UNITED REPUBLIC OF TANZANIA

MINISTRY OF LIVESTOCK DEVELOPMENT

DIRECTORATE OF VETERINARY SERVICES

CONTINGENCY PLAN
FOR
AVIAN INFLUENZA (VETERINARY)

MARCH 2006

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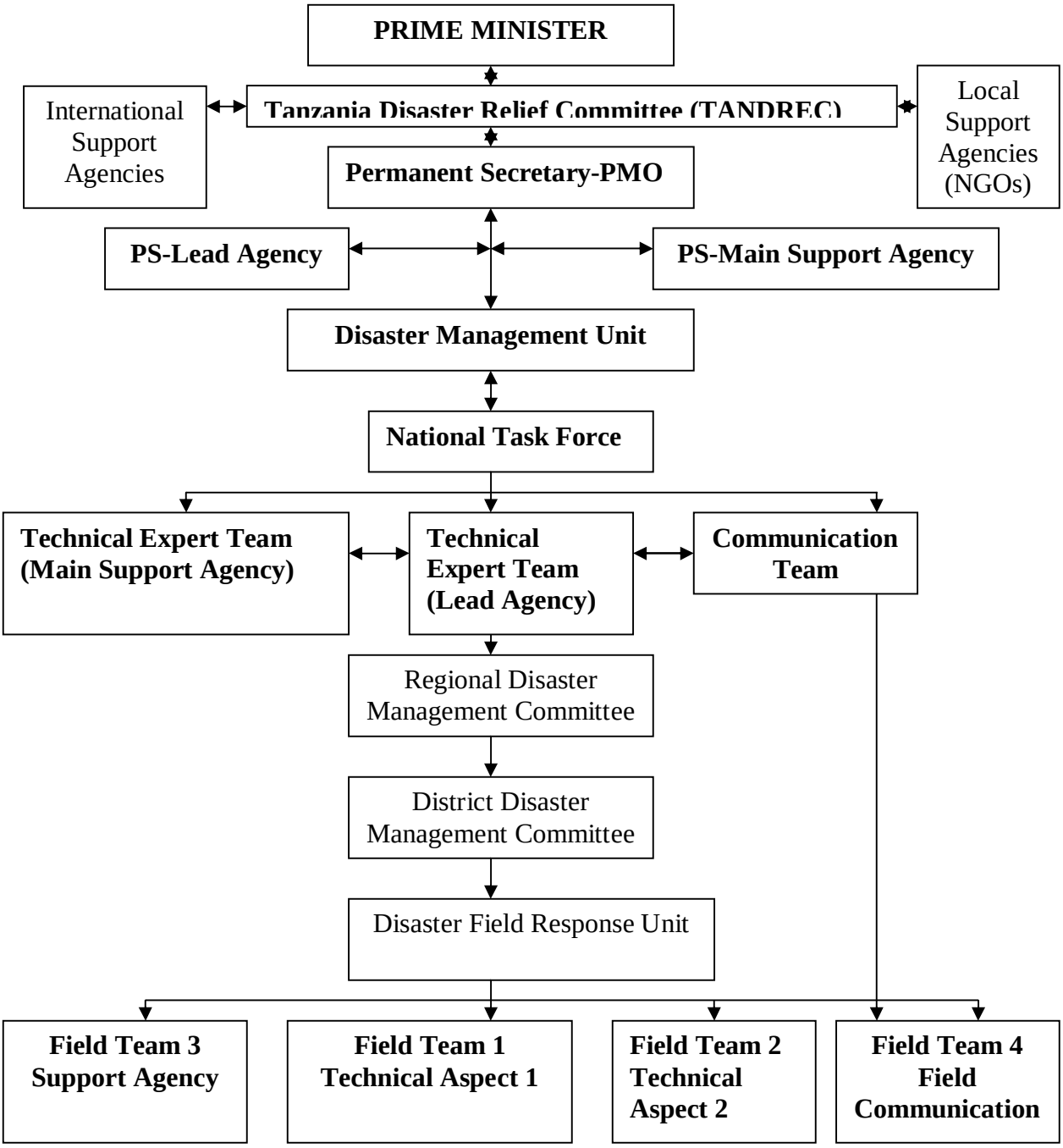
ABBREVIATIONS

AGID	Agar Gel Immunodiffusion Test
AI	Avian Influenza
CVO	Chief Veterinary Officer
DVO	District Veterinary Officer
DVS	Director of Veterinary Services
DCP	Disease Contact Premises
ELISA	Enzym Linked Immunosorbent Assay
EU	European Union
FAO	Food and Agricultural Organization
GIS	Geographical Information System
HA	Haemaglutination
HI	Haemaglutination Inhibition
HPAI	Highly Pathogenic Avian Influenza
IP	Infected Premises
IVPI	Intravenous Pathogenicity Index
LADECC	Local Animal Disease Emergency Command Center
NADECC	National Animal Disease Emergency Command Center
NVRL	National Veterinary Reference Laboratory
OIE	Office International des Epizooties
PBS	Phosphate Buffered Saline
PCR	Polymerase Chain Reaction
PZ	Protection Zone
RZ	Restriction Zone
RLA	Regional Livestock Advisor
RALG	Regional Administration and Local Government
SP	Suspected Premises
SPF	Specific Pathogen Free
SZ	Surveillance Zone
VIC	Veterinary Investigation Centre

Disaster Management Organization Structure

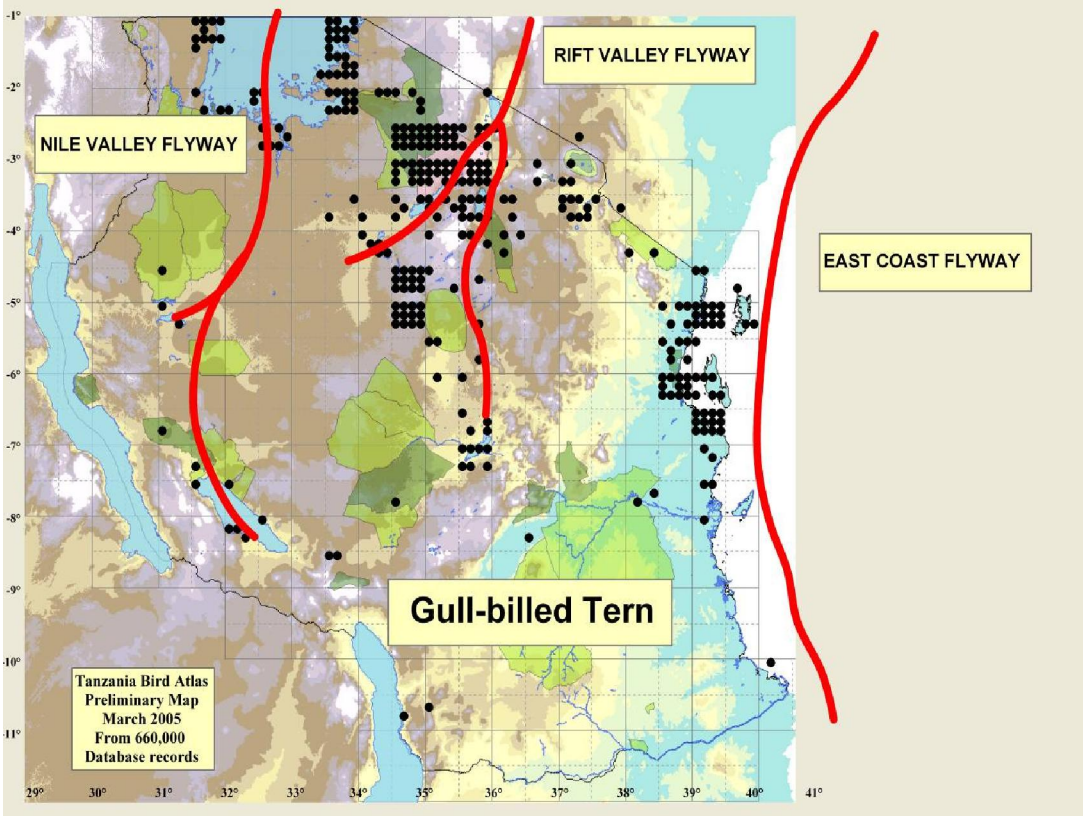


Introduction:

Tanzania is at high risk of Avian Influenza introduction and spreading based on the migratory birdfly ways passing through the country (Map 1) and interactions of people and goods through trade and other movements
This contingency plan for Highly Pathogenic Avian Influenza (HPAI) is prepared to specify containment events and activities that will be implemented in the event of a suspicious or confirmed outbreak of AI at local and national levels The measures adopted follow the recommendations , guidelines and procedures of the Food and Agriculture Organization (FAO),World Organization for Animal Helath (OIE) and the World Health Organization (WHO) for Avian Influenza Emergency Preparedness and Response that aim at maintaining a high level of avian influenza (AI) disease awereness,emergency preparedness and capacity to rapidily repond to disease outbreaks.

The contingency plan provides for the access of all facilities, equipment, personel and other appropriate materials necessary for the rapid and efficient eradication of an AI outbreak and encourage cooperation with neighbouring countries.The plan starts by stating the legal provisions that foster compliancy to disease contol measures in the Tanzania context and then goes on to describe essential areas that goes with Contigency Plans (a) recognizing the event:detection,investigation and reporting/notification (b) verifying the event: event assessment and immediate control measures (c) containing the event; the rapid response and containment operations and (d) operaational manual as an annex.

Map 1: Migratory Bird flyways passing throughTanzania



1-LEGAL PROVISIONS

Highly Pathogenic Avian Influenza (H5N1) is a notifiable disease under the Animal Disease Act Number 17 of 2003.
Statutory Powers for the control of AI are provided in Part III of the Act that spell out the measures for checking livestock diseases ,Part V on powers of Inspectors ,Part VI on Compensation and Part VIII on general provisions on control of animal diseases.

1.1. Notification of AI

Section 14 and 15 of the Act requires owners of animals to report to Inspectors death and diseases respectively of animals for the purpose of identifying the cause of death of the animal.The Inspector on the other hand is required by Section 16 to notify the Director of Veterinary Services and institute measures to control the spread of the disease through quarantine and slaughtering. The notification chanel and sequency of actions that will be executed are diagramatically erabolated in Annex 13

1.2. General measures to be taken at the outbreak area

Zoning to provide for Cordon,Infected ,Protection and Surveillance Areas
Movement control
Vaccination
Killing and destruction of animals
Cleansing and disinfection
Awereness Campaign
The Animal Disease Act No 17 of 2003 provides for the above measures.The Presidentiial Circular Number 1 of 2002 on Animal Disease Control also provides for these measures to be applied in the event of animal disease outbreak and general disease prevention measures.

1.3. Compensation

Compensation resulting from lawful orders for disease control made under the provision of the Animal Disease Act No 17 of 2003 is allowed and the process can be initiated in the bigining by the Inspector as stipulated in Section 16 Subsection 2 (b).The whole Compensation Scheme for disease control is covered under Section 40,41 and 42 of the Act and will be guided by the Compesantion Regulations (in draft).
Compesantion for slaughtered birds in AI outbreak containment as strongly advocated by the FAO/OIE/WHO recommendations is thus legally provided for in this Contigency Plan to encourage poultry farmers comply to disease control measures. A Compensation Regulation is being developed to provide a framework of organizing the Compensation Scheme for the purpose of Animal Disease Control.

1.4. Aid from the police and other persons

Under the National Guidelines for Disaster Management the police department and other stakeholders have a role to play as Support Agencies.The Ministry of Livestock Development is the Lead Agency while the Ministry of Health and Social Welfare and Ministry of Natural Resources and Tourism are Main Support Agencies.Other Support Agencies include the Police (Ministry of Public Safety and Security),Immigration Officers (Ministry of Internal Affairs),the Army (Ministry of Defence),the Judiciary (Ministry of Justice and Constitutional Affairs) and the Media (Prime Ministers Office- Information and Culture).The roles of each of these players are spelt out in Annex 14

2. FINANCIAL PROVISIONS

The contingency plan by the 2006 cost estimates require a minimum of Tsh----- (US$--------) to mount a rapid response to an Avian Influenza outbreak from first detection, confirmation to crisis and post crisis management of the animal disease emergency. The costs will involve personnel, equipment, disinfection, destruction and disposal, emergency vaccination and communication.

3.0 THE CHAIN OF COMMAND

3.1. Ministry of Livestock Development is responsible for the control of AI in livestock and the Ministry of Health and Social Welfare for Human Avian Influenza. The Directors of Veterinary Services in MLD and of Preventive Medicine in MHSW have delegated authority for the direction of control strategies.

3.2. The DVS and DPM in turn delegates AI contingency plan to the National Veterinary Epidemiologist and Emergency Preparedness and Response Unit respectively that will constitute the National Animal Disease Emergency Command Centre (NADECC) (name and address of the centre members are given in Annex1.

In the event of suspicion or an outbreak of AI, the centre will co-ordinate the national strategy, under the over-all direction of the DVS and DPM. The head of the NADECC is responsible for directing the disease emergency strategies of the Local Animal Disease Emergency Command Centres (LADECCs).The 20 Regional Livestock Advisors (RLAs)) are responsible for AI control measures in their areas. The list of RLAs and their contact information are given in Annex 2

3.3. Chain of command for contingency plan is given Annex 12

3.5. NADECC will co-ordinate all activities relating to the national response.

4. THE NATIONAL ANIMAL DISEASE EMERGENCY COMMAND CENTRE (NADECC)

4.1. Mandates of the NADECC

The National Animal Disease Emergency Command or Crisis Centre or Committee is the Steering Committee of Animal Disease Emergency Response Operations. It is charged with the responsibility of coordinating, supervising, resource mobilization and communication in animal disease emergency operations. The Director of Veterinary Services has a mandate to co-ordinate animal disease control operations and in the Contingency Plan is charged to ensure that the NADECC becomes as an acting centre at a short notice or according to risk assessment data provided by the National Epidemiology Unit. The responsibilities of the co-ordinator include the maintenance of checklist for people, equipment and facilities.

a) Maintaining disease preparedness and awareness during “peacetime”

Directing and monitoring of operations of LADECC and ensuring compliance with instructions

- Liaising with NVRL (National Veterinary Reference Laboratory)
- Liaising with agricultural and trading bodies, and the media
- Arranging financial provisions for the contingency plan
- Arranging training programmes

- Organization of simulation exercise
- Arranging disease awareness campaigns
- Managing controls at BIPs, ports and airports

b) Directing the national strategy in the event of suspicion or an outbreak of disease

- NADECC will immediately meet
- Directing, monitoring and supervision the operations of LADECCs

- designing the overall direction of AI control strategies using advices of national expert group
- deploying staff and other resources to the LADECCs,
- Collecting all information and data from LADECCs, local expert groups and NVRL and forward them to national expert group to be analyzed.
- Provide information to EAC SADC,AU-IBAR,OIE, FAO, WHO EU neighbouring countries national authorities including competent environmental authorities and bodies, as well as veterinary, agricultural and trading organisations and bodies;
- Organising an emergency vaccination campaign and also the delimitation of vaccination zones;
- Liaising with competent authorities to coordinate the actions on veterinary and environmental safety;
- managing farmer awareness and general publicity programmes, including press releases, and creating a public relations centre to liaise with the media;
- arranging training programmes
- arranging financial provision
- preparing international disease reports and, at the appropriate times, status for recognition of Zonal or national freedom from the disease;
- maintaining up-to-date lists of available personnel and other resources, and details of where further resources may be obtained;
- Liaison with the LADECCs in the establishment of protection zone (PZ) and surveillance zone (SZ) and vaccination zones if necessary.
- ordering and dispersing essential supplies, including vaccines if they are to be used;
- liaising with other groups involved in the emergency response, including those that may be activated as part of the Contingency Plan;

© Directing the Communication Team on what information materials should be prepared and disseminated to all stakeholders .The Communication Team can be drawn from the members of the National Technical Expert Committee or the National Task Force with a Team Leader who has adequate communication skills and others members from technical departments relevant for the emergency. The Communication Team can co-opt a communication Expert from UNICEF Country Office and the Media Association to tap their vast experience. For Avian Influenza the Communication Team can adopt a Communication Strategy that accompanies this document as an appended document-'Tanzania Avian Influenza & Human Influenza Pandemic Communication Strategy'

4.2. Members of NADECC

Chair: Director of Veterinary Services as the CVO

Acting Chair: Assistant Director Animal Health Services

Members:

Head of Transboundary Animal Disease Control Unit

Head of Zoonoses Control Unit

Head of Zoosanitary Inspection and Quarantine Services

Head of Public Health Department

Director Wildlife Services

Director Animal Production

Director Livestock Research

Director Policy and Planning

Director of National Veterinary Reference Laboratory

Press and Public relations consultancy,

National Expert Group

Representative of MLD Finance Division

Representative of MLD Personnel Division

Representative of Ministry of Internal Affairs

4.3. Facilities and physical resources in NADECC

The NADECC shall be fully equipped with a range of maps based on the regional and district structure for all parts of the country and with suitable communication equipment for liaison with LADECCs and expert groups, laboratories, etc. by telephone, mobile phones, e-mail and fax as appropriate.

a) maps that show animal population density
(b) all suitable means of communication including telephones, fax and if possible facilities for communication with the media;
(c) a communication system allowing exchange of information with the LADECCs, the laboratories and other relevant organisations, preferably computerised;
(e) a shared daily journal which shall be maintained to record in chronological order all the events associated with an outbreak of AI and allowing different activities to be linked and coordinated;
(f) lists of national and international organisations and laboratories that are interested in an outbreak of AI and shall be contacted in such an event;
(g) lists of staff and other persons who may be called upon immediately to serve at LADECCs or in expert groups provided for in the event of an outbreak of AI ;
(h) lists of competent environmental protection authorities and bodies to contact in the event of an outbreak of AI;
(i) maps identifying appropriate disposal areas;
(j) lists of treatment and processing undertakings authorised to treat or process animal carcasses and animal waste that could be commissioned in the event of an outbreak of AI, in particular, indicating their capacity, address and other contact details;
(k) lists of measures to monitor and control disinfectant run-off as well as body tissue and fluid displacement into the surrounding environment as a result of carcass decomposition, particularly into surface waters and groundwaters.

5. LOCAL ANIMAL DISEASE EMERGENCY COMMAND CENTER (LADECC)

AI preparedness and operational control at the local level is under the responsibility of the Regional Livestock Advisor at the regional level in the regional Administration and Local Government (RALGA).

In case of initial phase of an AI outbreak, the Regional Livestock Advisor at the regional level in the regional Administration and Local Government (RALGA) will become the LADECC. However, one or more Operational Units (Districts affected) will be established to provide support to the LADECC, in the case of getting difficulties (e.g. several outbreaks, infected premises located too far from the RLA to take under control outbreaks of AI.

5.1. Mandates of the LADECC:

The responsibilities and tasks of the LADECC are divided into:

a) Responsibilities and tasks to ensure preparedness
b) Responsibilities and tasks during actual (real time) situations

a) Responsibilities and tasks to ensure preparedness
   - Maintaining disease awareness and preparedness within the area, as instructed by the NADECC
   - Arranging disease control exercises locally and participating national simulation exercise
   - Maintaining links with other governmental departments for availability of equipment and personnel and disposal of carcases
   - Identifying all livestock units

b) The "real-time" responsibilities include:

   - Directing and implementing the control strategy in the event of an outbreak, in consultation with NADECC
   - Arranging immediate investigation of reported suspect disease and the transport of samples to the NVRL in accordance with standing instructions
   - notifying reports of disease suspect to the NADECC
   - liaising with the National Expert Group
   - declaring Infected Premises following confirmation of disease
   - liaising with the NADECC to determine the extent of the PZ and SZ and demarcation of the zones
   - carrying out immediate census of animals on holdings within the PZ and SZ, and of epidemiologically connected holdings
   - supervising procedures on the Infected Premises - quarantine of premises, valuation of susceptible livestock, slaughter and disposal of carcases, cleansing and disinfection
   - closing markets within the PZ and SZ and liaison with municipality guardance, police, gendarme and/or military forces to ensure control of livestock movements
   - liaising with the Local Expert Group to identify livestock units where risk of disease is highest and make recommendations to the NADECC for slaughter of potentially infected units
   - carrying out surveillance on livestock units in the PZ and SZ. All units in the PZ must be inspected as soon as possible after confirmation of the disease. Visits to be prioritised on guidance from the National Expert Group
   - tracing of movements off and into infected premises. Market tracing where necessary. Tracing movements of personnel, vehicles, etc.
   - liaising with National Expert Group and NADECC on required action.
   - recording data collected during the epidemiological investigations, movement licences issued, staff and equipment used, diary of events on infected holdings and in LADECC
   - liaising with Local Authority, regarding disposal of carcases and control of disinfectant run-off to minimise effects on the environment.

5.2. Members of LADECC

Chair: Regional Administrative secretary

Acting Chair: Regional Livestock Advisor

- Council Chairpersons& Mayor(s) where the outbreak has occurred
- District& Municipal Executive Directors
- Regional Police Commander
- Regional Security Officer
- District Veterinary Officers
- Regional Accountant
- Regional Planning Officer

Operational Unit:

Taking into consideration of following criteria's; an operational unit can be established at District Level-District Veterinary Office with the decision of LADECC.

- Several outbreaks located in more than one district
- Infected premises are far from the RLA,
- Population density is too high
- Poultry premises are too close in high populated area

5.3. Activities and Function:

- Operational unit will have the same task in accordance to the decisions given by LADECC

5.4. Facilities and physical resources in LADECC:

a) Office space is required for the exclusive use of those involved in the investigation until all tracing activities have been completed

b) A separate table should be provided within this room for viewing maps and for round-table group discussions

c) The room provided should have desk-space, desktop computers with internet connection and telephones for at least two people

d) A record system, preferably computer-based, connected to the NADECC and to all necessary databases, laboratories and other organisations;

e) A shared daily journal which shall be maintained to record in chronological order all the events associated with an outbreak of AI and allowing different activities to be linked and coordinated;

f) Up-to-date lists of persons, including private veterinarians, and local organisations in province who shall be contacted and may be involved in the event of an outbreak of AI;

g) A list of slaughterhouses and the area for burying

h) Detailed maps of the district and of the region on which village locations, topographical features and public roads are indicated (preferably in GIS format)

i) Up-to-date list of competent environmental authorities in the region, as well as other environmental bodies who must be contacted and are to be involved in the event of an outbreak of AI;

(j) Maps identifying suitable disposal sites for burial of carcasses that will not present a risk of harm to the environment, in particular to surface waters or groundwaters;

(k) List of treatment and disposal undertakings authorised to treat or dispose of animal carcasses and animal waste;

(l) List of measures to monitor and control disinfectant run-off as well as body tissue and fluid displacement into the surrounding environment as a result of carcass decomposition, particularly into surface waters and groundwaters.

m) Easy access to all records which are kept at district and/or provincial level on AI vaccination, livestock ownership and movement

n) Storage space for “sampling-kit” components and protective clothing

6.0. EXPERT GROUP

6.1. National Expert Group:

National expert group exists during “peacetime”. The group has skills and technical knowledge of the clinical signs and the epidemiology of AI and also of the methods of prevention and eradication of an outbreak of the disease.

Arrangements are in place to keep the relevant knowledge and expertise up to date. This includes education and regular training of the Expert Group.

The group comprises:

- 1 Virologist from the NVRL
- 1 Epidemiologist from DVS
- 1 Pathologist from NVRL
- 1 Disaster Focal Point Officer

In the event of a report of suspicion of AI, the National Expert Group will be alerted by the NADECC. The tasks of the group are to:

- Evaluate the clinical picture and the epidemiological situation
- Give advice regarding sampling and analyses needed in diagnosing AI
- Give advice regarding additional measures that need to be taken

In case of an outbreak of AI the National Expert Group will be responsible for:
(a) conducting at least in the index case and if necessary on the spot, an evaluation of the clinical picture and an analysis of the epidemiological inquiry in order to collect the necessary data for determining:
(i) the origin of the infection;
(ii) the date of introduction of the infectious agent;
(iii) the possible spread of the disease;

(b) reporting to the NADECC;
(c) giving advice on screening, sampling, test procedures, control and the other measures to be applied and on the strategy to be implemented, including advice on biosecurity measures on holdings or on premises and in relation to emergency vaccination;
(d) following up and guiding the epidemiological inquiry;
(e) supplementing the epidemiological data with geographical, meteorological and other necessary information;
(f) analysing the epidemiological data and perform risk assessments at regular intervals;
(g) assisting in ensuring that the processing of animal carcasses and animal waste is done with a minimum of detrimental effect on the environment.

- Evaluating the national disease situation and assess risks posed by different types of animal movement
- Providing advice that is consistent and proportional to the objective of preventing the introduction and spread of disease
- Advising the NADECC on any additional controls considered necessary for the different types of movement
- Reviewing movement controls as the situation evolves and provide up-to-date advice to the NADECC

- Advising on relaxation of controls and timeframe as the situation improves
- Evaluating codes of practice and protocols for various activities which may involve direct or indirect contact with animals and/or agricultural land and submit recommendations to the NADECC

6.2. Local Expert Group

A Local Expert Group will be established at the LADECC, when AI is confirmed.
The Group l comprises:

- 1 epidemiolgy expert from the VIC
- 1 veterinarian from the RLA or affected District
- 1 Labotory technician from VIC

The role of the group is to:

- Go to field where suspected cases have occurred.
- Investigate the origin and possible spread of the disease
- Prediction on the likely period of infection on the premises
- Describe the situation at the infected holding, the number and species of susceptible and other livestock, the method of husbandry,
- Describe the size and location of the holding and its relationships with other holdings, public roads etc
- Do risk assessment and categorise contact holdings
- Collect and sending the samples,
- Liaise with the experts from the National Expert Group, if necessary call them for investigation
- Examine and assess the surveillance and epidemiology reports
- Send reports and advices to LADECC, NADECC and national expert group.
- Advise to LADECC regarding preventive slaughtering and/or killing,
- Advise on limits of PZ and SZ, other control measures.

7. RESOURCES (Personnel, Laboratory, Equipment)

7.1. During an AI outbreak it will be necessary to deploy quickly a large number of staff and equipment to LADECC. This will be the responsibility of the DVS in collaboration with the DAP and RALG. These staff can be provided from other regions where outbreak is not occurred or private practitioners can be hired.

7.2.The NADECC maintains a list of veterinary staff that can be called upon if there is an outbreak of AI.

7.3. The NVRL maintains a list of laboratory staff experienced in AI sampling, packaging of samples for transport and transport arrangements for sample to be sent to the NVRL .

List of facilities to be maintained at the NADECC

- means for communication (telephone, fax)
- means of transport
- hardware and software (personal computer, printer, Microsoft windows software, Microsoft office software)
- Parking spaces for vehicles adjacent to the building
- Lists and contact numbers for all local organisations associated with livestock to be contacted in the event of AI outbreak.
- Lists of staff who may be called on in an emergency
- Facilities for cleaning and disinfecting personnel, clothing and vehicles
- Protective clothing
- Disinfectants against AI, detergents and soaps
- Slaughtering equipment - tranquillising drugs may also be required
- Equipment for post mortem examination and collection of diagnostic samples
- Sign posts with warning notices for use around infected areas
- Vaccination equipment
- Office equipment
- Outbreak investigation forms
- Tracing requests and reports (check lists)
- Movement permits
- Extra vehicles
- Combustible material
- Flame guns

7.4 Financial Resurces

This Contigency Plan requires Tsh----------equivalent to US$----------of which the government has committed Tsh----------(US$----------) and development parteners have been requested to bridge the gap.

A detailed cost break down of the planned activities is given in Table 1.

Table 1-Contigency Plan Budget

S/N	Activity	When	Resources	Quantity	Unit Cost	Value
A	Pre-Outbreak Activities					
A1	Training of NADECC					
A2	Review Meetings					
A3	Training of LADECC					
A4	Procurement of Kit 1 for NADECC					
A5	Procurement of Kit 2 for LADECC					
A5	Procurement of Kit 3 for VICs					
A6	Procurement of Kit 4 for NVRL					
A7	Training of VICs Lab Technologist in AITtests					
A8	Training of NVRL Lab Technologist in AITtests					
A9	Training of VICs VRO in AITtests					
A10	Training of NVRL VRO in AITtests					
A11	Training of DVOs in High Risk Areas & ZSIS Staff on AI Bissecurity					
A12	Public Aweeness					
A13	Stockpile Vaccine					
A14	Stockpile Disinfection Gear					
A15	Stockpile Destrction & Disposal gear					
A16	Surveillance Operations Livestock					
A17	Surveillance Operations Wildlife					
A18	Strengthen Epidemiology Unit					
A19	Strengthen Veterinary Laboratory System					
A20	Research on Risk Factors					
A21	Regional & International Collaboration meetings					
A22	Rapid Assessment of VS					
A23	Strategic Programme Formulation (Concept note,Appraisal & Funding Application/Negoations)					
	Subtotal Pre-Outbreak Operations					

S/N	Activity	When	Resources	Quantity	Unit Cost	Value
B	Management of Outbreak					
B1	Facilitate DVO Investigation & Notification					
B2	Facilitate LADECC – General Operations					
B3	Facilitate LADECC- Destruction and Disosals					
B4	Compesante for Destructions & Disposal					
B5	Facilitate LADECC Cleansing & Disinfection					
B6	Facilitate LADECC Emergency Vaccinations					
B7	Public Awereness Campaigns					
B8	Strengthen ZSIS at Entry & Exit Points					
B9	Facilitate VICs Field operations					
B10	Facilitate Expert Team Field Operations					
B11	Facilitate Expert Team Meetings					
B12	Facilitate NADECC Field Operations					
B13	Facilitate NADECC meetings					
B14	National stakeholders collaboration meeting					
B15	Public Awereness Campaign					
B16	Regional & International Collaboration Meeting					
	Subtotal Outbreak Management					
C	Post Outbreak Activities					
C1	Poultry Husbandry Extension					
C2	Facilitate Restocking					
C3	Research					
C4	Specialised Training to DVS on Contigency Planning & Management					
C5	Long Term Training of 3 young Vets for MSc Veterinary Epidemiology					
C6	Long Term Training of					

S/N	Activity	When	Resources	Quantity	Unit Cost	Value
	young Vets for MSc & PhD in Veterinary Epidemiology					
C7	Long Term Training of 1 young Vet for MSc & PhD in Veterinary Virology					
C8	Long Term Training of 2 young Vet for MSc & PhD in Veterinary Pathology					
C9	Long Term Training of 1 young Vet for MSc in Laboratory Technology					
C10	Short Term Training on Disaster Risk Management ,GIS & Modern Advances in Crisis Management to DVS & Senior Vets					
	Subtotal Post Outbreak Activities					
	Grand Total for the Contigency Plan					

8. OPERATIONAL MANUAL

8.1. INTRODUCTION

8.1.1. Definition

Avian influenza (AI) is a disease of viral etiology that ranges from a mild or even asymptomatic infection to an acute, fatal disease of chickens, turkeys, guinea fowls, and other avian species, especially migratory waterfowl.

8.1.2.Aetiology

Avian influenza viruses, constitute the virus family *Orthomyxoviridae,* genus A. Type A influenza viruses are serologically categorized into 15 hemaglutinin (H1-H15) and 9 neurominidase (N1-N9) subtypes. Each virus has one HA and one neurominidase antigen, apparently in any combination. All influenza A subtypes in the majority of possible combinations have been isolated from avian species. To date only viruses of H5 and H7 subtype have been shown to cause HPAI in susceptible species, but not all H5 and H7 viruses are virulent.

8.1.3. Epidemiology

8.1.3.1.World distribution

AI viruses are distributed throughout the world in many domestic birds, including turkeys, chickens, guine fowl, chukars, quail, pheasants, geese, ducks, and in wild species.

The AI viruses are often recovered from apperantly healthy migratory waterfowl, shore birds, and sea birds worldwide.

HPAI A viruses of the H5 and H7 subtypes have been isolated occasionally from free-living birds in Europe and elsewhere. Outbreaks due to HPAI were recorded in the Pennsylvania area, USA, in the years 1983-84. More recently outbreaks with HPAI have occurred in Australia *(H7),* Pakistan *(H7N3),* Mexico *(H5),* For detailed information on occurrence, see recent issues of World Animal Health and the OIE bulletin.

8.1.3.2. Recent Situation in Tanzania

Tanzania is free of this disease

8.1.3.3. Risk assessment of AI virus introduction into TANZANIA

- Legal or illegal imports of infected live poultry or products, from countries where AI infection is problem
- Infected birds movements
- Live bird markets
- Migratory routes
- Wild life
- Using untreated water from dams or creeks that have been contaminated with waterfowl faeces
- Village animals

8.1.3.4.Epidemiological remarks

8.1.3.4.1. Susceptible Species

AI virus is infective for almost all commercial, domestic and wild avian species. Infections in monkeys, pigs, ferrets, horses, cattle, seals and whales have been reported; the occurrence of a strain of virus lethal for humans that originated in chickens was reported from Hong Kong in 1997.

8.1.3.4.2. Virus Survival

8.1.3.4.2.1. In the environment: Influenza viruses can survive for a long periods of time in the environment, particularly under cool and moist conditions. Infectivity was retained in faecal material for as long as 30-35 days at 4 ^{0}C and for 7 days at 20 ^{0}C. Influenza viruses have been recovered from lake and pond water where there were large concentrations of waterfowl.

8.1.3.4.2.2. In the host (including pathogenesis of the diseases*)*: AI viruses exhibit a wide range of virulence, for chickens as well as other species. HPAI is a highly contagious, generalised viral disease that may cause high mortality in gallinaceous species of birds, in association with respiratory, gastrointestinal and/ or nervous signs. In other avian species, HPAI virus infection may range clinically from an in apparent to a highly lethal disease. Wild aquatic birds, such as waterfowl and seabirds, are recognised as important reservoirs for AI virus but rarely display clinical signs of infection.
Clinical signs result from the replication of virus in the respiratory tract and subsequent systemic replication in the visceral organs and brain. The viruses that are non-pathogenic replicate only on the surfaces of the respiratory and intestinal tracts.

8.1.3.4.2.3. Carcasses: AI virus survives for only several days in carcases at ambient temperatures, compared with up to 23 days at refrigeration temperatures. Birds' processes during the viraemic stage will contaminate other carcases with blood or faecal material containing virus. Packaging and the drips that develop during storage are also important as both can be contaminated with virus from infected carcases.

8.1.3.4.2.4. Meat products: Virus can persist in poultry meat products. For cooked chicken the inactivation times were found to vary between virus strains. The agreed minimum core temperatures for cooked poultry meat are considered sufficient to kill AI viruses;

- 70 ^{0}C for a minimum of 30 minutes;
- 75 ^{0}C for a minimum of 5 minutes or,
- 80 ^{0}C for a minimum of 1 minute

8.1.3.4.2.5. Table eggs and egg products*:* Although severely affected birds will stop laying, eggs laid in the early phase of the outbreak could contain AI virus in the albumen and yolk and / or on the surface. The virus can penetrate cracked or intact shells or, more significantly, contaminate the eggs fillers. The survival time on the eggs and fillers is sufficient to allow wide dissemination. Egg pulp products are another source of the virus.

8.1.3.4.2.6. Fertile eggs: AI virus has been isolated from eggs laid by infected breeding hens.

8.1.3.4.2.7. Poultry by-products*:* Poultry offal meal and pet foods are usually cooked at above 100 ^{0}C for several minutes to more than one hour, which is sufficient to kill AI virus. However, if the procedure is not carried out properly or cooked product is subsequently contaminated by unprocessed product, AI virus could persist in the by-product for several weeks.

8.1.3.4.2.8. Waste products AI virus has the potential to persist in waste products and could be disseminated by vehicles that transports them unless the products are treated before movement.

8.1.4. Disease transmission

Not all strains of AI viruses are highly transmissible for poultry; highly and lowly virulent viruses can have low transmissibility but, following passage through flocks, transmissibility as well as pathogenicity for the host can increase in the field.

8.1.4.1. Wild birds: Direct or indirect contact with migratory waterfowl is the most likely source of infection in poultry.

8.1.4.2. Live poultry*:* Transmissibility in poultry varies enourmously between AI virus strains. Contact with faeces or respiratory secretions is important while airborne spread is not considered significant. Field outbreaks are further complicated by having to distinguish between direct transmission and that of secondary spread by people and fomites.

8.1.4.3. Eggs: Vertical transmission via infected eggs has never been proved although AI virus has been detected on the shell surface and in the yolk and albumen of eggs, suggesting that the potential for spread exists. Optimal incubation temperatures of 37.2 - 37.7 ^{0}C in the early stages of embryo development may be lethal to AI virus, or infected embryos may be killed by the virus early during incubation. Persistance through the incubation process is most likely through shell contamination.

8.1.4.4. Fomites: "Persistence of the virus in faeces and respiratory secretions is of major importance and their sticky nature facilitates spread over a wide geographical area on footwear, clothing, equipment and other fomites. This comprises the major means of transmitting infection between premises.

8.1.4.5. People: The actions of humans in moving feedstuff, personnel, equipment and vehicles into and from premises that are contaminated with infected faeces or respiratory secretions.

8.1.4.6. Other vectors*:* There is no evidence to suggest that invertebrates are involved in the interepizootic maintenance of transmission. However, there is a possibility of mechanical transmission by either invertebrate or vertebrate vectors, although its occurance will be low priority.

8.1.5. Diseases Patterns

To introduction of the virus to previously free flocks, areas or countries is likely to lead to a very rapidly spreading epidemic with high morbidity and mortality rates.

8.1.5.1.Consequences

8.1.5.1.1. Public Health

H5N1 HPAI is a zoonotic disease. Humans may be infected via close contact with infected birds and by working in an environment that is heavily contaminated with HPAI viruses (e.g.people working in the slaughter of infected poultry or clean-up of infected premises). Infection has not been transmitted via handling or consumption of poultry products (meat and eggs). There is no evidence of sustained transmission of this virus between people.

8.1.5.1.2. Socio-economic

The main losses would be mortalities, which can be high, and losses due to decreased egg and meat production and reduced productivity. There would be further loss of income for an extended period due to the stamping out policy. The disruption to the flow of product and decreased consumption and production may cause job losses on farms, and in service and associated industries, depending on the time it takes to bring the outbreak under control. Even a small outbreak would result in dislocation of the industry and its normal marketing patterns. Infection in grandparent and foundation flocks would cause loss of some valuable genetic material.The eradication strategy and the movement controls that will need to be imposed and rigorously enforced if a zoning policy is to be pursued will likely result in severe disruption to many industry practices including breeding programs and sales of eggs, chicks, poults, pullets, turkeys and meat birds. Any delays beyond the marketing age of the various commodities can cause major increased production costs and losses over a short period, and affect the producers not directly involved in the outbreak through loss and disruption of supply.
Pet shops, aviaries and bird dealers may also be affected through the movement controls.

8.2. DIAGNOSIS

8.2.1. Clinical Signs

The incubation period is 3-5 days. Clinical findings; severe depression, inappetence, drastic decline in egg production, facial oedema with swollen and cyanotic combs and wattles, petechial haemorrhages on internal membrane surfaces, sudden deaths.

8.2.2. Incubation period

Incubation period is extremely variable for HPAI, from a few hours to 3 - 14 days. The OIE Code gives a maximum incubation period, for regulatory purposes, of 21 days

8.2.3. Clinical diagnosis

8.2.3.1. Symptoms

- Severe depression, in appetence
- Drastic decline in egg production
- Facial oedema with swollen and cyanotic combs and wattles
- Petechial haemorrhages on internal membrane surfaces
- Sudden deaths (mortality can reach 100%)

8.2.3.1.2. Lesions

Chickens

- Lesions may be absent in cases of sudden death
- Severe congestion of the musculature
- Dehydration
- Subcutaneous oedema of the head and neck area
- Nasal and oral cavity discharge
- Severe congestion of conjunctivae, sometimes with petechial

- Excessive mucous exudates in the lumen of the trachea, or severe haemorrhagic tracheitis
- Petechial on the inside of the sternum, on the serosa and abdominal fat, serosal surfaces and in the body cavity
- Severe kidney congestion, sometimes with urate deposits in the tubules
- Haemorrhages and degeneration of the ovary
- Haemorrhages on the mucosal surface of the proventriculus, particularly at the juncture with the gizzard
- Haemorrhages and erosions of the gizzard lining
- Haemorrhagic foci on the lymphoid tissues in the intestinal mucosa

8.2.4. Pathology

8.2.4.1. Gross lesions
In many cases, poultry dying from the peracute form of the disease lack visible gross lesions; such chickens die on day 1 or day 2 after infection.

With the acute form of infection, seen days 3 to 5 after inoculation, more diverse visible lesions are seen. Chickens have ruffled feathers, 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 gizzard.
In infections such as mildly pathogenic AI, lesions may be seen in the sinuses characterised by catarrhal, serofibrinous, mucopurulent or caseous inflammation. The tracheal mucosa may be oedematous with an exudate varying from serous to caseous. The air sacs may be thickened and have a fibrinous to caseous exudate. 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 Tanzanias. Exudates may be seen in the oviducts of laying birds.

8.2.4.2. Microscopic lesions (histopathology)
The 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.

8.2.5. Laboratory diagnosis

8.2.5.1. Specimens required / transport : Samples should be taken both from live, clinically-affected birds and from recently dead birds. Cloacal and tracheal swabs and/or fresh faeces and serum should be taken from live birds. From dead birds, alimentary tract tissues (proventriculus, intestine, caecal tonsil) and respiratory tract tissues (trachea, lung) should be collected. For details of sample collection, transport, storage and processing, see Annex 4

8.2.5.2. Laboratory tests: As pathological changes are not definitive for the disease, diagnosis needs to be confirmed by the isolation and characterisation of the causative virus. Bacteriology should be performed to

exclude bacterial septicaemias from the differential diagnosis, particularly to identify mixed infections with mildly pathogenic forms of AI.

Specimens should initially be sent to the VIC from where they will be forwarded to the NVRL for further characterization. Final identification is most commonly accomplished by one of the International AI reference laboratories or OIE influenza reference laboratories.

8.2.5.2.1. Virus isolation and identification*:* Methods for the isolation and identification of influenza viruses have been described in detail in OIE Manual for Diagnostic Test.

8.2.5.2.2. Tests for virus subtype *:* Influenza viruses can be typed as either A, B or C, but all AI viruses isolated from poultry have been type A. AI viruses are further subtyped on the basis of their haemagglutinin (H) and neuraminidase (N) structure.

8.2.5.2.3. Tests for pathogenicity*:* The virulence of an influenza virus isolated from a bird can be determined by any one or more of the following tests.

8.2.5.2.3.1.Chicken pathogenicity tests. A solution of virus in bacteria-free allantoic fluid is inoculated intravenously into eight 4–6-week-old specific pathogen free (SPF) chickens. If six or more chickens die within 10 days, the virus is considered to be highly pathogenic for chickens. Methods for the pathogenicity tests of influenza viruses have been described in detail in the OIE Manual of Diagnostic Tests.

8.2.5.2.3.2. Plaque test. Virus is added to cell cultures in the presence or absence of trypsin. If plaques form in the absence of trypsin and the isolate kills 1–5 chicks in the chicken pathogenicity test, the virus can be considered to be highly pathogenic for chickens. Methods for the plaque test of influenza viruses have been described in detail in the OIE Manual of Diagnostic Tests

8.2.5.2.3.3. Molecular pathotyping : The gene encoding the haemagglutinin protein of the virus at the cleavage site is sequenced. There are well-recognised differences between the gene sequence of high, mild, low and nonvirulent viruses. So far, only viruses of H5 and H7 subtype have been isolated from chickens with HPAI. As time is of the essence for diagnosis, molecular pathotyping is the preferred method of determining the pathogenicity of an AI virus. Once an outbreak virus has been characterised, immunohistochemistry, immunofluorescence and virus isolation can be used to confirm virulent infections. Molecular pathotyping should be performed on all AI viruses isolated from domestic poultry to ascertain their potential to develop virulence by mutation.

8.2.5.2.3.4. Serology : Evidence of previous AI infection can be obtained by testing for influenza A group specific antibodies using an agar gel immunodiffusion test (AGID) or enzyme-linked imunosorbent assay (ELISA), or subtype-specific antibodies to the haemagglutinin or neuraminidase antigens using a hemaglutination inhibition (HI), test or ELISA, respectively. Methods for the serologic tests which is used of influenza viruses antibody detecting have been described in detail in the OIE Manual of Diagnostic Tests

8.2.6. Differential diagnosis

AI infection of chickens and turkeys with various levels of pathogenicity are frequently indistinguishable on clinical and postmortem examination from:

- Newcastle disease virus,
- Avian pneumovirus and other paramyxoviruses,

- Infectious bronchitis,
- Infectious laryngotracheitis,
- Fowl cholera,
- *Escherichia coli* cellulitis of the head,
- Acute pasteurellosis,
- Infectious coryza,
- Chlamydia,
- Acute poisoning,
- misadventure causing high mortality (eg smothering, heat stress, dehydration)

Table 1: Diagnostic tests currently available for AI

Test	Specimen required	Test detects	T ime taken to obtain result
Immunofluorescence	fresh tissue (pancreas)	viral antigen	4 hours
Immunohistochemistry	formalin fixed tissues	viral antigen	2 days
Virus isolation	tissues	Virus	2–10 days
Virus identification by HI, EM/immune EM, neuraminidase test	virus isolate	Specific antigens	4 days
Serology on flocks and surrounding flocks: HI subtype specific, ELISA/ AGID groupspecific	serum	Antibody	1 day
Pathogenicity tests: bird challenge	virus isolate	Pathogen and pathogenicity of virus	2–8 days
PCR/gene sequencing	virus isolate	Virulence markers in RNA genome	2-3 days

8.3. MANAGEMENT OF OUTBREAK

8.3.1. Responsibility

As soon as the suspicion of AI is reported, the District Veterinari Officer (DVO) identifies the person who has reported the suspicion, and if the latter is the farmer, the DVO collects information concerning:
a) location, characteristics and number of birds and other animals on the farm,
b) presence of staff and vehicles,
c) recent movement of people, equipment, vehicles and animals,
d) the availability, on site of disinfectants and equipment for disinfecting premises.
Reporting the suspicion is also compulsory for the company veterinarian or private veterinarian, who must support the DVO in collecting information. If the suspicion is reported before the arrival of the DVO, the private veterinarian or practitioner must do everything in their power to prevent the infection from spreading. The vehicles

of the DVO and company/field veterinarian must be left outside the infected premises, and at a distance from the entrance of the farm.

The DVO coordinates at the farm level, in order to avoid movement of people, animals, equipment and vehicles from the suspected premises and in the meantime:

- Informs the Regional Livestock Advisor of the suspicion of AI,
- Provides himself with a kit No 1 (List of the equipments is given in Annex 8
- Informs the Director of Veterinary Services (DVS), and identifies the closest mobile disinfection unit, indicating the suspicion of AI,

8.3.2 Access

Access to the premises must take place following a complete change in clothing. Disposable gear, including head caps and shoe covers must be worn by all staff entering the farm. A changing room must be identified, and it should contain large plastic bags, cardboard boxes, latex gloves and a sufficient quantity of disinfecting solution. The remaining components of kit no: 1 are to be used inside the poultry house/shed.

The initial coordination tasks of the OV are to:

- Obtain a written declaration from the personnel on the farm stating that they will not visit any establishment containing live birds for 3 days; the OV and any other veterinarian must also comply to this general rule,
- Identify locations on the farm where vehicles leaving the farm can be properly washed and disinfected, and organize washing and disinfection procedures,
- Identify sites where staff may wash and disinfect and ensure that on leaving the premises, all staff leave their disposable gear inside the changing room, wash and disinfect exposed body parts and shoes and agree to wash their clothing as soon as they return home.

Washing and disinfecting of vehicles must take place internally and externally, and vehicles may leave the infected premises only if this is absolutely necessary. Care must be taken to avoid that contamination of natural or artificial water reservoirs may occur.

The VIC veterinarian should reach the premises equipped with kit no 2, accompanied by a driver, who must remain outside the premises and is responsible of the dispatch of the pathological samples to the laboratory. The Veterinarian must wear his protective gear in the changing room, and must leave the following items from kit no 2 in the changing room:

- The leak-proof water resistant container,
- The thermic container for carrying samples,
- Two pairs of latex gloves,
- Five autoclavable plastic bags,
- Five black rubbish bags,
- Disinfecting solution.

The remaining components of kit no 2 must be carried inside the house/shed (List of the equipments is given in Annex 9)

8.3.3. Preliminary investigation

The DVO and VIC collect the following information:

- Preliminary identification of the productive unit and subunits (topography of the farm), and identification of the unit for which the suspicion has been reported,
- Identification of staff directly involved with that unit,
- Anamnestic data.

8.3.3.1. Clinical investigation

The aim of the clinical investigation is to establish the clinical situation on the farm, including ill and suspect birds. The clinical investigation must be performed on all susceptible species present on the farm, and it must begin from the most peripheral units. Particular attention must be paid to any vaccinations performed. All this

information must be reported in the epidemiological inquiry. All the birds present per species must be identified, and for each species identified, a report containing the date of onset of clinical signs, description of clinical signs and reported percentage mortality must be prepared.

8.3.4. Collection of pathological samples

In cases of suspected AI the following pathological samples must be collected and sent to the laboratory:
- At least 5 moribund birds (for post mortem examination),
- Pooled tracheal and lung samples from at least 5 moribund birds,
- Pooled intestine samples from at least 5 moribund birds,
- Cloacal and tracheal swabs from healthy birds (also from waterfowl and ratites),
- At least 20 blood samples (acute sera).

Samples from different apparatuses must not be pooled. They must be packaged appropriately (in leak proof containers, wrapped in at least two plastic bags), to avoid dissemination of the infectious agent, and transported refrigerated to the laboratory. Sacrificed animals may be transported in a sealed autoclavable plastic bag, inserted inside a similar, sealed bag. All samples must be carried to the laboratory inside a polystyrene box containing icepacks. The polystyrene box must be appropriately disinfected before leaving the premises. The samples must be accompanied by the sample form.
The driver in charge of delivering the samples, must drive directly to the laboratory without any intermediate stops.

8.3.5. Epidemiological inquiry

The DVO and VIC are requested to carefully fill in the epidemiological inquiry form .With reference to the epidemiological inquiry, it is important that:
- Animal movements: animal movements should be recorded up to 20 days prior to the onset of the first clinical signs,
- People movements: all people (staff, relatives, servicing personnel, veterinarians) who had access to the farm must be recorded,
- Vehicle movements: all vehicles, regardless of their contact with animals, which have had access to the farm must be reported.

The epidemiological inquiry must be sent (possibly faxed) to the competent authorities as soon as it has been completed.

8.3.6 Exit

Following the clinical visit and the collection of samples the DVO and VIC, in the designated changing room, disinfect their protective gear and collect all sterilizable equipment in an autoclavable bag, which is sealed and inserted into a second bag, which is disinfected externally. All single use materials, sheets of paper, disposable gear, shoe-covers are put inside a plastic bag which is left on site.

8.3.7. Equipment

Equipment lists for the DVO and VIC are given in Annex 8 & 9

8.3.8. Management of Diagnosis

Any suspicion of AI must be reported to the VICs that will take action to collect convenient samples from the suspicion animals. During the taking samples it must be ensured to be correct sampling. The VIC must sent

samples immediately in suitable condition the NVRL companied with sample form described as sample transport article in Laboratory Contingency Planning. It must be ensured that the sample form is full-filled with necessary information and transportation must be the fastest route so that sample is delivered to laboratory as soon as possible the quickest way.

The VIC Officer in Charge must notify the NVRL about sample transportation timeline and way in order for the laboratory to prepare immediately extra-ordinary diagnosis action and also notify to related key staff at ADRI who can coordinate to gain speed diagnosis facilities and also can prepare in advance actions.

8.3.9. Epidemiology

8.3.9.1. Establishment of Expert Group

A small team of senior civil servants will be established in the early stages of an outbreak. Their remit will be to quality assure local structures and processes, and the capture of critical management information in LADECCs.

Head of the NADECC or LADECC, with together, must ensure to establish investigation teams or person in different category depending on purpose of outbreak tracing; such as initial investigation team, clinical inspection team, zone inspection and control team and the other actions of the related measures.

8.3.9.2. Tracing Back and Forward

8.3.9.2.1. Priorities of Tracing

Investigation team shall ensure that epidemiological investigation in relation to outbreaks of AI disease are carried out by specifically trained veterinarians on the basis of questionnaires, prepared within the framework of this contingency plans, to ensure standardised, speedy and targeted inquiries. It must be ensure that investigation would be planed precisely and determined its purpose in order to define strategy for combat disease and finally eradication in sort terms. For this reason, tracings are purposed as follows:

1. The origin of infection;
2. The date of introduction of the infectious agent
3. The possible spread of disease

During the planning of investigation, it must be taken into account the some priorities; it is the most important early in series of outbreaks to identify index case and find undiscovered cases e.g. other locations infected from the same source for tracing to source (tracing back) of disease. And also it still can be important later on to understand means of spread, so that appropriate control measures can be taken. It is the most important element as a priority that veterinary judgement on the most likely source of infection such as what moved on and when did it move on?

Urgent and meticulous trace-back and trace-forward of all contacts with infected animals and premises will be vital if the disease is to be effectively contained. It will be ensure that this investigation are done from 14 days before the appearance of the first lesion, until the date of taking control measures which bans all movements on and off the premises. The most urgent spread tracings are those from the time of the appearance of the first lesion onwards when virus shedding is likely to be highest.

Within these windows, tracings are prioritised as follows:

1. Poultry; Any poultry or the other animals (livestock) contact is high priority, a 'hot' tracing; all animals should be traced until found, and slaughtered as dangerous contacts.

2. Poultry vehicles, people; these should be traced two days before and after the date of contact with the IP; contact with livestock increases the priority.

3. Miscellaneous; other contacts such as chick, egg and feed Lorries. These should be traced at least a day before and after the date of contact with the Infected Premises (IP). Chick, egg and feed Lorries tracings might be extended to two days before and after contact if the degree of contamination is deemed to be high.

4. Circumstances may warrant variation of the tracing windows and/or the prioritisation of tracings within them: consideration must be given to the number and species of animal affected their location/distribution on the IP, the length of time over which virus may have been excreted, and the level of biosecurity on the farm.
5. Serological parameters may in some cases be deemed to be more reliable than lesion ageing in determining the likely date of introduction of infection to poultry flocks

8.3.9.2.2. Tools of Tracing

Scientists, epidemiologists and other investigation team members involved LADECC can decide which tracing tools are convenient to be used collection of epidemiological information; then they immediately will start actions to be done for rapid, successful and effective investigation. Some points regarding this purpose must be taken into account to determine convenient investigation methods and tools which are as follow:

8.3.9.2.2.1. Census

To define and establish the most convenient strategy, the team will need to be done census of whole animals which showed detailed of related whole population and animals products. Census details must be done as follow:

- a census is made of all categories of animals on the holding and that, in respect of each category of animals of susceptible species, the number of animals that are already dead and the animals suspected of being infected or of being contaminated, is recorded
- the census as referred to in point first paragraph in this subject is kept up to date to take account of those animals of susceptible species born or dying during the period of suspicion and such information is produced by the owner on request of the competent authority and is checked by that authority at each visit;
- all stocks of egg, meat, meat products, carcasses, slurry, manure as well as animal feed and litter on the holding are recorded and those records are maintained;
- no animals of susceptible species enter or leave the holding;

8.3.9.2.2.2. Checklist for gathering information

Using the checklist associated with epidemiological investigation and tracing can be more convenient and practical in order to gather data for determining disease extend and pattern. During the collection of information, some important points must be taken into account; such as starting with general questions then probing; using timelines and map; using several sources and following important hints respectively; talking to groups and individuals. These lists can form the basis of interviews with key informants and also the basis of data forms or report formats. However the best format can be developed into good interview prompt lists.

8.3.9.2.2.3. Tracing of Slaughterhouses and Animal Markets

Authorities of LADECC shall ensure that all slaughterhouses and animal markets within and around PZ and SZ shall traced backed and forwarded to obtained sources of spreading infection in terms of 15 day before defined disease and until the declaration of free of disease. Investigation team must ensure that it has carried out a full and fair inquiry assessing records of slaughterhouses and animal markets to develop logical information. Investigation team shall observed animals movement in and off on the slaughterhouses and animals markets.

8.3.9.2.2.4. Inspection and Clinical Surveillance and Sero-surveillance in and around IP and in PZ and SZ

Surveillance will be purposed for:

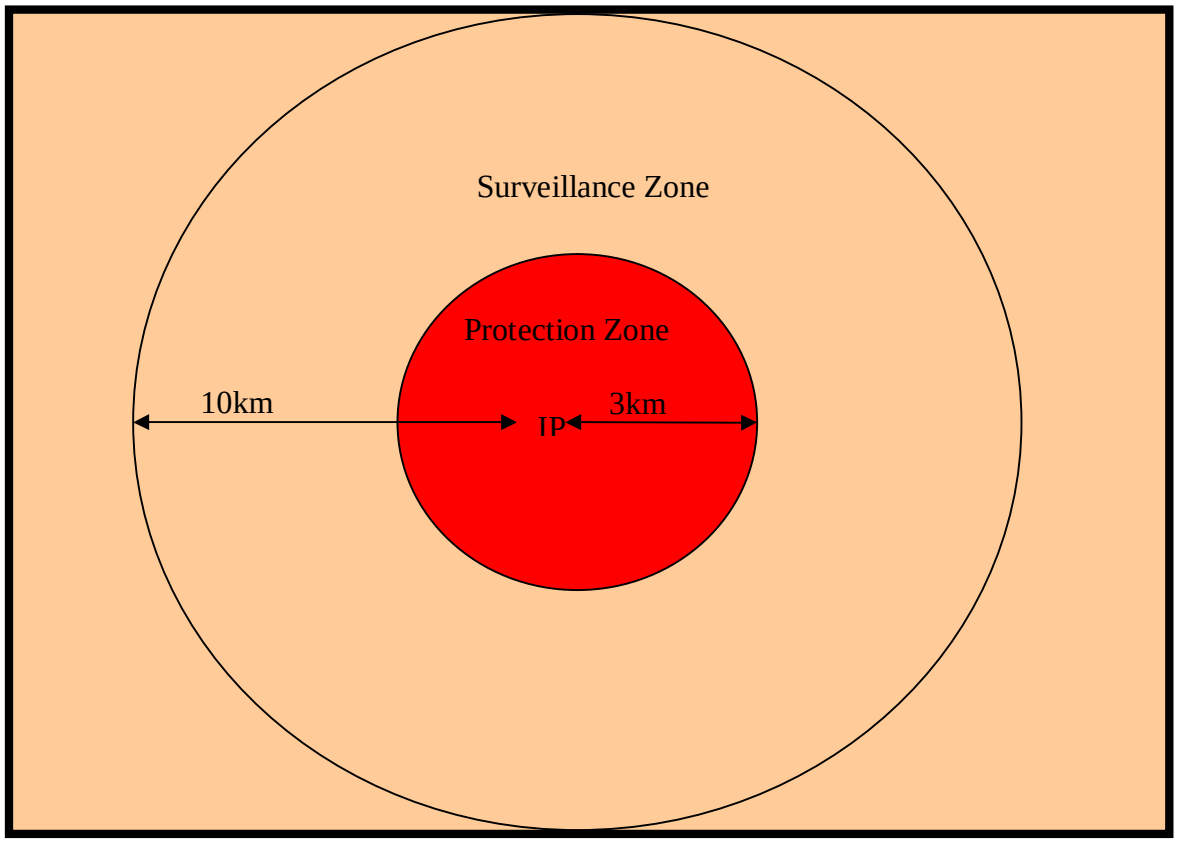
· defining the extent and pattern of the disease in PZ and SZ;
· detecting new outbreaks in PZ and SZ; and
· establishing disease-free zones.

The epidemiological investigation team shall establish a PZ based on a minimum radius of 3 km and a SZ based on a minimum radius of 10 km centred on the outbreak of AI disease. The geographical delimitation of those zones shall take account of administrative boundaries, natural barriers, supervision facilities and technological progress which makes it possible to predict the probable dispersion of the AI disease virus by air or any other means. That delimitation shall be reviewed, if necessary, in the light of such elements.

The PZ and SZ shall be marked by posting signs of sufficient size on roads entering the zones.

8.3.9.2.2.5. Diagram Showing PZ and SZ

These terms are in line with the OIE Terrestrial Animal Health Code



Within PZ described first paragraph, all holdings with animals of susceptible species shall periodically undergo a veterinary inspection, carried out in such a way as to avoid the spread of AI disease virus possibly present on the holdings, which shall recorded in special the relevant documentation.

Holdings must undergo clinical examinations of all animals of susceptible species for signs or symptoms of AI disease.

Special emphasis must be laid on animals which may have been exposed to AI disease virus with a high probability, notably transport from holdings at risk or close contact to persons or equipment that had close contact to holdings at risk.

Veterinary Inspectors shall usually target their search on the places where they think it most likely to find disease; targeted on animals likely to be infected

- traced contacts
- animals in proximity (PZ)

This can be done most likely to involve repeated clinical examinations, which is 'active surveillance' because it is actively done searching for disease; not waiting for disease to be reported by others.
Surveillance will be most intense in the Restriction zone (RZ) and will be driven by findings from the veterinary investigations unit. The intervals between inspections and surveys will depend on the observed incubation period, the resources available and the level of exposure risk. Suspect premises should be inspected at least every third day. Every effort must be made to educate producers about the clinical signs and to report lesions

Animals in SZ described first paragraph will be subjected to regular inspection and observation over an agreed period of at least 21 days. Tracing and surveillance will play an important role in identifying infected and in-contact animals to determine the extent of the RZ and free areas. During the regular inspection shall be taken care attention biosecurity for all action which decontamination will be used to limit spread of the virus.
Animal products and substances which are all stocks of poultry meat or carcases, or animal feed, implements, waste, manure, litter or anything liable to transmit AI be subject to authorization by the competent authority holding within SZ shall considered during the regular inspection; so that to prevent contact risk from any sources of contamination.

8.3.10. Serological Surveillance

In addition to the active clinical survey, sero-surveillance (serological) is also a suitable tool for tracing of outbreak, which is not only done to evidence for free of disease status post outbreak, but also it is done to define uncertain disease and dangerous contacts bring to low pathogenic infections.
During the outbreak investigation, sero-surveillance might be used to define infection in disease contact premises (DCPs) and suspected premises (SPs) using correct serum sampling. And also it is very useful especially of species which show clinical signs less overtly; such as duck.
Post-outbreak surveillance will be done to establish that all infection has been eradicated to evidence for free of disease.

Serological sampling shall be carried out:

a. according to the recommendations of the epidemiological team established within the expert group referred to in this Contingency Plan and

b. in support of tracing and the provision of evidence for the absence of previous infection.

c. Where sampling is carried out in the framework of disease surveillance after an outbreak, actions shall not commence before at least 30 days have elapsed since the elimination of susceptible animals on the infected holding(s) and the carrying out of preliminary cleaning and disinfecting.

d. Sampling of animals of susceptible species shall be carried out in according to the recommendations of the epidemiological team in each case where chick, poultry, duck, and goose, or other susceptible animals not displaying clear clinical signs are involved in the outbreak, and in particular where such animals have been isolated from infected birds.

8.3.10.1. Sampling on holdings

In holdings where the presence of AI disease is suspected but in the absence of clinical signs, duck and goose, and on recommendation of the epidemiological team other susceptible species, should be examined in accordance with a sampling protocol suitable to detect 5% prevalence with at least 95% level of confidence.

8.3.10.1.1. Sampling in PZ

All holdings within the perimeters of the PZ where poultry and the other birds have not been in direct and close contact with animals during a period of at least 21 days prior to taking the samples shall be examined in accordance with a sampling protocol suitable to detect 5% prevalence of disease with at least 95% level of confidence.

However, the competent authorities may decide where epidemiological circumstances allow and in particular in application of the measures that samples are taken not earlier than 15 days after the elimination of susceptible animals on the infected holding(s) and the carrying out of preliminary cleaning and disinfecting, under the condition that the sampling is carried out using statistical parameters to detect 2% prevalence of disease within the flock with at least 95% level of confidence.

8.3.10.1.2. Sampling in SZ

Holdings within the perimeters of the SZ where the presence of AI disease in the absence of clinical signs must be suspected, notably where duck, goose and the other birds are kept, shall be examined. For the purpose of this survey the model of a multistage sampling shall be sufficient, provided that those samples are taken:

From holdings in all administrative units within the perimeter of the zone where duck, goose and the other birds have not been in direct and close contact with bovine animals during a period of at least 30 days prior to taking the samples, and from as many holdings referred to above as necessary to detect with at least 95% level of confidence at least 1 infected holding if the estimated prevalence of the disease was 2% equally distributed throughout the zone, and from as many duck, goose and the other birds per holding as necessary to detect 5% prevalence of disease within the flock with at least 95% level of confidence.

8.3.11. Special Measures

8.3.11. 1. Restriction Zones (RZ)

The Chair person of LADECC will ensure that effective quarantine and movement controls are essential in the RZ. Movement controls increase the speed and likelihood of successful eradication by helping to prevent further spread of virus. Quarantine and movement controls should be imposed at several levels based on declared areas. Initially, the PZ and SZ referred at article 8.3.3.2.1 (a) in this Contingency Plan or the whole region where occurred outbreak depend on the situation related risk of spreading disease should be declared as the RZ, subject to action to be taken control measures and movement restrictions. These will be reviewed once the situation has been fully assessed.

Movement controls should be maintained to some degree until the disease is either eradicated or declared endemic. All IPs, DCPs and SPs will be quarantined with no movement into or out of the IPs and DCPs or the SPs while strict surveillance and inspections are being undertaken. Quarantine and movement controls will be imposed on appropriate risk enterprises to ensure any product from infected or in contact birds is disposed of and suspect product is detained.

Authorities shall ensure that there is no illegally transportation and trading on holdings off and in RZ; to inquiry this situation they can ask help to police and armed force.

People who exit from RZ particularly those who have been on or close to IPs, DCPs and SPs should avoid contact with livestock for a period of three days in order to prevent the risk of mechanical spread of virus.

Effluent of disease should be prevented from draining onto roads, stock routes, pastures, or into creeks and other watercourses.

Authorities shall ensure that all bird markets are closed in restricted zone and in any circumstances place where birds trading may contact infection must be closed and disinfected.

Birds of susceptible species may be transported directly under official supervision for the purpose of emergency slaughter to a slaughterhouse situated inside the same protection zone or, if that zone has no slaughterhouse to a slaughterhouse outside the zone designated by the competent authority in means of transport cleaned and disinfected under official control after each transport operation.

By way of derogation from risk assessments of LADECC authorities shall decided for transportation of birds to slaughterhouses subject to the following conditions:

a. the records referred to in Article 8.3.3.2.1 (a) have been subjected to official control, and the epidemiological situation of the holding does not indicate any suspect of infection or contamination with the AI virus, and

b. all the birds of susceptible species on the holding have been subjected with negative result to an inspection by the OV, and

c. a representative number of birds, taking into account the statistical parameters has been subjected to thorough clinical examination to rule out the presence or suspect of clinically infected birds, and

d. the slaughterhouse is designated by the competent authority and located as near to the SZ as possible.

8.3.12. Stamping Out

Stamping out is the strategy that will be implemented on all IPs and DCPs; birds on SPs will be subjected to regular inspection and observation over an agreed period of at least 21 days. Tracing and surveillance will play an important role in identifying infected and in-contact birds to determine the extent of the RZ and free areas.
Stamping out can only be achieved in association with other methods of control but these will be used to improve the effectiveness of the strategy and to ensure that infected stock are contained and destroyed and that unnecessary slaughter of birds does not occur. Birds that are considered to be most infective or at risk should be given priority in the destruction process. Taking consideration priorities of species susceptibility and more virus excreted species; authorities must give priority first more susceptible and excreted species then later the others; e.g. first poultry and later the other animals.

8.3.13. Cleaning and Disinfection

Contacts with infected birds and with their excrement also expose significant risks. Clothes, boots, vehicles and equipment can become contaminated and can carry disease from one premise to another. Diseases can also be spread by other means, such as wildlife, air or other vectors.
Authorities must take measures which will minimise the spread of diseases between different premises by contaminated clothes, boots, vehicles and equipment. If direct contact with farm birds cannot be prevented then it is best practice to clean and disinfect protective clothing, footwear, equipment, vehicles etc. before and after the contact with the birds, or use disposable protective clothing.

The owner or occupier of any premises where birds are kept shall maintain a footbath containing an approved disinfectant in some convenient place at every exit from those premises and renew the disinfectant as frequently as is necessary to maintain a clean solution and if so directed by an inspector.
Authorities of LADECC shall maintained that appropriate means of disinfection are used at the entrances and exits of buildings or places housing birds of susceptible species and of the holding itself.

No person shall enter or leave any livestock premises wearing clothing or boots which are visibly contaminated with mud, slurry, animal faeces, droppings or excretions or any other similar matter or without cleaning and disinfecting the outer surfaces of their footwear on entering or leaving those premises.

8.3.14. Transport Means

Any vehicle or trailer entering or leaving a premises must be cleaned and disinfected on the outside and underside [and include the tyres (including the whole circumference of their treads), wheel arches, mudguards and mud flaps of the vehicle]. Any parts of the vehicle or trailer where farm birds have come in contact must also be cleaned and disinfected. All visible traces of mud, slurry, animal faeces, droppings or excretions or other similar matter must be removed, including any inside the vehicle. This cleaning and disinfection also must be subjected at the entrance and exit of RZ.

Any equipment and materials must be subjected same cleaning and disinfection treatments at the entrance and exit of RZ.

9. EMERGENCY VACCINATION

The existance of a large number of virus subtypes together with the known variation of different strains within a subtype pose serious problems when selecting strains to produce influenza vaccines and to use vaccination as a routine tool for diseases prevention.
Vaccines also prevent development of the clinical symptoms but can not prevent emergence of the infection. In general, when animals carrying strains of low pathogenicity are vaccinated, the probability that these strains transform into highly pathogenic strains through mutation exists. The vaccinated animals continue spreading the virulent virus and at the same time hinder serological control.
In accordance with the OIE/FAO recommendations on AI outbreak containment, vaccination against AI may be used to supplement the control measures carried out after confirmation of diseases.

9.1. Vaccination Legal possibilities

According to the recommendation of the Expert Group , AI vaccinations are currently not allowed in Tanzania. The DVS determines if and with what vaccine any emergency or ring vaccination programme is to be undertaken. The decision to introduce vaccination to supplement control measures shall be taken by the DVS in consultation with the Director of the NVRL (ADRI)
This decision shall have particular regard to:
— the concentration of poultry in the affected area,
— the characteristics and composition of the vaccine to be used,
— the procedures for supervision of the distribution, storage and use of vaccines,
— the species and categories of poultry which shall be subject to vaccination,
— the areas in which vaccination shall be carried out.

9.2. Vaccine Stocks

There is no AI vaccine production in Tanzania. At this point there are no stocks of vaccine kept for emergency or ring vaccination. In case of an emergency situation registered vaccines may be used, allowed by the DVS

9.3. Vaccine Distribution

Given that it is not permitted there are no arrangements at present for distribution of emergency vaccine. If necessary, the DVS can set up this distribution within a few days.

9.4. Vaccine Administration

Vaccination is done exclusively by veterinarians.

10. Training of Staff

10.1. Training Needs

The staff should be regularly trained in procedures for diagnosing and dealing with AI.
The training programmes include;
• The clinical diagnosis of AI

• Procedures at infected premises and within PZ and SZ (sanitation and disposal)
- Epidemiological inquiries
- Diagnosis of AI
  - Procedures at LADECCs
  - Procedures at NADECC
  - Tracing exercises, record keeping
  - Notification and publicity procedures.

10.2. Training of different groups
- NADECC personnel
- LADECC personnel
- NVRL personnel
- VICs
- Expert group
- Field DVOs
- Private and company veterinarians
- Owners
- Farmers
- Salesman
- Police, gendarme and municipality guards

10.3. Implementation of training programme
- Theoretical
- Practical

A summary of the disease (pictures and video of clinical signs and example of how health and production records would change in flocks infected with AI virus), daily workshops, courses, etc

10.4. Time table of training staff
Special training programs on AI Contingency Plan and the dates will be arranged and announced when funds for the contigency plan are availed.

10.5. Resposibility for this program
DVS is responsible for implementation and training of AI Contingency Plan.

11.WORST CASE SCENARIO (MEASURES TO AVOID WORST CASE SITUATIONS)

In the result of the evaluation of a given situation, there is a danger of a development from a single outbreak to a point where it becomes an epidemic and it seems not possible to eradicate the disease in a short time.
Symptoms for such a danger could be;

- Uncontrolled spread,
- Outbreaks in high densely populated areas
- New outbreaks in a short time or in not expected areas,
- Unsuccessful control measures,
- Last results of risk assessment shows the tendency to the fast wide spread of disease.

In such situations it is necessary to reinforce and extend all control measures like;

- To intensify the epidemiological investigations and repeated risk assessments,
- To extend or change the measures concerning zoning or stand stil measures respectively,
- To check up the transport conditions of dead or live animals, animal products, feed, persons, conditions of transport means, equipment, devices etc.
- To improve the hygiene measures especially cleansing and disinfection and supervision of the animal health situation in the farms.
- To extend or adapt respectively the diagnostic system at the changed situation in particularly consideration of the border areas of RZs and possible new infected areas or premises.
- To support the extended measures by additional qualified staff.
- To work out a specified plan for Emergency Vaccination.

11.1. The First Existence Of The Diseases (Index case): The first epidemic can be originating from an ordinary flock, willage type poultry breeding, flock, any other commercial flock or an integrated enterprice. A certain time period is needed for the diagnosis and confirmation of the disease case with regaeds to AI.

11.2. Secondary Epidemic Following the first existance of the diseases, the primary epidemic is most likely to occur during the period that the diagnosis of the disease has been made and yet the laboratorie procedures have been continuing. Following the secondary epidemic other epidemics will occour in various flocks and intensive epidemics will be observed in very short time

11.3. General Epidemic: In this phase when the epidemics are numerous a lot of new epidemics will be observed. The diagnosis of the epidemic has already been done and serotyping of the agent virus is being done.
İn this phase the DVO is dealing with the combat studies and the samples and materials will be sent to the VICs for the diagnosis and confirmation. However; the procedures regarding the combat with this disease will be completed without waiting the diagnosis and confirmation results in newly occuring disease case.

11.4. The Actions Regarding The General Epidemic Period: The following actions will be taken when numerous epidemics occurr:
-The animal movements in the region will be ceased immediately by the RLA of the region.
-All types of animal and animal products are prohibited for coming in to ang going from the region.
-All kind of registered information and documents related with the infected flocks are prepared carefully.
-The condemnation of the infected and dead animals in the infected flocks will be completed immadiately.
- If the technical staff for the above mentioned procedures is insufficient, the MLD will provide additional staff from non infected regions.
-If needed private veterinarians will be used for the abovementioned actions.
-The epidemiologic studies regarding the filation of the disease will be done at each phase and the results of the this studies will be interpretated.

11.5. Laboratory Diagnosis: At the begining of the epidemic, the disease will be diagnosed for the first time by the regional institute before the detection of the infection. However the serotyping studies regarding AI will be done by the NVRL.
During the general epidemic period, if the facilties and working capacities of the regional laboratory are insufficient coordination between the NVRL and other VIC laboratories will be done and if needed all institutes will take part in diagnostic and combat activities.
The specimens and materyals will be sent to the laboratoreies as soon as possible and if needed private courrier services will be used and sampling and the submission of the samples will be done carefully.

11.6. The Situation at the Infection Area

11.6.1. The procedures prior to the laboratory diagnosis of the ınfection.

The Veteniarian from the DVO arrives at the infection site and ceases all the animal movements from and into the enterprice.

5 specimens from all newly dead birds are taken into plastic bags for being sent to the lab. Additionally 20 blood samples from live animals are sent to the laboratory in cold chain with special wehicles. Moreover; samples from food and water of the flock are also sent to the lab. The documents belonging to the above mentioned specimens will be prepared and sent with the samples. The protective clothings, masks, bonets, boots and glowes are used during the sampling and specimen collection. The disinfection of the hands and feet is carefully implemented while entering the henhouses.

The state veterinarian determines the infection in the flock and seperates the diseased suspected, healthy and dead animals. The veterinarians records the situation. The dead animals are destroyed immediately.

The inner and outer parts of the henhouse are cleaned and disinfected thouroughly , the waste materials are destroyed.. The dead animals are destroyed by burying into deep dimples at suitable sizes. Big amounts of lime are spreaded on the dead animals before burying. These procedures are repeated under the supervision of the official authorities everyday until the results of the laboratory analyses arrives at the enterprice. Meanwhile, any animals or animal products (including food , equipments,drugs and etc.) are let out of the enterprice by all means and also no new animals are let come into the enterprice. The animal movements are completely ceased.

These measures are continued rigorously untill the results of the laboratory analyses are received. Additionally, the veterinarians and the technical staff of this enterprice are prohibited for 5 days to visit any other henhouse or enterprice.

The lorries carrying food and eggs are prohibited to enter inside or leave the enterprise.Only exception for this case is when there is a necessity for the food of the birds, the lorry bringing the food is let to unload the feed without entering into the enterprise. The distribution of the food is done by the internal vehicles. The lorries are cleaned and disinfected rigorously.

11.6.2. The procedures following the laboratory diagnosis

All the animals in the flock should be condemned following the disease is determined after the disease is determined and confirmed by the laboratory.

The condemnation procedure: All animals in the flock are condemned by being suffocated by gas and then burried into deep dimples. The remaining food, sawdust, straw, waste and other equipments should also be destroyed. Following the completion of the destruction of all animals in the henhouse all the parts of the henhouse are cleaned and washed. The waste waters are collected in septic tanks without letting them spread out..The agent virus is killed and destroyed by chemicals. No animals are put into the hens for at least 3 months following all these procedures. The enterprice is kept for rest. The enterprise is let to have new animals by the approval of the official authorities after the disease is eradicated.

In the infected region all the enterprices within 3 km diameter except the infected one is accepted as infected region zone and within 10 km diameter is accepeted as SZ. All the poultry in the enterprices in the infected region zone and SZ are taken into epidemiological investigation and the current status and screening of the infection is observed.

11.7. The scenario of the procedures ın an ınfection of AI

1-) Day 1: The first signs of the AI infection in a flock or death occurence in one or more flocks of the enterprice

2-) Day 1: Notification of the situation to the district veterinary officer by the officials of the enterprice or the flock owner

3-) Days 1-2: The offical veterinarians of the district arrives at the infection site and collects the necessary specimens and samples. They send the samples and specimens to the NVRL.
4-) Days 1-2: The specimens and samples arrive at the NVRL.
5-) Day 1-2: The first examination of the samples and specimens is performed
(necropsy,collecting the isolation material, and blood etc.)
6-) Day 3: Performing the necessary serological tests. If the serum is available HI test and/or ELISA and AGID tests are performed. If there is material for virus isolation inoculation into the embryonated eggs are performed for the virus isolation.
7-) Day 3: The results of the HI and ELISA tests are obtained. In this phase if the material is brought to the laboratory on the 1st. day the results of the HI and ELISA tests are obtained. An important finding is obtained with regards to the presence of the infection in case of seropositivity.
8-) Day 3: Inoculations to the embryonated eggs are performed for the virus isolation if there are embryonated eggs available.
9-) Days 6-7: The results of the AGID test is obtained. If there is infection, according to the seropositivity an important finding can be obtained with regards to the presence of the infection.
10-) Days 7-14: The results of the first isolation for the inoculation into embryonated eggs are obtained. The results are notified to the related provincial directorate and Ministry as soon as the diagnostic procedures are completed.
11-) Days 11-18: The first results of the isolation in embryonated eggs are obtained.
12-) Day 19: If the result of the first isolation is positive and the virus isolation can be done succesfully, it is sent to the OIE reference laboratory at Weybridge for confirmation.
13-) Day 21: The isolate is received by the reference laboratory.
14-) Days 30-42: the results of the confirmation are obtained and prepared ready for the international decleration.
15-) Days 42-45: The infection is declared internationally.
Note: While the activities of inspection and diagnosis are going on, the DVS and PS MLD will be notified at every step through the communications ways such as telephone and internet.
This flow scheme consists the activities for the primary epidemy. At the following epidemies the procedures will be stopped at the 10 th step which is the diagnosisi of the disease.

11.8. The analysis of the disease situation that changes quickly :

Due to the characteristics of the disease to be spreading quickly, the geographic location of the infected flocks and the situation of the surrounding enterprices must be monitored rigorously and they will be monitored continously. Moreover; the records of these enterprices will be monitored continously. If needed the movements of animals and animal products will be prohibited.

11.9. Condemnation of the ınfects flocks:

The infected flock will be determined for the condemnation of the infected flocks. For this purpose:
- The condenmation of the infected animals will be performed by the workers or enterprice staff experienced for this procedure. While performing this procedure protective clothing, bonnet, special masks (able to prevent the the staff from the aeresol contagion), gloves and boots are used to prevent the health of the workers.

The condemnation procedure can be done in 2 ways:
1- The animals that will be condemned are put into a container designed particualrly for this purpose. The container is closed and a speciific gas for killing is given in to the container.
2- Using large nylon bags: For this purpose, nylon bags in which 15-20 birds can be used. (this amount varies according to the size of the birds. For example 250-300 for day old chiks)
3- Using plastic bags is practical in order to prevent contamination of the surrounding. The bags are tightened The animals die in a very short time due to lack of air.

The killing type varies according to the breeding type of the birds.

- Incubation houses: As the day old chicks are small in size a big amount of birds can be placed in nylon bags.
- In broiler flocks: The birds are collected at certain parts collecetd with hands and placed in plastic bags.
- Layer flocks: As the birds are kept in cages, they are collected from the cages 1 by 1 from the cages.

11.9.1. The destruction of the infected carcasses:

Both the dead and killed birds are loaded into dumping trucks or excavators or otherwise damper containers. Following the loading they are carried into the burying dimples and burried immediately

11.9.2. The burying of the carcasses:

The dead or killed carcasses burried into suitable sized dimples either inside or outside the enterprise.

The burying dimples will be away from streams or the places near water deposits. The depth of the dimples should not be less than 2 meters. If there are alot of birds to be burried the depth should be at least 3-5 meters.

The base of the dimples is covered with lime 1-2 cm thick. The lime is also poured on the birds that are being burried. And the soil level of the burrying must be at least 1 meter.

11.9. 3. The disposal of the wastes:

After the removal of the of the killed and dead birds and the dead carcasses from the henhouses, the basement covering material and manure are wetted with specially prepared disinfectants inorder to prevent the environmental contamination. After the completion of this procedure the basement covering material is collected into a truck .

11.10. Establishment of a disease control center:

For this purpose the foolwing units are established within the body of the Ministry:

-NADECC: All the procedures are carried out in accordance with the AI contingency plan.

-LADECCs are established at the field.. The support of the private sector is provided when necessary.

All the records of the birds are kept rigorously in the infected flocks. These records will be used as a base in the compensation payment for the condemned animals.

11.11. The compensations for the condemned animals:

According to the current legislation there is provision for compensation regarding AI.

11.12. Emergency Vaccinations:

The use of the inactivated vaccines against AI outbreaks will be done under the direction of the DVS. The stock of the vaccine should be prepared and the continuity of the stoks will be provided.

11.13. Disinfectants:

The provincial directotes are supposed to prepare the stocks of the appropriate disinfectants in order to combat against AI outbreaks.

11.14 .Readiness of the abbatiores :

The abbatoires that will b e used an AI epidemy will be chosen primarily from the infected zones. The birds except from the ones brought from the noninfected regions that will be slaughtered compulsorily will be sent to the other abbatoires. The number of the shift-work of the abbatoires will be increased if needed.

11.15.Bringing new animals to the hen houses:

21 days after the eradication of the infection in the region (the henhouses will be cleaned and disinfected) new birds will be placed in the henhouses under the control of the DVS (ZSIQS)

11.16.The monitoring procedures following an aı epidemy:

After the AI infection is ended and eradicated , control activities with the aim of monitoring will be performed. These control activties should be planned covering all the poultry population..

Broiler hen houses: 20 serum samples will be collected for each hen house during the slaughter.

Commercial layer enterprices: 20 serum samples will be collected for each hen house once every 3 months.

Backyard flocks: If necessary, 20 serum samples for each village will be collected from the risky regions.

12- removal of anımals and anımal products

12.1 Confirmation of AI

In case of confirmation of a primary outbreak of AI, all contingency procedures for the containment and eradication of AI are implemented.

The DVO must:

- activate the mobile disinfection unit that must be positioned at the only point of entrance/exit to the infected premises,
- reduce the number of vehicles and staff to the minimum necessary to extinguish the outbreak. Any staff that has access to the infected premises may only leave the farm after a complete change in clothing and possibly a shower. Staff involved in the depopulation of the farm must not have any contacts with susceptible species, for at least three days after the last contact with the infected premises,
- contact the depopulation crews, escavator operators (if the birds can be buried) , vehicles for transportation of dead animals (if the birds can be rendered), disinfection crews.

12.2. Depopulation and disposal of dead birds

12.2.1. General Concepts

The depopulation and disposal of infected birds must be done in the quickest time span to prevent spread of infection, in compliance with current legislation. Furthermore, they need to be performed with the doors of the shed/house closed to prevent access of wild birds and other animals to infected organic material. Generally speaking, where possible burial on site of infected birds is recommended, rather than burning or rendering. However this needs to be evaluated on the basis of where the outbreak has occurred.

12.2.2. Staff And Equipment Necessary For Depopulation And Disposal

- Wooden poles and plastic red-and white-tape to identify the infected premises and the entrance/exit to the farm
- Mobile disinfection units
- Night-time illumination devices
- Sufficient staff (depopulation crews and other staff) to avoid overworking
- Calculation of the number of vehicles necessary for carrying the carcasses out of the farm
- Identification of the route the vehicles carrying the dead birds
- Policemen or other social security service to escort the trucks to the disposal area
- Gase, drugs or devices to contain, sedate, stun and depopulate flocks (in case of ostriches captive bullet revolvers may be used)
- Appropriate containers for disposing of infected material

12.3. Depopulation

In compliance OIE Guidelines depopulation of infected flocks may take place using the following methods/drugs/systems:

- Electronarcosis by water dipping
- Decapitation and dislocation of the neck

- Gassing with carbon dioxide
- Vacuum tank
- Mechanical devices embryonated eggs and chicks

Other methods for depopulation of birds are listed below:

- Flocks consisting of a limited number of birds, intrapulmonary inoculation of drugs used for the euthanasia of pets may be used,
- Flocks consisting of considerable numbers of birds may be depopulated by gassing them inside sealed containers. The number of birds per m^3 of gas should not exceed 150 (mean weight 1.8 kg).
- Asphyxiation by carbon dioxide is not as effective in ducks and geese as in chickens. Physical methods such as cervical dislocation using cattle castration forceps are preferable for the humane destruction of waterfowl.

Gases used for depopulation of birds are listed below:

- Carbon dioxide (CO_2) 17.5 kg/1000 m^3: saturates the environment in 30 minutes, and death takes place in 15 minutes,
- Carbon monoxide (CO) 8 kg/1000 m^3: saturates the environment in 30 minutes, and death takes place in 15 minutes,
- Hydrogen cyanide (HCN) 3 kg/1000 m^3: saturates the environment in 30 minutes, and death takes place in 4 minutes extremely toxic, to be handled by trained staff only.

Drugs that may be used for depopulation of large flocks of birds:

- Alfa chloralose, mixed to feed in concentration of 2 %-6 %, causes loss of consciousness, and death can be obtained by suffocating birds in plastic bags. Can be used only if the birds are clinically ill and do not exhibit any loss of appetite,
- Sodium fenobarbital, dissolved in drinking water (80 mg in 55 mls), causes loss of consciousness in 4 hours, same recommendations as above.

12.4. Disposal of Birds

One of the major objectives of the eradication program is prompt and effective disposal of contaminated material that cannot be effectively treated (eg. dead birds, eggs, litter, manure, fresh and frozen carcases, plant and equipment and building materials). Available methods include burial, incineration, burning, rendering and composting. It is important to avoid contamination of water supplies, for reasons of animal, public and environmental health.

The removal of very large numbers of birds in a short time presents environmental and logistic problems. A shed full of meat chickens close to market weight represents about 40 tonnes of organic material of which 75% is water. The disposal of litter can also pose special problems as infective virus may be spread with the dust. It will be necessary to moisten the surface of the litter with a disinfectant and possibly heap it in mounds under plastic before removal

If infected material must be transported for disposal, particular attention should be paid to preventing the spread of the virus. For example, truck body trays must be waterproof and all loads carefully covered with tarpaulins to ensure that material cannot be blown out.

12.4.1.Burial

In areas which allow burial as a means of disposal, a pit must be prepared as soon as the diagnosis is confirmed. The size of the pit must be at least two meters wide by two meters deep, and this enables disposal of 300 birds (medium weight 1.8 kg) per 1.3 meters of surface. The number of birds can be doubled, each metre deeper the pit is made (3-6 meters). All non disinfectable, biodegradable material (wood, cardboard) must be buried with the

animals. The carcases must be covered with a layer of calcium hydroxide, and then with a layer of earth (at least 40 cm).

12.4.2.Rendering

Carcases to be rendered must be loaded onto suitable vehicles which must be completely leak-proof. Rendering must take place in establisments authorised for dealing with infectious material.

12.4.3.Incineration/ Burning

Incineration is a good method to safely dispose of infected material. However, incinerators are generally too small to be of any use and may not be near animal facilities. Burning has been used where no burial sites are available. Burning is an expensive method of disposal because of the high water content of carcases; it may also be environmentally unacceptable.

Incase of an outbreak of AI, the disposal method is choosen by the LADECC according to the situation of the site of the outbreak.

12.5. Disposal / Destruction Of Infected Materials

Waste, organic and all other non disinfectable material present on the farm must be destroyed, in particular the destruction of litter, eggs, egg products, hay, animal feedstuffs, feathers and egg-trays must be planned for.

Litter

Depending on the amounts present and on the characteristic of the farm, litter can be either buried in the pit with the animal carcases or piled in heaps to ensure maturation. The heap must be covered with a resistant sheet of plastic. In all cases, infected litter should not be moved from the infected farm prior to maturation.

Eggs and egg products

May be buried in the pit with the animal carcases or rendered.

Straw

Straw may be decontaminated by spraying its surface with an active disinfectant and covering up the stacks with a resistant sheet of plastic. Covered stacks must be left to decontaminate for at least 42 days. For time reasons, it could be more convenient to incinerate it.

Animal feed

Animal feed present on site must be decontaminated by fumigation and subsequently incinerated.

12.6. Disinfection of Infected Premises

A check-list and indications on the means of disinfecting infected premises is reported below:

- All units which are physically or functionally connected to the establishment (i.e. hatchery, egg storage rooms, packaging rooms, egg trolleys, egg product plants) must be properly disinfected. Vehicles, used for transporting live animals, eggs and animal feed must also be disinfected,
- Washing and disinfection of walls, floors and ceilings of the infected establishments must be performed aiming at the removal of all organic material, metal structures such as cages may be decontaminated by heat treatment,
- All equipment inside the house such as drinkers and food hoppers must be washed and treated with a disinfectant for at least 48 hours,
- Water reservoirs must also be emptied, washed and disinfected,
- Feed tanks (silos) need to be emptied, washed with a hot water-pressure pump and subsequently fumigated,
- After washing and disinfecting, all units must be fumigated twice with at least two weeks between fumigations.

12.6.1. A list of Disinfectants which are Active Against AI Virus, Their Concentration and Recommended Use is Reported Below:

1. Sodium hypochlorite: 2% active chlorine solution (disinfection of equipment)
2. Quaternary ammonium salts: 4% solution (treatment of walls, floors, ceilings and equipment)
3. Potassium peroxomonosulphate + sulphamic acid + sodium alkyl benzene sulphonate [as a ready-to use product] (treatment of floors, walls, ceilings and equipment)
4. Calcium Hydroxide: 3% solution (treatment of walls and floors)
5. Cresolic acid 2.2% solution: (treatment of floors)
6. Synthetic phenols 2% solution: (treatment of floors)
7. Formalin and permanganate: fumigation

For emergency cases enough stock and space must be available for disinfectans.

13. DISEASE AWERENESS AND PUBLIC RELATION

13.1. Introduction

All diseases related confidential information should only be directed to selected responsible adresses. All other information about diseases, economical and political importance relevant to human health can be very carefully prepared and published by the press department of the NADECC or LADECC respectively.

13.2. Reporting Requirement

The Animal Disease Control Act No 17 2003 states that if an animal shows symptoms of a contagious animal disease, this must be reported to the authorities by the livestock holder and veterinarian. To eliminate confusion, a national 24-hour telephone line has been opened.

13.3. Publicity

Through publications in the journal of MLD, articles in the farming press and local radio, Cafe visits and TV channels. Disease awareness campaigns targeted at farmers and professional personnel who regularly visit flocks are held when needed. Besides these publications, various Internet sites http://oie.int , http://www.who.int;http://www.mifugo.go.tz , and regular training programmes for farmers are used to maintain disease awareness.

13.4. Veterinary Education

During veterinary studies, clinical signs and epidemiology of AI are thoroughly studied. The control measures and notification procedures are discussed more generally for highly infectious animal diseases. In post-graduate veterinary medicine education, great attention is paid to the veterinarian's responsibility for the general good.

14.0 Budget Estimates

This Contigency Plan has a budget of Tsh----------equivalent to US$----------of which the government has committed Tsh----------(US$----------) and development parteners have been requested to bridge the gap.

A detailed cost break down of the planned activities is given in Annex 16.

ANNEX 1 (a) Members and contact details of NADECC

S/N	Substantive Post	Incumbent	Tel	Fax	Email
1	Permanent Secretary Ministry of Livestock Development	Dr. C.W. Nyamrunda			
2	Director of Veterinary Services	Dr.J.O.Mollei			
3	Director of Policy and Planning	Mrs.J.Catherine			
3	Director of Animal Production	Mrs.Njombe			
4	Director of Livestock Research	Dr.S.B.Meena	2863361		meenasbn@yahoo.co.uk
5	Director of Animal Disease Research Institute	Dr.Daas			
6	National Veterinary Epidemiologist	Dr.P.F.Mujuni			
7	National Veterinary Virologist	Dr.Yongolo			
8	National Veterinary Pathologist	Dr.Kapaga			
9	Assistant Director Animal Health Servces	Dr.P.Z.Njau			
10	Zoosanitary Inspection	Dr.G.Mwakasungula			
11	Chief Medical Officer	Dr.Upunda			
12	Director of Wildlife				
13	Representative of RALG				
14	Representative of Police				
15	Representative of Poultry Farmers				
16	Chief Accountant MLD				
17	Disaster Management	Dr.M.M.Bahari			

Annex 1 (b) National Avian Influenza Experts Team (Veterinary)

1	National Veterinary Epidemiologist	Dr .P.F.Mujuni			
2	National Veterinary Virologist	Dr.Yongolo			
3	National Veterinary Pathologist	Dr. Kapaga			
4	Disaster Management Focus	Dr.M.M.Bahari			

Annex 1 (c) International Contact Points

1	OIE-Director General	Dr B.Vallat			
2	FAO AGHAH	Dr.Lubrorth			
3	World Bank Africa Liv.Expert	Dr.F.Le Gall			
4	AU-IBAR	Dr.Modibo-keita			
5	SADC-OIE Desk	Dr.B.J.Mtei			
6	AVIS	Prof.Mark Rweyumamu			

ANNEX 1(d) List of VICs Officer In-charges and their contacts

S/N	VIC & National Veterinary Lab	Name	Tel	Fax	Email
1	Arusha				
2	Iringa				
3	Mpwapwa				
4	Mtwara				
5	Mwanza				
6	Tabora				
7	National Vet Lab(ADRI)Temeke				

ANNEX 2. Regional Livestock Advisors Names and contact particulars

S/N	**Region**	**Name**	Telefon No	Fax No	e-mail
1	Arusha				
2	Dar-es-Salaam				
3	Coast				
4	Dodoma				
5	Iringa				
6	Kagera				
7	Kigoma				
8	Kilimanjaro				
9	Lindi				
10	Manyara				
11	Mara				
12	Mbeya				
13	Morogoro				
14	Mtwara				
15	Mwanza				
16	Rukwa				
17	Ruvuma				
18	Singida				
19	Shinyanga				
20	Tabora				
21	Tanga				

Annex 3.Sampling and treatment of samples

Samples: In cases of suspected AI the following pathological samples must be collected and sent to the laboratory:

- At least 5 moribound birds (for post mortem examination),
- Pooled tracheal and lung samples from at least 5 moribound birds,
- Pooled intestine samples from at least 5 moribound birds,
- Cloacal and tracheal swabs from healthy birds (shouldn't be less than 30 birds) (also from waterfowl and ratites),
- Blood samples, 1 % of the flock (shouldn't be less than 20).

Samples from different apparatuses must not be pooled. They must be packaged appropriatly (in leak prof containers, wrapped in at least two plastic bags), to avoid dissemination of the infectious agent, and transported refrigarated to the laboratory. Sacrificed animals may be transported in a sealed autoclavable plastic bag, inserted inside a similar, sealed bag. All samples must be carried to the laboratory inside a polystyrene box containing icepacks. The polystyrene box must be appropriately disinfected before leaving the premises.
The driver in charge of delivering the saples, must drive directly to the laboratory without any intermediate stops.

Treatment of samples: The organs and tissues may be pooled, but separate treatment of faecal material is essential. Swabs should be placed in sufficient antibiotic medium to ensure full immersion. Faeces samples and organs should be homogenized (in an enclosed blender or using a pestle and mortar and steril sand) in antibiotic medium and a made to 10-20 % w/v suspensions in the medium. The suspensions should be left for about two hours at ambient temperature (or longer periods at 4^0C) and then clarified by centrifugation (e.g. 800 to 1000 x *g* for 10 minutes).

Antibiotic medium: High concentrations of antibiotics are required for faeces samples and a typical mixture is: 10 000 units/ml penicillin, 10 mg/ml streptomycin, 0,25 mg/ml gentamycin and 5 000 units/ml mycostatin in phosphate buffered saline (PBS). These levels can be reduced up to five-fold for tissues and tracheal swabs. For control of Chlamydia organisms 50 mg/ml oxytetracycline may be added. It is imperative when making the medium that the pH is checked after the addition of the antibiotics and readjusted to pH 7.0 – 7.4.

Annex 4.Isolation and identification of the agent

Virus Isolation in embryonated fowls' eggs: The clarified supernatant fluid should be inoculated in 0.1 – 0.2 ml amounts into the allantoic cavity of each of a minimum of five embryonated fowls' eggs which have been incubated for 9 to 11 days. Ideally, these eggs should be obtained from a SPF flock, or at least specific antibody negative flocks. The inoculated eggs are held at 37 ^{0}C and candled daily. Eggs dead or dying embryos as they arise, and all remaining eggs 7 days after inoculation should be chilled to 4 ^{0}C and the allantoic-amniotic fluids tested for HA activity. When HA is detected the presence of bacteria should be excluded by culture. If bacteria are present the fluids may be passed through a 450 nm membrane filter, further antibiotics added and inoculated into embryoneted eggs as above. Detection of HA activity indicates a high probability of the presence of an influenza A virus or of an avian paramyxovirus. Fluids that give a negative reaction should be passaged into at least one further batch of eggs.

The presence of influenza A virus can be confirmed in agar gel immunodiffusion (AGID) tests by demonstrating the presence of the nucleocapsid or matrix antigens both of which are common to all influenza A

viruses. The antigens may be prepared by concentrating the virus from infective allantoic fluid or extracting the infected chorioallantoic membranes; these are tested against known positive antisera. Virus may be concentrated from infective allantoic fluid by ultracentrifugation, or by precipitation under acid condition. The latter method consists of the addition of 1.0 M HCl to infective allantoic fluid until it is approximately pH 4.0. The mixture is placed in an ice bath for 1 hour and then clarified by centrifugation at 1000 *g* at 4 ^{0}C. The supernatant fluid is discarded. The virus concentrates are resuspended in glycine- sarcosyl buffer: this consist of 1 % (w/v) sodium lauroyl sarcosinate buffered to pH 9.0 with 0.5 M glycine. These concentrates contain both nucleocapsid and matrix polypeptides.

Preparations of nucleocapsid-rich antigen can also be obtained from chorioallantoic membranesfor use in the AGID test.

Preparation of nucleocapsid-rich antigen from chorioallantoic membranes removed from infected eggs;
This method involves: removal of the chorioallantoic membranes from infected eggs that have allantoic fluids with HA activity. The membranes are then homogenised or ground to a paste.
This is subjected to three freeze and thaw cycles, followed by centrifugation at 1 000 *g* for 10 minutes. The pellet is discarded and the supernatant is used as antigen following treatment with 0.1 % formalin.
Either of these two antigens may be used in immunodoublediffusion tests using 1% agarose, or agar gels containing 8.0 % (w/v) sodium chloride made up to 0.1 M phosphate buffer pH 7.2, poured to a tickness of 2-3 mm in Petri dishes or on microskope slides. Using a template and cutter, wells of approximetly 5 mm in diameter, and 2-5 mm apart, are cut in the agar. A pattern of wells must place each suspect serum adjacent to a known positive serum and antigen. This will make a continuous line of identity between the known positive, the suspect serum and the nucleocapsid antigen. Approximately 50 µl of each reagent should be added to each well.

Precipitin lines can be detected after approximately 24-48 hours, but this may be dependent on the concentrations of the antibody and the antigen. These lines are best observed against a dark background that is illuminated from behind. A specific, positive result is recorded when the precipitin line between the known positive control wells is continuous with the line between the antigen and the test well. Crossed lines are interpreted to be due to the test serum lacking identity with the antibodies in the positive control well.

Annex 5.Serological tests for AI virus antibodies*:*

During eradication programmes where the H subtype of the virus responsible is already known, or by using the homologous virus as antigen, serological monitoring for evidence of infection may be done using HI tests.
If the H subtype is not known, evidence for infection with influenza A viruses may be obtained by detecting antibodies directed to the group specific antigens.
For this purpose either an immunodoublediffusion test or an ELISA test may be used (a problem with ELISA is the host specificity of the test since it is dependent on the detection of host immunoglobulins). There is a sensitive and specific ELISA that demonstrates nucleoprotein of Type A influenza virus using a monoclonal antibody against Type A influenza nucleoprotein.

Waterfowl rarely give positive results in immunodoublediffusion tests and, unless the subtype is known, it is probably only practicably to examine such birds for the presence of antibodies to H5 and H7 subtypes.

Samples :

Blood samples should be taken from all birds if the flock size is less than 20 and from 20 birds from larger flocks (this will give a 99 % probability of detecting at least one positive serum if 25 % or more of the flock is positive, regardless of flock size). The blood should be allowed to clot and serum removed for testing.

Examination for antibodies :

Individual serum samples should be tested for their ability to inhibit influenza virus HI tests.

Variations of the procedures for HA and HI test are practised in different laboratories. The following recommended examples apply in the use of V-bottomed microwell plastic plates in which the final volume for both types of test is 0.075 ml.

Heamagglutination (HA) test Reagents

1- Isotonic saline buffered with phosphate (PBS) (0.05M) to pH 7.0-7.4.
2- Red blood cells (RBC) taken and pooled from a minimum of three specific pathogen free chickens (if not available blood may be taken from birds regularly monitored and shown to be free from antibodies to AI) into an equal volume of Alsever solution. Cells should be washed three times in PBS before use use as a 1 % (packed cell v/v) suspension.
3- H5 and H7 viruses of low virulence for use as standard antigens.
4- Positive and negative control antisera

Procedure ;

1- Dispense 0.025 ml PBS into each well of a plastic microtitre plate (V- bottemed wells should be used).
2- Place 0.025 ml of virus suspension (i.e infective allantoic fluid) in the first well.
3- Use a microtitration diluter to make two-fold dilutions (1:2 to 1:4.96) of virus across the plate.
4- Dispense a further 0.025 ml of PBS to each well.
5- Dispense 0.025 ml of 1 % red blood cells to each well.
6- Mix by tapping gently and then allow the Red Blood Cells to settle for about 40 minutes at room temperature, i.e. about 20 ^{0}C, or for 60 minutes at 4 ^{0}C if ambient temperatures are high, by which time control Red Blood Cells should be settled to a distinct button.
7- HA is determined by tilting the plate and observing the presence or absence of tear-shaped streaming of the Red Blood Cells. Wells with no HA should flow at the same rate as the control cells with no virus.
8- The HA titer is the highest dilution that causes agglutination of the Red Blood Cells. That dilution may be regarded as containing 1 HA unit (HAU). A more accurate method for determining the HA titer is to do HA tests on virus from a close range of initial dilutions i.e. 1:3, 1:4, 1:5, 1:6, etc. This is recommended for the accurate preparation of antigen for HI tests.

Heamagglutination Inhibition (HI) test Reagents

1- PBS
2- Virus containing allantoic fluid diluted with PBS to contain 4 or 8 HAU per 0.025 ml.
3- 1% chicken Red Blood Cells.
4- Negative and Positive control chicken serum.

Procedure:

1- Dispense 0.025 ml PBS into each well of a plastic V- bottemed microtitre plate.
2- Place 0.025 ml of serum into first well of plate.
3- Make two-fold dilutions of 0.025 ml volumes of the serum across the plate.
4- Add 4 HAU of virus / antigen in 0.025 ml to each well and leave for a minimum of 30 minutes at room temperature (i.e. about 20 ^{0}C) or 60 minutes at 4 ^{0}C.
5- Add 0.025 ml of 1% (v/v) chicken Red Blood Cells to each well and after gentle mixing, allow the Red Blood Cells to settle for about 40 minutes at room temperature, i.e. about 20 ^{0}C or for 60 minutes at 4 ^{0}C if ambient temperatures are high, by which time control Red Blood Cells should be settled to a distinct button.
6- The HI titer is the highest dilution of serum causing complete inhibition of 4HAU of antigen. The agglutination is assessed by tilting the plates. Only those wells in which the Red Blood Cells stream at the same rate as the

control wells (containing 0.025 ml Red Blood Cells and 0.05 ml PBS only) should be considered to show inhibition.

7- The validity of the results should be assessed against a negative control serum, which should not give a titre >1/4 (>2^2 or >$\log_2$ when expressed as the reciprocal), and positive control serum for which the titre should be within one dilution of the known titre.

HI titres may be regarded as being positive if there is inhibition at a serum dilution of 1/16 (2^4 or $\log_2 4$ when expressed as the reciprocal) or more against 4 HAU of antigen. Some laboratories prefer to use 8 HAU in HI tests. While this is permissible, it affects the interpretation of results to that a positive titre is 1/8 (2^3 or $\log_2 3$) or more.

Chicken sera rarely give nonspecific positive reactions in this test and any pretreatment of the sera is unnecessary. Sera from species other than chickens may sometimes cause agglutination of chicken Red Blood Cells, so this property should first be determined and then removed by adsorption of the serum with chicken Red Blood Cells. This is done by adding 0.025 ml of packed chicken Red Blood Cells to each of 0.5 ml of antisera, shaking gently and leaving for at least 30 minutes; the Red Blood Cells are then pelleted by centrifugation at 800 *g* for 2-5 minutes and the adsorbed sera are decanted. Alternatively, Red Blood Cells of the avian species under investigation could be used.

Agar Gel Immunodiffusion (AGID) Test

The preferred method to show the presence of influenza A virus is to demonstrate the possession of the nucleocapsid or matrix antigens which are shared by all influenza A virus. This fact enables the presence or absence of antibodies to any influenza A virus to be detected by AGID tests. This is generally done in AGID tests involving either concentrated virus preparations or extracts from infected chorioallantoic membranes.

Either of these two antigens may be used in immunodoublediffusion tests using 1% agarose, or agar gels containing 8.0 % (w/v) sodium chloride made up to 0.1 M phosphate buffer pH 7.2, poured to a tickness of 2-3 mm in Petri dishes or on microskope slides. Using a template and cutter, wells of approximetly 5 mm in diameter, and 2-5 mm apart, are cut in the agar. A pattern of wells must place each suspect serum adjacent to a known positive serum and antigen. This will make a continuous line of identity between the known positive, the suspect serum and the nucleocapsid antigen. Approximately 50 µl of each reagent should be added to each well.

Precipitin lines can be detected after approximately 24-48 hours, but this may be dependent on the concentrations of the antibody and the antigen. These lines are best observed against a dark background that is illuminated from behind. A specific, positive result is recorded when the precipitin line between the known positive control wells is continuous with the line between the antigen and the test well. Crossed lines are interpreted to be due to the test serum lacking identity with the antibodies in the positive control well.

Annex 6 Assessment of pathogenicity

The term HPAI implies the involvement of virulent strains of virus. It is used to describe a disease of chickens with clinical signs such as excessive lacrimation, respiratory distress, sinusitis, oedema of the head and face, cyanosis of the unfeathered skin, and diarrhoea. Sudden death may be the only sign. These signs may vary enormously depending on the host, age of the bird, presence of other organisms and environmental conditions. In addition, viruses that normally cause only a mild or no clinical disease may mimic highly pathogenic AI if exacerbating conditions exist.

Criteria for classifying an AI virus as highly pathogenic:

The intravenous pathogenicity index (IVPI) test was used as a method of assessing virulence. For the purposes of confirming disease and implementing the control measures, the following definition applies:
"an infection of poultry caused by an influenza A virus that has an IVPI in 6-week-old chickens > 1.2 or any infection with influenza A viruses of H5 or H7 subtype for which nucleotid sequencing has demonstrated the presence of multiple basic amino acids at the cleavage site of the haemagglutinin".

Intravenous Pathogenicity Index (IVPI) Test

1. Infective allantoic fluid from the lowest passage level available, preferably from the initial isolation without any selection, is diluted 1/10 in steril isotonic saline.
2. 0.1 ml diluted virus is injected intravenously into each of 10 six-week-old chickens (SPF birds should be used).
3. Birds are examined at 24 hour intervals for 10 days.
4. At each observation each bird is recorded normal (0), sick (1), severely sick (2) or dead (3). (The judgement of sick and severely sick birds is a subjective clinical assessment. Normally "sick" birds would show one of the following signs and "severly sick" more than one of the following signs: respiratory involvment, depression, diarrhoea, cyanosis of the exposed skin or wattles, oedema of the face and/or head, nervous signs. Dead individuals must be scored as 3 at each of the remaining daily observations after death).
5. The IVPI is the mean score Per bird Per observation over the 10-day periyod. An index of 3.00 means that all birds died within 24 hours, an index of 0.00 means that no bird showed any clinical sign during the 10-day observation periyod.

Assessment of plaque-forming ability:

1- It is usually best to use a dilution range of virus to ensure that an optimum number of plaques are present on the plate. Ten-fold dilutions up to 10^{-6} ; in PBS should be sufficient.
2- Confluent monolayers of chick embryo cells or a suitable cell line (Madin-Darby bovine kidney for example) are prepared in 5 cm diameter Petri Dishes.
3- 0.2 ml of each virus dilution is added to each two petri dishes and the virus allowed to absorb for 30 minutes.
4- After washing three times with PBS the infected cells are overlaid with the relevant medium containing 1% w/v agar and either 0.01 mg/ml trypsin or no trypsin. It is important that no serum is added to the overlay medium.
5- After 72 hours incubation at 37 ^{0}C the plaques should be of sufficient size. They are best seen by removing the agar overlay and staining the cell monolayer with crystal violet (0.5 % v/v) in 25 v/v ethanol.
6- All viruses should give clear plaques when incubated in the presence of typsin in the overlay. When trypsin is absent from the overlay only viruses virulent for chickens will produce plaques.

Annex 7 Preliminary differentiation

Because it is important that control measures aimed at limiting the spread of virus should be implemented as soon as possible, each regional laboratory should be in a position to identify any isolated haemagglutinating virus as influenza viruses of H5 or H7 subtype in addition to Newcastle disease virus. The haemagglutinating fluids should be used in a HI test as described.

Positive inhibition i.e. 2^4 (1:16) , or more, with polyclonal antisera specific for H5 or H7 subtypes of influenza A and of a titer of at least 2^9 would serve as preliminary identification enabling the imposition of interim control measures.

2- Confirmatory identification:

Since there are 15 haemagglutinin subtypes and 9 neuraminidase subtypes of influenza viruses and variations occur within each of these it is not practicable nor cost effective for each regional laboratory to hold antisera which will allow full antigenic characterization of influenza isolates. However, each regional laboratory should:

1. confirm that the isolate is an influenza A virus using an immunodoublediffusion test to detect group antigens as described (immunofluorescence or ELISA techniques to detect group antigens may be used if preferred by the regional laboratory);
2. determine wheter or not the isolate is of H5 or H7 subtype;
3. carry out an intravenous pathogenicity index test in six-week-old chickens as described. Intravenous pathogenicity indexes of grater than 1.2 indicate the presence of virus requiring a full implementation of control measures (it would be a useful exercise if national laboratories also carried out tests to determine the capacity of an isolate to produce plaques in cell cultures as specified)

Regional Laboratories should immediatly submit all AI and all H5 and H7 isolates to the NVRL for full characterization.

3- Further typing and characterization of isolates

The NVRL should receive all haemagglutinating viruses from the regional laboratories for further antigenic and genetic studies to enable a greater understanding of the epizootiology of the disease(s) and send them to the International Reference Laboratory within the European Community in keeping with the functions and duties of the reference laboratory.

In addition to these duties the NVRL shall carry out full antigenic typing for all influenza viruses received. For H5 and H7 viruses which do not have intravenous pathogenicity indices greater than 1.2, nucleotid sequencing of the haemmagglutinin gene to determine wheter or not there are multiple basic amino acids at the cleavage site of the haemagglutinin protein should also be carried out.

Infection with nonpathogenic viruses

- No clinical signs in infected birds however birds can seroconvert. Some of these viruses have the potential to become virulent by mutation of the genome.

Infection with lowly or mildly pathogenic viruses

- Clinical signs in chickens and Tanzanias 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 layers to 15 % in meat chickens.

- Egg production in layers can drop by up to 45 % with recovery to normal in 2 - 4 weeks.

- Mutation to virulence has been demonstrated in outbreaks.

Infection with highly pathogenic viruses

- Clinical signs in chickens and Tanzanias include severe respiratory signs with excessively watery eyes and sinusitis, cyanosis of the combs, wattle and shanks, oedema of the head, ruffled feathers, diarrhoea and nervous signs.
- The last eggs laid after the onset of ilness frequently have no shells.
- In acute cases involving sudden death (peracute), clinical signs may not be seen and 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. Overall mortality rates for peracute/acute cases of nearing 100 % have been reported.
- Some severely affected hens may recover, but rarely come back into lay.

The disease in Tanzanias is similar to that seen in chickens, but is often complicated by secondary infections such as fowl cholera, Tanzania coryza and colibacillosis.

Annex 8 Kit No 1- Equipment List for the DVO

1) Paper and pens
2) Epidemiological inquiry form
3) Equipment necessary for the clinical visit and sampling procedures:
a. 2 disposable suits
b. 5 pairs of disposable shoe-covers
c. 2 pairs of rubber gloves and 5 pairs of latex gloves
d. disposable caps and face masks
e. disposable cover- all
f. protective glasses (2 sides of the glasses should be closed)
g. medical waste bags which are resistant and impermeable
h. paper tissues
ı. 5 leak proof containers
j. 5 leak proof and water resistant plastic bags
k. electric torch
l. active disinfectant solution
m. 2 pens and a notepad
n. 100 syringes 2,5 mls with needle
o. blood tubes & tube holder
p. nebuliser
r. 100 thin, small plastic bags
s. 2 pairs of surgical scissors
t. 2 pairs of forceps
u. tape
v. 2 felt tip pens
y. 1 thermic container
z. 5 frozen icepacks
At least two of these kits should be prepared and available at the OV headquarters at all times.

Annex 9 Kit No 2 Equipment List for the VIC

a. 1 thermic container
b. 4 pairs of forceps

c. 2 pairs of surgical scissors
d. 1 knife
e. tape
f. labels and pens
g. 100 2.5 mls syringes with needle
h. sterile swabs
i. 50 test tubes containing virus transport media
j. 10 leak proof containers
k. 2 disposable suits
l. 5 pairs of disposable shoe-covers
m. 5 pairs of latex gloves
n. disposable caps and face masks
o. 10 black waste-bags
p. 50 rubber bands
q. disinfectant solution in a nebuliser
r. cardboard container

n. disinfectant, alcohol, soap, tissue
o. %10 neutral formalin in glass jars, 500ml, (for pathological inspections)
p. %50 serum physiological with glycerine in glass jars, 250ml, (for bacteriological inspections)
q. label
r. lam – lamel boxes
s. glass pencil
t. 10 bags to put the sample from the animal and from the feed

Annex 10 .AVIAN INFLUENZA EPIDEMIOLOGICAL INQUIRY FORM

Date ...../...../.....
Dr. ..... Phone number .....
Suspicion N. .....
Confirmation N. .....
Name of establishment .....
Address.....
Municipality ..... Province..... Phone .....
District .....
Farm code or identification number ☐☐☐ ☐☐
Owner
.....
Company .....
Address of the owner ..... Phone .....
Information provided by .....
.....
Farm Veterinarian Dr. ..... Present NO YES ☐ ☐

INFORMATION CONCERNING THE FARM
TYPE OF ESTABLISHMENT:
☐ Industrial ☐ Rural ☐ Dealer Retailer ☐
CATEGORY/PRODUCTION LINE:
Table-egg layers Meat birds

Type :Grandparents ☐
Parents
Pullets ☐
Meat-type (*broiler*) ☐
Layers ☐

NUMBER OF BIRDS AND SPECIES PRESENT

Chickens	☐	Meat	N° ..... Breeders	☐	N° ..... Layers	☐	N° .....
Turkeys	☐	Meat	N° ..... Breeders	☐	N°		
Guinea-fowl	☐	Meat	N° ..... Breeders	☐	N°		
Ducks	☐	Meat	N° ..... Breeders	☐	N°		
Pigeons	☐	Meat	N° ..... Breeders	☐	N°		
Pheasants	☐	Release	N° ..... Breeders	☐	N°		
Geese	☐	Meat	N° ..... Breeders	☐	N°		
Ducks	☐	Meat	N° ..... Breeders	☐	N°		
Quail	☐	Meat	N° ..... Breeders	☐	N°		
Partridges	☐	Release	N° ..... Breeders	☐	N°		

Other ....
....
Date of placing ...../...../..... Sex: ..... Age.....
...../...../..... Sex: ..... Age.....

HATCHERY OF ORIGIN

Company Hatchery NO ☐ YES ☐
Company ..... Address ..... N. .....
Municipality ..... Region..... Code ☐☐☐ ☐☐
Phone ..... Fax .....

Debeaking operations: Date ...../...../.....
Performed by: ☐Family members ☐mployed staff External staff☐ Other ☐.....
Remarks.....
.....

HOUSING SYSTEM

Sheds NO
YES N° .....

Tunnels NO
YES N° .....

Type of ventilation system:
Natural .....
Natural with fans .....
Artificial.....

Free-ranging system NO
YES m^2 .....
Bird proof nets NO YES
Possibility of contact with wild birds:
NO YES Species .....
.....
.....
Other birds present on site (captive or free)
NO YES Species .....
.....
.....
Presence of ponds or lakes :
NO YES .....
Other water reservoirs NO YES (specify) .....
Presence of pigs NO YES N° .....
Other animals NO YES (specify) .....
Remarks
.....

OTHER INFORMATION REQUIRED:

1. Topography of establishment

A map of the infected premises must be drawn, clearly indicating the productive units, the animals housed inside them, indicating the main routes of access to the premises.

Data on introduction/spread of infection: information necessary for the points a), b), c) etc., must be collected for all movements of animals/people and therefore may have to be repeated if necessary.

2. Movements of birds: information required

a) Introduction of birds from other establishments/hatcheries/farms NO YES
(Twenty days before the onset of the first clinical signs)

Date ...../...../..... N° ..... Species ..... Farm Hatchery
Name of Farm..... Code
Address..... N. .....
Municipality ..... Region .....
District.....

---

b) Introduction of birds from exhibitions/markets/fairs NO YES
(Twenty days before the onset of the first clinical signs)
Date ...../...../..... N. ..... Species .....
Origin: Fair Market Exhibition
Municipality..... Province .....
District .....

---

c) Exit of birds/eggs to other farms/establishments/hatcheries/abattoirs NO YES
(In the time span between 20 days before the onset of the first clinical signs and the date the farm was put under restriction)
Date ...../...../..... N. ..... Species .....
Destination: Other farm Hatchery Abattoir Other .....
Name of establishment..... Code
Address..... N. .....
Municipality ..... Region..... ..... District.....

d) Exit of birds/eggs to other fairs/markets/exhibitions NO YES
(In the time span between 20 days before the onset of the first clinical signs and the date the farm was put under restriction)
Date ...../...../..... N. ..... Species .....
Destination: Fair Market Exhibition Other .....
Address.....N. .....
Municipality ..... Region .... District.....

3.MOVEMENT OF PEOPLE: POSSIBLE MEANS OF INTRODUCTION OR OF SPREAD OF INFECTION (In the time span between 20 days before the onset of the first clinical signs and the date the farm was put under restriction)

NO YES

Date ...../...../..... Surname and first name .....
Veterinarian Technician Vaccinating crew Debeaker Other farmer Dealer Other (specify) .....
Address ..... N. .....
Municipality ..... Region .... District.....
Phone number .....
Previously visited farm: Name .....
Municipality ..... Region .... District.....

MOVEMENT OF VEHICLES:

(A) Transport of animals, (B) Transport of feed, (C) Transport of eggs, (D) Collection of dead animals, (E) Fuel/Gas, (Other) Specify (In the time span between 20 days before the onset of the first clinical signs and the date the farm was put under restriction)

Date of entry	Vehicle A/B/C/D /E/other	Name of company	Fax/Phone number	Vehicle plate tractor	Vehicle plate Trailer	Transporter (Company)	Driver	Phone number

a) INDIRECT CONTACTS WITH OTHER POULTRY ESTABLISHMENTS
NO YES
(Sharing of equipment, vehicles, feed, staff, etc. in the time span between 20 days before the onset of the first clinical signs and the date the farm was put under restriction)
Date of contact …../…../.....
Name of farm or establishment……..
Code ☐☐☐☐☐
Address ……..
Municipality ……..... Province ……..
District…….....
Species farmed …….. number …….
shared vehicle shared feed shared equipment shared staff collection/recycle of litter
other (specify) ……..
b) OTHER FARMS OWNED BY THE SAME OWNER NO YES
Name of farm or establishment……..
Code ☐☐☐☐☐
Address ……..
Municipality ……..... Province ……..
District…….....
Species farmed …….. number …….
Empty Full
---
c) POULTRY FARMS LOCATED NEAR THE OUTBREAK
NO YES Empty Full
Name of farm or establishment……
Code ..☐.☐.☐….☐.☐….
Address ……..
Municipality ……..... Province ……..
District……..... Distance in metres……..
Species farmed …….. number …….

ANAMNESTIC DATA

WEEKLY MORTALITY

NB: data concerning mortality rates recorded in the 6 weeks prior to the onset of clinical signs

WEEK FROM	TO	NUMBER ANIMALS DEAD

Remarks:

.....

.....

.....

.....

Date of onset of AI clinical signs ...../...../.....

Clinical signs observed by the farmer:

.....

.....

TOTAL NUMBER OF BIRDS Farm put under restriction (dead or alive)	Number of ill birds (Farm put under restriction)	Number of dead birds (Farm put under restriction)	Number of birds depopulated

NB: this information must refer to the data collected when the farm has been put under restriction with mortality and morbility referring to the suspicion of AI

VACCINATION of birds

Vaccination of birds is practised: NO YES

Date of vaccination	Type of vaccine (1)	Commercial name	Administration route
...../...../.....	.....	.....	
...../...../.....	.....	.....	
...../...../.....	.....		

(1) Live or inactivated

VACCINATING STAFF:

Family Employees External staff Other .....

Remarks.....

ADMINISTRATION OF DRUGS/MEDICAMENTS

In the last 15 days: NO YES (specify):

.....

STAFF WHO ADMINISTERED THE MEDICAMENT:

Family Employees External staff Other .....

Remarks.....

CLINICAL INVESTIGATION PER SPECIES

Species .....

Depression

Respiratory signs: mild
Severe
Drop or cessation of egg laying
Oedema, cyanosis or cutaneous haemorrhages
Diarrhoea
Nervous signs
Other.....

GROSS FINDINGS
Rhinitis and sinusitis
Tracheitis *catarrha*
haemorrhagic
Aersacculitis
Haemorrhages *epicardum*
Endocardium
proventriculus
ovarian follicles
Enteritis *catarrhal*
haemorrhagic
Pancreatitis
Other:
..... Remarks
……
……
……

Signature
……
……

Annex 11. AVIAN INFLUENZA SAMPLE SUBMISSION FORM

Region…… District ……Village----------
Veterinarian……
Phone……Fax……
……
Date …./…./….

Farm: Municipality ……Region……

Owner ……
Complete address ……

SPECIES AND CATEGORY
Turkey breeders (*) N …… Meat Turkeys (*) N ……
Broiler breeders N …… Layer breeders N ……
Layers N …… Broilers N ……
Other species (specify) …… N……
(*) Vaccinated against AI NO
YES ⇒ Number of vaccinations……

COLLECTION OF SAMPLE FROM / FOR
SUSPECT OUTBREAK → date of notification ……
Confirmed outbreak
Farm epidemiologically connected with outbreak → Name and farm code of outbreak ……
……
Farm located in protection zone → Name and farm code of outbreak ……
……
Farm located in surveillance zone → Name and farm code of outbreak
……
……
Testing for the movement of animals
Monitoring programme
Other ……

ANAMNESTIC DATA

Species and Category	Onset of clinical signs	Symptoms	% mortality	From/to

Species	Samples collected	N° samples	for detection of	
			Antibodies	Virus

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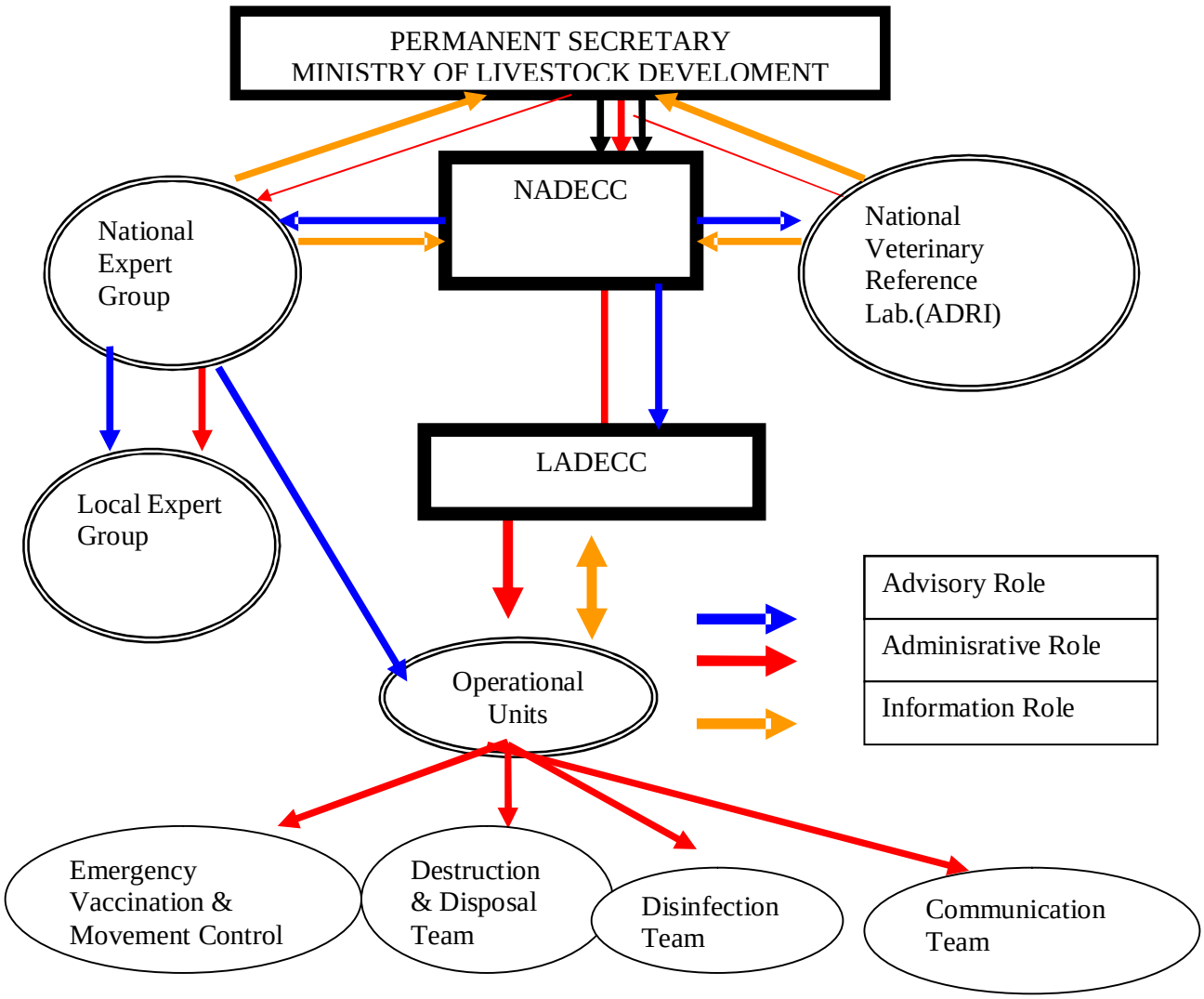
----------	----------	----------	
SAMPLE IDENTIFICATION			
N° house or shed	Species	Samples collected	

Signature

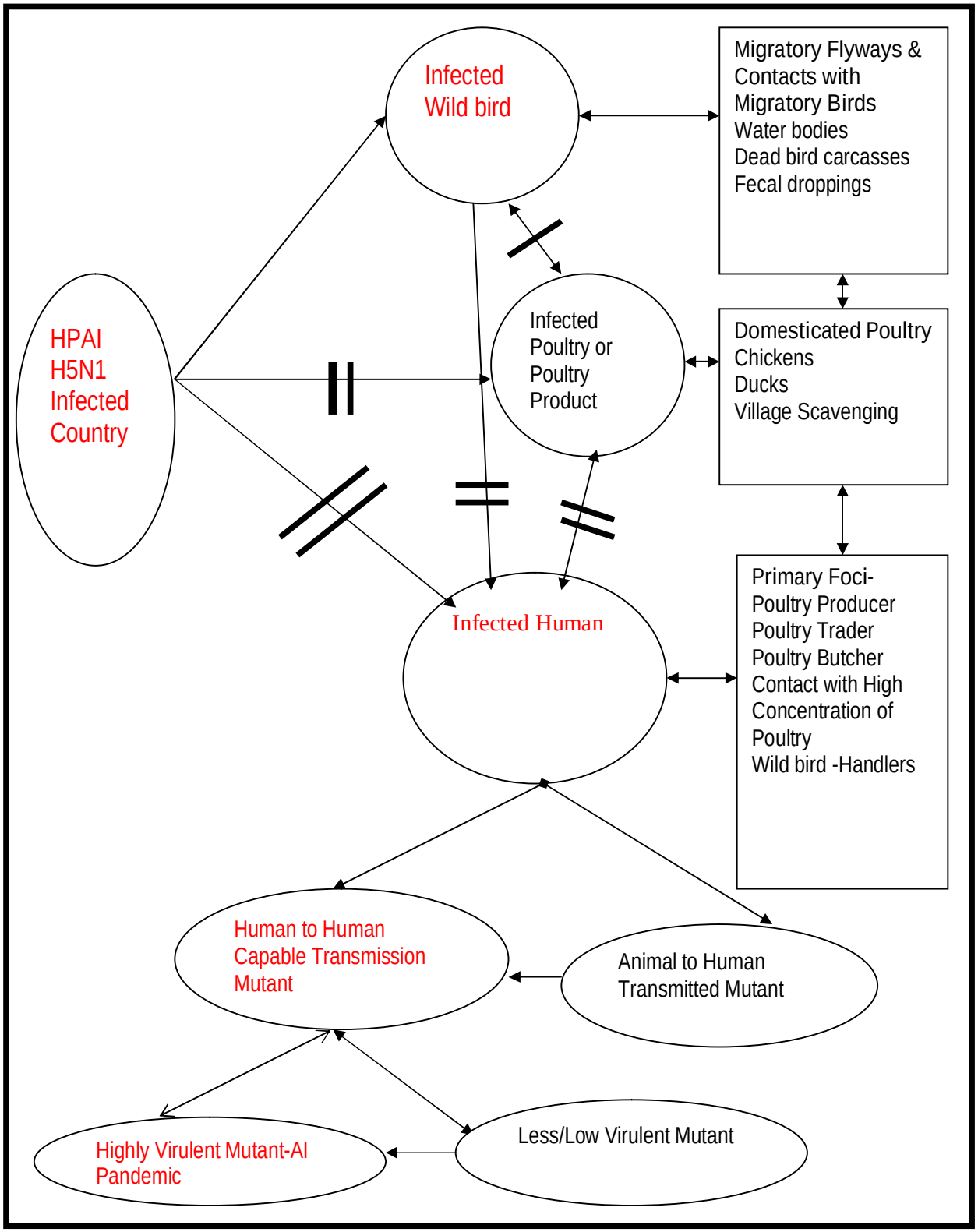
……

In case of given more addidional information, please attach an extra sheet.

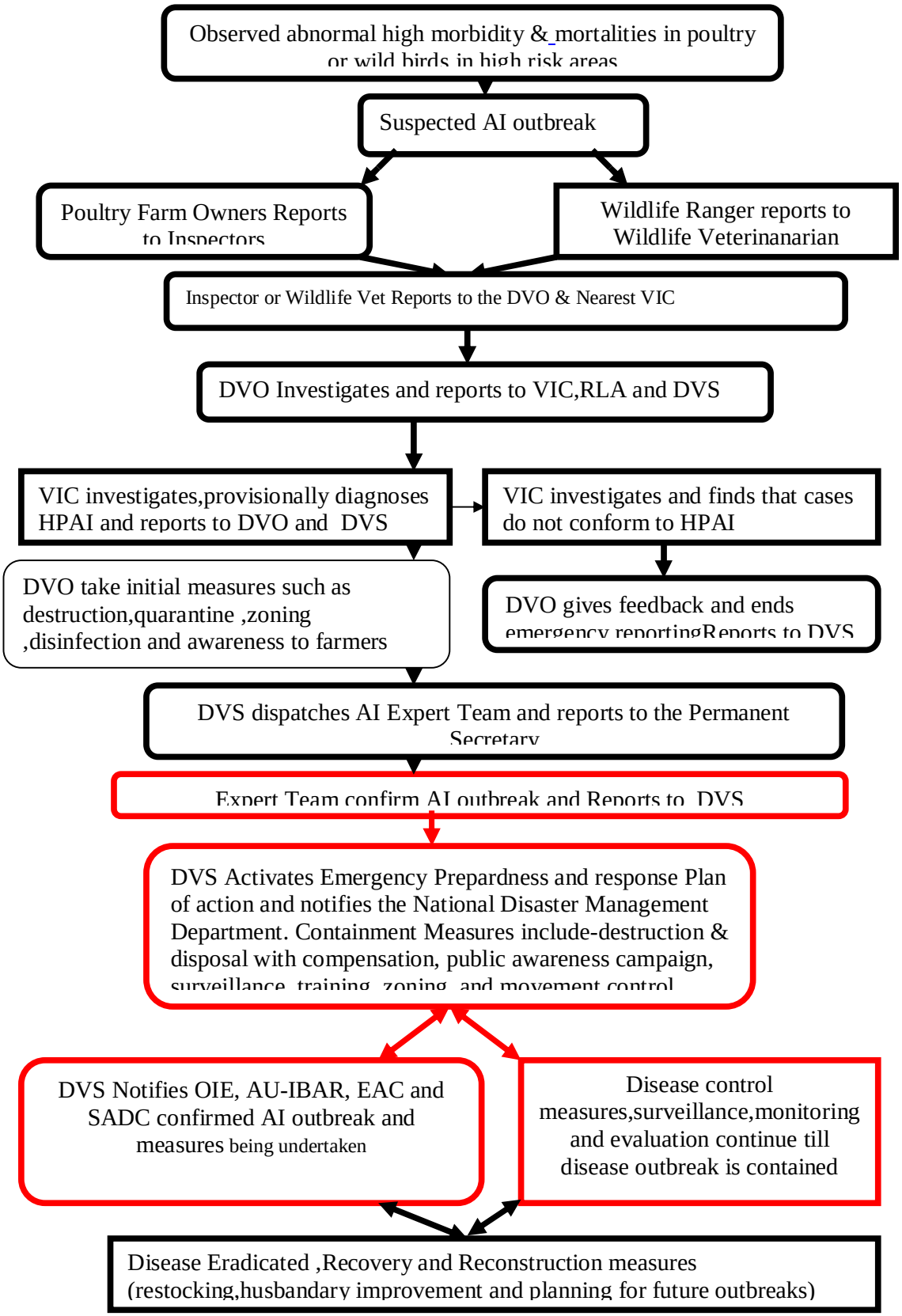
Annex 12 Chain of Command
Structure



Annex 13-Avian Influenza introduction scenario



Annex 14-Avian Influenza outbreak containment events.



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Annex 15-Avian Influenza Surveillance Strategy

Annex 16-Avian Influenza Contigency Plan (Veterinary) Detailed Budget