfected before use. Newly purchased birds should be housed in a separate enclosure for at least two weeks before allowing them to mix with birds already on the farm. The owners are advised that it is important to keep species separate, and not mix ducks with chickens, chickens with pigs, or ducks with pigs. Good practices also include not mixing of different aged animals together.

5.4 Use of clean equipment – keeping disease out

Poultry equipment, such as cages, egg crates, shovels or rakes, should not be shared between family or neighbours. Wooden pallets, wooden handles, or egg crates can be porous, and even though they can be treated with disinfectant, these items are difficult to ensure that indeed they are completely disinfected. Metal cages can be cleaned and disinfected; if these are borrowed because of necessity, they must be cleaned and disinfected by the owner of the birds before they are reused.

5.5 Report early signs of a problem that could be devastating

Many diseases of birds look similar. Early detection and prompt reporting would likely assist in stopping disease spread. The owner must know who and where to report abnormalities on the farm when they begin not when they end. Signs to be reported include: sudden death, depression and decreased appetite, diarrhoea, breathing difficulties such as coughing, sneezing, gasping, or nervous twitching or dropped wings or paralysis, swelling of the head with darkened combs, wattles, or legs.

Owners must be assured that early reporting of a problem will benefit them, their families and their village in the long run. In this regard, the government - in conjunction with the poultry industry - should be prepared to react and provide proper compensation (See section 4.3.3 above). Failure to provide an incentive for compensation for disease reporting will undoubtedly lead to disease spread.

5.6 A period of rest

One prevention measure that can be instituted, but requires planning and several enclosures, is the practice of “all-in all-out”. This method used in many countries encompasses the idea of having a complete growth cycle of chickens (or other species) from the moment of introduction – as in day day-old-chicks – all the way to marketing age. At no time are other birds introduced into the enclosure. Once sent to market, the floor is scrapped clean from faecal and feather debris and bedding and feed removed; cages and other equipment are cleaned and prepared for the introduction of young healthy birds. It would be wise to keep the enclosures – free of birds and other animals – for a rest period of say, seven days, before bringing in the next batch of poultry.

With duck operations, the “all-in all-out” operation may be more difficult, unless there is planning in the growth cycle between birds using the same pond and double netting used between age groups. Double netting – 2-3 metres apart – implies additional cost, but decreases the opportunity of pathogens from contacting susceptible ducklings. Although the ducks share the same water ponds, in which avian influenza viruses may survive, it still decreases the likelihood of disease transmission.
5.7 Vaccination against avian influenza or other diseases

Vaccination, in general, increases the resistance of poultry to disease but does not eliminate the possibility that infection may occur in flock. Prevention of disease and infection can only be accomplished with other aspects of Prevention and improved Biosecurity.

When poultry are to be vaccinated, it would be wise that the owner ensure that the vaccination team changes clothes and clean and disinfect their boots, gloves and equipment before entering the poultry enclosures. Should the vaccination team resist such instructions, the owner should document the failure of biosecurity measures to the appropriate veterinary authorities.

5.8 Compartmentalisation

In the terrestrial Animal Health Code, compartmentalisation refers to one or more establishments under a common biosecurity management system containing an animal subpopulation with a distinct health status with respect to a specific disease or specific diseases for which required surveillance, control and biosecurity measures have been applied for the purpose of international trade (more information available is chapter 1.3.5 on Zoning and compartmentalisation)

In countries where the disease may be present in some areas or confined to some production systems, this concept can be applied to poultry operations that will adopt strict biosecurity measures to prevent the introduction of the disease all along the production process.

In poultry operations that are tightly controlled by producers, a strict method of operation must be assured to prevent disease from entering the operation. Besides the measures mentioned in the above sections of this Manual, the operators need to constantly monitor areas or risk and practice “all-in all out” measures. For example: the origin of fertilised eggs, certified biosecured and reliable hatcheries and their incubators, certified feed sources, and transport companies must be registered, dated and documented. The poultry operation must register a complete account of their activities and sources, which include:

- Census of production – stages and location
- Protocols for training of operators
- Instructions to operators within the farm (clothing, cleaning, vaccination, feeding, reporting, etc.)
- Protocols for cleaning and disinfection
- Purchases and location of suppliers
- Vermin and insect control measures
- Egg crate circulation, management, and acquisition
- Employee profiles and responsibilities
- Transport control on and off the premises
- Employee and employee-family awareness
- Have registries open to frequent regulatory inspection

One area of concern to regulatory authorities is the reality that many highly developed production poultry operations have their own poultry diagnostic laboratories – that may surpass national potentials – and can carry out diagnostic assays not reported to the authorities. It is indispensable that such commercial associated laboratories and their managers be made aware of the importance of reporting of disease occurrence and consequences to international trade for the country and their enterprise.

The commercial operators should be linked with prevention, contingency, and emergency plans for national and regional success and health.
Annexes

Annex 1: Selected references for further information

1. Guiding principles for highly pathogenic avian influenza surveillance and diagnostic networks in Asian
2. FAO position paper on AI control strategy
3. Global strategy for the progressive control of highly pathogenic avian influenza (HPAI)
   Operational Procedures Manual – Disposal
5. FAO EMPRES Good Emergency Management Practice
   Australian Veterinarians. Bureau of Resource Sciences, Australian Government Publishing Service,
   Canberra.
   http://www.oie.int/eng/normes/mmanual/A_summary.htm
9. OIE Guidelines for HPAI surveillance
10. OIE Terrestrial Animal Health Code
Annex 2: Sample - Tender Document for Inactivated Avian Influenza vaccines

To be used for the purchase of inactivated avian influenza vaccine, by governments or donor organisations, for control of disease in infected countries.

Introduction

These are specifications for the purchase of inactivated avian influenza vaccine, for use in poultry, including chickens and ducks. Vaccination is used to provide protection in the face of possible exposure or to reduce the viral load in an infected environment. Vaccinated birds are generally not fully protected from infection but have increased resistance to infection, suffer less clinical disease and shed substantially less virus.

There are several options for vaccine. Conventional vaccine, for which this specification is written, is prepared from the allantoic fluid of infected eggs, which is inactivated and emulsified with adjuvant. Attenuated live influenza virus vaccines are not recommended, because of the risk that the vaccine virus could either mutate or re-assort with other influenza viruses to become pathogenic. However, recombinant vaccines have been produced, including fowl-pox virus with the influenza haemagglutinin gene inserted and haemagglutinin produced in a baculovirus expression system. This specification does not cover the requirements for recombinant vaccines.

The virus type used for vaccine production must be of the same haemagglutinin type as the outbreak virus. For maximum potency, it is preferable for the vaccine virus to be closely related to the outbreak strain. If post-vaccination monitoring depends on serology to determine whether antibody-positive birds have been infected or vaccinated (the DIVA test), the neuraminidase type should be different to that of the outbreak strain.

Tender Specification

General requirements

1. Vaccine manufacture must be undertaken in accordance with OIE Guidelines – Chapters 1.1.7 and 2.7.12 of the Manual of Diagnostic Tests and Vaccines for Terrestrial Animals, 5th edition 2004. It must be produced under Good Manufacturing Practice and under acceptable third-party audited quality assurance.
2. In assessing the acceptability of a vaccine, [FAO or other purchaser] may require documentation to be furnished to validate GMP and quality assurance practices and the production details for a specific vaccine batch. [The purchaser] may also seek to undertake an audit of the manufacturing plant(s).
3. The vaccine must be registered or otherwise acceptable for use, by the Government of [insert country].

Specific requirements

1. The requirement is for [insert number] of doses of vaccine for use in [insert species].
2. The vaccine must contain haemagglutinin antigen of H[insert type – for current SE Asia epidemic, H5] type. Evidence (challenge or VN test) should be provided that the vaccine protects against the virus strains currently circulating in [insert country/region].
3. Evidence should be provided that vaccine produced by the same means (i.e. not an individual batch requirement) in the same manufacturing plant significantly reduces virus transmission from vaccinated birds when subsequently infected.
4. The vaccine virus must be derived from an LPAI virus strain.
5. The virus should be grown in specific antibody negative or specific pathogen free eggs.
6. The virus is to be inactivated with formalin or beta-propiolactone.
7. The vaccine should be emulsified with a mineral oil adjuvant or with an alternative adjuvant with similar immuno-stimulating efficacy.
8. The vaccine must have undergone appropriate sterility, safety and potency tests in accordance with international standards.
9. The vaccine must have a minimum of one microgram per dose of haemagglutinin protein. Vaccine of a higher haemagglutinin concentration will be considered favourably. Alternatively, the potency of the batch may be demonstrated by live bird challenge with virulent virus or by a minimum HI antibody response of 1:32 in vaccinated birds.
10. Packaging of the vaccine should be in containers of [insert number of doses].
11. Labelling in [insert language/s] must indicate manufacturer, type of vaccine, batch identification, volume of contents, storage recommendations and expiry date. Package insert in [insert language/s], to include instructions for vaccinating poultry, recommended species to which the vaccine applies, vaccination regime and dose.
12. Vaccine to have a minimum of six months period prior to expiry, on delivery.
13. Vaccine must be delivered to cold storage in [insert place or country]. Verification will be required of continuity of appropriate storage of the vaccine from production to delivery.
Annex 3: Inactivated Avian Influenza Vaccine Suppliers

The companies below provide inactivated avian influenza vaccine (and some of them also recombinant vaccines). NB - By providing their contact details, FAO is not endorsing their product nor recommending them above other possible suppliers.

Harbin Veterinary Research Institute, China

- H5N2 - seed virus is A/Turkey/England/N-28/73 - Inactivated monovalent
- H5N1 – recombinant virus from A/Goose/Guangdong/1996 and human influenza vaccine virus
- H5N1 – Live recombinant avian pox virus vectored H5

Intervet – vaccines for chickens and ducks

http://www.intervet.com
- Nobilis Influenza H5 – for poultry, used in Vietnam - strain A/Chicken/Mexico/232/94/CPA
- Nobilis Influenza H5N2 – strain A/duck/Potsdam/1402/86
- Nobilis Influenza H5N6 – strain A/duck/Potsdam/2243/84
- Nobilis Influenza H7N1 – strain A/CK/Italy/473/99
- Nobilis Influenza H7N7 – strain A/duck/Potsdam/15/80

Merial – vaccine for chickens and ducks

http://www.merial.com
- Gallimune Flu H5N9 – for chickens, seed virus is A/turkey/Wisconsin/68 (H5N9)
- BioFlu H7N1 and H5N9 – for chickens, seed viruses are A/chicken/Italy/1067/99 (H7N1) and A/chicken/Italy/22A/98 (H5N9)

Lohmann

- Avian Influenza H1N1 – for turkey breeder hens
- Avian Influenza H2N
- Avian Influenza H3N
- Avian Influenza H4N
- Avian Influenza H5N
Annex 4: OIE/FAO Reference Laboratories and Experts for avian influenza

Dr Ortrud Werner
National Reference Laboratory for Highly pathogenic avian influenza and Newcastle disease, Institute of Diagnostic Virology, Federal Research Centre for Virus Diseases of Animals (BFAV)
Insel Riems, Boddenblick 5a, 17493 Greifswald - Insel Riems
GERMANY
Tel: (41) 383.517.152 Fax: (41) 383.517.151
Email: ortrud.werner@rie.bfav.de

Dr Ian Brown
VLA Weybridge
New Haw, Addlestone, Surrey KT15 3NB
UNITED KINGDOM
Tel: (44.1932) 34.11.11 Fax: (44.1932) 34.70.46
Email: i.h.brown@vla.defra.gsi.gov.uk

Dr Paul W. Selleck
CSIRO, Australian Animal Health Laboratory (AAHL)
5 Portarlington Road, Private Bag 24, Geelong 3220, Victoria
AUSTRALIA
Tel: (61.3) 52.27.50.00 Fax: (61.3) 52.27.55.55
Email: paul.selleck@csiro.au

Dr B. Panigrahy
National Veterinary Services Laboratories
P.O. Box 844, Ames, IA 50010
UNITED STATES OF AMERICA
Tel: (1.515) 663.75.51 Fax: (1.515) 663.73.48
Email: brundaban.panigrahy@aphis.usda.gov

Dr Ilaria Capua
Istituto Zooprofilattico Sperimentale delle Venezie, Laboratorio Virologia
Via Romea 14/A, 35020 Legnaro, Padova
ITALY
Tel: (39.049) 808.43.69 Fax: (39.049) 808.43.60
Email: icapua@izsvenezie.it

Dr Hiroshi Kida
Graduate School of Veterinary Medicine, Hokkaido University, Department of Disease Control
Kita-18, Nishi-9, Kita-ku, Sapporo 060-0818
JAPAN
Tel: (81.11) 706.52.07 Fax: (81.11) 706.52.73
Email: kida@vetmed.hokudai.ac.jp

For more information visit the OFFLU website www.offlu.net
Annex 5: Information for shipping international diagnostic specimens

To the OIE/FAO and National Reference Laboratory for Newcastle disease and Avian Influenza
Virology Department. Istituto Zooprofilattico Sperimentale (IZS) delle Venezie
(As of November 2005)

Types of specimen: Specimens submitted may be virus isolates (not via Marco Polo Airport, Venice) made in the submitting country or clinical specimen, such as tissues or swabs, collected from diseased birds.

Packaging requirements: All materials should be in a leak-proof containers. Packaging should be composed of (1) a primary receptacle, (2) a secondary packaging and (3) a rigid outer packaging. Packaging of “diagnostic samples” (coded UN3373 with IATA PI650 standard) and a “virus isolates” (coded UN2814 for HPAIV and UN2900 for NDV with IATA PI602 standard). Contact couriers to ascertain providing boxes complying with these requirements

Documents to be accompanied for clearing: Import permissions of the Italian Ministry of Health (formerly provided by the IZS) and 8 signed proforma invoices (8 originals with signature. No photocopy accepted. The format will be formerly provided by the IZS) should be attached firmly to the box.

Shipping modality: Air freight or couriers to Milan Malpensa Airport, Rome Fiumicino Airport or Venice Marco Polo Airport. Arrange for shipments to arrive in Italian airports from Monday to Thursday only.

Shipping Address:
Instituto Zooprofilattico Sperimentale delle Venezie
Virology Department
Viale dell’Università’10
35020 Legnaro
Padova, Italy

Notification of shipment: Before shipping, please notify the following information to the IZS contact person
- Embarkation date
- Airline name and the Flight number
- Name of the destination airport
- Date of arrival in Italy
- Airway bill number

Contact people at the IZS:
William Dundon E-mail: wdundon@izsvenezie.it
Phone: +39 041 8084371, Fax: +39 041 8084360
Giovanni Cattoli E-mail: gcattoli@izsvenezie.it
Alessandro Cristalli E-mail: acristalli@izsvenezie.it
Maria Serena Beato E-mail: msbeato@izsvenezie.it

Important: Contact the IZS in order to discuss testing and testing materials before shipping. Notify the contact person with whom the IZS will keep in touch.
To the **Australian Animal Health Laboratory (AAHL)**  
(As of November 2005)

**Type of specimens:** Specimens submitted to AAHL for disease diagnosis may be either virus isolate submitting country or clinical specimens, such as tissues or swabs, collected from diseased birds.

**Import permit and packing:** Copies of Australian import permits are available from AAHL by contacting aahl-accessions@csiro.au. All specimens must be packed in leak-proof containers in accordance with the appropriate IATA regulation and appropriately labelled. Suitable transport containers, packing instructions are also available from AAHL by contacting aahl-accessions@csiro.au. Copies of the import permit and other consignment details should be attached to the outside of the package to expedite clearance through Australian customs.

**Notification of shipment:** If submitting specimens please notify the accessions clerk on accessions@csiro.au, the Duty Veterinarian on dutyvet@csiro.au or Dr. Peter Daniels on +61 3 5227 5000 of the consignment details so that the specimens can be collected upon arrival in Australia. Alternatively send the information by facsimile to +61 3 5227 5555. Consignment details include the consignment note/air weigh bill number, courier/airline and expected arrival date.

**Shipping address:**

The Director  
Australian Animal Health Laboratory  
5 Portarlington Road  
Geelong, 3220  
Australia  
Telephone +61 3 5227 5000  
Facsimile +61 3 5227 5555  

**Contact for Avian Influenza:** You may also wish to discuss the testing required with Peter Daniels (peter.daniels@csiro.au) or Paul Selleck (paul.selleck@csiro.au) on +61 3 5227 5000 prior to submitting the specimens.
To the Avian Virology Laboratory, Veterinary Laboratories Agency, Weybridge, UK
From outside the EU
(As of November 2005)

Packaging requirements. All materials should be in leak-proof containers. At least two layers of packaging should be used and the inner layer treated lightly with disinfectant.

The outer packaging must be marked as follows:

ANIMAL PATHOGEN – PACKAGE ONLY TO BE OPENED AT THE AVIAN Virology SECTION, VAL, WEYBRIDGE. IMPORTATION AUTHORISED BY LICENCE NUMBER …*… ISSUED UNDER THE IMPORTATION OF ANIMAL PATHOGENS ORDER.

*Insert one of the following LICENCE NUMBERS:
For Newcastle disease, avian influenza and other viruses: AHZ/2232/2002/5
For tissues and other materials: AHZ/963A/99/2

Shipping address: Avian Virology
VLA Weybridge, New Haw, Adelstone, Surrey KT15 3NB, United Kingdom

Packages should be sent by AIR MAIL or AIR FREIGHT. If sending by AIR FREIGHT it is essential that the AIRWAY BILL NUMBER is given to us by FAX, telephone or Email before the arrival of the materials. Packages sent by air freight should be clearly marked: CARE OF TRANSGLOBAL to ensure rapid processing at the airport.

Notification of shipment: Please notify the VLA-Weybridge, Avian Virology Laboratory of the shipment details before dispatch
Contact:
Dr. I. H. Brown
Direct TEL: +44 01932 357 339; Direct FAX: 01932 357 239; E-mail: i.h.brown@vla.defra.gsi.gov.uk
Dr D. J. Alexander
Direct TEL: +44 01932 357 466; Direct FAX: 01932 357 856; E-mail: d.j.alexander@vla.defra.gsi.gov.uk
To the National Veterinary Services Laboratories (NVSL), Ames, Iowa, USA

**Import permit:** Packages containing diagnostic specimens or organisms (infectious materials) imported from foreign locations into the United States must be accompanied by a permit issued by the U.S. Department of Agriculture. The importation permit, with proper packaging and labelling, will expedite clearance of the package through U.S. Customs. One copy of the permit should be attached to the outside of the shipping container and a second copy placed just inside the lid of the outer shipping container. The importation permit can be obtained from the laboratory (NSVL, Ames, Iowa).

**Packaging requirements:** All materials should be in leak-proof containers and packaged to withstand breakage. All materials should be properly labelled.

**Shipping address:**

Director,
National Veterinary Services Laboratories
Diagnostic Virology Laboratory
1800 Dayton Avenue, Ames, Iowa, USA 50010

**Notification of shipment:** Please notify the Diagnostic Virology Laboratory with shipping information (date of arrival, airline/courier, weigh bill number, etc.) as soon as it is available. Fax information to +1(515) 663-7348 or telephone +1(515) 663-7551.

**Contact for Avian Influenza:**

Dr. Beverly J Schmitt
Direct Tel +1 515/663-7532
Direct Fax +1 515/663-7348
E-mail: beverly.j.schmitt@usda.gov
Annex 6: Criteria for defining infected areas and disease control zones

Infected places

A place classified as an IP will be a defined area (village, farm) in which highly pathogenic avian influenza (HPAI) disease or a highly virulent strain of AI virus exists, or is believed to exist. An IP will be subject to quarantine. A mildly or lowly virulent AI virus may be declared an agent for eradication if it has the potential to mutate into virulent AI virus.

Dangerous contact places

Places classified as Dangerous contact places will be those that contain birds, poultry products, poultry waste that have recently been introduced from an IP (usually up to 21 days before the premises were declared infected) and are likely to be infected or contaminated or any of these items that may have been in substantial contact with people, vehicles and equipment that have been associated with an infected premises within three days of visiting the Dangerous contact places.

Suspect places

Places classified as suspect places will be those that contain birds that have possibly been exposed to an AI virus, such that quarantine and surveillance, but not pre-emptive slaughter, are warranted; or birds not known to have been exposed to an AI virus but showing clinical signs requiring differential diagnosis. The classification ‘suspect place’ is a temporary classification and should be treated as infected until determined otherwise. High priority should be given to clarifying the status of the suspect birds so that the suspect place can be reclassified as either an infected premise (IP) and appropriate quarantine and movement controls implemented, or as free from disease, in which case no further disease control measures are required.

Restricted area

An RA will be a relatively small declared area (compared to a control area) around infected places that are subject to intense surveillance and movement controls. Movement out of the area will, in general, be prohibited, while movement into the area would only be by allowed. Multiple RAs may exist within one CA. The RA does not need to be circular but can have an irregular perimeter provided the boundary is initially an appropriate distance from the nearest IP, DCP or SP. This distance will vary with the size and nature of the potential source of virus, but will be approximately 1–5 km around the IP, depending on the density of poultry premises. The boundary could be the perimeter fence of the IP if the IP is in an isolated location. The boundary in a densely populated area will take into account the distribution of susceptible birds and traffic patterns to markets, service areas, abattoirs and areas that constitute natural barriers to movement. If possible hatcheries should be kept out of the RA.

Control area

The CA will be a larger declared area around the RA(s) and, initially, possibly as large as a Province where restrictions will reduce the risk of disease spreading from the RA(s). The boundary of the CA will be adjusted as confidence about the extent of the outbreak becomes clearer but must remain consistent with the OIE Code chapters on surveillance and zoning (Chapters 1.3.4 and 1.3.5; see Appendix 3). In general, surveillance and movement controls will be less intense and animals and products may be permitted to move under permit from the area.
The declaration of a CA also helps to control the spread of the outbreak from within the RA. The CA is a buffer zone between the RA and the rest of the country. The boundary does not have to be circular or parallel to that of the RA but should be 2–10 km from the boundary of the RA. In general, the movement of possibly contaminated things and materials within the CA is allowed but movement out of the CA is prohibited without CVO approval. This type of control area allows reasonable commercial activities to continue.

**NB When declaring RAs and CAs, the areas must not be larger than necessary, thus restricting the number of properties to be quarantined to only those deemed prudent. If flocks in a quarantine area are not depopulated, then the cost of keeping the birds beyond their normal market age could be substantial.**

**International considerations**

Under OIE Code definitions, an *infected zone* means a clearly defined territory in which a disease (listed in the Code) has been diagnosed. This area must be clearly defined and decreed by the veterinary authorities in accordance with the environment, the different ecological and geographical factors as well as all the epidemiological factors and the type of husbandry being practised. The territory in question should have a radius from the centre or centres of the disease of at least 10 km, in areas with intensive livestock raising, and 50 km, in areas where extensive livestock raising is practised.

In June 1993, the European Union published a decision laying down the criteria for classifying Third Countries with regard to avian influenza and Newcastle disease. Annex C point 4 of this decision states: Around confirmed outbreaks of disease a protection zone with a minimum radius of 3 km and a surveillance zone with a minimum radius of 10 km shall be implemented. In these zones stand still measures and controlled movements of poultry shall be in force until at least 21 days after the end of disinfection operations on the infected holding. Before lifting the measures in these zones the authorities shall carry out the necessary inquiries and sampling of the poultry holdings to confirm that disease is no longer present in the region concerned. The practicality of declaring a zone, the intensity of the industry and the transmissibility of virus causing an outbreak might mean a decision is taken to declare larger areas than those used by the EU.
Annex 7: Poster example

BIRD FLU
(Highly Pathogenic Avian Influenza)
The purpose of this leaflet is to inform the public, especially poultry farmers and those responsible for meat markets, with basic information on Avian Influenza, commonly known as Bird Flu. This leaflet also informs them about the rights and responsibilities in case of eventual disease outbreaks.

WHAT IS BIRD FLU

Bird Flu (avian influenza) is a highly contagious viral disease of poultry and other birds. According to the virulence (ability and degree to cause disease), the bird flu virus is often be characterized as:

- Highly pathogenic – causing illness with high a death rate (over 75%)
- Low pathogenic – causing mild signs of disease, but in case of secondary infections it can cause serious problems with death rates of up to 50 %.

Some low pathogenic types can change over time to become highly pathogenic.

A current strain of avian flu virus, known as H5N1, is circulating in many countries in Southeast Asia, and has recently been confirmed also in Russia, Romania, Croatia and Turkey. The H5N1 can cause disease in humans, too.

WHO CAN BE INFECTED BY BIRD FLU?

Domestic poultry – chickens and turkeys - are most often affected, while ducks and geese often develop only mild signs of the disease. Wild birds, especially wild ducks may be naturally resistant to the infection and do not show clinical signs of the disease.

Birds that do not show signs of the disease can still carry the virus and represent a danger for the introduction of the virus in poultry operations.

HOW IS THE DISEASE SPREADING?

The main sources of infection in poultry are the following:

- Live infected birds even if they don’t appear sick
- Faecal droppings and discharges from sick birds
- Dead Birds
- Contaminated objects, including equipment (egg crates, cages), shoes or clothing, and contaminated ground

It is believed that the disease can be spread over long distances by migrating birds (especially wild ducks and geese, but possibly other water or shore birds). If infected wild birds have contact with domestic poultry during their resting times along their migrating routes, transmission of the virus could occur.

Within the country the disease is most commonly spread in poultry through the movement of people, birds and goods in an infected area and marketing practices used when infected poultry come into contact with healthy birds. Humans play a very important role in spreading the disease since the virus can easily be carried on dirty clothes, shoes, contaminated equipment, vehicles and in the transportation of sick poultry.

Though rare, the disease can be introduced by importing healthy live poultry or poultry products (meat, unprocessed feathers, laying eggs etc.). The virus can also be introduced to an unaffected area or country through illegal trade, especially live birds.

HOW TO RECOGNIZE THE DISEASE IN THE FLOCK

Bird flu spreads very quickly within the flock, so almost all units will be infected in a very short time and birds may die within days. The spread of the disease is likely to be slower in layers - chickens that are used to produce eggs – since they are often in cages and do not mix with each other.

Signs of disease are:

- Depression and lack of appetite
- Drastic drop in egg production
- Swollen head and neck
- Dark and swollen wattles and combs
- Bleeding under the skin
- Sudden death which can reach 100 % of the flock

If any of your birds show these signs you should immediately report to the nearest veterinary station or to the veterinary inspector. Failure to notify the authorities could put more animals - and even humans - at risk of becoming infected.
HOW TO PREVENT BIRD FLU

Basic prevention measures include:

• Decrease the opportunity for wild birds coming into contact with domestic poultry through the use of protective nets, or keeping the poultry in enclosed and protected buildings.

• Apply bio-safety measures:
  o Fence the farms to keep unwanted animals and visitors out. Lock doors!
  o Disinfect vehicles before they enter the farm.
  o Prohibit the entrance of unauthorized people to the farm.
  o Establish disinfection areas (foot-baths) before entering the farm or in each of the poultry houses.
  o Use boots and outer clothing that can be cleaned or changed between houses or farms. Use rubber gloves as well.
  o Clean and disinfect all surfaces regularly (cages, walls, poultry eating and watering areas).
  o Do not borrow equipment from other farms, as these may be contaminated.
  o Disinfect with detergents or hypochlorite solutions

• Replace animals from within the flock or from controlled and healthy flocks.

• Apply the principle of “all in – all out”. This means that all animals in a poultry house are taken off the farm at the same time (for the market), the ground and house must be cleaned and disinfected, and only then can young stock be introduced into the cleaned house. Lock doors!

• Avoiding keeping ducks, chickens and turkeys in the same yard.

• In case of an outbreak on your farm, you should immediately report to the veterinary authorities. Do not sell your animals. Do not eat or feed sick or dead birds to other animals.

• prohibition for movement of all kinds of live poultry, meat and other poultry products from contaminated settlement areas

• increased control for movement of people, mechanical equipment, vehicles, etc. from contaminated yards, farms and settlement areas

• other measures ordered by the veterinary inspectors.

Owners will be compensated for slaughtered birds as part of the measures for preventing the spread of the disease. Compensation will also be paid for destroyed poultry products and equipment according to their market value.

IS BIRD FLU A THREAT FOR HUMAN HEALTH?

The bird flu virus rarely infects people. If it does, the disease is usually not serious and is often characterized as reddening of the eye (conjunctivitis) with mild respiratory symptoms.

However, the highly pathogenic avian influenza H5N1 strain can be a serious health problem, and can kill people that are infected. Avoid contact with birds that are thought to be affected.

HOW DO PEOPLE BECOME INFECTED?

Avian influenza is not a food-borne disease. The bird flu virus is killed by the heat of normal cooking. There is no risk of getting avian influenza from properly cooked poultry meat and eggs. However, sick chickens should not be eaten, as a sick bird often releases toxins and has other microorganisms that may pose a danger to you and your family.

WHICH GROUPS OF PEOPLE ARE AT RISK?

People that are at higher risk are those who work with poultry (breeders, buyers, transporters, slaughterhouse workers and also veterinarians), who are in contact with infected poultry material or are employed in disease control activities (veterinarians employed in laboratories). Hunters and ornithologists are only at risk if they are handling sick or animals that have died due to avian influenza virus.

HOW PEOPLE CAN BE PROTECTED?

Currently, there is no medicine that could fully protect people from bird flu. Present seasonal vaccines against human flu are only effective against known circulating human influenza. Should you or anyone in your family have a fever with flu-like symptoms, it is advised to see medical attention immediately.

WHO DO I CONTACT?

This area to be used for information for local or regional veterinary authority, diagnostic laboratory, help desk, or call-free number

NB: this poster was designed by authorities of Macedonia